

EFFECT OF BULK- FILLING, SNOWPLOW AND INJECTION-MOLDED TECHNIQUES ON NANOLEAKAGE OF CLASS II COMPOSITE RESTORATIONS IN PERMANENT MOLARS USING DIFFERENT ETCHING MODES (AN IN VITRO STUDY)

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### ABSTRACT

**Purpose :** The Objective of this in vitro study was to compare nanoleakage in permanent molars restored with bulk-filling, snowplow and injection-molded composite application techniques using a universal adhesive with two different etching modes.

**Materials and Methods:** A total of twenty four human permanent molars were used. Class II box was prepared on the proximal surfaces of molars. Forty eight specimens were randomly divided into three main equal groups according to resin composite application techniques, bulk-filling, snowplow and injection-molded techniques. Each group was further divided into two subgroups according to etching mode; etch and rinse mode and self -etch mode. Each specimen was sectioned mesiodistally and immersed in 50% freshly prepared silver nitrate solution for 24 hours followed by washing under running water and were kept for 8 hours in a photo-developer solution. The specimens and the silver nitrate amount were analyzed in (ESEM) within the adhesive layer, hybrid layer and the resin tags.

**Results:** The results showed that there is a statistically significant difference in nanoleakage regarding to the three different composite application techniques. Bulk filling and injection molded techniques have mean values that are close to each other, while these mean values are greater than the ones snowplow have. There is a statistically significance difference between etch and rinse mode and self-etch mode. Also, the effect of etching mode on nanoleakage is more than twice as much as that of composite application technique.

**Conclusion** Using the "snowplow" technique with etch and rinse mode of universal adhesive, can decrease nanoleakage rate, moreover etching mode could influence the nanoleakage of universal adhesives.

**KEYWORDS:** Nanoleakage, Bulk-filling technique, Snow-plow-technique, Injected molded technique, Composite restoration, Universal adhesive.

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# INTRODUCTION

Class II resin composite restorations present different challenges to the operator. The resin composite traditional application techniques leave the restorations liable to micro-leaking, marginal chipping, fracture and recurrent caries leading to their failure in higher rates when compared to amalgam<sup>(1)</sup>. In addition, polymerization shrinkage of resin composite creates stresses at the toothrestoration interface that may be displayed as nanoleakage which affects bond strength and leading to pulpal inflammation, microleakage and discoloration<sup>(2,3)</sup>. The volumetric shrinkage of resin composites is mainly depending on the formulation and curing condition<sup>(4,5)</sup>. To counter the problems often associated with resin composite, many application techniques were developed, one of them was called injection-molded composite technique, which decreases the possibility for voids and fault lines but at the same time maintaining the integrity of the tooth structure. This application technique implicates the proper combination of paste and flowable composites to create esthetic and strong restorations (6).

However, a different method for resin composite application technique was described known as "snowplow technique." This technique is depending on squeezing of the flowable composite out of the sides of the restoration by being pressed by application of packable composite. The researchers have found that, this technique can promote resin composite adaptation in addition to minimize the chance of marginal leakage<sup>(7)</sup> In deep and wide cavities, it is recommended to use the bulk-fill technique to save time and simplify the procedure<sup>(8)</sup>. Previous studies reported low polymerization shrinkage, good marginal adaptation, fast application steps and reduced working time for bulkfill restoration<sup>(9,10)</sup>. Since their introduction, several bulkfill products have been launched into the market <sup>(11,12)</sup>.

In 1995, Sano et al, described a specific test for testing leakage through using silver nitrate as a

dye for detecting nanometer porosities within and adjacent to the hybrid layer<sup>(13,14)</sup>. The nanometric spaces were attributed to inefficient resin infiltration or inadequate polymerization. Nanoleakage were evaluated by measuring penetration of silver grains due to inadequate polymerization or presence of submicron defects in resin infiltration. Therefore, nanoleakage assessment is considered an essential indicator of a restorative material sealability and quality of hybrid layer and affects the longevity of the restoration<sup>(15)</sup>. The purpose of this in vitro study was to evaluate nanoleakage in permanent molars restored with bulk-filling, snowplow and injection-molded composite application techniques using universal adhesive with two different etching modes.

