Sys Rev Pharm 2020; 11(1): 521 – 528
A multifaceted review journal in the field of pharmacy

SEM Evaluation of Depth of Penetration and Lateral Wall Adaptations of Different Resin Based Pit and Fissure Sealants

Sazgar M. Sabir[1,*], Dara H. Rasheed[2]
[1] Department of Peadodontic, Orthodontic and Preventive Dentistry, College of Dentistry, Hawler Medical University.
[2] Department of Conservative Dentistry, College of Dentistry, Hawler Medical University.
*Corresponding Author: sazgar.muhammed83@gmail.com

Article History: Submitted: 20.10.2019 Revised: 16.12.2019 Accepted: 05.01.2020

ABSTRACT
Aim: Tend to evaluate Depth of Penetration and Lateral Wall Adaptations of three types of resin based pit and fissure sealants.
Methods: The present in-vitro study was done on 30 extracted sound human upper permanent first premolars, to evaluate the lateral adaptation and assess the penetration ability of three types of fissure sealants Helioseal Clear® (HC), Helioseal F® (HF) (Ivoclar Vivadent AG, Schaan, Liechtenstein) and experimental material (EM) (Helioseal Clear with addition of 10% of silanized dentin chips) on molars, divided into 3 equal groups of 10 each, with further sectioning of each sample into 3 parts (1 mesial, 1 middle and 1 distal sections) giving 30 samples per group, following embedding of teeth in auto-polymerizing acrylic resin, evaluation of the sections was done for depth of penetration and lateral wall adaptation by Scanning Electron Microscope (SEM).
Results: Showed that the EM sealant had more penetration depth, with a statistically significant increase compared to HC and HF, while according to the lateral adaptation, EM slightly higher than HF and HC, followed by HC and HF, respectively, with no statistically significant difference.
Conclusion: Within the limits of this study, the addition of dentin chips to fissure sealants increased the penetration depth, which made them more resistant to wear, and no statistically difference in related to the wall adaptation.
Keywords: Fissure Sealant, Caries, Dentin Chips, Depth of Penetration and Lateral Wall Adaptation.
Correspondence:
Sazgar M. SabirDepartment of Peadodontic, Orthodontic and Preventive Dentistry, College of Dentistry, Hawler Medical University
E-mail: sazgar.muhammed83@gmail.com
DOI: 10.5530/srp.2020.1.65

BACKGROUND
Dental caries is the most prevalent disease in the world, affecting children and adults, and representing a series public health issue (1). The term fissure caries was earlier used to describe the caries lesions found in pits and fissures. This definition was based on the assumption that the high incidence of caries lesions in molar pits and fissures was directly related to the poor cleaning accessibility to these surfaces (2).
The history of occlusal caries dates back for more than 100 years when Black (3) reported that more than 40% of all caries in the permanent teeth occurs in the occlusal pit and fissure surfaces. The high susceptibility of occlusal surface of molars and premolars to dental decay may be attributed to the complex morphology of pits and fissures, which are considered to be an ideal site for retention of bacteria and food remnants (4).
In order to prevent dental caries, several methods have been used including dietary control to restrict the intake of cariogenic food and induce the intake of non-cariogenic food, tooth brushing instruction to effectively remove dental plaque, and use of pit and fissure sealants to protect the tooth areas most susceptible to caries (5).
The occlusal surfaces of the newly erupted posterior teeth are particularly susceptible to carious lesions due to local conditions such as incomplete maturation of the enamel, infra-occlusion and very complex occlusal anatomy. Removing the bacterial plaque under such conditions is difficult, and those surfaces are, consequently, the most affected by caries (6).
The cariostatic properties of sealants are ascribed to their ability to obstruct pits and fissures preventing bacterial colonization and fermentation of carbohydrates to produce acids at cariogenic concentration (7).
Adaptation of the restorative material to cavity margins and internal cavity surfaces is of great importance for the long-term performance of the restoration. Adequate adhesion at the interface between hard dental tissues and restorative materials is crucial for achieving good clinical performance and durable restoration (8). Penetration of the sealant into the complete depths of pits and fissures, its lateral wall adaptation and subsequent retention are the key factors in the longevity of these restorations (14).
The aim of this in vitro study was to evaluate the interfacial microgaps generating between three types of fissure sealants [Helioseal Clear® (HC), Helioseal F® (HF) (Ivoclar Vivadent AG, Schaan, Liechtenstein) and experimental material (EM) (Helioseal Clear with addition of 10% of salinized dentin chips)] and dentin after polymerization, and depth of penetration, using Scanning Electron Microscope (SEM).

