

Original Article

Evaluation of the marginal integrity of various pit and fissure sealantsBilal Yaşa¹⁾, Özlem Erçin¹⁾, and Hüseyin Hatırlı²⁾¹⁾Department of Restorative Dentistry, Faculty of Dentistry, Istanbul Kent University, Istanbul, Turkey²⁾Department of Restorative Dentistry, Faculty of Dentistry, Gaziosmanpaşa University, Tokat, Turkey

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Abstract**Purpose:** To evaluate the marginal integrity of various pit and fissure sealants subjected to different application methods.**Methods:** A total of 253 non-carious human third molars extracted and randomly divided into two groups according to the preparation method employed: invasive or non-invasive. Eight fissure sealant materials were tested: nano-filled flowable composite (Filtek Ultimate Flow), nanohybrid flowable composite (GrandioSo Flow), micro-hybrid flowable composite (Majesty Flow), resin-based unfilled fissure sealant (ClinPro Sealant), resin-based filled fissure sealant (Fissurit FX), resin-based highly filled fissure sealant (GrandioSeal), giomer-based fissure sealant (BeautiSealant), and glass ionomer-based fissure sealant (Fuji Triage). Samples were subjected to two-year cyclic thermo-mechanical and brushing simulations. Two observers quantitatively evaluated the restoration margins and classified them as either “permanent restoration edge”, or if a gap larger than 250 µm was evident, “gapping at the restoration edge”. The extent of the gap was recorded as a percentage relative to the total length of the restoration edge.**Results:** The baseline marginal adaptation had no significant effect on the marginal adaptation ($P > 0.05$). However, the preparation method and type of fissure sealant material had a significant impact on the marginal adaptation ($P < 0.05$).**Conclusion:** On the basis of quantitative analysis, the highest marginal integrity was observed for flowable composites, whereas the lowest was observed for glass ionomer-based fissure sealant.

Keywords: marginal integrity, pit and fissure sealants, thermal aging

Introduction

Dental caries is a multifactorial disease caused by a change in the composition of the bacterial biofilm, leading to an imbalance between the remineralization and demineralization processes, and manifested by the formation of carious lesions in primary and permanent teeth [1]. Dental caries usually occurs on the occlusal surfaces of posterior teeth because pits and fissures are suitable for plaque retention and are difficult to clean [2]. In recent years, dentistry has focused on non-invasive conservative techniques for reducing the risk of caries, taking preventive measures, and protecting dental structures [3]. Pit and fissure sealants are one of several treatment choices for prevention of plaque accumulation and formation of occlusal caries [4].

Several techniques for tooth preparation before fissure sealant application have been reported. Invasive or non-invasive techniques are available for placement of pit and fissure sealants. Invasive fissure sealant application is for teeth with deep and narrow occlusal pits and fissures, which are abraded and enlarged with a tapered fissure diamond bur 0.5-0.6 mm in diameter to remove organic material, plaque, and the thin superficial prismless enamel layer. With this technique, acid and sealants can

penetrate deeper into the fissures and increase the enamel surface area. Enameloplasty (or fissurotomy) and air abrasion are some of the invasive procedures employed [5]. In the non-invasive technique, the fissures are mechanically cleaned using polishing brush. Deciding whether an invasive or a non-invasive technique should be used before fissure sealant application is a controversial issue [6].

The use of flowable resin composites as pit and fissure sealants has many advantages because of their favorable properties such as a low modulus of elasticity, low viscosity, and ease of handling. In addition, flowable resin composites have more filler particles than resin-based fissure sealants, resulting in less polymerization shrinkage and better wear resistance [7-9].

Marginal integrity between a tooth and restorative materials is an important factor for the longevity of restorations [10]. The aim of the present study was to evaluate the marginal integrity of different pit and fissure sealants employing two different application methods. The null hypothesis was that there would be no difference in marginal integrity between the fissure sealant materials and preparation methods employed.

Materials and Methods

This study was approved by the Izmir Katip Celebi University Ethics Committee (Protocol number 171). The study materials were chosen from among 253 extracted non-carious human third molars, which were examined with a stereomicroscope at $\times 10$ magnification (Stemi 2000 C, Zeiss, Oberkochen, Germany), and 160 teeth with intermediate deep fissures but without defects, hypoplasia, or cracks were included. The teeth were cleaned with a scaler and slurry pumice, placed in 0.5% chloramine T solution for 48 h, and then kept in distilled water until used.

