THE EFFECT OF PIT AND FISSURE MORPHOLOGY AND SEALANT VISCOSITY ON SEALANT PENETRATION AND MICROLEAKAGE

by

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Submitted to the Graduate Faculty of the School of Dentistry
in partial fulfillment of the requirements for the degree of
Master of Science in Dentistry, Indiana University School
of Dentistry, 2017.

Thesis accepted by the Departments of Cariology, Operative Dentistry, and Dental Public Health, Indiana University School of Dentistry, in partial fulfillment of the requirements for the degree of Master of Science in Dentistry.

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DEDICATION

This work is humbly dedicated to all those in my life who made this possible.

To my husband, Wajdi, whose unwavering support, encouragement, and sacrifice made it possible for me to complete this work. His faith in me has been my motivation. He has gotten me through the challenges and was the first to cheer my accomplishments.

To my loving parents, the reason for who I am today. They instilled in me a love of learning from a young age and encouraged me all the way. Their unconditional love and support have guided me through difficulties, and they have always believed in me.

To my dear brother, Dr. Yacin, who welcomed my family and me with open arms to share life each step of the way, and whose care and advice eased my burdens with a shoulder to lean on.

To my brothers, Taha and Assadi. Taha's kind words from afar always comforted me and gave me a sense of peace. Assadi would keep me guessing every year when the next surprise visit would come to give me a much needed stress relief.

To my younger sisters, Rasha and Safa, whose friendship more than anything I value and hold dear. Their vocal pride in me gives me confidence and encouragement.

To the loves of my life, Ibrahim, Enji, and Mohammed, my children. My love for you has made me a better person and given me a sense of fulfillment in life. All I do is for you.

ACKNOWLEDGMENTS

First and foremost, I would like to thank Dr. Andrea Zandona for accepting me into the Preventive Dentistry Program at Indiana University. Dr. Anderson Hara, my program director, I would like to thank you for your support, advice, guidance, knowledge, and skills that I will hold forever.

I would like to acknowledge the other members of my committee: Dr. Armando Soto, my research advisor, and Dr. N. Blaine Cook.

Dr. Soto helped me to get through a difficult phase of my life. He showed me that when one door closes, another opens, and he placed opportunities in front me that I thought were not possible. I truly appreciate his effort and work in guiding me into the program and the work I have done.

Dr. Cook's expertise, tolerance, patience and wisdom have given me knowledge and skills that I will forever treasure.

Finally, and most important, highest praise is owed to my husband, Wajdi, for his patience, encouragement, and love.

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INTRODUCTION

Dental caries is still a common chronic disease in childhood.[1] Many studies have been documented that the occlusal surfaces of posterior permanent teeth are known to be the most susceptible areas for the development of dental caries lesions as they have pits and fissures.[2] Pits and fissures are considered as a shelter and rich incubator for microorganisms and food.[2] Indeed, pit and fissure sealants have become one of the biggest breakthroughs in terms of dental prevention.[3,4,5] Many studies have emphasized the effectiveness of correctly placed sealants in preventing caries for permanent teeth.[3,4,6] They have been found not only to prevent occlusal caries, but also to arrest the progression of incipient lesions.[4]

Dental sealants were developed and studied by Dr. Buonocore in the early 1970s[7,8] Various studies in the early 1980s reported that pit and fissure sealants reduced caries lesions in developed countries[3,5] The advantages of sealing the occlusal surfaces are the decrease in caries prevalence when compared with non-sealed teeth, and the associated lower cost compared with the cost of placing a restoration.[3,5]

Nowadays, there are two types of pit and fissure sealants on the market: resin-based and glass ionomer-based. Resin-based sealants are categorized into generations according to their mechanism of polymerization or their content.[9,10] Different types of dental sealants are available, such as, light activated, self-cured, chemically cured, and fluoride-releasing sealant.[9,10-12] Resin-based sealants are also classified into filled and unfilled. This consists of the presence or absence of filler particles.[10,13] Filled sealants contain microscopic glass beads, quartz particles, and other fillers used in composite resins.[10,13,14] Glass ionomer cement was introduced in 1974 by McLean and Wilson,[10,15] as an alternative sealant material, especially in situations where moisture control is difficult due to its hydrophilic properties.[10] Glass ionomer also has good adhesion, biocompatibility, and fluoride release.[16]

The retention of dental sealants is one of the most important features of success in preventing dental caries.[17,18] The longer the material remains bonded to the occlusal surfaces, the more protection provided to the tooth. Several factors may be considered when evaluating the occlusal surfaces where the sealant is to be placed that may influence its retention. One of the most important factors in sealant retention may be the morphology of pits and fissures.[19,20,21]

According to Nagano's classification, there are five major types of occlusal pits and fissures described as V, U, I, IK and Inverted Y[19] (Figure 1). Nagano's study showed that fissure morphology was related to the susceptibility of the tooth to caries; the author reported that V- and U-type fissures are self-cleansing and have less risk of developing caries than other types (I, IK, and Y).[19] Results of a study by Selectman et al.[20] using a different fissure morphology description showed that 29 percent of fissures were "U" types, 37% "Y", 25% were Y1 and Y2 fissures in morphology (Figure 2). However, Nagano's description showed different percentages with the U-type (14%), V-type (34%), I-type (19%), IK-type (26%), and Inverted Y-type was 7%.[19] Grewal and Chopra et al. (2008), reported that a fissure's shape was highly significant for sealant penetration; V- and U-shaped fissures were found to have the greatest penetration while IK types and I types showed the lowest penetration of the dental sealant.[21] Another factor that may affect sealant retention or the penetrability of the sealant is the viscosity of the material.

Bottenberg et al. (1989) found that the viscosity of the dental sealant and the surfactant content influenced the penetration ability of the sealant material into the bottom of the fissure on an *in-vitro* study.[22] Therefore, penetration of the sealant into the porous etched enamel may depend on the viscosity of the material and the fissure's morphology.

Marginal leakage of the oral fluid into the space between the tooth surfaces and the sealant material interfaces may increase the risk of developing caries below the sealant.[23,24-26] By proper filling of the pit and fissures with the sealant material, the access to fermentable substrates is blocked, and this is only true as long as the sealant is retained.[27] Therefore, microleakage is an unwanted outcome from an improper sealant placement or a defective bond of the dental sealant.[25,26] Various clinical studies have documented that the resin-based sealants have shown lesser microleakage rates compared with glass-ionomer dental sealant materials.[10,15,27] Several *in-vitro* studies reported that low viscous dental sealant produced significantly less microleakage than high-viscosity dental sealant.[28,29]

AIM OF STUDY

The purpose of this *in-vitro* study was to assess the influence of fissure morphology and the sealant viscosity on sealant penetration and microleakage at the sealant-enamel interface.

Null Hypothesis

The viscosity of the dental sealant and the fissure morphology will not have an effect on the sealant penetration ability and microleakage.

Alternative Hypothesis

The viscosity of the dental sealant and the fissure morphology will have an effect on the sealant penetration ability and microleakage.

REVIEW OF LITERATURE

Tooth decay remains as one of the most common chronic childhood diseases.[1] Indeed, around 75 percent to 80 percent of the occlusal caries lesions in children ages 5 to 17 years occur in a small segment of the population (20 percent to 25 percent) [1,10] Additionally, dental caries is considered a major problem in dentistry that should receive significant attention, not only from the standpoint of restorative procedures, but also in terms of preventive measures designed to reduce caries.[4,30] Dental caries is defined as a site-specific disease that manifests itself primarily in pits and fissures.[4,31] Pits and fissures of posterior permanent teeth are vulnerable sites for dental caries due to their morphology.[2,10] The microorganisms (bacteria) and food (carbohydrates) are commonly retained in the pits and fissures. Bacteria convert these carbohydrates into acids and then lead to de-mineralization of the enamel.[2,10] Therefore, the occlusal surface (pits and fissures) has been reported to be the most at risk areas for caries to develop.[32-34] Around 90 percent of dental caries lesions are found in the pits and fissures of occlusal surfaces of permanent teeth.[2]

The most efficient way of preventing and arresting caries in primary and permanent molars in children and adolescents is by effectively sealing these pits and fissures with a material (pit and fissure sealants).[10,11]

HISTORY OF PIT AND FISSURE SEALANTS

For more than thirty years, pit and fissure sealants have been recommended for caries prevention. [1, 35,36] By sealing the pits and fissures with a resin based material, thus a mechanical barrier (protective layer) is created between enamel surface and biofilm, to reduce the accumulation of dental plaque, food, and micro-organisms and prevent caries development.[37,38] Several studies have shown that the sealant must completely seal the pits and fissure system from the fermentable food substrates and bacteria to be effective in the prevention and reduction of dental caries.[3,21, 39] Subsequently, a pit and fissure sealant provides a smooth and cleansable surface for saliva and the toothbrush bristles when cleaning these surfaces.[40]

Dr. Buonocore advocated the benefits of etching the enamel surface with 85-percent phosphoric acid for 60 sec in 1950s.[7,8,41,42] Several studies demonstrated that the resin-based material could be bonded to the etched enamel mainly through a micro-mechanical retention. This improved the marginal integrity of the resin restorative material.[22,33,41] In the late 1970s and early 1980s, pit and fissure sealants played an important role in the drop in the caries indices prevalence.[5,7,8] The advantage of sealing the occlusal surfaces is associated with the decrease in caries prevalence when compared with non-sealed teeth, and sealing has a lower cost compared with placing a restoration.[3,5,24,40]

Nowadays, there are two types of pit and fissure sealants in the market: resin-based and glass-ionomer-based.[43,44]

RESIN-BASED PIT AND FISSURE SEALANTS

Resin-based materials are widely used as pit and fissure sealants.[38,45] By definition, a pit and fissure sealant is an organic polymer (resin) that flows into the pit and fissure that bonds to the prepared enamel surface (etched) mainly by micro-mechanical retention following the acid etch process.[46] By mid 1960s, the first sealant material that utilized the acid etch-technique was a methyl cyanoacrylate material.[47,48] Later, Bowen in 1965 reported that a viscous resin material known as BIS-GMA was found to be resistant to bacterial degeneration and produced a tenacious bond with the treated enamel surface.[5, 48-50] By 1967, Boununocre and Cueto suggested using an adhesive resin material for the sealing of pit and fissures.[47,50] The first commercial dental pit and fissure dental sealant was introduced in 1971; it was Nuva-Seal (LD Gaulk, Milford, Del).[20,38,50]

Resin-based sealants are categorized into generations according to their mechanism of polymerization or their contents.[44] The development of sealants has progressed from the first generation sealants that were activated using an ultraviolet light, through the second and third generation sealants of auto-polymerized and visible light activated sealants, to the fourth generation containing fluoride.[11,44] The first generation sealants are no longer in the market. The first sealant material introduced was a cyanoacrylate that utilized the acid-etch technique.[44] The second-generation sealants are BIS-GMA di-meth-acrylates; these can be self-cured and chemically cured.[11, 44] During the sealant application, the etched enamel surfaces might be contaminated with water and oral fluids, and this leads to incomplete penetration of acrylic resin and proper bonding due to the hydrophilic property of BIS-GMA.[52,53] This is one of the most frequently cited reason for sealant failure.[52,53] The third generation of dental sealants is the photo-activated resin; a visible light source that is required to initiate the polymerization process.[44] The last generation is a fluoride-releasing sealant.[12]

Resin-based dental sealant materials are also classified into filled and unfilled resin systems based on the presence or absence of filler particles on the system.[13] Filled sealants contain microscopic glass beads, quartz particles, and other fillers used in composite resins.[13] Sealants present different colors; the first colored sealant that was introduced to the US market was white (3MTM ESPE TM Concise TM).[11] Different types of sealants are also available such as clear, tinted, and opaque dental sealants.[14] Certain types of sealant have the capability of changing color through light activation such as the Helioseal Clear Chroma and Ivoclar Vivadent that change from clear to green color after light curing.[14] The 3MTMESPETM Clinpro Sealant changes from pink to white opaque after photo-polymerization.[14]

GLASS IONOMER-BASED PIT AND FISSURE SEALANTS

In 1968 Smith introduced the first poly-carboxylate cement that bonded chemically to the enamel surface.[54] Later, in 1974 McLean and Wilson introduced the glass ionomer material as a restorative material, base, and cementing agent.[15,55] Glass ionomer cement is less sensitive to moisture contamination due to its hydrophilic properties and rapid setting.[10,15,56] For this reason, it is the best alternative sealant that could be used in patients in which moisture is difficult to control.[10,15,56] The additional advantage of glass ionomer cement over the conventional resin based sealants is that it releases fluoride.[56] Subsequently, it is reported that glass ionomer increases the resistance of the fissure to demineralization and may prevent occlusal caries even after the sealant has failed.[9,31,41,48,57,58] In addition to this, many studies that show that glass ionomer prevented dentin caries lesions better than resin-based sealants.[59, 60]

MORPHOLOGY OF PITS AND FISSURES

Several factors may be considered when clinicians evaluate the occlusal surfaces where the sealant will be placed, because this may have an influence on its ability to seal the pit and fissure system. One of the most important factors could be the morphology of pits and fissures.