METHODS
Two Commercial resin based fissure sealants, {Helioseal F fissure sealant (HF) (as the control group), which is a light-curing, white shaded fissure sealant featuring fluoride release, and Helioseal Clear fissure sealant (HC) (as the reference group), which is a light-curing, transparent fissure sealant}, were used in this study. Dentin chips coated with silane coupling agent was added to the HC fissure sealant (EM) (as the experimental material). The commercial name,

composition and manufacture of all materials used in this study are listed in Table (1)

Table 1: The commercial name, composition and manufacturer of the fissure sealants used

Materials	Composition	Lot number	Manufacturer
Helioseal F (Control)	The monomer matrix consists of Bis-GMA, urethane dimethacrylate,and triethylene glycol dimethacrylate (58.6 wt%).The fillers are highly dispersed silicon dioxide and fluorosilicate glass (40.5 wt%). Additional contents are titanium dioxide, stabilizers and catalysts (< 1 wt%).	V25115	Ivoclar Vivadent AG/ Schaan/ Liechtenstien Exp: 2020
Helioseal Clear (As reference)	Helioseal Clear consists of Bis-GMA and tri e thylene glycol dimethacrylate (> 99 wt%). Additional contents are stabilizers and catalysts (<1 wt%).	W01582	Ivoclar Vivadent AG/ Schaan/ Liechtenstien Exp: 2020

Preparation of Dentin Chips

Twenty sound permanent third molar teeth were used to obtain the dentin powder. They were cleaned with ultrasonic scaler (Delta, Turkey). Each tooth was cut 1mm above cemento enamel junction, by using a slow speed conventional straight hand-piece with diamond disc (Komt, German), and was performed in such a way that the disc was perpendicular to the long axis of the tooth and abundance of water spray. A new disc had been used after every five samples [9].

All intra root dentin was removed with long-neck #4 tapered fissure carbide burs (KG-Sorensen, S˜ao Paulo, SP, Brazil) at low speed and without refrigeration, the prepared dentin chips were placed on glass slide and grinded immediately for 5 minutes using a domestic grinder and then with Mortar and Pestle[10], then sieved with different size of sieves, then sterilized by autoclaving with 121 c for 21 min [23], and immediately stored in vacuum sealing plastic bags after air tighting with Silvercrest Vacuum Sealer device.

The prepared dentin chips coated with silane coupling agent on a sterilized mixing pad with different proportions of dentin chips and silane coupling agent, by mixing them with a spatula and ultra-sonication

The final result is that the proportion for mixing of dentin chips with silane coupling agent was 1/10 scoop/drops for 40 minutes' ultrasonication, that were tested with infrared (IR-300) Spectrometer, revealed that the mixing was done without change in the chemical properties of both materials.

Preparation of pit and fissure sealant

The pretreated dentin chips with silane, then weighed on a sterilized mixing pad and added to the HC types of fissure sealant with the ratio of 10%. The mixture thoroughly blended by speed mixture device, the mixing procedure was carried out before all the experiments, in order to ensure a uniform mixture [11].

Homogeneity evaluation: Scanning electron microscope (SEM) analysis was performed to confirm the homogeneity of the distribution of the dentine chips in the fissure sealant. Therefore, two disc-shaped sample from experimental group was prepared for SEM analysis (Fig 1) Samples were broken in to two pieces with a chisel like blade, and the broken surfaces were gold sputter coated in a thin 15-nm layer to prevent the surface of the samples from burning during SEM observation. The broken surfaces of each samples were observed with a SEM [11].

