Future Dental Journal

Volume 4 | Issue 2 Article 26

2018

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Recommended Citation

Hassan Alia, Rasha; Niazy, Maha Ahmed; Abdel Naguib, Essam; and Abdel Ghany, Osama Saleh (2018) "Effect of application technique and mode of curing on nano-leakage of universal adhesive system," *Future Dental Journal*: Vol. 4 : Iss. 2 , PP 253-261.
Available at: https://digitalcommons.aaru.edu.jo/fdj/vol4/iss2/26

Future Dental Journal 4 (2018) 253–261

Contents lists available at ScienceDirect

Future Dental Journal

journal homepage: www.elsevier.com/locate/fdj

Effect of application technique and mode of curing on nano-leakage of universal adhesive system

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ABSTRACT

This study was carried out to Investigate the effect of application technique (total etch &self etch) and curing mode (regular & soft start) on nanoleakage of universal adhesive system in coronal and root dentine. A total number of 80 freshly extracted lower molars were used. The teeth were divided into two groups (40 teeth each) according to the application technique of the adhesive (either total or self-etch mode). Each group was subdivided into two subgroups (20 teeth each) according to the mode of curing that was used either regular or soft start mode. Each subgroup was further divided into two divisions (10 teeth each) according to the surface to which the adhesive will be applied either coronal or root dentin. The roots of all teeth were cut off at the cemento-enamel junction. The occlusal enamel of the coronal portion was removed to the depth of the central fossa to expose dentine. For the root portion, the cervical 1/3 of the buccal surface was flattened by wet grinding on a bench grinder machine. The adhesive was applied according to manufacturer instructions. Resin composite was incrementally packed and light cured either in the regular continuous mode for 20 s or cured in the ramp soft-start mode. The specimens were sectioned into beams of 1 mm thick. The beams were immersed in ammonical silver nitrate for 24 h, rinsed with distilled water, and immersed in a photo-developing solution for 8 h. Nanoleakage patterns were detected using an environmental scanning electron microscope (ESEM), and the amount of silver nitrate penetration (Wt%) was analyzed using the EDAX. The results revealed that, regardless the mode of curing in coronal or root dentine, self-etch application technique showed statistically significantly higher nanoleakage mean values compared to total-etch in all the groups. In all the groups regular curing mode showed statistically significantly higher nanoleakage mean values than soft start mode, except in coronal dentine with the self-etch application technique there was no statistically significant difference between nanoleakage mean values of the two curing modes. Regardless the curing mode with self-etch application technique, there was no statistically significant difference between nanoleakage mean value of coronal and root dentin, while for total etch technique with regular curing mode, root dentin showed statistically significantly higher nanoleakage % mean value than coronal dentin. It could be concluded that, Nanoleakage varied with technique of application of the universal adhesive system being greater with the self-etch technique. Total etch technique using soft start curing mode in coronal and root dentine presented the lowest nanoleakage between the groups.

1. Introduction

Resin composite filling materials and adhesive techniques have changed dramatically nowadays dental practice. Regardless how impressive the shape and color, a perfect composite restoration doesn't last without good bonding [1]. The current pattern for bonding to dentine is based upon the diffusion and micromechanical retention of monomers within the collagen meshwork [2].

Bonding of resin composites to dentin can be achieved by many adhesive systems. Current adhesive systems can be categorized according to their etching technique into two groups: total-etch and self-etch. The first group is the total-etch technique, which relies on the removal of the smear layer by etching, followed by the application of a self-priming agent that incorporates the primer and the adhesive resin into one bottle. The second group is the self-etch, in which the etching step is excluded. The self-etching products may be one-step or two-step. Two-step self-etch adhesives are formed of step 1:self-etching primer application and step 2: the bonding agent application, while the one-step self-etch adhesives are formed of two liquids which are applied to the tooth after mixing. Self-etching adhesives in which the etchant, primer, and bond are supplied in one bottle have been introduced lately [3].