According to Nagano's classification, five major types of occlusal pits and fissures are described as V, U, I, IK and Inverted Y.[19] As for the percentage distribution pattern of fissure morphology, the results of Nagano's study were V-type (34%), U-type (14%), I-type (19%), IK-type (26%), and inverted Y-type (7%).[19] Nagano's study concluded that the Vand U types were self-cleansing and caries-resistant; however, K-type fissures were very susceptible to caries.[19] Results of a study by Selectman et al. using a different fissure morphology description showed that 37 percent of fissures were U types, 25 percent V, 8 percent were Y1 and Y2 fissures in morphology.[20] In addition, no significant differences between fissure types were found for microleakage and penetrability in this study.[20] Another study by Grewal and Chopra reported that the fissure's shape was highly significant for sealant penetration; V- and U-shaped fissures were found to have the greatest penetration, while IK types and I types showed the lowest penetration of the dental sealant.[21,27] The sealant penetration ability to the base of shallow fissures occurred more frequently than in a deep fissure system.[21,27] This may be because in the shallow pit and fissure systems it is more likely to be completely clean from any residual impurities, and this may contribute to a better sealant penetration into the base of the pit and fissure.[21,27] On the contrary, in deep fissure systems these may not be completely dry prior to the sealant placement.[21,27]

VISCOSITY OF PIT AND FISSURE SEALANTS

Another factor that may affect sealant penetration into the fissures is the viscosity of the material. The viscosity of material has been defined as the measurement of a liquid's internal resistance to flow.[63] Some studies showed that sealant viscosity may impact the ability of sealants to seal and prevent caries.[64, 65] Bottenberg et al. (1989) found that the viscosity of the dental sealant and the surfactant content influenced the penetration ability of the sealant material into the bottom of the fissure on an *in-vitro* study. [21] There are some investigations that have shown that the low-viscosity sealant penetrated fully into the pit and fissure system demonstrating a better marginal seal.[28,66] On the other hand, the high viscosity sealant did not penetrate as well into the fissure depth and therefore did not exhibit a better marginal adaptation.[28,65-68] Subsequently, the penetration of the sealant into the porous etched enamel may depend on the viscosity of the material and the fissure's morphology.[28, 65, 66]

MICROLEAKAGE EVALUATION

The term microleakage is used to describe a leakage of minute amounts of oral fluid and bacteria through the microscopic space at the tooth structure and dental restoration interface.[23, 24] Consequently, bacterial leakage beneath the dental restoration can have adverse effects such as: enamel demineralization, discoloration, and decreased bond strength.[25,26]

The marginal seal of the pit and fissure sealant can be measured in *in-vitro* studies through evaluation of dye penetration. This is the most common method used.[18,64,69] There are various types of dye with different concentrations that have been used for microleakage evaluation of dental sealants such as; methylene blue, [72] silver nitrate,[73] radioactive isotopes,[74] alcohol gentian violet, [26] basic fuchsin,[75] rhodamine,[76] and erythrosine.[77]

Different methodologies have been used to measure microleakage *in-vitro* by assessing the dye penetration along the tooth structure-dental restoration interface. Some studies assess the dye penetration by measuring the percentage of dye penetration along the enamel-sealant interface.[71] Other studies have scored the degree of microleakage by using the criteria developed by Colley et al.[78,79] which is: score (0) no marginal dye penetration; (1) marginal penetration along the enamel-sealant interface; (2) dye penetration to the depth of sealant/fissure. A ranked scale method described by Grande et al. has also been used by some studies to measure microleakage.[80] The rank is described as: (0) no dye penetration; (1) dye penetration into the occlusal third of enamel-sealant interface; (2) dye penetration into the middle third of enamel-sealant interface; (3) dye penetration into the apical third of the interface.[80]

EVALUATION OF SEALANT PENETRATION

The most predominate factor governing the longevity of a dental sealant is its penetration depth along the pit and fissure system.[27] Subsequently, boosting the ability of the sealant to penetrate deeply and fully into the fissure should improve the sealant retention.[27] Indeed, the ability of sealant penetration along the pit and fissure system may depend on various factors. These factors could be pit and fissures configuration, the clinical technique, and the physical and chemical properties of the dental sealant.[21,27] Some studies reported that the complete penetration of dental sealants into the deep and narrow fissures is hard to achieve, and that this is due to the phenomenon of closed-end capillaries or isolated capillaries.[29,81]

There are different methods that have been employed to assess the sealant penetration into the fissure. Some studies have used a simple recording of the penetration as a complete or incomplete penetration regardless of fissure length.[82] Some other studies have recorded penetration as a percentage of sealant depth to evaluate sealant penetration.[83] Other studies used a ranked scale system to evaluate sealant penetration depth, such as a scale described by Hosoya et al.[84] The rank is: (0) no penetration; (1) sealant penetrated into the outer half of the fissure; (2) sealant penetrated into inner half of the fissure; (3) penetration restricted into almost all fissure but one minor failure of adaptation or penetration; (4) complete penetration and adaptation of sealant into the fissure. Some studies have assessed the penetration of sealant as a presence or absence of unfilled areas below the sealant material, or by measuring the proportion of unfilled to filled areas.[85,86]

AGING CONDITION (THERMOCYCLING)

The thermocycling process is the most common method used on *in-vitro* studies to assess the influence of thermal stresses on the bond strength of dental materials and their durability.[87] During eating, drinking, and breathing, teeth are exposed to thermal changes, so the thermocycling method could mimic these thermal changes and provide more information about the aging process of dental materials in an *in-vitro* situation.[88] Due to the mismatch of coefficient of thermal expansion (CTE) between the restorative material and tooth structures, this may results in an unfavorable effect on the margins of the restoration (enamel is around $11.4x10^{-6\circ}C^{-1}$, dentin $8.0x10^{-6\circ}C^{-1}$, while resin composite is17-$50x10^{-6\circ}$).[89,90,91] Therefore, these thermal stresses could affect bond strength of the material directly, which can increase the crack propagation along the bonded interface, cause bond failure, and may lead to microleakage along the tooth-restoration interface.[91]

Many investigators have shown that thermocycling regimens used different numbers of cycles, temperature, and dwell times. Many studies documented a range of cycling from 100 cycles to 500 cycles, while the temperature extremes range from 4°C to 15°C for the cold bath, and up to 45°C to 60°C for the hot bath. Although the dwell time is different, some used 15 seconds, while others used 30 seconds or 60 seconds.[92] Moreover, ISO standard (ISO TR 11450) proposed a regimen of 500 cycles.[93]

MATERIAL AND METHODS

EXPERIMENTAL DESIGN

This was an *in-vitro* study on permanent molars. Two types of sealants were studied: A: a low viscosity (Delton Light-Curing Direct Delivery System Opaque, US DENTSPLY), and B: a high viscosity sealant (UltraSeal XT Plus, Hydrophobic pit and sealant, US ULTRADENT). Extracted teeth were sealed; dental sealants were placed, and specimens were thermocycled (500 cycles for 24 h). The penetration of sealant was measured in microns, and digital images were obtained to measure the dye leakage.

Dental Sealant Viscosity Measuring

The viscosity of the sealants was measured using a rheometer (Model cS, Bohlin, East Brunswick, NJ) in cone-and-plate setting at shear rates of 1-1000s^{-1}.[94] The gap size was set at 200 μm, and the steady shear viscosity was measured at each shear rate.[94]

Teeth Selection and Imaging

One hundred and fifty extracted teeth were obtained from Oral Health Research Institute (OHRI) (IRB#0306-64). An inclusion criterion for selection of teeth for this study included only permanent first and second molars, with an ICDAS code 0-1. The exclusion criteria were teeth with histological and morphological defects, the presence of a restoration, or the presence of large caries lesions. Teeth were cleaned using a toothbrush and no invasive technique (e.g. enamelplasty) was used prior to sealant placement. The teeth were washed with water for 15 seconds and stored in 0.1-percent thymol all the time. Fissure morphology of the teeth was assessed. The teeth were separated visually into three subgroups based on fissure anatomy (Group one: V-fissure shape, n = 19), (Group two: U-fissure shape n = 19); (Group three: for convenience had both fissure types I, and inverted Y, n = 38). Digital images of the occlusal surfaces were obtained using a stereomicroscope with X20 magnification (Nikon SMZ1500, Nikon, Tokyo, Japan). Teeth were randomly assigned to two groups: Group A was sealed with the Delton sealant material, and group B was sealed with the UltraSeal XT plus.

Sealant Placement

Selected teeth were randomly assigned to treatment groups. Sealant placement sequence followed a randomized block design. The occlusal surface of each tooth was etched with 35-percent phosphoric acid for 30 seconds, rinsed with an air/water spray for 15 seconds, and then dried with air spray until a matte chalky surface was obtained. For group A (Delton), and for group B (UltraSeal XT Plus), a small drop of sealant was applied to the deepest anatomy of fissure using a scrubbing motion with a micro-brush. Additional sealant material was expressed until the pits and fissures system was covered.

Light Curing

The sealant was light-cured for 30 seconds according to manufacturer's instructions. The curing light used was a Poly-wave LED LCU (VALO Ultradent, South, Utah). A built-in digital radiometer was used to measure the precise output monitoring. Calibration for output was maintained at 450mW/cm$^{3.}$ The light output of the curing light was monitored once for each group of 15 dental sealants/teeth.

Thermocycling

Following sealant placement, specimens were thermocycled for 500 cycles between two water baths with a 40°C temperature differential. An 8°C and 48°C bath was used with a 30-second dwell time and transfer time of 10 seconds[95] (Figure 4).

Specimens Dying and Sectioning

Two layers of nail varnish were applied to non-occlusal surfaces of the teeth. The teeth apices were sealed with wax to avoid the dye going from the apex to the pulp chamber. After this the specimens were immersed in 1.0-percent methylene blue dye at 37°C for 24 hours to allow dye penetration into possible gaps at the tooth-sealant interface. The specimens were cleaned with water. The specimens were allowed to dry for 24 hours before sectioning. The roots were removed by using an Isomet Low Speed Saw (Buehler, Lake Bluff, Illinois, US)(DiamonD Wafering blade, 15.2cm x 0.5 mm). The specimens were mounted on plastic rods. Each tooth was sectioned longitudinally in a bucco-lingual direction through the sealant over the occlusal centered area. Four to five slices were made per tooth (1.5-mm apart), depending on the tooth size. After cutting the specimens, each section was evaluated and assessed; digital images for well-defined fissure systems were obtained and analyzed using a stereomicroscope with X20 magnification (Nikon SMZL1500, Nikon and Tokyo, Japan). The fissure type was correlated with the fissure classification given to the teeth visually and fissures U, V, I, I and inverted Y. Fissure morphology of the samples was studied and then separated into four subgroups based on fissure anatomy; the sample size was 76 per group, 19 per subgroup (Table I).

Micro-leakage/ Dye Penetration Evaluation

Measurements for depth penetration of the sealant material were made perpendicular to the occlusal plane; if the fissure had a different angulation, then the fissure image was adjusted to be on 90-degrees to the occlusal plane and was measured from a 200-μm point at the opening of the fissure. If the fissure was above the angulation threshold, it was adjusted so measurements were made on a 90-degree plane (Figure 5).

The penetration of the sealant material into the pits and fissures was expressed as a percentage of the total length of fissure, according to Bottenberg et al.[22] The fissure depth was measured from the point where the width of the fissure orifice becomes smaller than 200 μm down to the bottom of the fissure. To assess the sealant penetration, the penetration depth was measured from the same point down to the deepest edge of the sealing material.[83]

For the microleakage score, a ranked scale method described by Grande et al. was used.[80] The rank is: (0) no dye penetration; (1) dye penetration into the occlusal third of the enamel-sealant interface;(2) dye penetration into the middle third of the interface; and (3) dye penetration into the apical third of the interface.

STATISTICAL ANALYSIS

The effect of morphology of fissures and viscosity of sealant on penetration and microleakage of sealant was assessed using analysis of variance (ANOVA). The ANOVA included factors for morphology and sealant type, and interactions among factors. The ANOVA test included a random effect for specimen to account for multiple sections analyzed from each specimen. A transformation of the calculated penetration and microleakage percentages was analyzed to satisfy the homogenous variance assumption for the ANOVA: arcsine (percentage $^{1/2}$). Pair-wise comparisons between groups were performed if the overall test for any difference among groups is significant.

Sample Size

Based on the study by Gerwal and Chopra,[21] the within-group standard deviation of the sealant penetration measurements was accepted to be 15 percent. A sample size of 76 per group shall provide 80-percent power to detect a difference of 10 percent for sealant penetration, assuming a two-sided test conducted at a 5-percent significance level. This study had two viscosity levels, and 150 teeth were needed for this study.

RESULTS

MICROLEAKAGE RESULTS

Tables XI and XII show summary data collected for the microleakage scores for each group investigated. The viscosity of the dental sealant and the morphology of fissures did not have significant effect on microleakage ($p = 0.5891$ and $p = 0.4857$). The interaction between the viscosity of the dental sealant studied and the morphology of fissures on extracted teeth was not significant ($p = 0.6657$).

SEALANT PENETRATION RESULTS

Tables XIII, XIV, and XV present the results for the percentage mean of sealant penetration for each group investigated. The viscosity of the dental sealant and the morphology of fissures had a significant effect on sealant penetration ($p < 0.001$). The interaction between the viscosity of the dental sealant and the morphology of fissures was not significant ($p = 0.4236$). The sealant penetration average for the Delton sealant material was significantly greater than the UltraSeal XT Plus ($p < 0.0001$). The sealant penetration average for the fissure I-type was significantly lower than U-fissure type and the V-type ($p < 0.0001$). The sealant penetration for Y-type was significantly lower than the U-type and V-type ($p < 0.0001$).

VISCOSITY OF DENTAL SEALANTS

The viscosity of the dental sealants studied was measured using a Brook's filed viscometer unit. The viscosity is the resistance of a material to flow. Viscosity is measured in units mPa (mega Pascal) per second or cP (centipoise). A high viscous material flows slowly and may cause incomplete penetration.[28] The viscosity value obtained for the Delton Sealant material was 213.16 mPa. s. The viscosity obtained for the UltraSeal XT Plus Sealant material was 2817.37 mPa. s. Water has a viscosity of 1.002 cPa; therefore, the Delton sealant is more viscous than water, and Ultra Seal XT Plus had a much higher value than the Delton Sealant.