Figure 1: Scanning Electron Microscope (Field Emission SEM, S-4160, HITACHI, Tokyo, JAPAN)

Sample Collection: The present study was conducted on 30 extracted first premolars, which had been extracted due to orthodontic reason were selected, based on following criteria.

Inclusion criteria of sample selection:

- Intact buccal surface that is free of carious and restoration,
- Unbroken buccal surface, no cracks present due to trauma and/ or extraction forceps,
- Buccal surface free from erosion, fluorosis and hypoplastic enamel deformities

The collected teeth were stored in distilled water in a refrigerator at 4°C up to 3 months to avoid dehydration until the time of the experiment. Also, the distilled water was change every week to prevent bacterial growth and to minimize deterioration according to International Standardization Organization, TS 11405 [12].

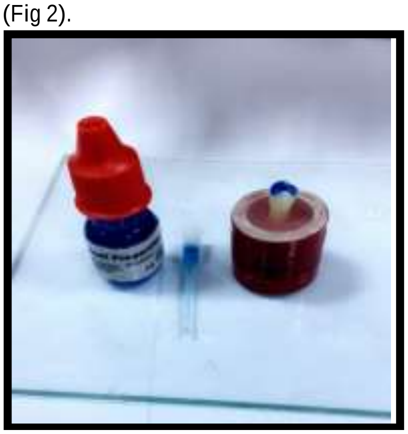
Sample Preparation

The teeth were individually mounted and placed at center of plastic tube, with the dimension of (2cm in diameter and 2.5 in height), filled with self-cure acrylic resin, covering the tooth at the level less than 2mm below the cemento enamel junction (CEJ), in such a way that the occlusal surface of the tooth was always parallel to the adjustable table of the surveyor that was previously adjust in zero angled horizontal planes using balancing instrument, high speed straight hand piece attached to the surveyor arm [13].

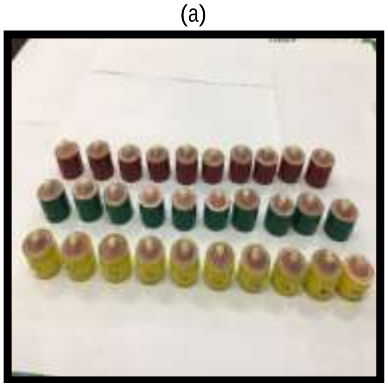
Steps in placement of sealant

All fissures in each tooth were cleaned for 15 seconds with aqueous slurry of 5g pumice/4ml water using a rubber cup in a slow-speed contra-angle hand piece. The teeth were rinsed with air-water spray. Teeth in all groups, were etched with 37% phosphoric acid gel (Email Preparator Blue; Vivadent), and let it react for 30 seconds, then were rinsed thoroughly with water for 20 seconds and lightly dried with water- oil free air. The etched enamel should have a mat white appearance. After etching and drying, pit and fissure sealant was applied directly with the disposable cannula. Sharp explorer was used to remove any air bubbles and uniform flow of the material was ensured. Waited for approximately 15 seconds the sealant was cured with a bluephase polymerization light for 20 seconds, according to manufacturer's instructions [18].

Each tooth was checked with an explorer for complete coverage and retention of the sealant, after that the restored teeth were stored in distilled water for 7 days at 37°C in incubator (Fig 2).



(a)



(b)

Figure 2: (a) Etching occlusal surface of teeth with 37% phosphoric acid gel (Email Preparator), (b) Placement of fissure sealants on occlusal surface of teeth.

Thermocycling

After placement of fissure sealant, all the restored teeth were subjected to thermocycling, the purpose of thermocycling was to simulate the oral environment.

All specimen was immersed alternately in water baths at 5+-2°C followed by 55+-2°C for 600 cycles, with at dwell time of 60 seconds in each bath and a transfer time of 15 seconds [14].