The null hypothesis of this study is that no significant effect on nanoleakage of resin composite restorations applied by the three different techniques using either etch and rinse or self etch modes of the universal adhesive.

#### MATERIALS AND METHODS

#### Selection and grouping of the teeth

The study was an in vitro study conducted in Faculty of dental medicine Ahram Canadian University. A total of twenty four human permanent molars were used for Nanoleakage evaluation. Randomly selected teeth were free from caries, cracks, fracture, previous restorations or structural defects. Teeth were cleaned from debris and blood stains, polished with slurry of pumice and water, and then stored in distilled water at room temperature until cavity preparation. Class II proximal box cavities were prepared on the mesial and distal sides of each permanent molar following the guidelines for resin-based composite restoration.

Forty eight specimens were randomly divided into three main equal groups according to application techniques of composite restoration used, 16 specimens each (Group one: bulk-filling technique, Group two: snowplow technique and Group three: injection-molded technique). Each main group was further divided into two subgroups (n=8) according to etching modes (etch and rinse mode and self etch mode)using universal adhesive. There were six subgroups, Subgroup A: bulk- filling application technique with etch and rinse mode , Subgroup B: bulk filling application technique with self etch mode, Subgroup C: snowplow application technique with etch and rinse etching mode, Subgroup D: snowplow application technique with self etch mode, Subgroup E: injection-molded application technique with etch and rinse mode , Subgroup F: injection-molded technique with self-etch mode.

### **Teeth preparation:**

Class II proximal box cavities were prepared on the proximal surfaces of the mesial and distal sides of each permanent molar after fixation of the teeth in the cast and following the guidelines for resinbased composite restoration (3mm in width buccolingually, 5mm in depth). Cavity preparation was carried out using #330, #245 short shank carbide burs, 0.8mm diameter mounted in a high-speed water-cooled handpiece<sup>\*</sup>.

#### Adhesive system application and teeth restoration:

The prepared class II proximal box cavities were restored after application of sectional matrix bands for permanent molars<sup>\*\*</sup>. The bands were adapted and secured in place using plastic wedges<sup>(16)</sup>

Adhesive system was applied using universal adhesive in etch and rinse mode and self-etch mode at the same manner for group one and two as follow, for the etch and rise mode, the acid etching step was done before applying the dentin bonding systems, etching of the cavity enamel surface for

30 seconds by using 37% phosphoric acid\*\*\* and etching the rest of the cavity for 15 seconds, then rinsing for 10 seconds. Air drying was done gently to remove excess water and to preserve the dentine in moist condition. Universal adhesive\*\*\*\* was applied to the cavity in two coats by rubbing microbrush for 15 seconds and air drying again as before, then curing of adhesive for 20 seconds according to manufacture instructions using a LED curing unit\*\*\*\*\* at zero distance<sup>(17)</sup>. No light curing was applied between coats. While for the self-etch mode, the dentin bonding system was applied directly without etching step as mentioned in the etch and rinse mode following manufacturer's instructions. The standard composition of adhesive and resin composites are shown in table (1).

TABLE (1) The standard composition of adhesive and resin composites investigated in the present study <sup>(18-21)</sup>.

Tested materials	Composition
Bond Futurabond U	-Mild 2.3 HEMA, Bis-GMA, HEDMA, acidic adhesive monomer, urethane dimethacrylate, catalyst, silica nanoparticles, ethanol
EtchingGel Attaque Gel	Orthophosforic Acid, Methylparaben, Blue Deying(Cl42015) ,Thickening and Deionized Water
(Grandio) Universal Nano- hybrid composite	-BIS GMA,Triethylene glycol dimethacrylate
Flowable Composite)	BIS GMA,UDMA, TEGDMA, BHT
X-tra-base Nanohybrid composite (bulk- fill flowable)	-BIS GMA, Aliphatic dimethacrylate methacrylates, BHT ,Inorganic fillers (75 wt%)

\*\*\* Attaque Gel- Spesifikasi Biodinamica- Brazil)

\*\*\*\* Futurabond U, VOCO GmbH,Germany

\*\*\*\*\* LED.F 1600 mW/cm<sup>2</sup> High Intensity, WOODPECKER)

<sup>\*</sup> Brasseler USA Dental Instrumentation, Savannah, GA, USA)

<sup>\*\*</sup> TOR –VM sectional contoured metal matrix Kit NO 1.398-hard 0.035 Alex Dent

Regarding group one (Bulk-Filling technique), the prepared cavities were restored by inserting the composite in a single increment 4-5 mm using the X-tra-base Nanohybrid composite then the resin composite was cured for 40 seconds following manufacturer's instructions<sup>\*</sup>.