FIGURES AND TABLES

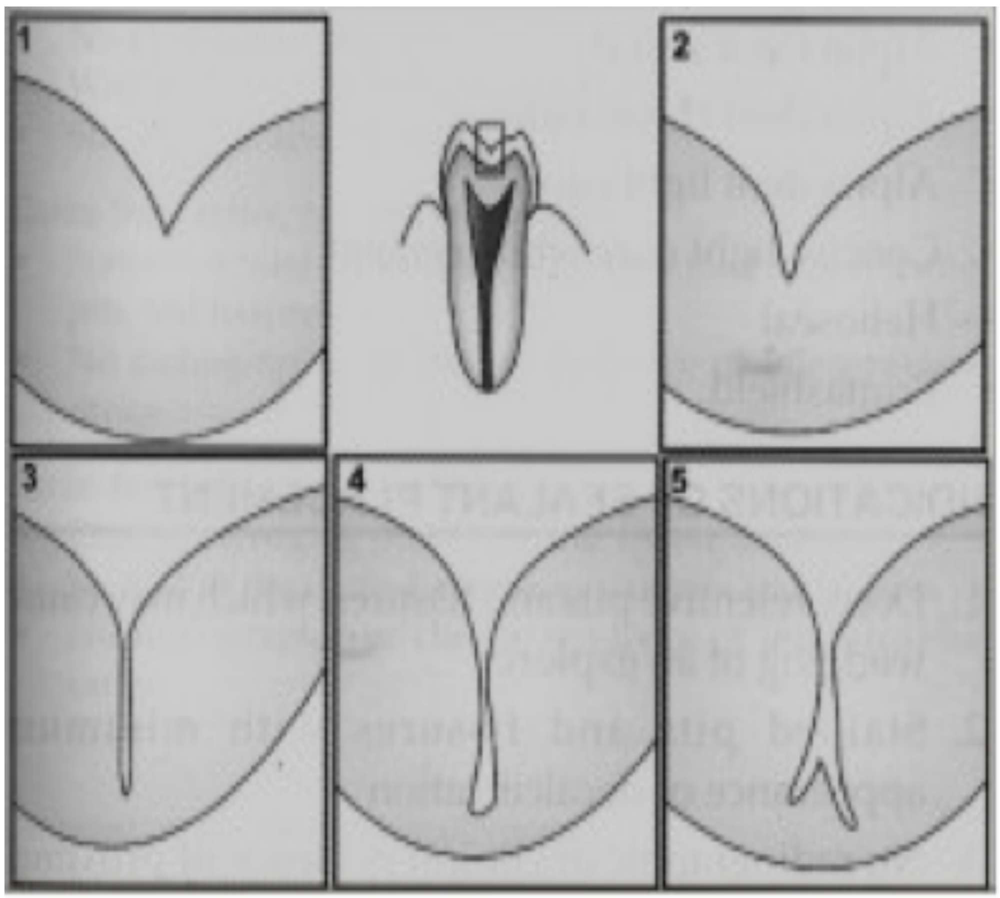

FIGURE 1. Diagram showing pit and fissure morphology: 1:V-type, 2: U type, 3: I type, 4: Y-type. 5: IK-type.

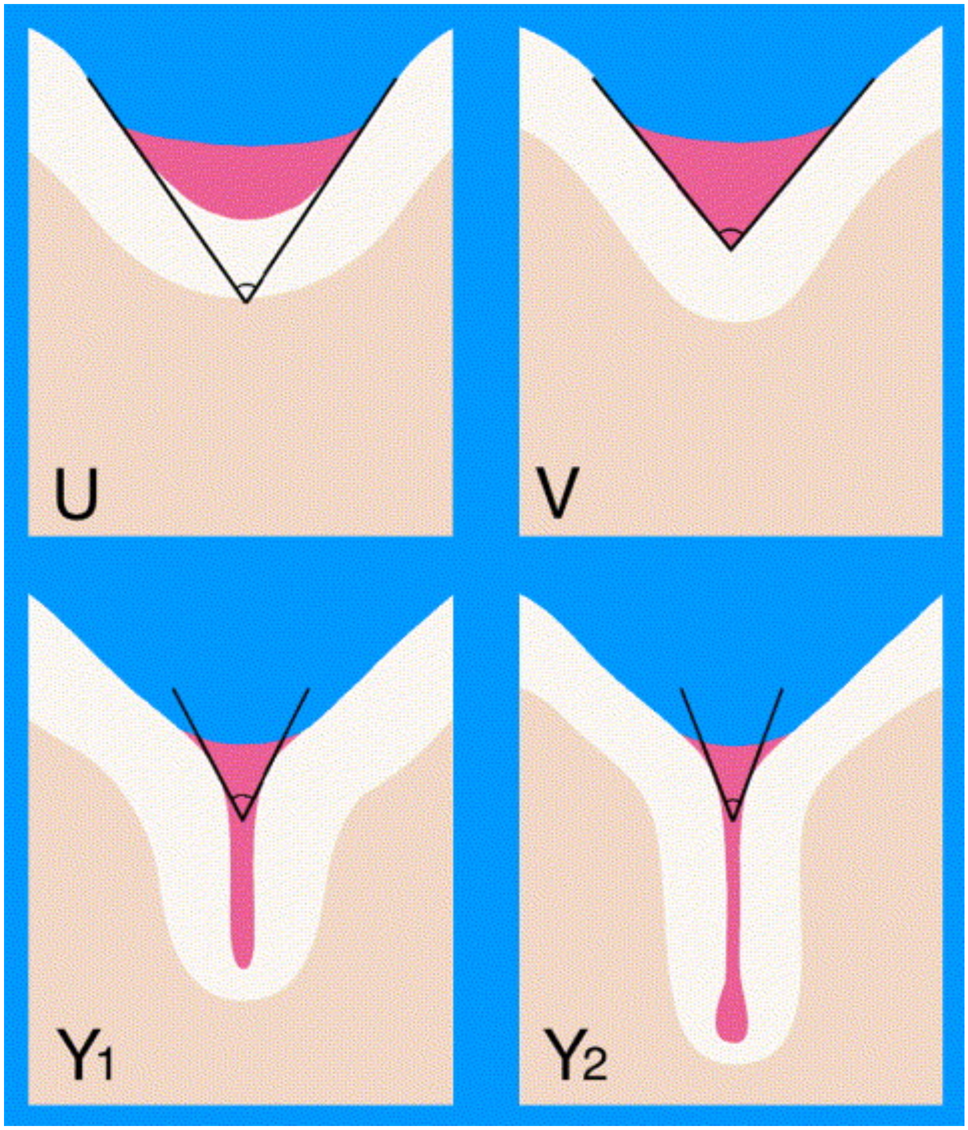

FIGURE 2. Diagram showing pit and fissure morphology Y1 and Y2 shapes.

FIGURE 3. Image of Brook’s field viscometer unit.

FIGURE 4. Image of thermo-cycling unit.

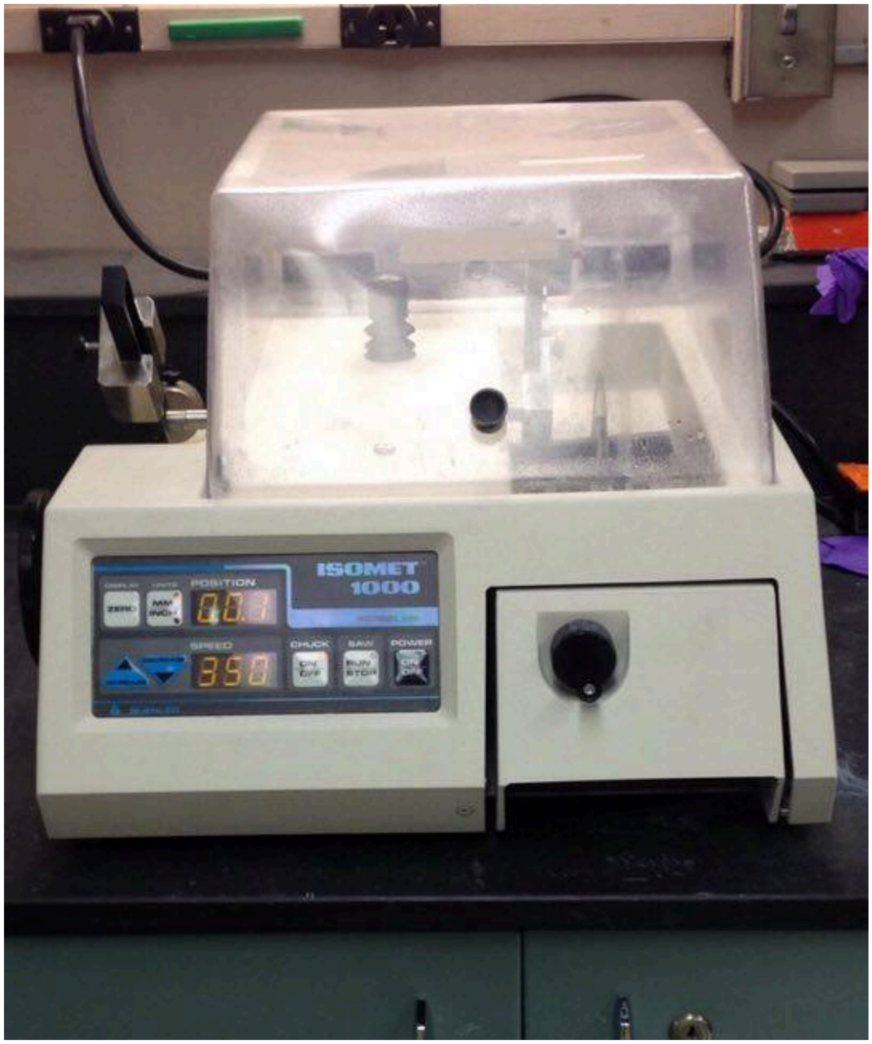

FIGURE 5. Image of Isomet Low Speed Saw unit.

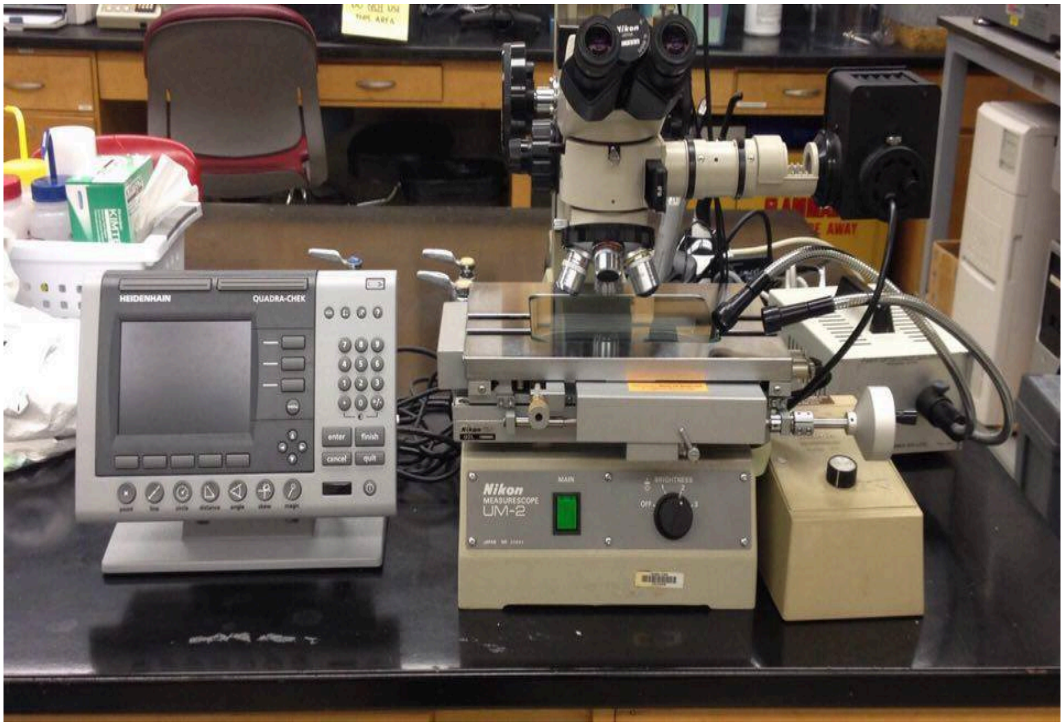

FIGURE 6. Image of Nikon Measure-scope unit.

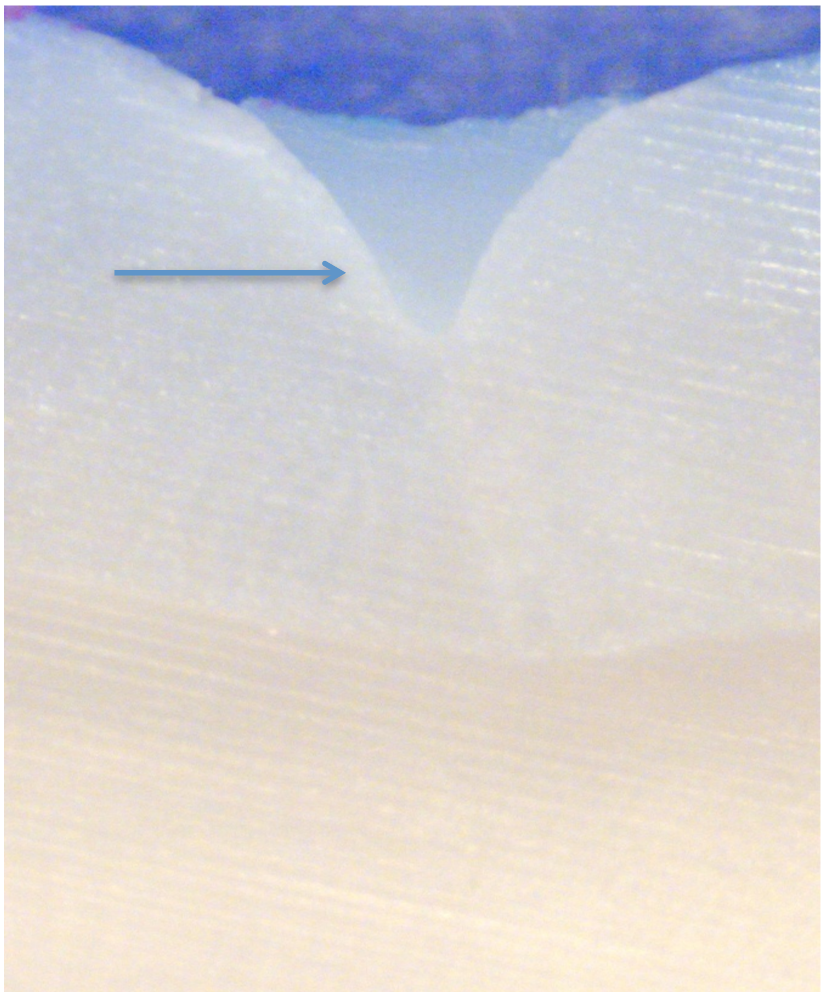

FIGURE 7. Digital image of fissure labeled as V-shape.

