Two Adhesive Systems-effect on Adhesion to Tooth Structure

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This study evaluates and compares by dye penetration method and SEM photomicrographs the sealing obtained using two different classes of adhesive systems (etch-and-rinse and self-etch with selective etching) with SDR (Dentsply) bulk fill composite. 84 class V cavities were prepared on oral and vestibular face of 42 intact, freshly extracted wisdom teeth. The cavities were randomly divided in two groups and restored: Group 1 with prime&bond one select (Dentsply) and SDR (Dentsply) and Group 2 with prime&bond one Etch&Rinse (Dentsply) and SDR (Dentsply). Prime&bond one Select (Dentsply) is a single component adhesive and can be used in self etch mode, in selective enamel etch mode, or in etch-and-rinse mode. We chosen for this study the selective etch of the enamel mode. Prime&bond one Etch Rinse (Dentsply) is a universal etch-and-rinse one-bottle dental adhesive, designed to be used in two steps. The bulk fill composites are commonly used in modern dentistry due to their properties of low polymerization shrinkage and curing in layer of 4 mm depth, offering the practitioner a fast clinical procedure with good results. The results showed a good sealing at enamel and dentin margins with no statistically significant difference between adhesives, even though the mean of enamel infiltration was smaller for Group1. Furthermore the results show that there were differences between the two groups, for the infiltrations at the enamel, the values of microleakage being arithmetically higher for Group 1, but with no statistically difference between the two groups.SEM images showed for both groups a good adhesion surface with the tooth, but the hybrid layer of the total-etch adhesives is different from the hybrid layer formed by self etch adhesives, in terms of thickness, uniformity. In conclusion both adhesive systems have equivalent sealing qualities and can be successfully used with SDR.

Keywords: adhesive systems, polymerization shrinkage, infiltrations

Resin based composites are universally used for all types of cavities in anterior and posterior teeth. They are considered to be the main option of countless dentists in direct restoration of carious lesions, because of their advantages as high strength, low thermal conductivity, modulus elasticity, hardness [1, 2]. However, the drawbacks of this material are well known among the practitioners. Two of the principal limitations of composites are the cure depth and polymerization shrinkage. Consequently it is suggested the use of a layering technique, in which the layers should not exceed 2.5 mm, in order to obtain an appropriate polymerization [3]. Nowadays dentists use materials that decrease the chair time, reduce microleakage and increase patients compliance. In literature microleakage is defined as the clinically undetectable passage of bacteria, fluids, molecules or ions between a cavity wall and the restorative material [4-7]. Therefore the bulk-fill composites have been developed, in order to minimize the time of work required by the incremental techniques and decrease micro-leakage [2, 4].

Bulk-fill composites can be applied in layers of 4-5 mm, because of their high reactivity to light curing and reduced polymerization stress. They have larger filler size, less filler content, grater translucency than conventional composites and excellent bond strength regardless of the method of filling and cavity form. Depending on their consistence, there are available two types of bulk-fill composites: flowable and regular. With the regular type one can restore the whole cavity, while the flowable type must be completed by a 1.5 mm layer of conventional composite, in order to increase the aesthetics and the resistance to masticator forces [1, 4, 8].

The shrinkage that accompanies the polymerization of the composite resins causes stress at the interface toothrestoration that can lead to microleakage, secondary or recurrent caries and pulpal irritation [3, 10, 11]. The resulted stress should be absorbed by the adhesive systems. The development of adhesive materials provided an important step in restorative dentistry, that led to what is called today, minimal invasive dentistry [9, 12, 13]. Over the years dental adhesives have been classified in numerous ways, based on generations, on the components, on the number of steps or on the clinical strategy. Contemporary classifications depend on the clinical approach and divide adhesive systems into two groups: *etch-and-rinse adhesives* and *self-etch adhesives* [13-15].

Etch-and-rinse adhesive systems have two versions used in practice: in three steps and in two steps. In the three steps version, the primary components (etchant, primer and bonding) are packaged, each one, in different bottles and the application is made in sequences, whereas the two steps version is a simplified version, that combines two of the components (primer and bonding) in one bottle,

All authors have participated equally in developing this study.

but the etching component is still applied in an independent phase [13, 15]. Disregarding the number of steps etchand-rinse adhesives are generally accepted as being the golden standard in dental adhesion [13, 15, 16]. Moreover etch-and-rinse systems have an excellent adhesion to enamel because of the etching with the phosphoric acid, that creates a favorable micromechanical interlocking (macro and micro resin tags) and convenient physical properties for the adhesion to this dental tissue [9, 15]. However the complicated protocol of adhesion is susceptible to mistakes and there is the peril of dehydrating too much the demineralised dentin and generating the collapse of collagen. This can lead after a period of time to serious clinical consequences as adhesion failure or postoperative sensitivity [9].