Regarding group two (Snowplow technique), the flowable composite was applied over pulpal floor and axial wall of the prepared cavity in a thin layer<sup>\*\*</sup>. This layer was not cured. Then pushing a heavily filled nanohybrid resin composite into the uncured flowable composite<sup>8</sup>. Displacement of the most of the flowable composite by the effect of restorative resin composite was done and subsequently removed from the cavity. A little amount of flowable composite was remained in those areas where the higher-viscosity composite was adapted completely to the cavity walls. The combined layer of flowable and restorative composites were then cured using LED curing unit for 40 seconds .

Regarding Group three (Injection molded technique), composite syringes and compules were preheated using a composite heating device (Ena Heat, Micerium S.p.A. Avegno GE, Italy) in T2 mode at 55°C for 10 minutes (22,23) Adhesive system was applied using universal adhesive in etch and rinse mode and self-etch mode. In case of etch and rinse mode, the tooth was etched (while in case of self-etch mode ,no etching was done) and then a universal adhesive was massaged . First, a small amount of adhesive is splashed into the cavity for 15 seconds and air thinned for five seconds, then immediately injecting a layer of heated flowable composite and followed by injecting a layer of heated resin composite, then all the three layers were co-cured as one monolithic mass using LED curing light for 40 seconds, and the rest of the cavity was injected by preheated nanohybrid resin composite and cured. Finally, composite

restorations were finished and polished. Specimens were stored in distilled water until their preparation for nanoleakage evaluation<sup>5,7,8</sup>

#### Specimen preparation for Nanoleakage evaluation

Before sectioning of teeth, root surfaces were dipped in melted wax up to 2.0 mm below the cemento-enamel junction, resulting in a 0.2- to 0.3-mm-thick wax layer (\*\*\*,24) All the specimens were sectioned mesiodistally into two halves using high-speed diamond disc under water coolant\*\*\*\*. Two halves of each tooth were used. Nanoleakage patterns were detected ultra-morphologically and also the amount of silver nitrate penetration was detected within the resin dentin interface. Then the distilled water was used to store the specimens for 24hours. Then immersing the specimens in 50% freshly prepared silver nitrate solution was done for 24 hours in darkness followed by washing under running water for 5 minutes \*\*\*\*\*. A photo-developer solution was used to keep the specimens for 8 hours under fluorescent light to converse silver ions into metallic silver\*\*\*\*\*. Finally, The specimens were rinsed under running water for 5 minutes to remove traces of photo-developer (25)

#### ESEM evaluation of Nano-leakage

Resin dentin interface of each specimen was analyzed using environmental scanning electron microscope; ESEM operated with back-scattered electron mode \*\*\*\*\*\*\*. The silver nitrate uptake was expressed as a nano-leakage percentage of the total area evaluated <sup>(26)</sup>. In each specimen, measuring the amount of silver nitrate within the adhesive layer, hybrid layer and the resin tags was done in an area (200µmx200µm). The entire interface was scanned

- \*\*\*\*\*\* Kodak, Rochester, NY, USA
- \*\*\*\*\*\*\* FEG Quanta 250, EMRA France