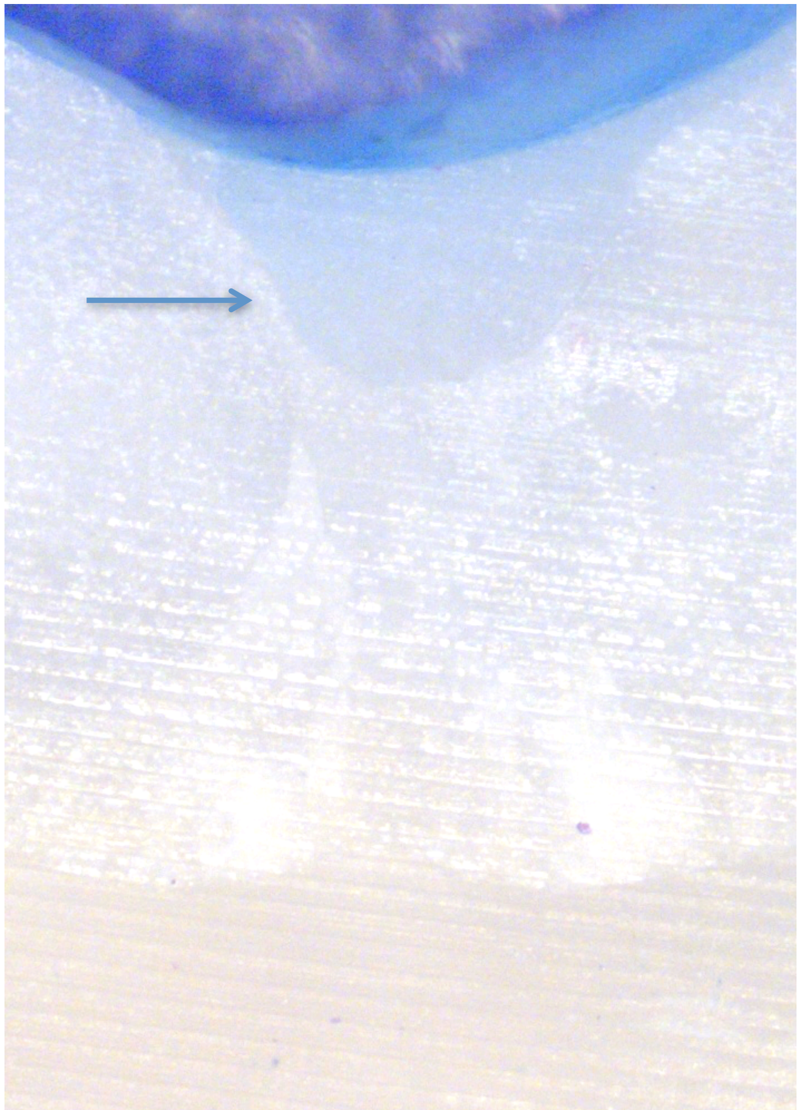

FIGURE 8. Digital image of fissure labeled as U-shape.

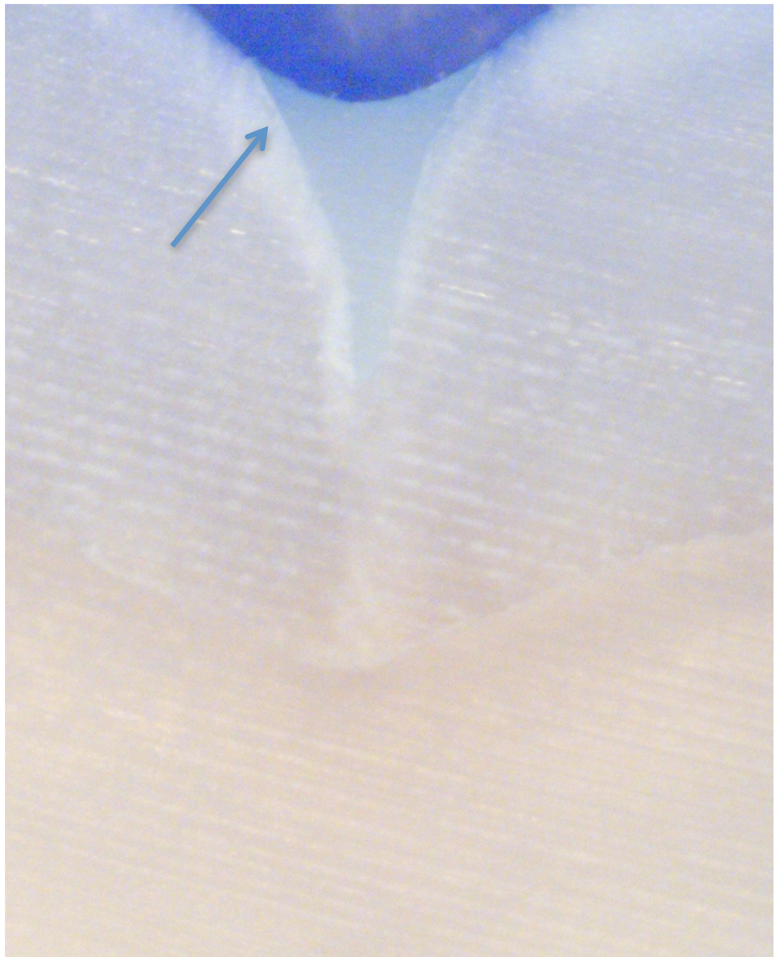

FIGURE 9. Digital image of fissure labeled as Y-shape.

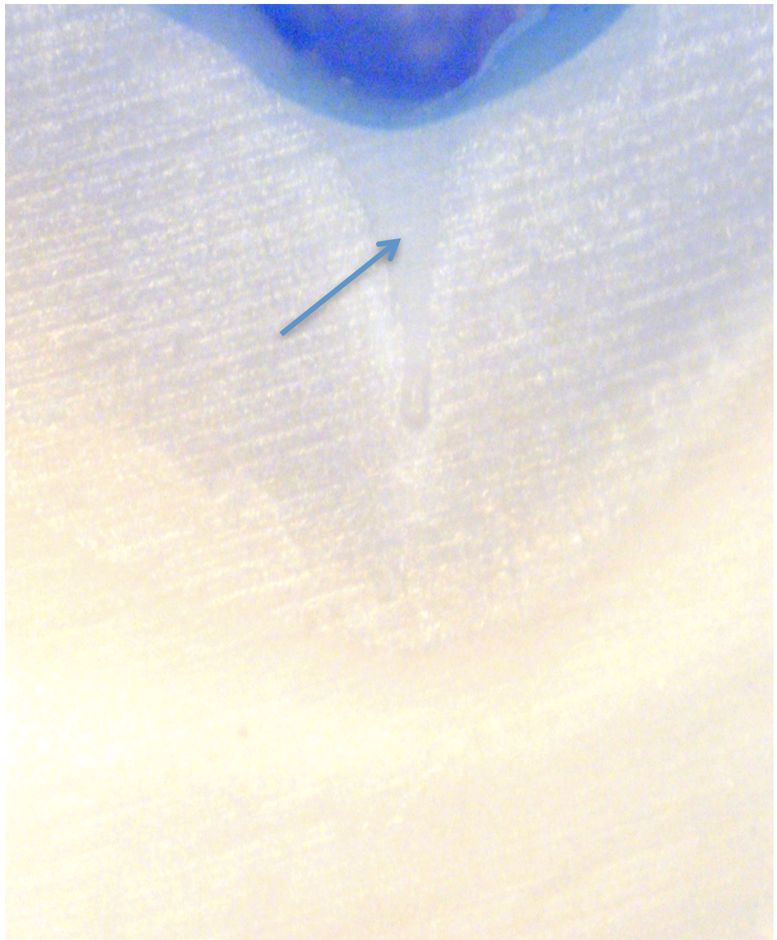

FIGURE 10. Digital image of fissure labeled as I-shape.

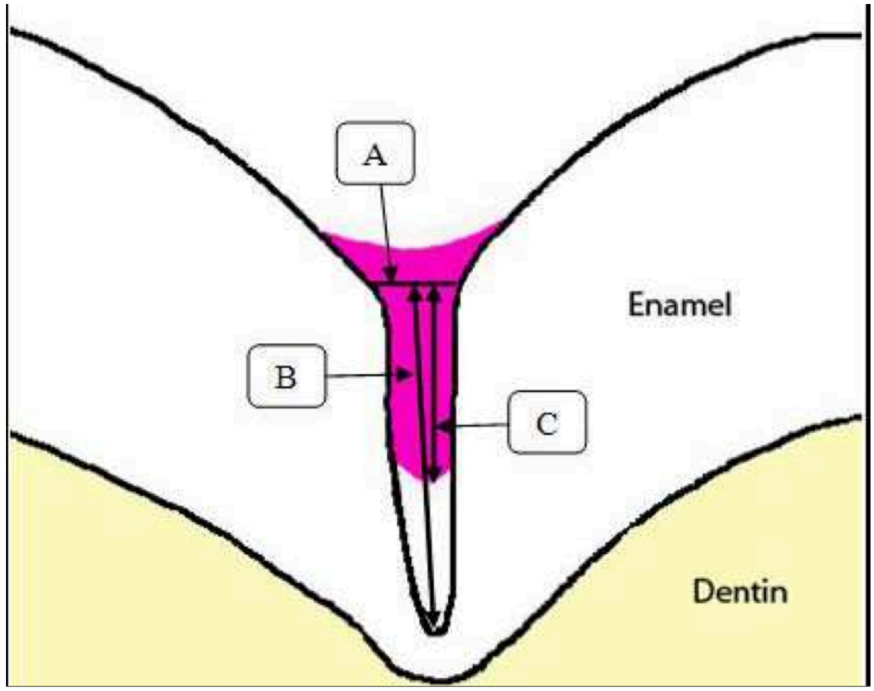

FIGURE 11. Diagram of the method used for quantifying sealant penetration: A) Point where the width of fissure is 200 μ; B) Fissure depth from the width of 200 μM to the base of fissure; C) Sealant penetration. Percent of sealant penetration was calculated using the formula AC/ABx100.

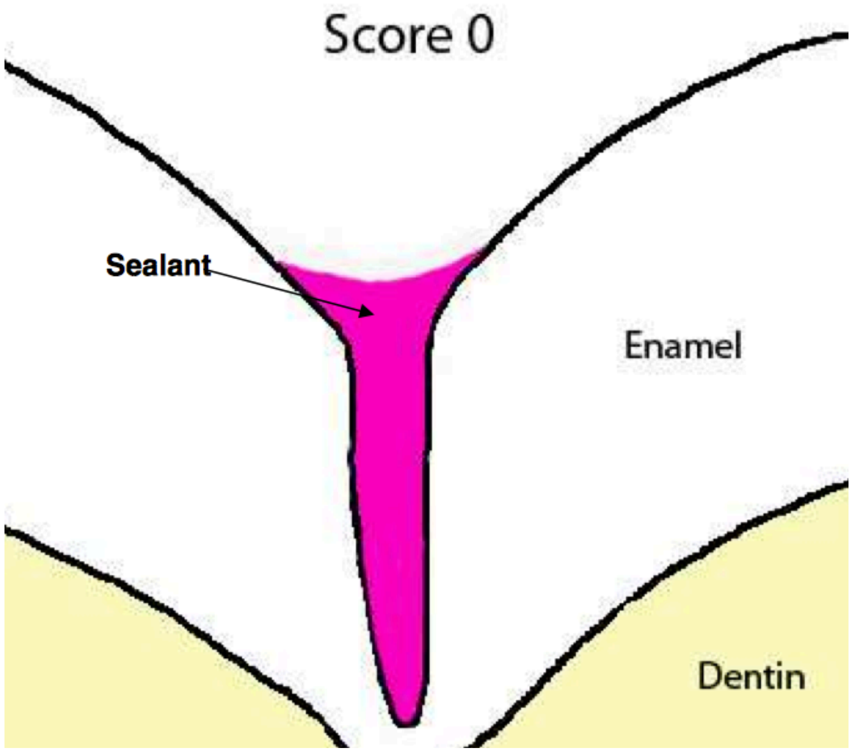

FIGURE 12 . Diagram showing micro-leakage Score 0, and no dye penetration.

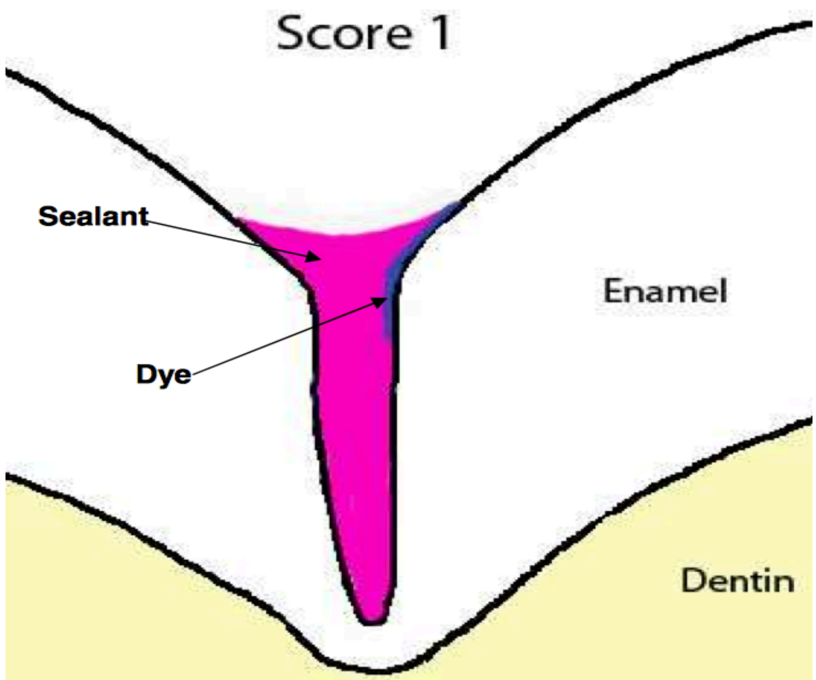

FIGURE 13. Diagram showing microleakage score 1, dye penetration into the occlusal third of enamel-sealant interface.