Self-etch adhesives systems appeared in order to overcome the drawbacks of the etch-and-rinse systems [15]. These types of adhesives shorten the protocol and make it more intelligible. Distinctive from the etch-andrinse systems, self-etch systems do not use a separate acid to do the conditioning of dental tissues, which is made by acidic monomers contained in the adhesive solution [13]. Regrettably self-etch adhesives used in a single step manner offer lower bond strength than the other systems available on the market and also poor marginal adaptation to enamel [9, 13]. The version of selective etching of enamel, when using a self-etch system, can enhance its adhesive properties, by creating a retentive morphology of the enamel, but a great care should be taken in not etching the dentin also and ruining bonding effectiveness to dentin [9]

The aim of this study is to asses and compare the sealing obtained for the two different classes of adhesive systems (etch-and-rinse in two steps and self-etch in one step used with selective etching) used with SDR bulk fill composite in order to obtain information that will help the practitioners choosing the adhesive system.

Experimental part

Materials and methods

42 freshly extracted human maxillary and mandibular wisdom teeth, intact, with no decays, were included in the study. The teeth were cleaned from all the calculus, osseous tissue and residual soft tissue, using a scaler. They were stored in distilled water. Standard class V cavities were prepared on vestibular and oral faces of each tooth with a width of 4mm, height of 3mm and depth of 1.5 mm having the occlusal margin in enamel and the cervical margin in cement (dentin). All cavities were prepared using a0.10 round diamond bur(Komet) for turbine and a 0.10 inverted cone carbide bur(Komet) for dental clinic Contra Angle Handpiece.

The cavities were randomly divided into 2 groups:

-Group 1- restored with SDR using prime&bond one Select;

-Group 2- restored with SDR using prime&bond one Etch&Rinse Universal.

The protocol used for Group 1 included etching the enamel for 15 seconds using the 37% phosphoric acid then rinse vigorously for 15 s and gently air-dried for 5 seconds in order to remove the excess water. Next step involves applying the adhesive on the entire surface of the cavity using a microbrush for 20 s then thoroughly drying for 5 s to evaporate the solvent according to the manufacturers recommendations (table 1), followed by light-curing for 10 s using LEDEX WL-090+-DENTMATE. SDR composite was the injected in the cavities using a gun applier and light-cured for 20 s. Then the restoration was finished and





Fig. 1. Schematic view of the tooth preparation

Fig. 2. Optical microscopy of buccolinguallyslice with two restorations

polished using paper discs (Kerr; OptiDisc, Medium 40µm, Fine 20µm) and rubber points (Enhance, Dentsply).

The protocol used for Group 2 included etching the enamel for 30 s and the dentin for 15 s. Then it was rinsed thoroughly for 15 s and gently air-dried for 5 s, to dry the dentin, taking care not to over dry it. The adhesive was applied next, for 20 s with a microbrush, on the whole surface of the cavity, then the solvent was evaporated by thoroughly air drying for 5 seconds, as the manufacturer recommended (Table 1), followed by light-curing for 10 s using LEDEX WL-090+-DENTMATE. After this the application of the restoration material was similar to that realized for Group 1 [9].

The teeth were then subjected to thermo cycling at 5-55°C for 500 cycles with a dwell time of 30 s [17]. The apices were occluded with composite resin and two coats of nail varnish were then applied on the tooth surface except 1mm around the restorations. The teeth were then immersed in 2% methylene blue for 24 h [18]. Following that the teeth were rinsed under running water, in order to remove surface dye and kept in distilled water until sectioning. Each tooth was embedded in acrylic resin and then sectioned through the middle of both restorations into slices of 1mm, using a microtom Isomet (Buehler Ltd., IL, SUA). The slices were examined at the optic microscope and one slice was selected from each tooth, the one with the highest infiltration. The selected slices were examined by 2 operators (dentists) using the Olympus KC301, Olympus America Inc. optic microscope. At the toothrestoration interface the length of dye penetration was measured using QuickPhoto Micro 2.2 software (Olympus Inc.) The values obtained for the enamel and the dentin margins were transformed into percentages of dye penetration length and statistically analyzed.