After thermocycling, all teeth were placed in water at 37°C in incubator until they were sectioned. The whole crown, then sectioned buccolingually, three sections were obtained from each tooth (1 mesial, 1 middle and 1 distal sections) by using a slow speed conventional straight hand piece with diamond disc (Komt, Germany), at a speed range from 8000-12000 rpm with water coolant, and was performed in such a way that the disc was parallel to the long axis of the tooth) also using surveyor for standardization [14].

Scanning Electron Microscope evaluation

The specimens were allowed to dry for 24 h before subjecting them to gold sputtering. For this, the specimens were mounted on aluminum stubs using double-sided adhesive tape; they were mounted in such a way that area to be studied faced upward, then coated with a thin layer (25 nm thickness) of pure gold using an ion sputtering unit. The stubs were then placed in the vacuum chamber of the SEM. The accelerating voltage, angle of tilt and the aperture were prepared to suit the specimen to optimize the quality of the micrograph. Under different magnifications (×13 to ×1500), the surfaces were scanned and observed on the screen [15].

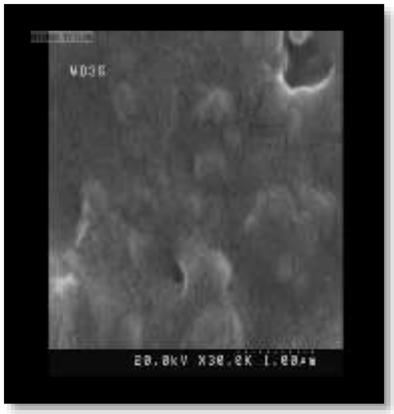
Parameters that were studied were:

1. Analysis was performed to confirm the homogeneity of the distribution of the dentine chips in the fissure sealant.
2. Sealant penetration depth (μ) calculated as length measured (μ) from the deepest point on concavity of the upper margin of the occlusal sealant to the base of the sealant.
3. Total length of fissure (μ) calculated as length measured (μ) from deepest point on the upper margin of the sealant to the base of the fissure.
4. Penetrability (%) = $\frac{\text{Sealant Penetration Depth}}{\text{Total Length of Fissure}} \times 100$
5. Sealant adaptation was expressed as the gap width. The gaps between the sealant and tooth surface were measured by placing the two indicator marks at two extremes of the gaps along the lateral walls and the distance between them was recorded as given by the computer under higher magnification (×300 to ×1500). the mean of the minimum and the maximum gaps was calculated for each section in μm [14].

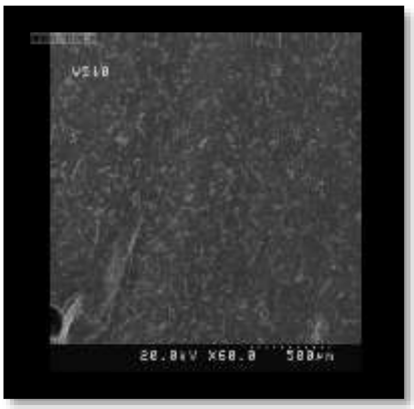
All the linear measurements were made with the help of software (ImageJ, National Institutes of Health, Bethesda, USA) and calculated in microns (μ).

RESULTS

SEM images of experimental material (EM) showed individual dentin chips particles that are embedded in the resin fissure sealant along with other fillers and there are homogenous distributions of the filler inside HC fissure sealants, as shown in Figure (3), and average particle size is between 160.0 nm – 2460 nm as shown in Figure (4).