<sup>\*</sup> bulk-fill flowable, VOCO GmbH,Germany

<sup>\*\*</sup> Amaris, VOCO GmbH Germany

<sup>\*\*\*</sup> Epoxiglass; Diadema, SP, Brazil

<sup>\*\*\*\*</sup> MTI Corporation, Richmond, USA

<sup>\*\*\*\*\*</sup> Cennabras Indŝtria e Comércio Ltda,Guarulhos, São Paulo, Brazil

at magnification 500% directly on the ESEM for quantitative analysis<sup>(27)</sup>. In each specimen, selected area of nanoleakage was scanned and a photo was captured as shown in figures(4-8). One photomicrograph was taken from each specimen. Ultra-morphological nano-leakage patterns were detected and analyzed using ImageJ/Fiji1.51j8 software program as shown in figure(1) (\*). Opening the photos was done inside the program and the scale was set by measuring the line of the scale bar of each photo. A line was drawn to record the full length of the interface. Recording the length of the nano-leakage at the resin-dentin interface was done by drawing another line. Separate nano-leakage areas were measured using separate lines and the total length of the nano-leakage at the interface was calculated by the sum of all separate measuring lines <sup>(28)</sup>. Photos were calibrated by converting pixel dimensions to the desired measuring unit of the photo. This was done by converting the scale at the information section in the photo to the desired unit which in this case was micrometers.

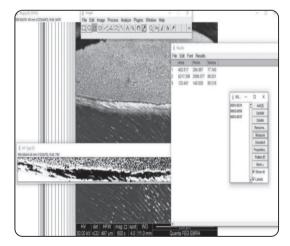


Fig. (1) ImageJ program used for analysis of ESEM images

The nano-leakage percentage for each photo was calculated according to the following equation:

The length of nano-leakageThe total length of the interfaceX 100

The final results were then obtained and subjected to statistical analysis

#### **Statistical analysis**

The normal distribution of data (Shapiro-Wilk's test) and the homogeneity of the variances (leven's test) were assessed. The two-way ANOVA was performed to highlight the effect of composite application technique (A) and etching mode (B) and the interaction between both variables (A\*B). The results were expressed as the mean ± standard deviation (SD). Statistical analysis was performed with IBM® SPSS® (SPSS Inc., IBM Corporation, NY, USA) Statistics Version 21 for Windows.

### RESULTS

The silver nitrate penetration method, combined with high magnification SEM by means of secondary electron or back-scattered electron mode, can provide much better information concerning the sealing ability of the restorations and the percentage of nanoleakage.<sup>(25)</sup> Results showed that Nano-leakage for bulk filling technique has the highest values 9.05 and 16.55um for the total-etch mode and selfetch mode respectively followed by injection molded technique with mean values 8.82 and 12.50um. Lastly, snowplow technique with mean values 3.10 and 4.51um as shown in table (2) and figure (2).

In general, there are no large differences between the mean values and median values, in addition both mean and median values by composite application techniques have the same pattern despite the used etching mode where it can be easily noted that bulk filling and injection molded have closer mean and median values while both are much greater than snowplow technique, besides no clear pattern can be spotted regarding the values of standard deviation inter-quartile range.

<sup>\*</sup> Wayne Rasband, National institutes of health, USA, <u>http://imagej.nih.gov/ij</u>, Java 1.8.0\_112 (64-bit), 4212K of 2921MB (<1%)</p>

Etching mode	Composite application technique	Ν	M (SD)	Mdn (IQR)	$\left(\frac{M-Mdn}{M}\right)\%$
	Bulk filling	8	9.05 (2.31)	9.07 (1.53)	-0.22
Total-etch	Snowplow	8	3.10 (1.66)	3.17 (2.86)	-2.26
	Injection-molded	8	8.82 (5.03)	8.78 (4.45)	0.45
Self-etch	Bulk filling	8	16.55 (5.96)	17.35 (10.40)	-4.83
	Snowplow	8	4.51 (1.27)	4.47 (0.73)	0.89
	Injection-molded	8	12.50 (5.83)	11.97 (6.11)	4.24

TABLE (2) Nano-leakage descriptive statistics by etching mode and composite application technique.

N: Number of observations, M: Mean, SD: Standard deviation, Mdn: Median, IQR: Inter-quartile range

The values of percentage difference between mean and median suggest the lack of outliers and skewness in the data since they ranged between (-4.83% to 4.24%).

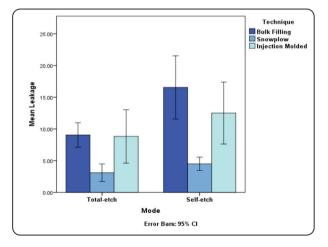


Fig. (2) Nano-leakage error bar according to etching mode and composite application technique.