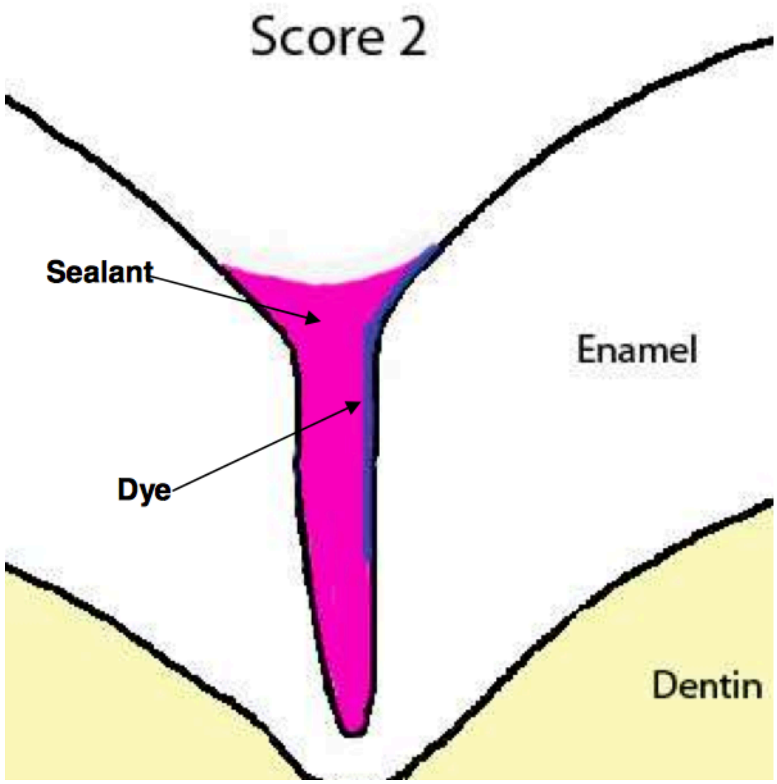

FIGURE 14. Diagram showing microleakage score 2, dye penetration into the middle third of the interface.

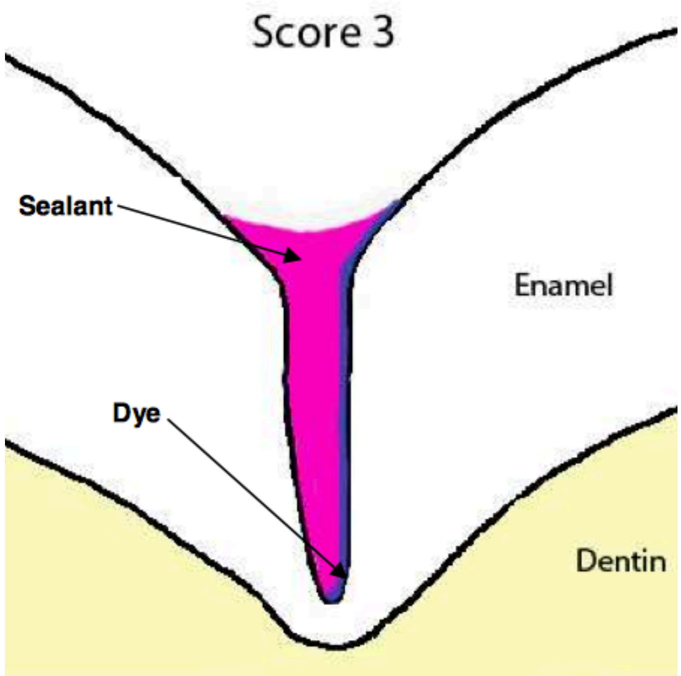

FIGURE 15. Diagram showing microleakage score 3.

FIGURE 16. Digital image of complete fissure penetration of Delton sealant and micro-leakage score 0.

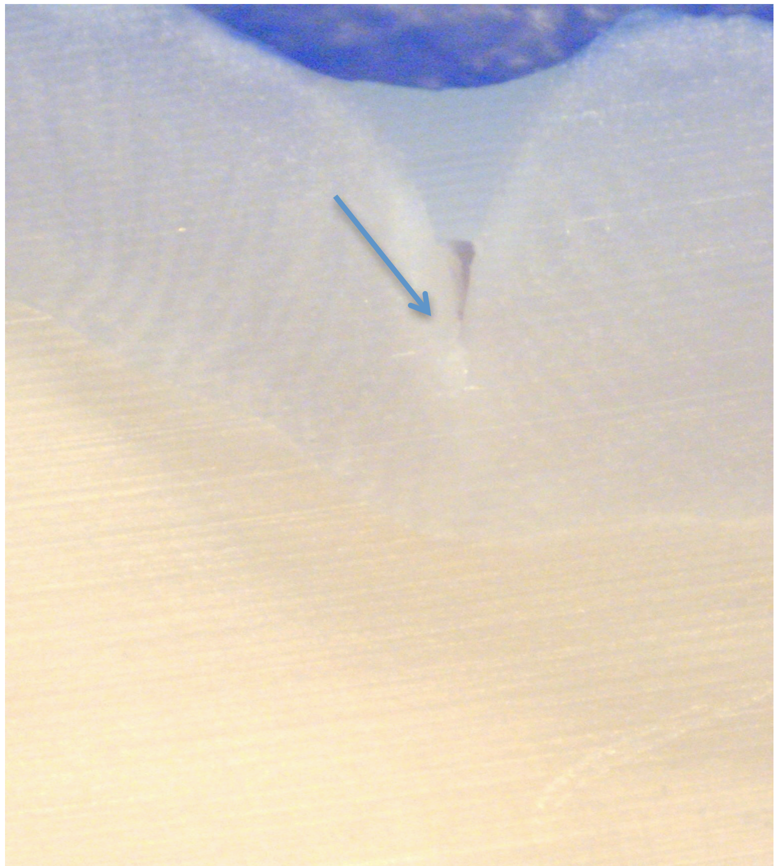

FIGURE 17. Digital image of UltraSeal XT Plus sealant, incomplete penetration and microleakage score 0.

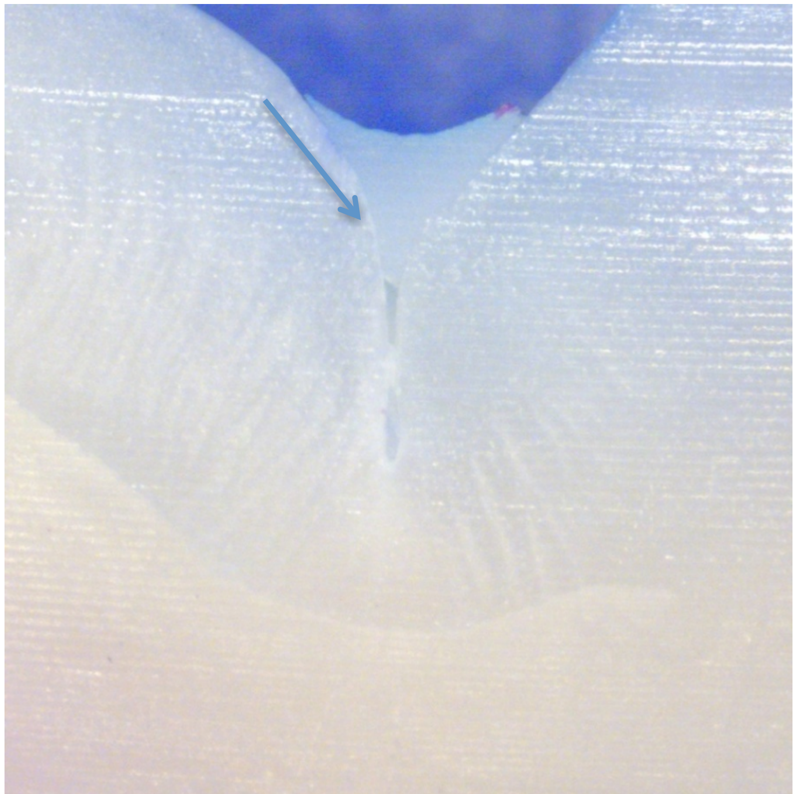

FIGURE 18. Digital image of dye penetration throughout the sealant and micro-leakage score 1 and incomplete sealant penetration.

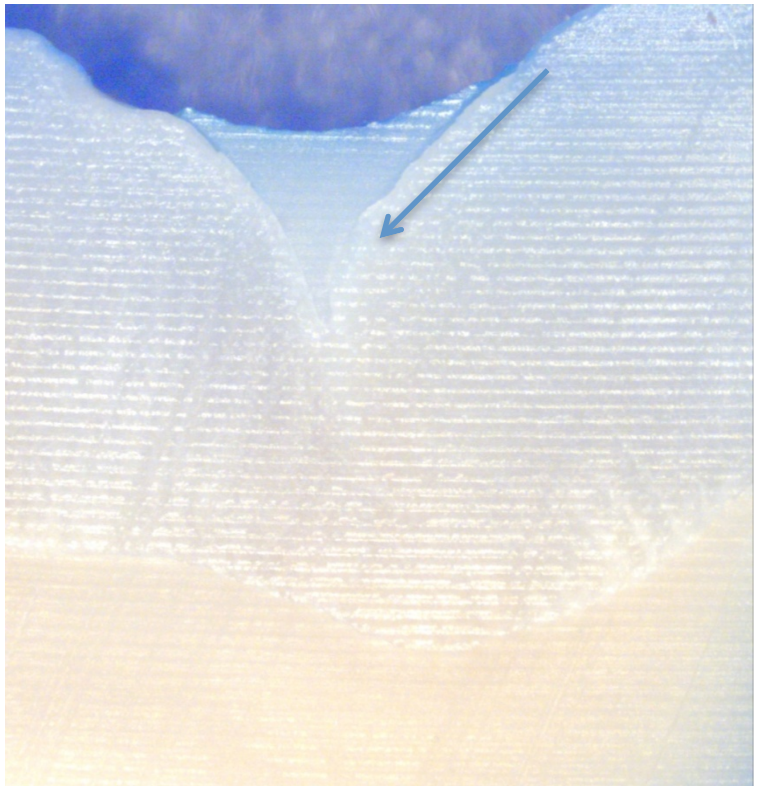

FIGURE 19. Digital image of dye penetration into the middle third of interface and microleakage score 2.

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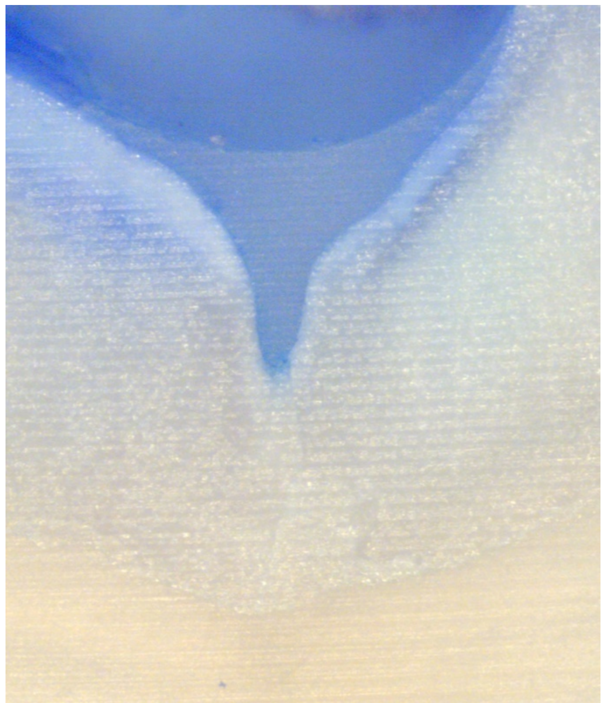
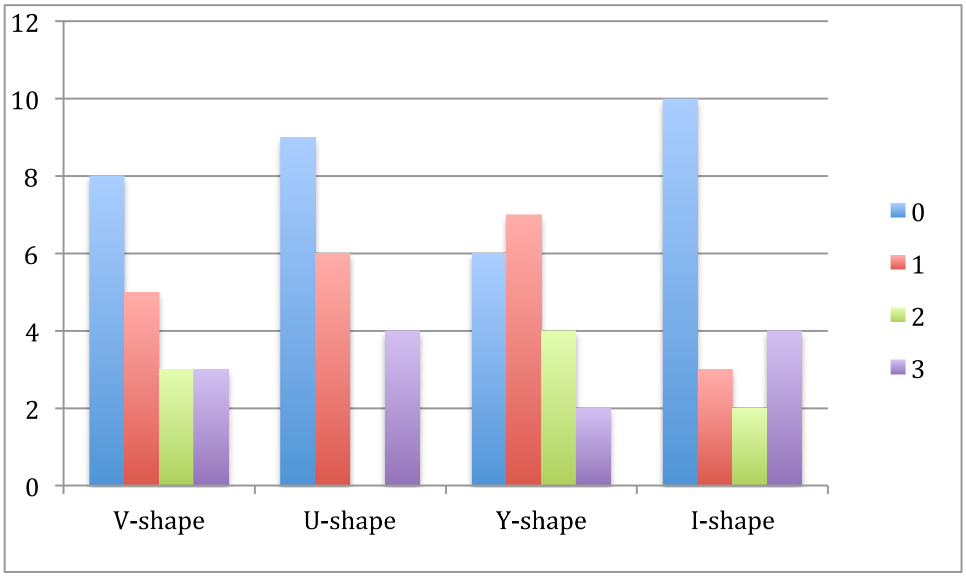
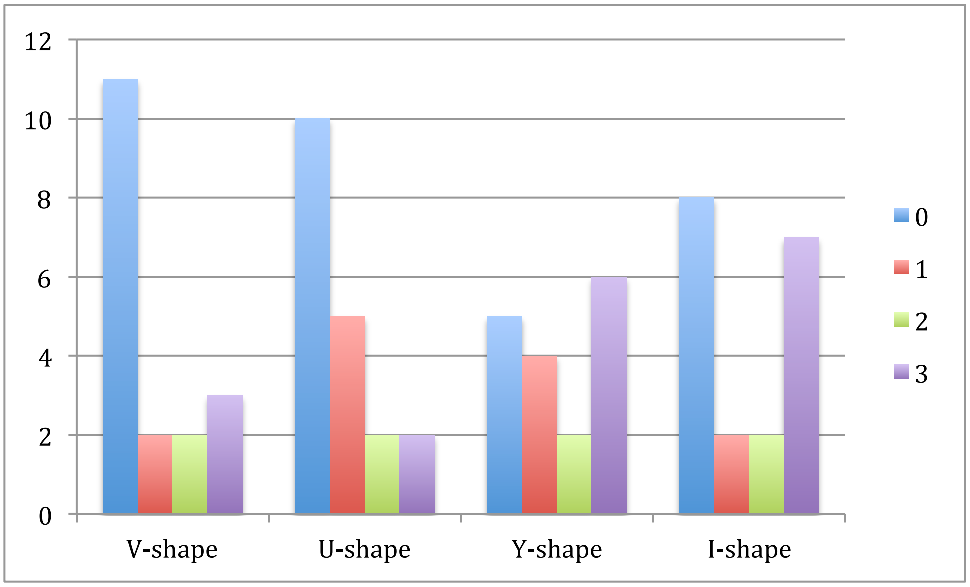


FIGURE 20. Digital image of dye penetration into the apical third of enamel-sealant interface.