The primer and adhesive resin may not always fully infiltrate the demineralized dentin. The difference between depth of dentin demineralization and depth of resin infiltration leads to the formation of micro pores underneath and within the hybrid layer which was observed using silver nitrate [4,5] in a phenomenon named nanoleakage. Nanoleakage may be also due to microvoids which were resulted from incomplete water removal from the resin–dentin interface [6–8].

Clinicians are confused between total and self-etch adhesives. Thus

Peer review under responsibility of Faculty of Oral & Dental Medicine, Future University.
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https://doi.org/10.1016/j.fdj.2018.04.001
Received 29 April 2017; Accepted 5 April 2018
Available online 13 October 2018

the universal adhesive incorporates the advantages of both, giving dentists one bottle for a wide range of indications. It can be used with total-etch, self-etch, and selective etch. This universal adhesive eliminate the variances currently occur in materials we are using and make the procedures much easier. So this new commercial product will be examined in vitro in terms of nanoleakage.

2. Materials and methods

2.1. Selection and grouping of teeth

A total number of 80 freshly extracted sound human lower molars which were extracted for periodontal reasons from patients with age range of 40–50 years were selected for this study. Teeth were cleaned, debrided of soft tissue remnants, scaled with ultrasonic scaler (Cavitron, NSK-Varios 550, Japan), then stored in saline solution at 4 °C (Venturini et al., in 2006), within one month after extraction until use. The teeth were divided into two groups (40 teeth each) according to the application technique of universal bond system (A) i.e. either self-etch (A_1) or total-etch mode (A_2). Each group was subdivided into two subgroups (20 teeth each) according to mode of curing (B) that was used either regular (B_1) or soft start mode (B_2). Each subgroup was further divided into two divisions (10 teeth each) according to the surface to which the adhesive was applied (C) either coronal (C_1) or root dentin (C_2).

2.2. Preparation of teeth

The roots of all teeth were cut off at the cemento-enamel junction using a double sided diamond disc (Edental Golden S.A.W., Switzerland) mounted to low speed handpiece (W&H, WA-66A, dentalwerk, Austria) under copious water coolant. For the coronal dentine specimen: The occlusal enamel was removed to the depth of the central fossa using a diamond disc mounted to low speed handpiece under copious water coolant. The ground occlusal surface was examined using magnifying lens (Bausch and Lomb, Opt. Rochester, USA) at 6 × magnification to verify that all enamel was removed. To standardize flat occlusal dentin surface a round bur (komet, Germany) was used to make a central indentation in dentin till the head of the bur disappeared. Then, the occlusal surface was flattened by wet grinding on a bench grinder machine with 240 grit Silicon (SiC) paper (waterproof silicon carbide paper, Atlas brand, England) under copious water coolant. The cervical 1/3 of the buccal surface of the root was flattened by wet grinding on a bench grinder machine with 240 grit SiC paper under copious water coolant. Then, the grinded coronal and root dentin surfaces were finished for 1 min by wet grinding with a 600-grit SiC paper to achieve a standardized smear layer just prior to the bonding procedure [9].

2.3. Restoration of teeth

The teeth were divided into two groups. Group $A_{1:}$ The Single bond universal adhesive system was applied in self-etch technique to the dentine following the manufacturer instructions. The adhesive was applied to the dentin surface and was rubbed for 20 s using a disposable applicator, gently air thinned with compressed air For 5 s until it no longer moved. Then, it was light cured for 10 s using (bluephase C8 LED, Ivoclar vivadent, Liechtenstein, Austria) light curing unit at wavelength of 440–480 nm range. For group A_2: Scotchbond etchant gel was applied to the dentin surface first for 15 s following the manufacturer instructions. Then, the acid was rinsed with water using a three way syringe for 10 s, and excess water was blotted using a piece of gauze until the surface appeared glistening without pooling of water. Immediately after blotting, the adhesive was applied to the etched dentin and was rubbed for 20 s using a disposable applicator, gently air thinned with compressed air for 5 s until it was no longer moved to completely evaporate the solvents. Then, it was light cured for 10 s. Dentine samples were then restored with nanohybrid resin composite, shade A_4 (Filtek™ Z250XT, 3 M ESPE, St Paul, MN, USA, Lot No. N403554). Five increments of resin composite, 1 mm thickness each, were built up and individually light cured and Light activation was performed by exposing the entire composite surface to the (bluephase C8) light curing unit either with continuous mode for 20 s with the full intensity 800 mW/cm^2 or cured in the ramp soft-start mode where the curing starts with 0–800 mw/cm^2 for 5 s and continue afterwards automatically at 800mW/cm2 for another 15 s. The light curing tip was placed to the surface at close range of (0–1 mm).