The two-way ANOVA showed that there is a statistically significance difference according to the composite application technique and etching mode, whilst there is no significant interaction between them. Also, the effect of etching mode on nano-leakage is more than twice as much as that of composite application technique as shown in table 3 and figure 3.

The effect size (partial  $\eta^2$ ) value of composite application technique 0.224 suggests a high effect on nano-leakage where 22.4% of variations in nanoleakage can be explained by change in composite application technique. Also, the effect size of etching mode 0.491 indicates a high effect on nano-leakage where 49.1% of variations in nano-leakage can be explained by change in etching mode. Moreover, the effect of the interaction between the variables 0.094 implies a medium effect on nano-leakage where only 9.4% of total variations in nano-leakage attributes to the interaction between etching mode and composite application technique.

The mean values for the composite application techniques have the same pattern despite the etching mode where bulk filling and injection molded have mean values that are close to each other (especially when total-etch mode is used, they are almost identical) while these mean values are greater than the ones snowplow have as seen in the profile plot (figure 3)

	SS	Df	MS	F-statistic	Р	Partial $\eta^2$
Etching Mode	706.434	2	353.217	20.279	0.000	0.491
Application Technique	211.417	1	211.417	12.138	0.001	0.224
Mode × Technique	75.998	2	37.999	2.182	0.125	0.094
Error	731.537	42	17.418			

TABLE(3) Two-Way ANOVA of nano-leakage by etching mode and composite application technique.

SS: Sum squares of errors, df: degrees of freedom, MS: Mean squares of errors, P: p-value, partial n2: effect size, level of

significance is 0.05

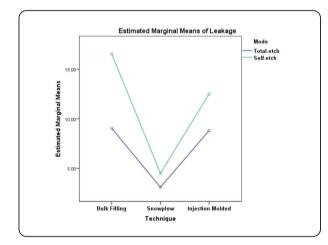


Fig. (3) Profile plot of marginal means for nano-leakage (%) based on composite application technique and etching mode.

Sheffe test was used to determine the significance of the difference between all the pairs of the three composite application techniques. There is no significant difference in nano-leakage between bulk filling and injection molded whilst snowplow is significantly different from any other technique,  $p \le 0.005$  as shown in table 4.

 TABLE (4) Sheffe test for nano-leakage based on application technique and etching mode

Te	echnique	Mean	Р	
(I)	(J)	Difference		
Bulk filling	Snowplow	8.99	0.000	
Bulk filling	Injection molded	2.14	0.453	
Snowplow	Injection molded	-6.86	0.001	

P: p-value, level of significance is 0.05

#### **ESEM Evaluation**

In all the specimens, nanoleakage was manifested by silver penetration that had different patterns and different degrees

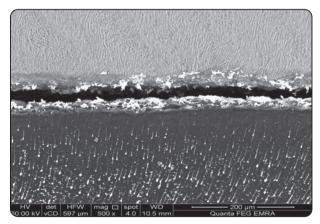


Fig. (4) SEM photomicrograph of nanoleakage (mag. 500x) of bulk-filling technique with self-etch mode of universal adhesive, the pattern of nanoleakage showed heavy silver deposition along the dentine resin interface

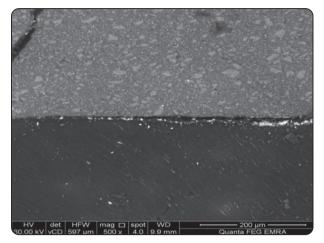


Fig. (5) SEM photomicrograph of nanoleakage (mag. 500x) of snow-plow technique with total-etch mode of universal adhesive, the pattern of nanoleakage showed silver deposition in the form of spots at the dentine resin interface.

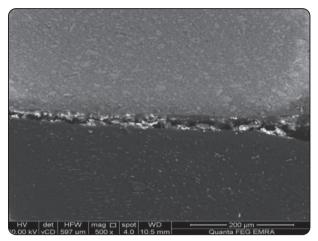


Fig. (6) SEM photomicrograph of nanoleakage (mag. 500x) of snowplow technique with self-etch mode of universal adhesive, the pattern of nanoleakage showed less dense silver deposition along the dentine resin interface.