Score 0 (no dye penetration)
Score 1 (dye penetration into the occlusal third of enamel-sealant interface)
Score 2 (dye penetration into the middle third of the enamel sealant interface)
Score 3 (dye penetration into the apical third of the interface)

FIGURE 21. Bar graphs showing the distribution of microleakage scores of the different morphology groups presented by number of specimens for each group that were sealed using the Delton sealant.



Score 0 (no dye penetration).
Score 1 (dye penetration into the occlusal third of enamel-sealant interface).
Score 2 (dye penetration into the middle third of the enamel sealant interface).
Score 3 (dye penetration into the apical third of the interface.

FIGURE 22. Bar graphs showing the distribution of micro-leakage scores of the different groups presented by number of specimens for each group that used a UltraSeal XT Plus sealant.

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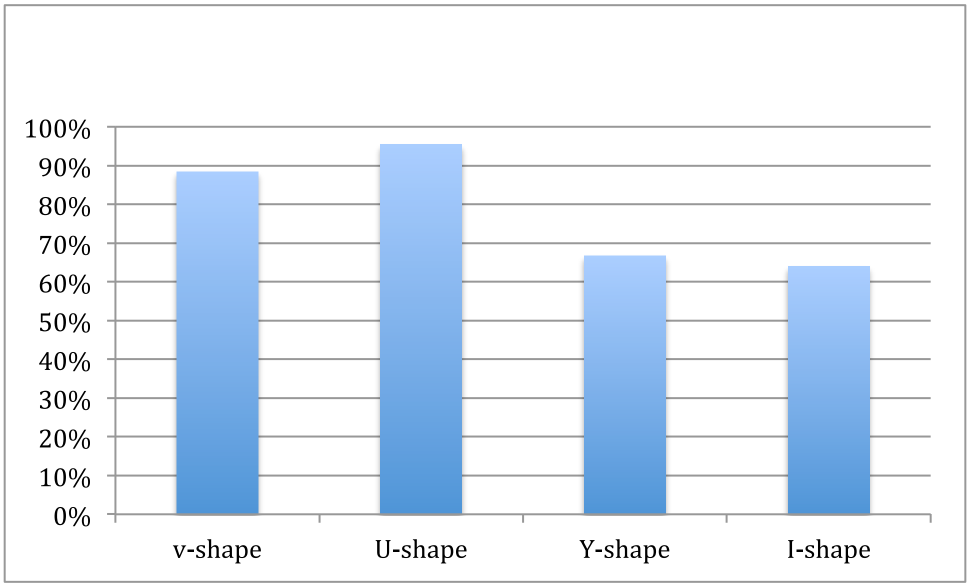

FIGURE 23. Bar graphs showing the average sealant penetration percentage of different groups of pits and fissures morphology sealed with UltraSeal XT Plus sealant material.

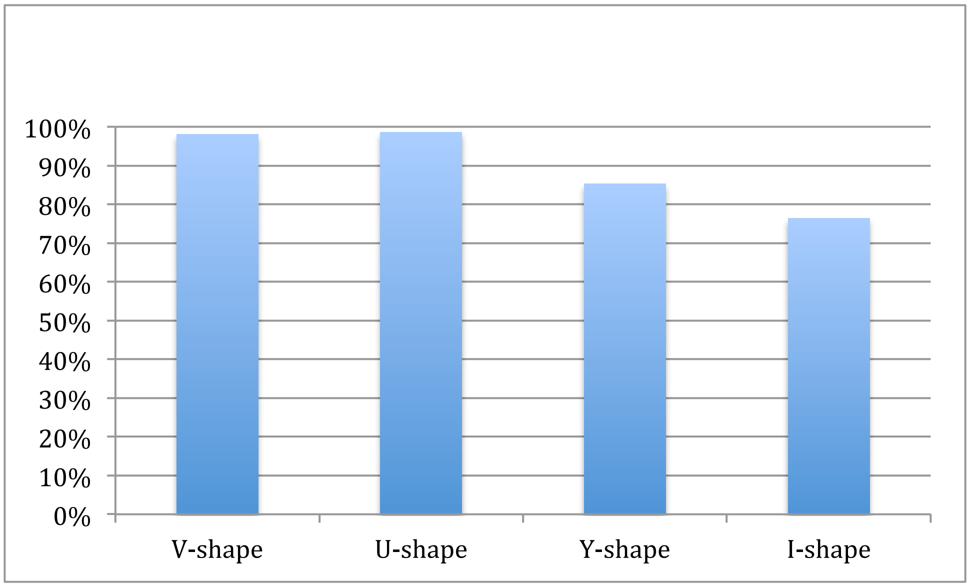

FIGURE 24. Bar graph showing the average of sealant penetration percentage of the different morphology of pits and fissures sealed with Delton sealant material.

TABLE I

Experimental groups description

Group/Sealant	Sub-group	Shape of Fissure	Sample Size (n)
A	I	V-shape	19
A	II	U-shape	19
A	III	I-shape	19
A	IV	Y-shape	19
B	I	V-shape	19
B	II	U-shape	19
B	III	I-shape	19
B	IV	Y-shape	19

TABLE II

Summary of experimental protocol for applying the dental sealant.

Group	Sealant Type*	Conditioning Method	Conditioning Time	Light curing Time
A	Delton	35% Phosphoric acid	15 Sec	30 Sec
B	UltraSeal XT Plus	35% phosphoric acid	15 Sec	30 Sec

*Delton Sealant: Low viscosity
UltraSeal XT Plus: High viscosity

TABLE III

Microleakage scores and penetration percentages*

Sub-Group (Number of Sample)	Microleakage Score	Penetration Percentage
1	0	99.63%
2	0	100%
3	1	100%
4	1	100%
5	3	100%
6	0	100%
7	0	89.14%
8	0	93.28%
9	3	96.71%
10	3	84.55%
11	2	100%
12	1	100%
13	1	100%
14	2	100%
15	0	100%
16	0	100%
17	0	100%
18	1	100%
19	2	100%

1- Score 0 (no dye penetration)

2- Score 1 (dye penetration into the occlusal third of enamel-sealant interface)

3- Score 2(dye penetration into the middle third of the enamel-sealant interface)

4- Score 3 (dye penetration into the apical third of the interface).

*Sealant Type: Delton
Fissure Type: V-type

TABLE IV

Microleakage scores and penetration percentages*

Sub-group (Number of Samples)	Microleakage Score	Penetration Percentage
1	0	100%
2	0	100%
3	1	100%
4	0	100%
5	1	85.71%
6	1	100%
7	0	100%
8	1	100%
9	3	100%
10	0	100%
11	3	100%
12	0	100%
13	0	100%
14	3	100%
15	1	88.69%
16	1	100%
17	0	100%
18	0	100%
19	3	100%

*Sealant Type: Delton
Fissure Type: U-Type

1- Score 0 (no dye penetration).
2- Score 1 (dye penetration into the occlusal third of enamel-sealant interface).
3- Score 2(dye penetration into the middle third of the enamel-sealant interface).
4- Score 3 (dye penetration into the apical third of the interface).
5- Sealant Type: Delton.
6- Fissure Type: U-Type.

TABLE V

Microleakage scores and penetration percentages*

Sub-groups (Number of Samples)	Microleakage Score	Penetration Percentage
1	0	82.45%
2	3	100%
3	3	93.86%
4	2	56.92%
5	0	87.78%
6	1	88.66%
7	1	81.81%
8	1	69.27%
9	0	100%
10	1	100%
11	1	100%
12	0	100%
13	2	100%
14	1	56.08%
15	0	81.77%
16	1	100%
17	2	64.29%
18	2	77.03%
19	0	79.63%

*Sealant Type: Delton
Fissure Type: Y-Type

Score 0 (no dye penetration).
Score 1 (dye penetration into the occlusal third of enamel-sealant interface).
Score 2(dye penetration into the middle third of the enamel-sealant interface).
Score 3 (dye penetration into the apical third of the interface).

TABLE VI

Microleakage scores and penetration percentages*

Sub-group (Number of Sample)	Microleakage Score	Penetration Percentage
1	0	47.59%
2	1	76.74%
3	3	89.31%
4	3	68.14%
5	0	66.18%
6	2	72.93%
7	3	100%
8	2	64.95%
9	3	100%
10	0	44.19%
11	1	61.16%
12	0	100%
13	1	65.63%
14	0	84.64%
15	0	87.55%
16	0	76.15%
17	0	61.90%
18	0	100%
19	0	84.66%

*Sealant Type: Delton

Fissure Type: I-Type

Score 0 (no dye penetration).
Score 1 (dye penetration into the occlusal third of enamel-sealant interface).
Score 2 (dye penetration into the middle third of the enamel-sealant interface).
Score 3 (dye penetration into the apical third of the interface).

TABLE VII

Microleakage scores and penetration percentages

Sub-group (Number of Samples)	Microleakage Score	Penetration percentage
1	0	100%
2	0	100%
3	1	100%
4	0	100%
5	1	88.77%
6	2	100%
7	0	100%
8	0	100%
9	1	100%
10	0	100%
11	0	100%
12	0	100%
13	2	100%
14	3	100%
15	3	84.49%
16	0	62.67%
17	0	90.70%
18	1	94.85%
19	1	91.86%

*Sealant Type: UltraSeal XT Plus

Fissure Type: U-Type

Score 0 (no dye penetration).
Score 1 (dye penetration into the occlusal third of enamel-sealant interface).
Score 2(dye penetration into the middle third of the enamel-sealant interface).
Score 3 (dye penetration into the apical third of the interface).

TABLE VIII

Microleakage scores and penetration percentages

Sub-group (Number of Sample)	Microleakage Score	Penetration Percentage
1	0	100%
2	0	77.59%
3	0	69.55%
4	0	84.32%
5	0	100%
6	0	100%
7	2	42.96%
8	3	83.05%
9	3	100%
10	3	100%
11	0	91.49%
12	0	100%
13	1	100%
14	3	100%
15	0	83.76%
16	1	100%
17	0	69.04%
18	2	91.66%
19	0	87.12%

Sealant Type: UltraSeal XT Plus
Fissure Type: V-Type

Score 0 (no dye penetration).
Score 1 (dye penetration into the occlusal third of enamel-sealant interface).
Score 2(dye penetration into the middle third of the enamel-sealant interface).
Score 3 (dye penetration into the apical third of the interface.

TABLE IX

Microleakage scores and penetration percentages

Sub-group (Number of Sample)	Microleakage Score	Penetration Percentage
1	0	66.41%
2	1	37.31%
3	2	84.68%
4	0	82.05%
5	0	100%
6	3	59.46%
7	3	86.64%
8	3	95.55%
9	2	78.75%
10	0	100%
11	1	50%
12	3	96.29%
13	3	63.88%
14	0	17.45%
15	0	30.45%
16	3	44.49%
17	1	56.43%
18	0	60.51%
19	1	66.40%

Sealant Type: UltraSeal XT Plus
Fissure Type: Y-Type

Score 0 (no dye penetration).
Score 1 (dye penetration into the occlusal third of enamel-sealant interface).
Score 2(dye penetration into the middle third of the enamel-sealant interface).
Score 3 (dye penetration into the apical third of the interface).

TABLE X

Microleakage scores and penetration percentages

Sub-group (Number of Samples)	Microleakage Score	Penetration Percentage
1	0	63.16%
2	3	65.19%
3	2	100%
4	3	42.83%
5	3	40.91%
6	3	55.73%
7	1	71.84%
8	3	37.87%
9	1	71.92%
10	0	87.91%
11	0	70.73%
12	0	75.04%
13	0	71.88%
14	3	48.11%
15	0	40.94%
16	0	65.54%
17	0	42.99%
18	2	73.03%
19	3	90.44%

Sealant Type: UltraSeal XT Plus
Fissure Type: I –Type

Score 0 (no dye penetration).
Score 1 (dye penetration into the occlusal third of enamel-sealant interface).
Score 2 (dye penetration into the middle third of the enamel-sealant interface).
Score 3 (dye penetration into the apical third of the interface.).

TABLE XI

Statistical analysis for microleakage showing p-value

Effect	p-value
Viscosity of Sealant	0.5891
Shape of Fissures	0.4857
Viscosity* Shape	0.6657

*. The interaction between the sealant viscosity and shape of fissure.

TABLE XII

Results of statistical analysis for micro-leakage showing means of study groups

Sealant	Type	N	Mean
Delton	I-_type	19	1.0
	Y-type	19	1.1
	U-type	19	0.9
	V-type	19	1.1
UltraSeal XT Plus	I-type	19	1.4
	Y-type	19	1.4
	U-type	19	0.8
	V-type	19	0.9

TABLE XIII

Statistical analysis for sealant penetration showing p-value

Effect	p-value
Sealant Viscosity	<0.0001
Type of fissure	<0.0001
Sealant*type	0.4236

* The interaction between the sealant viscosity and fissure type.