2.4. Beam preparation for nanoleakage evaluation

Each specimen were placed in the cutting machine (Bronwill, E. McGrath Inc.)vertically and sectioned into a series of 1 mm thick slabs under water cooling then they were mounted horizontally and again sectioned longitudinally into a series of 1 mm thickness. The sectioning was performed using a diamond disc (MTI Corporation, Richmond, USA) of 4 diameter x 0.3 mm thickness with water-resistant Titanium coating. Three sticks were selected from each tooth for ultamorphological detection of nanoleakage patterns and detection of the amount of silver nitrate penetration within the bonded interface. The specimens were stored in distilled water until subjected to the nanoleakage evaluation after 24 h. The sticks were coated with two layers of acid resistant varnish, except for a 1 mm width around the adhesive layer. Specimens were immersed in the prepared 50% ammoniacal silver nitrate (pH = 9.5) solution for 24 h. Specimens were then thoroughly rinsed in distilled water for 5 min and immersed in a photo-developer solutionfor 8 h under fluorescent light in order to reduce the silver ions to metalic silver. After removal from the developing solution, the specimens were placed under running water for 5 min.

2.5. Nanoleakage evaluation

Resin dentin interfaces were analyzed using an environmental scanning electron microscope ESEM (FEI Quanta 200, France), operated with backscattered electron. Ultramorphological nanoleakage patterns were detected and the amount of silver nitrate penetration (Wt%) was analyzed using the EDAX (energy dispersive analytical x-ray). The amount of silver nitrate within the adhesive layer, hybrid layer and the resin tags, in each specimen was measured in an area (199μmx199 μm) at 1500 × magnification directly on the ESEM microscope monitor at 3 regions within three different sticks in each tooth. The mean of the three regions was calculated. The silver nitrate uptake was expressed as a weight percentage of the total area evaluated.

2.6. Statistical analysis

Numerical data were presented as mean and standard deviation (SD) values. Data were explored for normality using Kolmogorov-Smirnov and Shapiro-Wilk tests. Nanoleakage showed non-parametric distribution so, Mann-Whitney *U* test was used to compare between teeth tissues, adhesive systems, curing modes. Kruskal-Wallis test was used to compare between more than two groups. Mann-Whitney *U* test was used for pair-wise comparisons when Kruskal-Wallis test is significant. The significance level was set at $P \le 0.05$. Statistical analysis was performed with IBM – SPSS (IBM Corporation NY, USA) Statistics Version 20 for Windows.

3. Results

The data in Table (1) revealed that, in coronal dentine either with regular curing mode or soft start mode; there was a statistically significant difference between nanoleakage % mean values of the two adhesive techniques at P-value = 0.014 and *P*-value < 0.001,

Table 1
Nanoleakage % of self-etch and total-etch techniques of single bond universal at coronal dentin using different modes of curing.

Curing mode	Adhesive application technique		
	Self-etch	Total-etch	*P*-value
Regular	8.8 ± 2.6	6 ± 1.2	0.014[a]
Soft start	8.1 ± 1.8	3.1 ± 1.3	< 0.001[a]

[a] Significant at P ≤ 0.05.

Table 2
Nanoleakage % of self-etch and total-etch adhesive techniques at root dentine with different curing mode.

Curing mode	Adhesive application technique		
	Self-etch	Total-etch	*P*-value
Regular	10 ± 2.7	8.2 ± 2	0.049[a]
Soft start	8.2 ± 0.7	2.7 ± 0.7	< 0.001[a]

[a] Significant at P ≤ 0.05.

respectively. Self-etch showed statistically significantly higher nanoleakage% mean values compared to total-etch.