Sample preparation

The teeth were embedded in acrylic resin up to 1 mm below the cemento-enamel junction using a PVC mold (3 cm diameter and 1.5 cm height). Fissures were cleaned using an air abrasion machine (Gnatus Prophyl Jet, Sao Paulo, Brazil) with sodium bicarbonate powder (Clinpro Prophyl Powder, 3M ESPE, St. Paul, MN, USA) and then examined for the presence of any debris under a stereomicroscope at $\times 10$ magnification.

Study design

The teeth were randomly divided into two preparation groups (invasive treatment and non-invasive treatment) and eight fissure sealant material subgroups ($n = 10$). The eight fissure sealant materials tested were: a nano-filled flowable composite (Filtek Ultimate Flow, 3M ESPE) a nanohybrid flowable composite (GrandioSo Flow, Voco GmbH, Cuxhaven, Germany), a microhybrid flowable composite (Majesty Flow, Kuraray Noritake Dental, Tokyo, Japan), a resin-based unfilled fissure sealant (ClinPro Sealant, 3M ESPE), a resin-based filled fissure sealant (Fissurit FX, Voco GmbH), a resin-based highly filled fissure sealant (GrandioSeal, Voco GmbH), a giomer-based fissure sealant (BeautiSealant, Shofu Inc., Kyoto, Japan) and a glass ionomer-based fissure sealant (Fuji Triage, GC Corp., Tokyo, Japan) (Table 1).

In the invasive treatment group, enameloplasty was applied to the teeth with a micro narrow taper fissure carbide bur (Fissurotomy Micro NTF, SS White, Lakewood, NJ, USA). The preparation was standardized by extending the fissure entrance to the diameter and half-length of the fissure carbide bur. No bur preparation was performed in the non-invasive group.

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Table 1 Fissure sealant materials used in this study

Material	Composition		Manufacturer
Filtek Ultimate Flow	nanofilled flowable composite	Bis-GMA, UDMA, TEGDMA, prokriat resin, ytterbium trifluoride, silica nano-fillers, zirconia nano-fillers	3M ESPE, St Paul, MN, USA
GrandioSo Flow	nanohybrid flowable composite	HEDMA, Bis-GMA, TEGDMA, glass ceramics, silicon dioxide	Voco GmbH, Cuxhaven, Germany
Majesty Flow	microhybrid flowable composite	TEGDMA, hydrophobic aromatic dimethacrylate, silanized colloidal silica and barium glass	Kuraray Noritake Dental, Tokyo, Japan
ClinProSealant	resin-based unfilled fissure sealant	Bis-GMA, EDMAB, TEGDMA, TBATFB, BHT, diphenylodoniohexafluorophosphate	3M ESPE
Fissurit FX	resin-based filled fissure sealant	Bis-GMA, UDMA, TEGDMA, BHT, benzotriazole derivatives, inorganic glass ionomer filler, NaF	Voco GmbH
GrandioSeal	resin-based highly filled fissure sealant	Bis-GMA, TEGDMA, nano fillers	Voco GmbH
BeautiSealant	giomer-based fissure sealant	Bis-GMA, UDMA, S-PRG fillers	Shofu Inc., Kyoto, Japan
Fuji Triage	glass ionomer-based fissure sealant	powder: alumina-fluoro-silicate glass (amorphous) liquid: polyacrylic acid, specific component	GC Corp., Tokyo, Japan

Bis-GMA, bisphenol-A-diglycidyl methacrylate; UDMA, urethane dimethacrylate; TEGDMA, triethyleneglycol dimethacrylate; HEMA, 2-hydroxyethyl methacrylate; EDMAB, ethyl P-dimethylamino benzoate; BHT, butylhydroxytoluene; TBATFB, tetrabutyl dimethacrylate; S-PRG, surface-pre reacted glass

Fissure Sealant Application in the Flowable Composite Groups

An etchant containing 35% phosphoric acid was applied to the fissures for 30 s with ultrasonic vibration from the buccal surfaces of the teeth to increase the penetration, followed by rinsing with water and drying with an air syringe. For optimal drying, a solution containing 60% ethanol was applied to the surfaces before air drying. An adhesive (Scotchbond Universal, 3M ESPE) was applied to the fissures for 20 s using a microbrush. Excess solvent was removed by thorough air drying with an air syringe and then polymerization was performed with a LED light (Valo Cordless, Ultradent, South Jordan, UT, USA) for 10 s. Flowable composites were applied to the fissures with ultrasonic vibration for better penetration and polymerized for 20 s. Fissure sealant surfaces were coated with glycerin and polymerized again for 60 s.