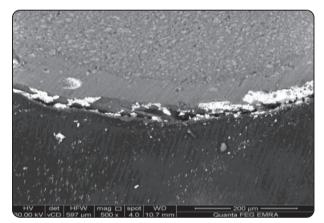


Fig. (7) SEM photomicrograph of nanoleakage (mag. 500x) of Injected –molded technique with total-etch mode of universal adhesive, the pattern of nanoleakage showed water trees silver deposition along the dentine resin interface.

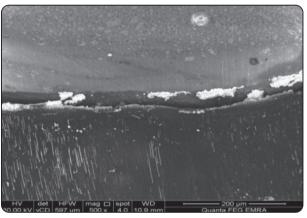


Fig. (8) SEM photomicrograph of nanoleakage (mag. 500x) of Injected–molded technique with self-etch mode of universal adhesive, the nanoleakage pattern showed more dense of silver deposition along the resin dentine interface.

### DISCUSSION

The type of leakage that exist in the dentin margins of the resin restoration is termed nanoleakage. Nanoleakage allows oral and pulpal liquids to penetrate into porosities within or adjacent to the hybrid layer. The type of bonding agent and resin composite application techniques affect amount of penetration occur that consequently affect the stability of adhesive bond between dentin and restorative material.<sup>(15)</sup> Degradation of the bond will lead to multiple clinical problem such as hypersensitivity, recurrent caries, discoloration near restoration margins or loss of the restoration. So, there is some techniques are introduced in restorative dentistry to overcome the problems of class II as bulk filling, snowplow, injection molding techniques. In addition, the microleakage and the nanoleakage evaluation is important for assessment the defective adaptation of the resinous material. In the present study, ammonical silver nitrate solution was used because it has small diameter molecule (0.059 nm) which enables the solution to migrate within the interface zone <sup>(29,30)</sup>.Silver nitrate solution is highly reactive to stain after binding to exposed collagen fibrils makes it the most appropriate agent to detect the nanoporosities within the hybrid layer <sup>(31)</sup>. Moreover, silver nitrate is providing a sharp picture (figure 4,5) as it induces an electron microscopic measurable contrast for the degree of penetration into the interface. Also, it has the potential to immobilize, which prevents further penetration during specimen preparation <sup>(14)</sup>.

The null hypothesis of current study was totally rejected as the results showed that there is a statistically significant difference in nanoleakage regarding to the three different composite application techniques. Also, there is a statistically significance difference between etch and rinse mode and selfetch mode. The highest nanoleakage mean value was recorded in bulk-filling technique with selfetch mode of universal adhesive, whereas the least Nanoleakage mean value was recorded in snowplow technique with etch and rinse mode of universal adhesive, this mean values were higher than those reported by other studies reported by Al-Agha et al (26) in which the mean value of silver deposition was (2.013%) for Class V resin restorations using nanofilled self-etch adhesive. Regardless the etching mode, the study showed that nanoleakage was greater with bulk filling tech, than other techniqus, might be due to that the polymerization shrinkage of the bulk-fill materials was higher than that of a conventional resin composite with higher nanoleakage values. Benetti et al, 2015 concluded that (x-tra base) showed higher polymerization contraction values as it is low-viscosity bulkfill resin composites containing lower filler volume . In contrast, (SonicFill and Tetric EvoCeram Bulk Fill) high-viscosity bulk-fill resin composites with higher filler fraction, demonstrated polymerization contraction values closer to the conventional resin composite<sup>(8)</sup>. An increase in the filler content can,