The data in Table (2), revealed that, either with regular curing mode or soft start mode; there was a statistically significant difference between nanoleakage% mean values of the two adhesive techniques at P-value = 0.049 and P-value < 0.001, respectively. Self-etch showed statistically significantly higher nanoleakage % mean value than total-etch.

The data in Table (3), revealed that, with self-etch application technique; there was no statistically significant difference between nanoleakage% mean values of the two curing modes at P-value = 0.450. However, with total-etch application technique; there was a statistically significant difference between nanoleakage % mean values of the two curing modes at P-value < 0.001. Regular curing mode showed statistically significantly higher nanoleakage % mean value than soft start mode.

The data in Table (4), showed that, either with self-etch or total-etch adhesive technique; there was a statistically significant difference between nanoleakage % mean values of the two curing modes at P-value = 0.034 and P-value < 0.001, respectively. Regular curing mode showed statistically significantly higher nanoleakage % mean value than soft start mode.

The data in Table (5), revealed that, either with regular curing mode or soft start mode; there was no statistically significant difference between nanoleakage % mean value at coronal and root dentin at P-value = 0.367, and P-value = 0.733, respectively.

The data in Table (6), revealed that, with regular curing mode; there was a statistically significant difference between nanoleakage % mean value at coronal and root dentin at P-value = 0.011. Root dentin showed statistically significantly higher nanoleakage % mean value than coronal dentin. Regarding the means and standard deviation (SD) of nanoleakage with soft start mode, there was no statistically significant difference between nanoleakage% mean value at coronal and root dentin at P-value = 0.307.

Table 3
Nanoleakage % of the two curing modes at coronal dentin.

Adhesive	Curing mode		
	Regular	Soft start	*P*-value
Self-etch	8.8 ± 2.6	8.1 ± 1.8	0.450
Total-etch	6 ± 1.2	3.1 ± 1.3	< 0.001[a]

[a] Significant at P ≤ 0.05.

Table 4
Nanoleakage % of the two curing modes at root dentin.

Adhesive	Curing mode		
	Regular	Soft start	*P*-value
Self-etch	10 ± 2.7	8.2 ± 0.7	0.034*
Total-etch	8.2 ± 2	2.7 ± 0.7	< 0.001*

Table 5
Nanoleakage% at coronal and root dentin with self-etch technique using different curing modes.

Curing mode	Dentine surface		
	Coronal	Root	*P*-value
Regular	8.8 ± 2.6	10 ± 2.7	0.364
Soft start	8.1 ± 1.8	8.2 ± 0.7	0.733

*: Significant at P ≤ 0.05.

Table 6
Nanoleakage % at coronal and root dentin with total-etch technique using different modes of curing.

Curing mode	Dentine surface		
	Coronal	Root	*P*-value
Regular	6 ± 1.2	8.2 ± 2	0.011[a]
Soft start	3.1 ± 1.3	2.7 ± 0.7	0.307

[a] Significant at P ≤ 0.05.

Figures 1–14 show the observation of the EDAX histogram and ESEM photomicrographs resulting from backscattered electron mode at magnification of 1500×. In all the specimens nanoleakage was manifested by silver penetration that had different patterns and different degrees.

4. Discussion

In the present in-vitro study, flat, smooth dentin surfaces were used as the adhesion substrate; such surfaces have been shown to produce less stress on resin-dentin bond than those seen in conventional cavity preparations. It has also been shown that as the ratio of bonded to unbounded surface area increases, the stress developed during polymerization increases, so that more stress would be generated in a cavity preparation than on flat surface [10].