Fissure Sealant Application in the Resin-Based Fissure Sealant Groups

A solution containing phosphoric acid and ethanol was applied to the fissures as described above. Resin-based fissure sealants were applied to the fissures with ultrasonic vibration, polymerized for 20 s then coated with glycerin and polymerized again for 60 s.

Fissure Sealant Application in the Giomer-Based Group

A self-etch Beauti-Primer was applied to the fissures for 5 s with an applicator in accordance with the manufacturer's instructions, and then dried thoroughly. Beauti-Sealant was then applied with ultrasonic vibration, polymerized for 20 s, then coated with glycerin and polymerized again for 60 s.

Fissure Sealant Application in the Glass Ionomer-Based Group

In accordance with the manufacturer's instructions, GC Cavity Conditioner was applied to the fissures for 10 s then dried thoroughly. Pre-capsulated Fuji Triage was triturated for 10 s in a high-speed amalgamator, then applied to the fissures. The Fuji Triage was then coated with Fuji Coat LC and polymerized for 10 s in accordance with the manufacturer's instructions.

The use of dehydration agents is recommended for removal of moisture from fissures that have been etched-washed and air-dried, to increase the bonding of hydrophobic fissure sealants [11]. Therefore, in this study, ethanol solution (60%) was applied for optimum drying after the etchant had been washed away, and also after air drying.

All samples except for the glass ionomer groups were kept in distilled water for 24 h in a darkroom at 37°C for completion of the polymerization. Samples treated with glass ionomer fissure sealant were kept in saline solution for 7 days. After that, all the occlusal surfaces of the samples were duplicated using polyether impression material (Impregum Penta H Duo-soft and Garant L Duo-soft, 3M ESPE) and epoxy resin.

Artificial aging

Samples were subjected to two-year cyclic thermo-mechanical and brushing simulations. The samples were aged by a chewing machine under a force of 49 N and a frequency of 1.7 Hz (3 mm vertical, 1.5 mm lateral

movement) with 240,000 cycles to simulate one year [12]. Thermal cycling was applied for 1,000 cycles with a dwell time of 30 s between 5-55°C. For the brushing procedure, deionized water and a daily used dentifrice was prepared at a ratio of 2:1, and the slurry was changed every 1,000 cycles. Medium-hard toothbrushes were used under a load of 200 g, a frequency of 1 Hz, and 10,000 strokes to simulate one year [13,14]. The toothbrushes were changed after every 10,000 strokes. This cycle of brushing and chewing simulation was repeated twice to simulate two years of aging.

Scanning electron microscopy (SEM) evaluation

After the artificial aging, the occlusal surfaces of the samples were duplicated as described above. Images of the duplicated samples were taken with a scanning electron microscope (SEM LS-10, Zeiss) at $\times 100$ magnification and, if necessary, at $\times 200$ and $\times 400$. Two observers quantitatively evaluated the restoration margins on images using ImageJ software (National Institutes of Health, Bethesda, MD, USA) and classified them as either a "permanent restoration edge" or, if a gap larger than 250 μm was evident, as "gapping at the restoration edge". The extent of the gap was then expressed as a percentage relative to the total length of the restoration edge.

Statistical analyses were performed using SPSS 20.0 for Windows (IBM, Armonk, NY, USA). The Kolmogorov-Smirnov and Levene tests were used to test the normality and homogeneity of variance distribution. Normally distributed and homogeneous data were subjected to analysis of covariance (ANCOVA) and *post-hoc* tests, whereas non-normally distributed and non-homogeneous data were analyzed using the Kruskal Wallis and Mann-Whitney *U* tests at a significance level of $P < 0.05$.

Results

The marginal adaptation tests revealed that artificial aging led to decreases in the marginal adaptation of the test materials, the percentages differing according to the preparation methods and materials employed (Table 2). ANCOVA showed that whereas the baseline marginal adaptation had no significant effect on the marginal adaptation ($P > 0.05$), the preparation method and type of fissure sealant material significantly affected the marginal adaptation ($P < 0.05$). A significant effect of the preparation method was observed only in the glass ionomer fissure sealant group ($P < 0.05$).