(a)



(b)

Figure 3: SEM image of homogenous distribution of dentin chips in HC fissure sealant

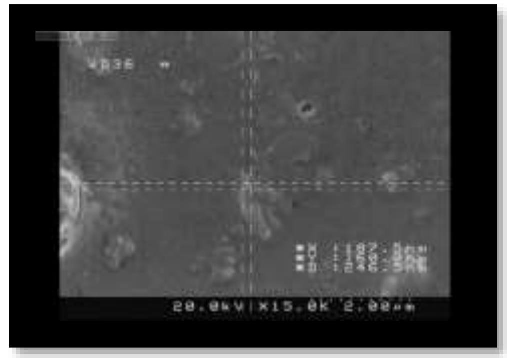


Figure 4: SEM image of experimental materials

The means and standard deviations of the depth of penetration values of the tested materials are shown in Table (2). One-way ANOVA test Table (3) revealed that a statistically significant difference ($P<0.05$) were found among the groups, in which the Helioseal F (HF) and Helioseal C (HC) groups showed the lowest penetration mean value with statistically no significant difference between them. While the Experimental material (EM) group showed the highest penetration value and there were statistically significant differences between (HF, EM) groups and (HC, EM) groups. The result showed that the addition of dentin chips to the Helioseal Clear resin based fissure sealant significantly increased the penetration value of such fissure sealant.

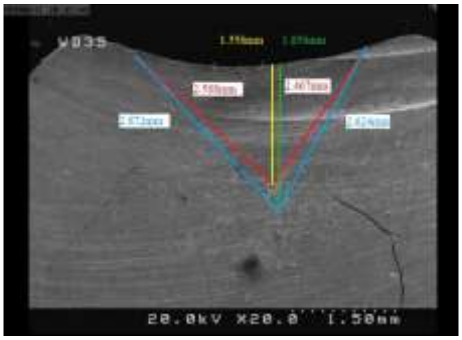
Table 2: Mean values, standard deviations, standard errors and 95% confidence intervals for depth of penetration data.

Groups	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bond	Upper Bond		
HF	30	87.33	13.40	2.45	82.33	92.34	60.00	100.00
HC	30	87.43	10.34	1.89	83.57	91.30	60.00	100.00
EM	30	94.00	8.78	1.60	90.72	97.28	68.00	100.00

Table 3: ANOVA test for difference among groups for depth of penetration

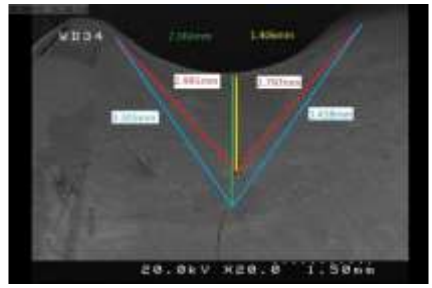
Source of Variation	Sum of Squares	df	Mean Square	F	P-value	Sig.
Between Groups	875.756	2	437.878	3.612	0.0311	.0
Within Groups	10546.033	87	151.219			
Total	11421.789	89				

The variation of the depth of penetration of materials in to the fissures by scanning electron microscope shown in (Fig 5)



(a)

(b)



(c)

Figure 5: Scanning Electron Microscope pictures showing penetration of sealant into fissures in different groups; (a) HF, (b) HC, (c) EM

All groups showed some degree of adaptation failure as shown the means and standard deviations of the lateral adaptation values of the tested materials as shown in Table (4). One-way ANOVA test Table (5). EM group showed the lowest mean values (highest degree of adaptation), followed by HC groups, and then followed by HF groups with statistically no significant difference was found between them.

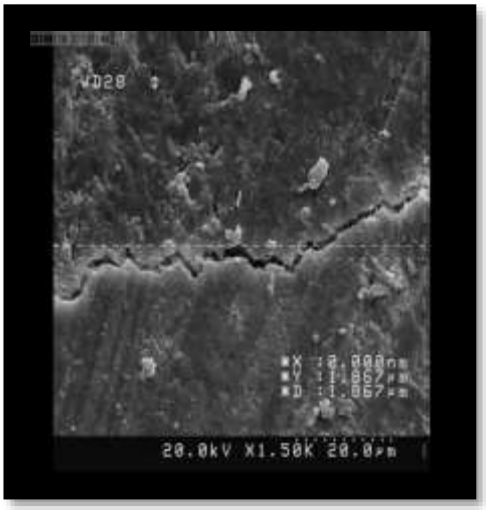
Table 4: Mean values, standard deviations, standard errors and 95% confidence intervals for lateral adaptation data.