The interface obtained using the investigated two classes of adhesive systems was examine with an scanning electron microscopy Philips XL 30 ESEM.

Statistical analysis: The differences in the microleakage percentages were compared between the two groups. We tested the normality of the distributions of the data with the test Shapiro-Wilk. The Mann-Whitney U-test was used to investigate the differences between the two groups of dental adhesives, regarding the infiltration of enamel, as well as the infiltration of the dentin, separately. The Wilcox on test for paired samples was used to assess the differences within the groups between the percentage of dye penetration length between the enamel and the dentin. The results were considered statistically significant when p < 0.05.

Materials	Composition	Application mode ^a			
		Application of 34%-36% phosphoric acid conditioner			
prime&bond one Select Self-	Bifunctional acrylate,	Selective enamel-etch mode			
Etch and Etch&Rinse	Acidic acrylate,	1. Apply the respective conditioner to the enamel surfaces of			
Adhesive; Dentsply, DeTrey,	Functionalized phosphoric	the cavity.			
Konstanz, Germany	acid ester .	For best results, condition enamel for at least 15 seconds.			
, ,	Water	Application and curing			
	Tertiary butanol	1 Using the applicator tip apply the product sufficiently			
	Initistor	(larger cavities may require 2 or more applications) wetting all			
	Stabiliza	(anger cavities may require 2 or more applications) weiting an			
	Stabilize	2. Then contactes uniformity. Avoid pooling.			
		2. Then gently agitate the adnesive for 20 seconds.			
		3. Evaporate solvent thoroughly. Begin with a moderate stream			
		of clean, dry air from a dental syringe close and strong enough			
		to keep the adhesive layer slightly moving. Continue			
		evaporation with more forceful air until there is no more			
		movement of the adhesive, but for at least 5 seconds. Use a			
		vacuum aspirator to prevent spatter of excess adhesive onto the			
		mucosa.			
		4. Surface should have a uniform, glossy appearance. If not,			
		apply prime&bond one Select adhesive once more and repeat			
		step 3 Surface should not show areas of excessive adhesive			
		thickness or pooling			
		5 Cure prime&bond one Select adhesive for 10 seconds			
		Ensure uniform empeure of all surfaces			
		Application of 340% 360% phosphoric said conditioner			
uning Schoud and Etch & Dings	Carbomilia said modified	Application of 54%-50% phosphoric acid conditioner			
prime& bond one Etch&Rinse	Carboxylic acid modified	1. Apply the respective conditioner to the cavity surfaces			
Universal Etch&Rinse	dimethacrylate (ICB resin),	starting at the enamel margins.			
Adhesive;	Phosphoric acid modified	2. For best results, condition enamel for at least 15 seconds			
Dentsply, DeTrey, Konstanz,	acrylate resin (PENTA),	and dentin for 10 seconds.			
Germany	Urethane dimethacrylate	Rinsing and blot drying			
	(UDMA),	1. Remove conditioner with aspirator tube and/or vigorous			
	Triethyleneglycol	water spray and rinse conditioned areas thoroughly for at least			
	dimethacrylate (TEGDMA),	15 seconds.			
	2-Hydroxy-	2. Remove rinsing water completely by blowing gently with an			
	ethylmethacrylate (HEMA),	air syringe or by blot drying with a cotton pellet. Do not			
	Butvlated benzenediol	desiccate dentin.			
	(stabilizer).	3. Proceed immediately to application of prime&bond one			
	Ethyl-4(dimethyl-	Etch&Rinse adhesive			
	amino)benzoate	Application and curing			
	Camphorquinone	1 Using the applicator tip, apply the product wetting all cavity			
	Eurotionalised amorphous	surfaces uniformly. Avoid nooling. This surface should remain			
	ailian	undisturbed for 20 seconds			
	Tartiana hatar	2 Englished for 20 seconds.			
	Tertiary outan	2. Evaporate solvent by thoroughly drying with clean, dry air			
		from a dental syringe for at least 5 seconds. Use a vacuum			
		aspirator to prevent spatter of the adnesive onto the mucosa.			
		Surrace should have a uniform glossy appearance. If not, repeat			
		application and air dry.			
		3. Cure prime&bond one Etch&Rinse adhesive for 10 seconds			
		using a curing light.			
SDR TM Smart Dentin	SDR™ patented urethane	 Dispense SDR™ material directly into preparation site using 			
Replacement	di-methacrylate resin,	slow, steady pressure. Begin dispensing at the deepest portion			
-	Di-methacrylate resin.	of the cavity, keeping tip close to cavity floor. Gradually			
	Di- functional diluents	withdraw tip as cavity is filled Avoid lifting the tip out of			
	Barium and strontium	dispensed material while dispensing to minimize air			
	alumino-fluoro-silicate	entrapment At the completion of dispensing wine tin against			
	glasses (68% by pt 45%	cauity wall while withdrawing from the operative field			
	hy wol) Photoinitisting	2 Curing SDRIM material is designed to be sured in increments			
	System	2. Comingsion material is designed to be cured in increments			
	System,	up to a 4 mm deptn/tnickness. Light-cure each area of the			
	Colorants	restauration surface with a suitable visible light-curing unit			
		designed to cure materials containing camphorquinone initiator.			
l	ļ	Minimum light output must be at least 550mW/cm ² exposure			
for at least 20 seconds.					
Abbreviations: TCB resin, PENTA, UDMA, TEGMA, HEMA					
 According to the manufacturer's instructions. 					