Non-invasive groups

After the artificial aging, Filtek Ultimate Flow (FUF) showed the highest marginal adaptation (91.41 ± 1.46) whereas Fuji Triage (FJT) showed the lowest (65.59 ± 4.50) (Table 3). All of the flowable composites showed higher marginal adaptation and the Glass Ionomer fissure sealant showed lower marginal adaptation than the other tested materials ($P < 0.05$). Among the resin-based fissure sealants, the highest marginal adaptation was observed for Clinpro sealant (CPS), and the lowest was observed for Grandio Seal (GDS) (Fig. 1).

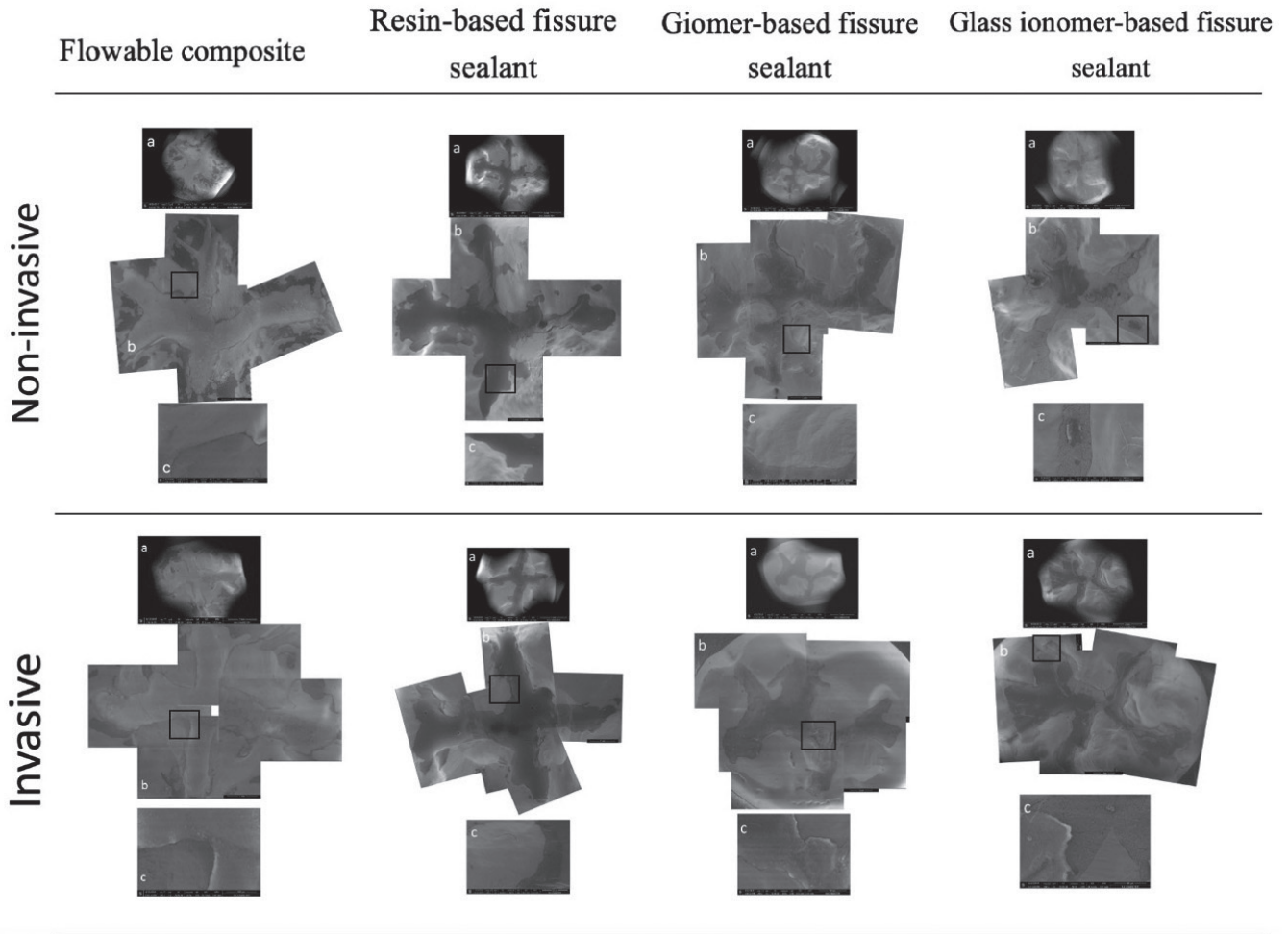


Fig. 1 Representative FE-SEM images of the tested materials. a: whole occlusal surfaces of the teeth, b: merged FE-SEM images taken at $\times 100$ magnification, c: FE-SEM images taken at $\times 200$ magnification