Groups	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bond	Upper Bond		
HF	30	3.84	1.52	0.28	3.27	4.40	1.07	7.33
HC	30	3.62	1.41	0.26	3.09	4.14	1.27	6.67
EM	30	3.42	1.91	0.35	2.70	4.13	0.65	9.80

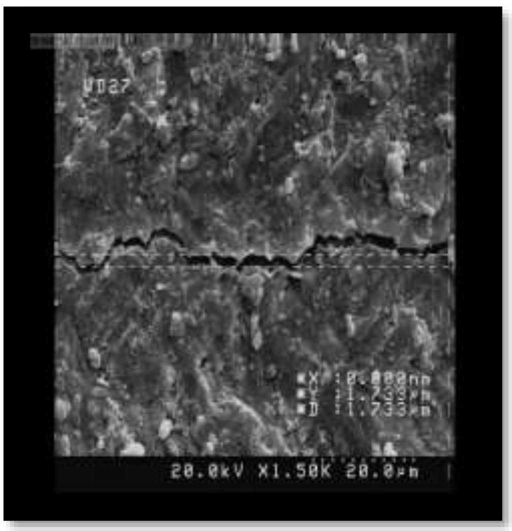
Table 5: ANOVA test for difference among groups for lateral adaptation

Source of Variation	Sum of Squares	df	Mean Square	F	P-value	Sig.
Between Groups	2.617	2	1.3082	0.4934	0.6122	
Within Groups	230.674	87	2.6514			
Total	233.290	89				

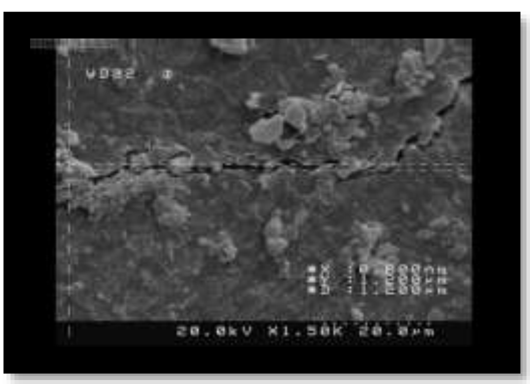
The lateral wall adaptation of materials in to the fissures by scanning electron microscope showed in (Fig 6).



(a)



(b)



(c)

Figure 6: Scanning Electron Microscope pictures showing gap along lateral wall of fissures in different groups; (a) HF, (b) HC, (c) EM

DISCUSSION

Scanning electron microscope (SEM) images of experimental helioseal clear resin based fissure sealant with 10% dentin chips (pretreated with silane coupling agent) added, showed individual particles that are embedded in the resin based fissure sealant along with other filers and there are homogenous distributions of the dentin chips within the sealant, this might be due to the fact that the added fillers were silane coated which attribute to the good bonding between the nano-sized particles and the matrix and can efficiently enhance uniform dispersion of the fillers in nano-scale in composite resin [22].

In this study, penetration and lateral wall adaptation of resin-based pit and fissure sealants were evaluated using SEM analysis.

The use of SEM provides a mean of direct visual observation of penetration and adaptation of sealant materials to enamel walls due to its focusing in depth and magnification. Using of computer software for calculating the measurements, gives reflecting results better than personal estimation using a scoring system.

Regarding sealant penetration, EM showed better penetration than HF and HC, which might be due to the combination of low viscosity.

At times, the size of the filler particles may be larger than the porosities of the enamel. Therefore, faster penetration rates are found with larger holes, but slower rates are found with fluids that are more viscous. The addition of filler particles that alter the flow of the sealant and the size of the filler particles used, are influence the depth of penetration of the pit and fissure sealant [14]. Barnes et al has been studied that the sealants containing fluoride tend to be thicker than those without fluoride [21].