Table 1 EXPERIMENTAL MATERIALS COMPOSITIONS AND APPLICATION MODE

Results and discussions

Results and discussions The current study assess the effect of a multi-mode adhesive system (prime & bond one Select) and a two-step etch & rinse adhesive (prime & bond one Etch & Rinse) on the microleakage of class V resin composite restorations. The procedures of restoration were realised strictly as planned, without any alteration of the method. The data collected from the study were subjected to statistical analysis using SPSS Software.

Table2 PROPORTION OF MICROLEAKAGE

, ! !	Group 1		Group 2	
	Mean	Standard Deviation	Mean	Standard Deviation
Enamel	13.5098	23.5417	7.4382	13.3598
Dentin	14.9651	16.0231	15.3985	17.8359



The microleakage of the enamel had smaller values, that the one present in dentin, for Group 1 (fig. 3). For Group 2, the infiltration affected, as well, both the enamel and the dentin, but the values for the dentin were two times higher than the values present at the enamel (fig. 4).

The microleakage was present both in enamel and at the dentin margins, but the extent at the enamel margins, was smaller compared to the dentin margins. The explanation for this can be the different morphology of the two dental structures.

When demineralised, enamel forms highly stable bond strength, because of his high mineral percentage and low water content, while dentin has an lower mineral content and a higher water and organic percentage compared with enamel [19].

The infiltration at the enamel had two times higher values for Group 1, when compared with Group 2. When referring to the microleakage at the dentin, both groups show similar values of infiltration. The multi-mode approach of the seventh generation allows the practitioner to decide the modality of usage he considers the most suitable for his clinical case. The selective enamel etching was designed in order to obtain a better marginal adaptation in enamel, superior to that achieved when using only self-etch adhesive systems[20]. Phosphoric acid pre-etching allows the resin to infiltrate better into the enamel, increasing the enamel surface by creating porosities on its surface [21].

Although the work protocol allowed the selective etching of the enamel, for increasing the adhesion of the all-in-one adhesive system, the results of this study showed a superior sealing when using the 2 steps etch-and-rinse adhesive system.

When statistical analysis was made, firstly were analyzed the values of infiltration at enamel and dentin (cement) for each adhesive system, to investigate if there is a difference on the adhesion performances on enamel compared with on dentin. For this analysis the Related-Samples Wilcoxon Signed Rank Test was used, but no statistically difference was showed for neither one of the adhesive systems (p = 0.199 for prime&bond one Select and p = 0.145 for prime&bond one Etch&Rinse).

Then were compared the infiltrations of the two adhesive systems in enamel and then in dentin. Although there was an arithmetic difference of the means of percentages of infiltration for enamel (table 2), the MannFig. 3. Optical microscopy of some different microleakage lenghts at enamel (E) and dentin (D) margins: a) (D) for Group 1; b) (E) for Group 1; the red line marks the lengths of microleakage in enamel and in dentin (cement) the yellow line marks the total length of the interface toothcomposite restoration (axial and lateral wall)

Fig. 4. Optical microscopy of some different microleakage lengths at enamel (E) and dentin (D) margins: a) (D) for Group 2; b) (E) for Group 2; the red line marks the lengths of microleakage in enamel and in dentin (cement); the yellow line marks the total length of the interface tooth-composite restoration (axial and lateral wall)