Invasive groups

After artificial aging, FUF showed the highest marginal adaptation (95.06 ± 2.40) and FJT showed the lowest (74.27 ± 2.76) (Table 3). Among the Flowable composite resins, FUF showed significantly higher adaptation than the nanofilled flowable composite, GSF ($P < 0.05$). All of the flowable composites showed higher marginal adaptation than the other tested fissure sealant materials ($P < 0.05$) (Fig. 1). Among the resin-based fissure sealants, Fissurit FX showed the highest marginal adaptation (83.47 ± 4.28) and CPS showed the lowest (80.71 ± 9.50), but the difference was not significant ($P > 0.05$).

Discussion

As a result of modern preventive dentistry practices, both the incidence and progression rate of dental caries have decreased [15]. Epidemiological studies have shown that the incidence of flat surface caries has decreased, although the incidence of occlusal surface caries is still high [16]. Although occlusal surfaces constitute only 12.5% of total tooth surfaces, more than 2/3 of dental caries occur on occlusal surfaces [17]. Pit and fissure sealant application is a scientifically proven and accepted preventive method for protection of pits and fissures that are prone to caries formation.

Various *in vitro* aging methods can be applied to predict the problems that restorations face over time in intraoral conditions. Fissure sealants applied to occlusal surfaces are exposed to many different types of stress in the mouth, including mechanical, thermal, and chemical. As a result of such stresses, marginal integrity problems, wear, loss of retention and partial or total loss of restoration may be seen in fissure sealants over time. Most previous studies evaluating fissure sealants have applied only thermal cycling for aging [18-20]. In only two studies, chewing simulation with thermal cycling was used as the aging method, and microleakage and mar-

Table 2 Mean and standard deviations according to application method employed

	Application method	n	Mean % (\pm SD)
Before aging	non-invasive	80	93.55 (± 4.04)
	invasive	80	93.27 (± 4.33)
After aging	non-invasive	80	81.09 (± 8.93)
	invasive	80	84.34 (± 7.69)

Table 3 Mean and standard deviations between materials

Application method	Material	n	Before aging mean % (\pm SD)	After aging mean % (\pm SD)	P (<0.005)	
Non-invasive	FUF	10	96.25 (± 1.26)	91.41 (± 1.46)	a, b	
	GSF	10	95.73 (± 2.07)	88.04 (± 3.22)	b, c, d	
	MJF	10	98.10 (± 0.73)	91.13 (± 1.69)	a, b	
	CPS	10	94.42 (± 2.35)	81.89 (± 3.02)	e, f	
	FFX	10	94.03 (± 2.27)	79.60 (± 3.14)	e, f, g, h	
	GDS	10	91.96 (± 2.55)	76.30 (± 3.48)	f, g, h	
	BTS	10	91.69 (± 2.67)	74.78 (± 2.97)	g, h	
	FJT	10	86.20 (± 2.84)	65.59 (± 4.50)	i	
	Invasive	FUF	10	97.39 (± 1.87)	95.06 (± 2.40)	a
		GSF	10	94.36 (± 1.87)	88.32 (± 2.99)	b, c
MJF		10	95.86 (± 1.38)	91.24 (± 2.46)	a, b	
CPS		10	94.87 (± 2.22)	80.71 (± 9.50)	e, f, g	
FFX		10	94.94 (± 2.42)	83.47 (± 4.28)	c, d, e	
GDS		10	90.91 (± 3.44)	82.25 (± 3.22)	d, e, f	
BTS		10	90.90 (± 4.21)	79.38 (± 3.58)	e, f, g, h	
FJT		10	86.95 (± 5.02)	74.27 (± 2.76)	h	

FUF, Filtek Ultimate Flow; GSF, GrandioSo Flow; MJF, Majesty Flow; CPS, Clinpro Sealant; FFX, Fissurit FX; GDS, Grandio Seal; BTS, Beuti Sealant; FJT, Fuji Triage. Different letters indicate statistically significant differences ($P < 0.05$).

ginal integrity of resin-containing fissure sealants were evaluated [21,22]. A two-year cyclic thermo-mechanical chewing and brushing simulation was performed in these studies to evaluate the effect of intraoral conditions on the fissure sealants. This type of approach can predict the possible clinical success of sealants.