TABLE XIV

P-values for comparisons among groups

Comparison Group	Results	p-value
Sealant Viscosity	Delton > UltraSeal XT Plus	<0.0001
Type of fissures	I-_type & Y-_type	0.0821
Type of fissures	I-type < U-type	<0.0001
Type of fissures	I-type < V-type	<0.0001
Type of fissures	Y-type < U-type	<0.0001
Type of fissures	Y-type < V-type	<0.0001
Type of fissures	U-type & V-type	0.1452

TABLE XV

Summary of statistical analysis for sealant penetration resulted from the interaction between study variables

Sealant	Type	N	Mean
Delton	I-type	19	76.4
	Y-type	19	85.2
	U-type	19	98.7
	V-type	19	98.1
Ultra Seal XT Plus	I-type	19	64.0
	Y-type	19	66.7
	U-type	19	95.4
	V-type	19	88.4

DISCUSSION

JUSTIFICATIONS FOR EXPERIMENTAL PARAMETERS

The purpose of this *in-vitro* study was to assess the influence of different sealant viscosities and various types of pit and fissure morphologies on sealant penetration and microleakage. To investigate the study question, two different types of sealants with different viscosities were used. The Delton sealant material was considered the low viscosity (213,16 mPa. S), while the UltraSeal XT Plus sealant material as the high viscosity (2817,37 mPa. S). One hundred and fifty teeth were selected and separated visually into three subgroups based on fissure anatomy. Group one V-fissure shape (n = 19); Group two U-fissure shape (n = 19), and Group three for convenience had fissure types I and inverted Y (n = 38). Dental sealants were placed according to manufacturers' instructions. Specimens were thermocycled for 500 cycles between two water baths having a 40°C temperature differential. [95] All specimens were immersed in 1-percent methylene blue dye at 37°C for 24 hours. Specimens were sectioned, photographed, and analyzed. The efficiency of sealant viscosity and pit and fissure morphology on penetration and microleakage were analyzed using two-way ANOVA. According to microleakage evaluation, we used a ranked scale described by Grande et al. For the sealant penetration measurement, we used a percent of sealant penetration.[77]

The success of dental sealants depends on several factors; among those are the effect of sealant viscosity and the pit and fissure morphology.

THE EFFECT OF PITS AND FISSURES MORPHOLOGY

The morphology of pit and fissure and the viscosity of the dental sealant material influenced the ability of sealant to penetrate. In this study, the fissure morphology did not have a significant effect on the microleakage ($p = 0.4857$), and nor was there a significant interaction between the morphology and the sealant viscosity ($p = 0.6657$). These results agree with previous findings of Selectman et al. study, which found no significant differences between fissure types with the microleakage.[20]

The present study showed that the morphology of pit and fissures had a significant effect on sealant penetration ($p < 0.001$), in which the sealant penetration averages for the U- and V-type fissures were higher than the Y- and I-type fissures. From our study findings, it can be assumed that the viscosity of the sealant material and the morphology of pit and fissure may influence the ability of sealant penetration. In their *in-vitro* studies, Selectman et al. and Grewal and Chopra et al. reported that their sealant material had a higher penetration in shallow pit and fissures in comparison with the lower penetration into deep pit and fissures.[20,21,34] Finally, on scanning electron microscope (SEM) studies by Durmusoglu et al.[96] and Ramya et al.[97] reported that the U-type fissures showed the greatest mean percentage penetration. These findings are in agreement with our *in-vitro* study results.

The present *in-vitro* study showed that the sealant penetration for I-type fissures was significantly lower than U- and V-type fissures; the sealant penetration for Y-type was significantly lower than U- and V-type fissures. This finding is somewhat similar to the study by Selectman et al., who reported that morphology was not a significant factor regarding microleakage, but that it had significant impact on sealant penetrability. With

U-type fissures displaying the greatest value of penetrability, no correlation was found between the extents of microleakage with the sealant penetrability.[20]

Grewal et al. reported that fissure shape was highly significant for sealant penetration; V- and U-shaped fissures were found to have the greatest penetration while IK types and I types showed the lowest penetration of the dental sealant.[21] The results were in agreement with our present study. From another study, statistically significant differences between the fissure morphology and the depth of sealant penetration were reported by Ramya et al. Their results indicated that the U-fissure type showed the highest mean percentage penetration, and the poorest percentage penetration was reported for the IK- type fissure.[97]

In a study by Nicola et al., teeth were sealed with three types of dental sealants: Admira, Fotoseal, and Fuji. Teeth were thermocycled for 1000 cyles. This study concluded that pit morphology plays a role in the depth penetration of sealants; again, the U type showed better penetration compared with fissures V-shape, Y, and IK-shape.[98] This may be because shallow pit and fissure systems are more likely to be completely clean from any residual impurities that influence sealant penetration into the base of pit and fissure.[21,27] On the contrary, deep pit and fissure systems may not be completely clean and dry prior to sealant penetration.[21,27]

THE EFFECT OF SEALANT VISCOSITY

In the present study, we analyzed the relation among dental sealant viscosity and pit and fissure morphology and the ability of the sealant penetration. The results of the present study showed that the low-viscosity sealant (Delton) had a greater percentage of penetration than the high-viscosity sealant (UltraSeal XT Plus). On the other hand, the findings showed that the viscosity of the dental sealant did not have a significant effect on microleakage (p = 0.5891). Nevertheless, the interaction between the viscosity of sealant and the pit and fissure morphology was not significant in this study.

Furthermore, Mehrabkhani et al.[65] found no significant differences between the viscosity of the dental sealant and the microleakage scores. The results of their study cannot be compared with the present study, due to their use of a bonding agent, which indicated a different sealant placement protocol.

Parbhakar et al.[28] concluded that the low viscosity sealant penetrated fully into the etched enamel surface; nevertheless, the high viscosity sealant could not penetrate enough to ensure good marginal depth seal. [28] Irinoda et al. reported that the low viscosity sealant Teethmate penetrated more in comparison with the high viscosity sealant (Prisma-Shield and Concise White Sealant).[66] In addition, Prabhakar et al. documented that the low-viscosity sealant spread more rapidly over the surface and penetrated better than the high-viscosity sealant.[28] This may cause a poor adaptation and incomplete penetration to the bottom and then affect their retention adversely.[28] These studies cannot be compared with ours as they studied the penetrability of sealants into etched enamel and measured tags using SEM.[28,65,66]

Contrary to our results, some studies reported that the viscosity of dental sealant does not affect the penetration ability of sealant.[99] Barnes et al. used five commercial light-cured fissure sealants to evaluate the relation among viscosity, sealant penetrability and leakage.[99] Study results may be in disagreement with our present study as they found that the viscosity had an effect on the penetration but no effect on sealing ability.[99]

In this study, we attempted to reproduce clinical conditions; we used human permanent molars and mimicked the thermal changes that occur in the oral cavity from eating, drinking, and breathing by thermocycling the specimens.[95] Furthermore, many different methods have been used to measure microleakage through assessing the dye penetration along the tooth's structure-dental restoration interface in *in-vitro* studies. The majority of *in-vitro* studies used ranked scale methods described by Grande et al.[72, 80] In the present *in-vitro* study to evaluate sealant penetration we recorded penetration as a percentage of sealant depth.[22,83]

In summary, the present findings suggest that the pit and fissures morphology and dental viscosity might have an effect on sealant penetration, but do not effect micro-leakage. The low viscosity dental sealant (Delton) exhibited better penetration than the high-viscosity dental sealant (UltraSeal XT Plus) and U- and V-type fissures had a statistically greater penetrability than Y-shape and I-shape fissure types.

SUMMARY AND CONCLUSION

Dental caries is considered a major public health issue especially for children. Subsequently, it should receive significant attention, not only from the standpoint of restorative procedures, but also in terms of preventive measures designed to reduce caries. Pit and fissure sealants have been found not only to prevent occlusal caries, but also to arrest the progression of incipient lesions. The advantages of sealing the occlusal surfaces are associated with the decrease in caries prevalence when compared with non-sealed teeth. Moreover, sealing is associated with lower cost when compared with the cost of placing restorations.

The retention of sealant and the ability to seal are important factors in the success of sealants in preventing dental caries. Indeed, the longer the material remains bonded to the occlusal surface, the greater the protective benefit it provides to the teeth. In addition, the success of preventing leakage between the sealant and the enamel tooth surface is considered to be an important feature of the success of fissure sealants.

The purpose of this *in-vitro* study was to evaluate if the pit morphology and sealant viscosity could affect sealant penetration and the microleakage.

Within the parameters of this *in-vitro* study the following conclusion can be drawn:

The viscosity of the dental sealant and the morphology of pits and fissures had a significant effect on sealant penetration ability.

The sealant penetration for the low viscosity sealant (Delton) was significantly greater than the high-viscosity (UltraSeal XT plus) dental sealant.

The sealant penetration ability into I-type fissures was significantly less than for U and V types (both sealants).

The sealant penetration for Y-type fissures was significantly less than for fissures U and V types (both sealants).

REFERENCES

1. Eckert GJ, Jackson R, Fontana M. Sociodemographic variation of caries risk factors in toddlers and caregivers. Int J Dent 2010.

2. Li Y, Caufield PW. The fidelity of initial acquisition of mutans streptococci by infants from their mothers. J Dent Res 1995;74(2):681-5.

3. Griffin SO, Oong E, Kohn W, et al. The effectiveness of sealants in managing caries lesions. J Dent Res 2008;87:169-74.

4. Beauchamp J, Caufield PW, Crall JJ, et al. Evidence-based clinical recommendations for the use of pit-and-fissure sealants: a report of the American Dental Association Council on Scientific Affairs. J Am Dent Assoc 2008;139(3):257-68.

5. Ahovuo-Saloranta A, Forss H, Walsh T, et al. Sealants for preventing dental decay in the permanent teeth. Cochrane database of systematic reviews 2013;3,1-139.

6. Robert J F. Sealants and preventive restorations: review of effect iveness and clinical changes for improvement. Pediatr Dent 1998;20(2):85-92.

7. Grande RH, Reis A, Loguercio AD, Singer Jda M, Shellard E, Neto PC. Adhesive systems used for sealing contaminated surfaces: a microleakage evaluation. Brazil Oral Res 2005;19(1):17-22.

8. Mertz-Fairhurst EJ, Schuster GS, Fairhurst CW. Arresting caries by sealants: results of a clinical study. J Am Dent Assoc 1986;112(2):194-7.

9. Pinkham J Casamassimo P. Pediatric dentistry. Infancy through adolescence. 4th ed. New Delhi:Saunders; 2005.

10. Babu.G, Mallikarjun S,Wilson B, Permkumar C. Pit and fissure sealants in pediatric dentistry. SRM J Res Dent Sci 2014;5(4):253-7.

1. Donly KJ, Garcia-Godoy F. The use of resin-based composite in children: an update. Pediatr Dent 2015;37(2):136-43.

12. Cohen L, Sheiham A. Importance of variables affecting pit and fissure sealant use in the United Kingdom. Community Dent Oral Epidemiol 1988;16(6):317-20.

13. Kervanto-Seppala S, Pietila I, Meurman JH, Kerosuo E. Pit and fissure sealants in dental public health -- application criteria and general policy in Finland. BMC Oral Health 2009;9:5.

14. Lussi A. Validity of diagnostic and treatment decisions of fissure caries. Caries Res 1991;25(4):296-303.

15. McLean JW Wilson AD. Fissure sealing and filling with an adhesive glass ionomer cement. Br Dent J 1974;25(4):269-76.

16. Siegal M. Promotion and use of pit and fissure sealants: an introduction to the special issue. J Public Health Dent 1995;55 (5 Spec No):259-60.

17. Muller-Bolla M, Lupi-Pegurier L, Tardieu C, Velly AM, Antomarchi C. Retention of resin-based pit and fissure sealants: A systematic review. Community Dent Oral Epidemiol 2006;34:321-36.

18. Aguilar FG, Drubi-Filho B, Casemiro LA, Watanabe MG, Pires-de-Souza FC. Retention and penetration of a conventional resin-based sealant and a photochromatic flowable composite resin placed on occlusal pits and fissures. J Indian Soc Pedod Prev Dent 2007;25(4):169-73.

19. Nagano T. Relation between the form of pit and fissure and the primary lesion of caries. Dent Abstr 1961;6:426.

20. Selecman JB, Owens BM, Johnson WW. Effect of preparation technique, fissure morphology, and material characteristics on the in vitro margin permeability and penetrability of pit and fissure sealants. Pediatr Dent 2007;29(4):308-14.

21. Grewal N, Chopra R. The effect of fissure morphology and eruption time on penetration and adaptation of pit and fissure sealants: An SEM study. J Indian Soc Pedod Prev Dent 2008;26(2):59-63.

22. Bottenberg P, Graber HG, Lampert F. Penetration of etching agents and its influence on sealer penetration into fissures in vitro. Dent Mater 1996;12(2):96-102.

23. Larson TD . The clinical significance and managment of microleakage (Pt1). Northwest Dent 2005;84(1):23-5,28-9,31.

24. Miller-Keane encyclopedia and dictionary of medicine N, and allied health. 7th ed., 2003.

25. Trowbridge HO. Model systems for determining biologic effects of microleakage Oper Dent 1987;12(4):164-72.

26. Ganesh M Shobha T. Comparative evaluation of the marginal sealing ability of Fuji VII and Concise as pit and fissure sealants. J Contemp Dent Pract. 2007;8(4):10-8.

27. Symons AL Chu CY, Meyers IA. The effect of fissure morphology and pretreatment of the enamel surface on penetration and adhesion of fissure sealants. J Oral Rehabil 1996;23(12):791-8.

28. Prabhakar AR Murthy SA, Sugandhan S. Comparative evaluation of the length of resin tags, viscosity and microleakage of pit and fissure sealants – An in vitro scanning electron microscope study. Contemp Clin Dent 2011;2:324-30.

29. Duangthip D Lussi A. Variables contributing to the quality of fissure sealants used by general dental practitioners. Oper Dent 2003;28 (6):756-64.

30. McDonald SP, Sheiham A. The distribution of caries on different tooth surfaces at varying levels of caries--a compilation of data from 18 previous studies. Community Dent Health 1992;9(1):39-48.

31. Schour I Massler M. Studies in tooth development: The growth pattern of human teeth. J Am Dent Assoc 1940; 27:1918.

32. Disney JA Graves RC, Stamm JW, Bohannan HM, Abernathy JR, Zack DD. The University of North Carolina Caries Risk Assessment study: future developments in caries risk prediction. Community Dent Oral Epidemiol 1992;20(2):64-75.

33. Feigal RJ. Donly KJ. The use of pit and fissure sealants. Pediatr Dent 2006;28(2):143-50.

34. Mejare I Kallestal C, Stenlund H, Johansson H.Caries Development from 11 to 22 years of age: a prospective radiographic study. Prevalence and distribution. Caries Res 1998;32(1):10-6.