Sridhar et al [18] evaluated the more penetrability of Clinpro fissure sealant than Helioseal F fissure sealant attributed to the lesser filler loading of 6% in Clinpro fissure sealant when compared to filler loading as high as 43% in Helioseal F fissure sealant. Lesser filler loading contributes to low viscosity which enables better penetrability into pits and fissures.

Thus, placement of pit and fissure sealant can be considered as an effective treatment modality in preventive dentistry. Though all the three sealants have shown the depth of penetration of more than 60% but due to better results obtained with EM sealant (94.00%) versus HF Sealant (87.33%) and HC Sealant (87.43%).

The properties of the pit and fissure sealants such as surface tension and viscosity are the important factors that affect penetration of the sealants. Cleaning and conditioning of the fissures can remarkably influence the penetration behavior of the sealants [16]. Blackwood JA et al, showed that the least microleakage was seen with the conventional pumice prophylaxis compared to enameloplasty [20].

The materials used in the present study were applied without enameloplasty in order to observe the behavior of these materials without removal of tooth substance, as done in the in vitro studies by Prabhakar et al and Sridhar et al [19,18], Even though the sealing efficiency of pit and fissures and the penetration into the etched enamel is of prime importance in caries prevention [17]. The depth of penetration of the EM was found to be superior followed by HC and HF.

The results of the present study revealed a good performance of EM in relation to its adaptation to the fissure walls with no significant differences in compared to HC and HF, it is important to note that during HF application, the inclusion of air bubbles was very common, could have occurred in consequence of the injector tip of HF. It was observed that the HF tip has greater diameter than that HC. After the material use, the syringe's plunger was always pulled back and this will effect on the adaptation, this superficial defect occurrence was also verified in the study of Kobayashi et al [24], who observed the inclusion of air bubbles are more in HF sealant, as well as those of Koch et al [25] who reported that materials with fillers have a significantly higher deficiency in the marginal adaptation than those without fillers.

To the best of our knowledge, Helioseal Clear and Helioseal F (Ivoclar Vivadent) resin-based had never been compared in vitro studies before.

CONCLUSION

The present study concluded that the sealants used in this study showed some degree of failure in adaptation, the more adaptation was seen with EM sealant.

The major disadvantage of resin-based sealants is the technical sensitivity during clinical applications.

Continuous improvement of materials and clinical techniques has led to better marginal adaptation and penetration.

The present study was conducted in an in-vitro environment; care was taken to replicate the oral conditions in terms of thermocycling.