to a certain extent, reduce the polymerization contraction <sup>(32,33)</sup>. Although, the present study showed that the least nanoleakage value in all groups was recorded in snow-plow technique with etch and rinse mode, the use of snowplow technique is still controversial as some studies concluded that co-curing the flowable liner with the composite restoration did not improve marginal seal. This may be explained that flowable composite displaced the main bulk of the composite, which leads to a heterogeous increase in contents of resinous material of the main bulk of the composite restoration and therefore, an increase of polymerization shrinkage. This may also be due to polymerization shrinkage of the resin composite, which causes contraction forces and dislodges the bond of uncured flowable composite (34,35). In contrast, other studies found that co-curing the flowable with the hybrid composite can decrease the nano-leakage in ClassII composite restorations compared to the other group, hence deceasing nanoleakage value due to improvement in sealing ability at the margin of the overlying viscous composite which is attributed to improve in penetration of uncured liner<sup>(7,36)</sup> The current study approved these results. Also the results showed that bulk filling and injection molded application techniques have closer mean and median values regarding Nanoleakage, while both are much greater than snowplow technique, and the results revealed that there is no effect of preheating of resin composite in case of injection molded application technique on nanoleakage values. Botros et al, agreed with current study results, in which they approved that the pre-heating temperatures of resin composite did not affect nanoleakage score. Pre-heating of composite improved its Degree of conversion %.<sup>(37)</sup> The universal adhesive was used in all the groups to standardize process of the study<sup>(38)</sup> and to merge the advantages of self-etch (SE) adhesives, such as rapid application and low susceptibility to differences in operator technique with those of etch-and-rinse (ER) adhesives, such as effective and stable bonding to enamel (39-41)

Regardless the composite application techniques used, the results showed high significant nanoleakage mean value between etch and rinse and self etch in all the study groups, the self-etch mode showed higher nano-leakage mean value than that of etch and rinse mode. This might be attributed to that, in etch & rinse approach, the smear layer is removed after preliminary etching with phosphoric acid in and subsequently increases impregnation by the adhesive, allowing the creation of a well impregnated hybrid layer <sup>(42-44)</sup>. These results are in agreement with Zhao et al who reported that, regarding to the etch-and-rinse approach, the bond strength is improved using universal adhesives in comparison to the self-etch strategy<sup>(28)</sup>.

Yamauchi et al, disagreed with the study results. They concluded that the universal adhesives in etch-and-rinse mode showed no significant difference when compared to that in self-etch mode (45). This might be related to the different types of materials used, tooth structure, location of the restorations, type and size of the cavities, operator factors, and research methodologies (34). Our results also support previous findings showing that when selective enamel etch technique was used, phosphoric acid extends to dentine, the application of a self-etch adhesive results in higher micro-tensile bond strength deceasing nanoleakage value compared with its application on smear layercovered dentine<sup>(46,47)</sup>. This is also in agreement with many in vitro studies using transmission Electron Microscopy (TEM) and the Field Emission- SEM (FE-SEM), in short-term periods of observation. and reported that, a certain amount of nanoleakage was more pronounced for the self-etch adhesives when compared to etch and rinse adhesives (27).

ESEM images indicated presence of nanoleakage in all samples bonded with different etching modes of universal adhesive as manifested by the infiltration of silver ions around the collagen fibrils and at the adhesive/dentine interface. These results might

be attributed to shrinkage of resin at the time of its polymerization (48,49) Moreover improper wetting of the resin to dentin and collagen surfaces could exist due to the wet nature of dentin tissues and bonding resin viscosity. The previously mentioned factors could create nanogaps between the dentin surfaces and bonding resin due to presence of areas of imperfect resin infiltration, retained water or other solvent poor polymerization or phase separation (50,51) In addition, various degrees of silver penetration were obvious between samples of different groups indicating different degrees of nanoleakage. This result could be related to the chemical nature of the universal adhesive that is reflected on the expected degree of polymerization shrinkage and the degree of water sorption (dye solution)<sup>(52)</sup>

Addressing the limitation of this study, one concern was that it was conducted in-vitro, where different oral environmental parameters were not tested. Also, specimens were stored in distilled water and were not subjected to thermocycling.

## CONCLUSION

Considering the limitations of an in vitro study, our findings showed that:

- Using the "snow-plow" technique with total etch mode of universal adhesive, can decrease nano-leakage rate.
- Using the universal adhesive system for bonding composite restorations with etch and rinse mode can minimize the expected degree of nanoleakage.
- 3. Etching mode could influence the nanoleakage of universal adhesives.

Finally, further long-term research programmes, particularly clinical, are warranted to demonstrate the influence of physical factors on the new injected molded technique

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