Fig. 5. SEM photomicrographs: a) obturation from Group 1, magnification 25X; b) obturation from Group 2, magnification 25X;c) interface enamel (E) - adhesive- composite(C),Group 1,magnification 500X;d) interface enamel(E) -adhesive- composite (C), Group 2, magnification 500X;e) interface dentin(D) -adhesivecomposite (C), Group 1, magnification 500X;f) interface dentin(D) adhesive - composite (C), Group 2,magnification 500X;g) interface dentin(D) -adhesive- composite (C), Group 1, magnification 1000X;h) interface dentin(D) -adhesive- composite (C), Group 2,

magnification 1000X

Whitney U- test showed no statistical difference between the two adhesives (p=0.380). Applied for the dentin values, the test Mann-Whitney U illustrated as well no statistical difference between the two adhesives (p=0.910). The statistic difference between the two adhesive systems was not significant.

Although Lopez et al. [9]showed that, when prime&bond one Select Self-Etch and Etch&Rinse Adhesive is used with the selective etching of the enamel, it increases their performance in clinical trial, our study shown that in comparison with prime&bond one Etch&Rinse, it had a lower performance.

Clinically, bond failure can happen for various causes as moisture contamination, incomplete infiltration of resin into the demineralised layer, excessive etching or over-drying. The risk of making an error increases with the number of steps of an adhesive system. The present study shows that the bonding process can be simplified by using systems that combine the primer and adhesive or even by using all in one adhesive systems. Both systems provided low microleakage values, although the process is still technique sensitive [22].

The adhesion provided shown, at magnification 500X, a continuous interface without voids or gaps, as we can observe in the SEM images, with a good interconnection of the three substrates involved. This proves that a two step etch-and-rinse adhesive system can perform similar as a two step self etch with selective etching on enamel, when strictly respecting the clinical protocol. At magnification 1000X it can be clearly noticed the penetration of the resin into the dental canaliculi and the formation of the hybrid layer and resin filaments. One can observe a very well formed hybrid layer, in close contact with the dentin, which sends ramification to the dental canaliculi, along the line of junction between dentin and adhesive. The presence of alcohol in the composition of total-etch adhesive systems is extremely important, because it helps the penetration of the hydrophobic monomers in the demineralised dental tissue. However, the sensitivity of the adhesive to water is increased, by the presence of alcohol in their composition. An extended acid etching lead to, a decrease of the adhesion strength, therefore the correct acid treatment is vital for the totaletch adhesive systems. The hybrid layer is susceptible to the hydrolysis reaction, which can affect the capacity of sealing of the dental canaliculi and generates a decrease in adhesion strength. For the self-etch adhesive systems, the penetration of the dentin is reduced compared with total-etch adhesives, because the smear-layer is not removed. The SEM studies expose the fact that, for the adhesive systems, there is no standard hybridisation. The hybrid layer of the total-etch adhesives are different from the hybrid layer formed by self etch adhesives, in terms of thickness, uniformity, number and length of resin tags [23].

The possibilities of methods available in order to asses microleakage are direct visual examination, microscopic examination, scanning electron microscopic examination, dye penetration, air pressure, the use of radioactive isotope tracer, the use of a chemical tracer, electrochemical methodologies, the artificial caries method, the measure of bacteria penetration, neutron activation analysis and three-dimensional image analysis [24]. The methods elected to be used in this study were microscopic examination, scanning electron microscopic examination and dye penetration. The examination at the optic microscope, as well as, dye penetration is the most notorious methods because they are easy to handle [25-40]. However the result from the dye penetration method depends on the dye-marker used, on the concentration of the substance used as dye and on the time of immersion. This method is used in small cavities, as class V, combined with SEM for a complete examination, in order to complete scanning electron microscopic examination. Although SEM is considered golden standard in microleakage evaluation, it is dependent of having the technical equipment and well trained man in this domain. These methods are working together in offering a complex vision of the interface tooth-restoration material [10, 26, 39].

The actual study is an *in vitro* study, in which class V cavities restored by using different types of adhesive systems were investigated in terms of microleakage [22]. *In vitro* studies remain an indispensable approach in the initial testing of dental materials, as an indicator to the theoretical amount of leakage that can possibly appear *in vivo* [40]. However, *in vitro* studies cannot imitate the oral environment adequately and clinical conditions may influence the long-term microleakage of restorations. Consequently, *in vivo* studies should be made in order to support the result of this study.

Conclusion

The examined samples revealed subtle differences between the distinctive adhesive systems showing that there is no standard hybridization. Within the limitations of this study, it can be concluded that both adhesive systems offer a good sealing to the restorations and can be used successfully with SDR.

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