REFERENCES

1. Ribeiro A. Biological Features of Dental Caries. JSM Dent 4(3): 1065, 2016.
2. Kervanto-Seppälä S. Arresting occlusal enamel caries lesions with pit and fissure sealants. M.Sc. thesis, University of Helsinki, Faculty of Medicine, in Auditorium, Biomedicum, Helsinki; Finland, 2009.
3. Black GV. A work on operative dentistry. The technical procedures in filling teeth. Chicago Medico – Dental Publishing; 2:1908: 63.
4. Nirwan M, Nigam GA, Marwah N, Nayak UA, Bansal A, Gahlot MS. A comparative evaluation of retention of pit and fissure sealant bonded using sixth-, seventh-, and eighth- generation adhesives: An in vivo study. J Indian Soc Pedod Prev Dent; 35: 359- 66, 2017.
5. Oong EM, Griffi n SO, Kohn WG, Gooch BF, Caufi eld PW. The effect of dental sealants on bacteria levels in caries lesions: a review of evidence. J Am Dent Assoc. 139: 271- 8, 2008.
6. Brown LJ, Kaste L, Selwitz R, Furman. Dental caries and sealant usages in U. S. children, 1988-1991: selected findings from the Third National Health and Nutrition Examination Survey. J Am Dent Assoc. 127: 335- 43, 1996.
7. Naaman R, El-Housseiny AA, Alamoudi N. The Use of Pit and Fissure Sealants-A Literature Review. Dent J (Basel); 5 (4): 34, 2017.
8. Gorseta K, Borzabadi-Farahani A, Vrazic T and Glavina D. An In- Vitro Analysis of Microleakage of Self-Adhesive Fissure Sealant vs. Conventional and GIC Fissure Sealants. Dent J; 7 (32), 2019.
9. Mahdi SF. Evaluation the effect of different irrigation solutions and techniques on smear layer removal and erosion of root canal dentin (In vitro comparative study). M.Sc. thesis. Hawler Medical University, College of Dentistry, Iraq, 2019.
10. Kim ES. Autogenous fresh demineralized tooth graft prepared at chairside for dental implant. Kim Maxillo Plast and Reconstruct Surg: 37:8, 2015.
11. Saleem SS. Modification of Microhybrid Composite Resin by addition of Zinc oxide and Calcium Carbonate Nanofillers. Ph.D. thesis. Hawler Medical University, College of Dentistry, Iraq, 2013.
12. International Standards Organization. Technical Specification ISO 11405. Dentistry- Testing of adhesion to tooth structure, 2015.
13. Shahswar RS, Saleem SS. Compressive Strength of Three Types of Pit and Fissure Sealants (An In-Vitro Comparative Study). JPAS; 30 (6): 107-117, 2018.
14. Garg N, Indushekar KR, Saraf BG, Sheoran N, Sardana D. Comparative Evaluation of Penetration Ability of Three Pit and Fissure Sealants and Their Relationship with Fissure Patterns. J Dent Shiraz Univ Med Sci; 19(2): 92-9, 2018.
15. Al-Jobair A. Scanning electron microscope analysis of sealant penetration and adaptation in contaminated fissures. J Indian Soc Pedod Prev Dent; 31:169- 74, 2013.
16. Burrow MF, Burrow JF, Makinson OF. Pits and fissures: etch resistance in prismless enamel walls. Aust Dent J; 46: 258-262, 2001.
17. Kakaboura L, Mathaiou L, Papagiannoulis. In vitro study of penetration of flowable resin composite and compomer into occlusal fissures. Europ J Pedi Dent; 4; 205- 9, 2002.
18. Sridhar LP, Moses J, Rangeeth BN, Sivakumar S. Comparative Evaluation of the Marginal Sealing Ability of two Commercially Available Pit and Fissure Sealants. *J Clinic Diag Res*; 10(9): ZC01-ZC04, 2016.
19. Prabakar AR, Sankriti A. Murthy, Sugandhan S. Comparative evaluation of the length of resin tags, viscosity and microleakage of pit and fissure sealants – an invitro scanning electron microscope study. Cont Clin Dent; 2(4): 324- 30, 2011.
20. Blackwood JA, Dilley DC, Roberts MW, Jr Swift EJ. Evaluation of pumice fissure enameloplasty and air

abrasion on sealant microleakage. Pediatr Dent; 24:199-203, 2002.

21. Barnes DM, Kihn P, von Fraunhofer JA, Elsabach A. Flow characteristics and sealing ability of fissure sealants. Oper Dent; 25: 306- 10, 2000.
22. Mortazavi V, Fathi MH, Ataei E, Khodaeian N & Askari N. Shear Bond Strengths and Morphological Evaluation of Filled and Unfilled Adhesive Interfaces to Enamel and Dentine. Inter J Dent: 1-9, 2012.
23. Rafael D, Andrade F, Martinez-Trucharte F, Basas J, Seras-Franzoso J, Palau M, et al. Sterilization Procedure for Temperature-Sensitive Hydrogels Loaded with Silver Nanoparticles for Clinical Applications. Nanomaterials: 9, 1-14, 2019.
24. Kobayashi TY, Rios D, Machado MA, Oliveira TM, Silva SM. A Two-Year Clinical Evaluation of fluoride and non-fluoride resin-based pit-and-fissure sealants. Braz Dent J; 26: 678- 4, 2015.
25. Koch MJ, Garcia-Godoy F, Mayer T, Staehle HJ. Clinical evaluation of Helioseal F fissure sealant. Clin Oral Investig; 1:199-202, 1997.