Studies of fissure sealants have shown that no material can completely seal a fissure or guarantee maintenance of marginal integrity, and that the main reason for deterioration of marginal integrity is differences in the expansion coefficient with temperature [23]. However, marginal integrity has also been reported to be an important factor when evaluating the clinical success of fissure sealants [24]. As secondary caries may occur due to loss of retention of fissure sealants due to deterioration of marginal integrity, strong restoration-enamel adhesion is recommended [25].

Use of SEM images for evaluating the marginal integrity of restorations is known to be reliable and accurate [26]. This allows quantitative data to be obtained by evaluating all restoration edges without specimen deterioration [27]. In addition, the initial and final data can be compared using epoxy replicas of samples. Since the samples are not damaged when replicas are used, the imaging process can be repeated if necessary [28]. In this study, the marginal integrity of fissure sealants was investigated by obtaining SEM images of epoxy replicas and evaluating them using software.

It has been reported that stresses caused by polymerization shrinkage and the modulus of elasticity are the main factors affecting the marginal integrity of resin-containing materials [26]. It has been reported that enlargement of fissures using invasive techniques necessitates a larger volume of material for sealing, and thus a higher rate of polymerization shrinkage will occur [29], and this in turn will lead to deterioration of marginal integrity [18]. In this study, however, no significant difference of marginal integrity was observed between invasive and non-invasive application methods before artificial aging.

A reduction of continuous marginal integrity after aging with thermo-mechanical brushing and chewing simulation was observed for all the fissure sealants used in this study. After aging, the non-invasive group had a higher degree of gapping at the restoration margin than the invasive group. Similarly, it has been reported that organic residues in fissures as well as the prismless enamel layer are removed when enlargements are made with a bur, thus creating a restoration-tooth interface that is more resistant to mechanical and thermal stresses [30,31]. Xalabarde et al. [32] evaluated the application of fissure sealants with or without bur preparation and found that after thermal aging, enameloplasty with a bur increased the degree of marginal integrity.

The present analysis of SEM images showed that flowable composites had the highest marginal integrity after aging (Fig. 1). For both application methods, the marginal integrity of flowable composites was significantly higher than for resin, giomer and glass ionomer-containing fissure sealant materials. Therefore, the null hypothesis was partially rejected. It has been reported previously that flowable composites have a low modulus of elasticity and a thermal expansion coefficient similar to that of hard dental tissues [33]. Flowable composites can show elasticity against occlusal stress, respond to temperature changes by expanding and contracting at rates similar to tooth tissues, and thus can retain continuous marginal integrity. In addition, as glass ionomer and resin-containing fissure sealants provide longer wear than flowable composites, sub-margination occurring at the restoration margins could be considered as a deterioration of marginal integrity.

It has also been reported that polymerization shrinkage is higher for resin-containing materials with a low filler content [34] and that marginal integrity may be affected by this high shrinkage stress [35]. However, initial and post-aging marginal integrity analysis demonstrated that Clinpro Sealant had values similar to those of the other resin-containing fissure sealants used. On the other hand, it has also been reported that the modulus of elasticity, and ultimately polymerization shrinkage stress, increase with higher filler ratios in resin-containing materials [35,36]. It is thought that the low modulus of elasticity of Clinpro Sealant could be effective for monitoring the marginal integrity of resin-based fissure sealants containing other fillers.

Filtek Ultimate Flow showed the highest marginal integrity in both the invasive and non-invasive groups. SEM analysis demonstrated the lowest percentage of marginal integrity in the non-invasive glass ionomer (FJT)

group before and after aging. Vineet et al. [37] evaluated the marginal integrity of resin and glass ionomer-containing fissure sealants applied with the use of invasive or non-invasive techniques, and observed a higher rate of marginal integrity for resin-containing fissure sealants than was the case for the FJT group, similarly to the results obtained in the present study. Furthermore, Gunjal et al. [38] reported a significantly higher marginal integrity for Clinpro Sealant than for Fuji Triage. After aging, significantly higher marginal integrity was observed in the invasive FJT group than in the non-invasive FJT group, although no differences were evident between the groups for other materials, irrespective of the application technique.

In summary, it was demonstrated that the use of invasive abrasion of the tooth enamel did not adversely affect the performance of fissure sealant materials. Supporting the quantitative analysis, visual evaluation of marginal integrity showed that flowable composites had the highest marginal integrity while glass ionomer fissure sealant had the lowest.

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Conflict of Interest

The authors have no competing interests to declare.

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