35. Ripa LW. Sealants revisited: an update of the effectiveness of pit-and-fissure sealants. Caries Res 1993;27 (Suppl 1):77-82.

36. Simonsen. RJ Pit and fissure sealants. In: Clinical application of acid etch technique. 1st ed. Chicago: Quintessence Publishing Co., Inc. 1978: 19-42.

37. Harris N, Garcia-Godoy F. Primary preventive dentistry. 6th edition. New Jersey, 2004.

38. Simonsen RJ. Pit and fissure sealant. Review of literature. Pediatr Dent 2002; 24: 393-414.

39. Oong EM, Griffin SO, Kohn WG, Gooch BF, Caufield PW. The effect of dental sealants on bacteria levels in caries lesions: a review of the evidence. J Am Dent Assoc 2008;139(3):271-8;quiz 357-8.

40. Welbury R Raadal M, Lygidakis N, . Guidelines on the use of pit and fissure Sealants in pediatric dentistry. An EAPD policy document. Eur J Pediar Dent. 2004;3:179-84.

41. Avinash J, Marya CM, Dhingra S, et al. Pit and fissure sealants: an unused caries prevention tool. J Oral Health Community Dent 2010;4(1):1-6.

42. Mertz-Fairhurst EJ, Fairhurst CW, Williams JE, Della Giustina VE, Brooks JD. A comparative clinical study of two pit and fissure sealants: 7-years results in Augusta, Georgia. J Am Dent Assoc 1984;109(2):252-5.

43. Romcke RG,Lewis DW, Maze BD, Vickerson RA. Retention and maintenance of fissure sealants over 10 years. J Can Dent Assoc 1990;56(3):235-7.

44. Pinkham J Casamassimo P. Pediatric dentistry. infancy through adolescence. 4th ed. New Delhi: Saunders; 2005.

45. Adair SM. The role of sealants in caries prevention programs. J Calif Dent Assoc 2003;31(3):221-7.

46. Darby, Walsh. Dental hygiene theory and practice. 2nd ed. Saunders,2003.

47. Cueto EL, Buonocore MG. Sealing pits and fissures with an adhesive resin. Its use in caries prevention. J Am Dent Assoc 1967;75(1):121-8.

48. Varun S, Aswini YB. Missed, concealed and obscured aspect of caries prevention-legacy for the future. J Dent Sci 2011;3(2):44-9.

49. Zero DT. How the introduction of acid-etch technique revolutionized dnetal practice. J Am Dent Assoc 2013;144(9):990-4.

50. Gwinnett AJ Buonocore MG. Adhesive and caries prevention. A preliminary report. Br Dent J 1965;119:77-80.

51. Buonocore MG. A simple method of increasing the adhesion of acrylic filling materials to enamel surfaces. J Dent Res 1955;34:849-53.

52. Peutzfeldt A. Resin Composites in dentistry: the monomer systems. Eur J Oral Sci 1997;105(2):97-116.

53. Silverstone LM.. State of the art on sealant research and priorities for further research. J Dent Educ 1984; 48 (2 Suppl):107-18.

54. Smith DC. A new dental cement. Br Dent J 1968;124(9):381-4.

55. Welbury R Raadal M, Lygidakis NA,. Guidlelines on the use of pit and fissures sealants in pediatric dentistry. An EAPD policy document. Eur J Pediar Dent. 2004;5(3):179-84.

56. Lindemeyer RG . The use of glass ionomer sealants on newly erupting permanent molars. J Can Dent Assoc 2007;73(2):131-4.

57. Weintraub JA. The effectivness of pit and fissure sealants. J Public Health Dent 1989;49(5 Spec No):317-30.

58. Eronat N Bardakci Y, Sipahi M. Effect of different preparation techniques on microleakage of compomer and resin fissure sealants. J Dent Child (Chi) 2003;70:250-3.

59. Arrow P Riordan PJ. Retention and caries preventive effects of a GIC and a resin-based fissure sealants. Community dent oral epidemiol 1995;23(5):282-5.

60. Beiruti N Frencken jE, van't Hof MA, Taifour D, Van Palentein Helderman WH. Caries-preventive effect of a one-time application of composite resin and glass ionomer sealants after 5 years. Caries Res 2006;40(1):52-9.

61. Komatsu H Shimokobe H, Kawakami S, Yoshimura M. Caries-preventive effect of glass ionomer sealant reapplication: study presents three-year results. J Am Dent Assoc 1994;125(5):543-9.

62. Rubenstein LK, Dinius A. Dental sealant usage in Virginia. J Public Health Dent 1986(46):147-51.

63. Dabir S, Viswanath T, Ghosh K, et al. Viscosity of liquids: theory, estimation, experiment, and data. Dordrecht, The Netherlands: Springer, 2010.

64. Yamada Y Hossain M, Shimizu Y, et al. Analysis of surface roughness and microleakage of fissure sealants following organic debris removal with Carisolv. J Dent 2008;36:130-7.

65. Maryam M, Mazhari F, Sadeghi S, Ebrahimi M. Effects of sealant, viscosity, and bonding agents on microleakage of fissure sealants: An in vitro study. Eur J Dent. 2015;9(4):558-63.

66. Irinoda Y, Matsumura Y, Kito H, et al. Effect of sealant viscosity on the penetration of resin into etched human enamel. Oper Dent 2000;25:274-82. .

67. Dukic W Dukic OL, Milardovic S. The influence of Healozone on microleakage and fissure penetration of different sealing materials. Coll Antropol 2009;33:157-62.

68. Papacchini F Cury AH, Goracci C, et al. Noninvasive pit and fissure sealing: Microtensile bond strength to intact bovine enamel of different pit and fissure sealants in a simplified fissure model. J Adhes Dent 2006;8:375-80.

69. Birkenfeld LH, Schulman A. Enhanced retention of glass-ionomer sealant etching: a microleakage and scanning electron microscopic study. Quintessence Int 1999;30(10):712-8.

70. perez-Lajarin L Cortes-Lillo O, Garcia-Ballesta C, Cozar-Hidalgo A. Marginal micro-leakage of two fissure sealants: a comparative study. J Dent Child 2003;70(1):24-8.

71. Manhart J Huth KC, Chen HY, Hickel R. Influence of the pretreatment of occlusal pits and fissures on the retention of a fissure sealant. Am J Dent 2004;17(1):12-8.

72. Hatibovic-Kofman S Wright GZ, Braverman I. Microleakage of sealants after conventional, bur, and air-abrasion preparation of pits and fissures. Pediatr Dent 1998;20(3):173-6.

73. Youssef MN Youssef FA, Souza-Zaroni WC, Turbino ML, Vieira MM. Effect of enamel preparation method on in vitro marginal microleakage of flowable composite used as pit and fissure sealant. Int J Pediatr Dent 2006;16(5):342-7.

74. Swartz ML Phillips Rw. In vitro studies on marginal leakage of restorative materials. J Am Dent Assoc 1961(62):141-51.

75. Theodoridou-Pahini S Tolidis K, Papadogiannis Y. Degree of microleakage of some pit and fissure sealants: an in vitro study. Int J Pediatr Dent 1996;6:173-6.

76. Borsatto MC, Corona SA, Dibb RG, Ramos RP, Pecora JD. Microleakage of a resin sealant after acid-etching, Er: YAG laser irradiation and air-abrasion of pits and fissures. J Clin Laser Med Surg 2001;19(2):83-7.

77. Montanari M Pitzolu G, Felline C, Piana G. Marginal seal evaluation of different resin sealants used in pits and fissures. An in vitro study. Eur J Paediatr Dent 2008;9(3):125-31.

78. Keyur J, Bhavna D, Niyanta J, Rajashekhara B, Jobanputra L, Khushbu Y. Comparative evaluation of two different pit and fissure sealants and a restorative material to check their microleakage. An in vitro study. J Int Oral Health 2013; 5(4):35-9.

79. Cooley R, James W, Huddleston H, Camedes P. Evaluation of fluoride containing sealant by SEM, microleakage and fluoride release Pediatr Dent 1990;12:38-42.

80. Grande RH, Ballester R, Singer Jda M, Santos JF. MPicroleakage of universal adhesive used as a fissure sealant. Am J Dent 1998;11 (3):109-13.

81. O'Brien WJ. Capillary action around dental structures. J Dent Res 1973;52(3):544-50.

82. Moore BK, Ewoldsen N, Winkler MM. Laboratory testing of light-cured glass ionomers as pit and fissure sealants. Gen Dent 1995;43(2):176-80.

83. Kersten S, Lutz F, Schupbach P. Fissure sealing: Optimization of sealant penetation and sealing properties. Am J Dent 2001;14(3):127-31.

84. Hosoya Y Garcia-Godoy F, Summitt JB. Microleakage and sealant penetration using a vibrating probe. Am J Dent 2004;17(6):427-32.

85. Hevinga MA Opdam NJ, Frencken JE, Bronkhorst EM, Truin GJ. Microleakage and sealant penetration in contaminated carious fissures. J Dent 2007;35(12):909-14.

86. Celiberti P Lussi A. Penetration ability and microleakage of fissure sealant applied on artificial and natural enamel fissure caries. J Dent 2007;35(1):59-67.

87. Dos Santos PA, Garcia PP, Palma-Dibb RG. Shear bond strenght of adhesive systems to enamel and dentin. Thermocycling influnce. J Mater Sci Mater Med. 2005;16(8):727-32.

88. Gale MS, Darvell BW. Thermal cycling. Procedures for laboratory testing of dental restorations. J Dent 1999;27(2):89-99.

89. Craig R G, Powers J. Restorative dental materials. 11^{th} ed. St. Louis:Mosby, 2001.

90. Souder WH, Peters GC. Circular of the National Bureau of Standards C433: Physical properties of dental materials. Washington: U.S. Department of Commerce, National Bureau of Standards, 1942.

91. De Munck J, Van Landuyt K, Peumans M, et al. A critical review of the durability of adhesion to tooth tissue: methods and results. J Dent Res 2005;84(2):118-32.

92. Amaral FL, Colucci V, Palma-Dibb RG, Corona SA. Assessment of in vitro methods used to promote adhesive interface degradation: a critical review. J Esthet Restor Dent 2007; 19(6):340-53; discussion 54.

93. ISO. Guidance on testing of adhesion to tooth structure. International Organization for Standardization. TR 11405,1-14. Geneva (Switzerland). 1994.

94. Chu G, Halloran J. High-temperature flow behavior of ceramic suspensions. J Am Dent Assoc 2000;83(9):2189-95.

95. Palasuk J, Platt JA, Cho SD, Levon JA, Brown DT, Hovijitra S. Effect of surface treatments on microtensile bond strength of repaired aged silorane resin composite. Oper Dent 2013;38(1):91-9.

96. Durmusoglu O, Tagtekin DA, Ozyoney G, Bozzkurt FO, Yanikoglu FC. Light microscopy and SEM evaluation of three different fissure sealant.OHDMBSC 2005;4(4);26-32

97. Iyer RR, Gopalakrishnapillai AC, Kalantharakth T. Comparisons of in-vitro penetration and adaptation of moisture tolerant resin sealant and conventional resin sealant in different fissure types. Chin J Dent Res 2013;16(2):127-36

98. Nicola C, Muntean A, Bondor C, et al. Influnce of pit morphology on sealants penetration depth, 2010 (abstract).

99 Barnes DM Kihn P, Von Fraunhofer JA, Elsabach A. Flow characteristics and sealing ability of fissure sealants. Oper Dent 2000;25:306-10.

ABSTRACT

THE EFFECT OF PIT AND FISSURE MORPHOLOGY AND SEALANT VISCOSITY ON SEALANT PENETRATION AND MICROLEAKAGE.

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Background: The ability of sealants to prevent caries is directly related to the sealant being retained in teeth. The longer the material remains bonded to the occlusal surface, the more protection it provides to the tooth.

Objective: The aim of this *in-vitro* study was to evaluate the influence of pit and fissure morphologies and sealant viscosity on sealant penetration and micro-leakage.

Study Hypothesis: The low viscosity dental sealant will express better penetration ability and less microleakage in permanent molars with any pit and fissure morphology than the high viscosity sealant.

Material and methods: Permanent extracted molars (n = 150) were distributed into two groups based on two types of sealant (high and low viscosity) Permanent extracted molars (n = 150) were selected using the International Caries Detection Assessment system (ICDAS) criteria 0-1. Teeth were stored in 0.1-percent thymol and distilled water. Teeth were assigned to three subgroups according to the fissure's morphology. Enamel was etched with 35-percent phosphoric acid for 30 seconds; two different light cured sealants were placed, Group A: Delton and Group B: Ultra X Plus. Specimens were thermocycled for 500 cycles between two water baths, having a 40°C temperature differential (4°C to 48°C). Teeth were coated with nail varnish and wax, except in the occlusal areas. All specimens were immersed in 1-percent methylene blue dye at 37°C for 24 hours. Specimens were sectioned longitudinally in a bucco-lingual direction, and the sections were photographed and analyzed by a previously trained examiner for fissure morphology, sealant penetration, and microleakage using a standardized grading system. Data were entered and statistically analyzed, at the 5-percent significance level.

Results: Viscosity of sealant and morphology of fissures had significant effects on sealant penetration ($p < 0.001$). The interaction between viscosity of sealant and morphology of fissures was not significant ($p = 0.4236$). The sealant penetration for Delton was significantly higher than the UltraSeal XT Plus ($p < 0.0001$). The sealant penetration for fissure I-type was significantly lower than fissures U and V-types ($p < 0.0001$). Sealant penetration for Y-type was significantly lower than U and V-types (p <

0.0001). However, the viscosity of sealant and morphology of fissures did not have significant effect on microleakage ($p = 0.5891$ and $p = 0.4857$). The interaction between the viscosity of the sealant material and the morphology of pit and fissures was not significant ($p = 0.6657$).

Conclusion: The results of the present study indicated the viscosity of the sealant and the morphology did not affect the microleakage. On the other hand, the viscosity of sealant affected the penetration ability of dental sealant. The low viscosity dental sealant (Delton) exhibited a better penetration than the high viscosity sealant (UltraSeal XT Plus). As the morphology of pit and fissure directly affected the penetration ability, the fissures types U and V exhibited a better penetration than fissure types Y and I.

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