Ammonical silver nitrate solution [Ag(NH3)2]+ 50% (wt/vol) (PH = 9.5) was used because it is the most commonly used material for nanoleakage evaluation as it easily migrates within the interface zone due to its extremely small diameter molecule (0.059 nm) [11]; and [12]. This small size and high reactivity to stain after binding tightly to any exposed collagen fibrils that are not enveloped by the adhesive resin makes sliver nitrate the most appropriate agent to detect the nanoporosities within the hybrid layer [13]. Moreover, silver nitrate induces an electron microscopic measurable contrast providing a sharp picture of the degree of penetration into the interface. Following its penetration, it has the potential to immobilize, which prevents further penetration during specimen preparation [14].

There is a possibility, however, that the acidic conventional silver nitrate (PH = 3.4) may over-demineralize dentin and create artificial paths along the dentin-adhesive interface, this may produce artifactual microporosities that give rise to false positive results which may mask the nanoleakage results [15]. Therefore, in this study the modified silver staining technique was used. It utilizes 50 wt% ammonical silver nitrate having PH measured at 9.5 aiming to eliminate the possibility of

Element	Wt %	At %
O K	36.39	55.69
SiK	22.80	19.87
P K	14.63	11.56
AgL	08.11	01.84
CaK	18.08	11.04

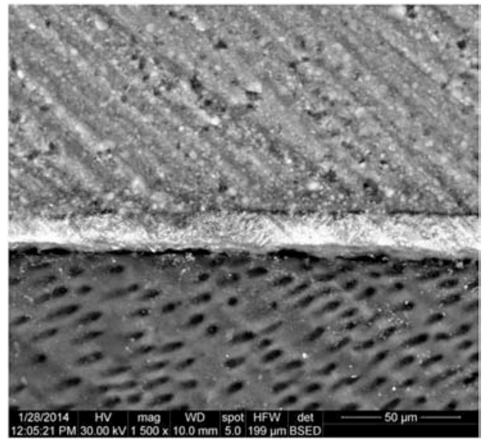

Fig. 1. EDAX element analysis and ESEM photomicrograph of single bond universal nanoleakage for self-etch technique with regular mode of curing in coronal dentine. The nanoleakage pattern showed a thick continuous silver deposition along the resin-dentin interface.

Element	Wt %	At %
O K	40.98	61.78
SiK	17.89	15.36
P K	12.67	09.87
AgL	10.95	02.45
CaK	17.51	10.54

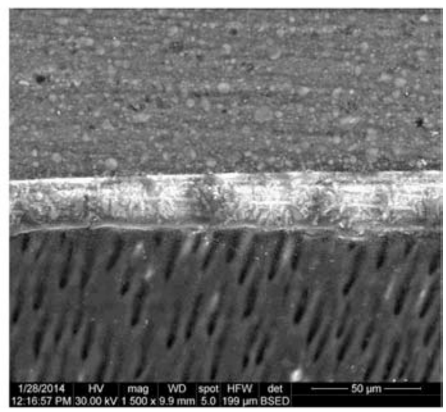

Fig. 2. EDAX element analysis and ESEM photomicrograph of nanoleakage of single bond universal using self-etch technique and regular curing mode in root dentine. The nanoleakage showed silver deposition resembling the water trees along the resin dentine interface.

Element	Wt %	At %
C K	37.83	55.34
O K	25.07	27.54
SiK	10.14	06.34
P K	07.43	04.21
AgL	07.25	01.18
CaK	12.28	05.38

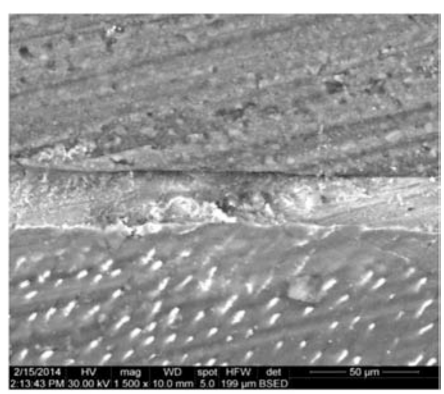

Fig. 3. EDAX element analysis and ESEM photomicrograph of nanoleakage of single bond universal self-etch technique and soft start curing mode in coronal dentine. The nanoleakage pattern showed dense silver deposition along the resin-dentin interface and within the dentinal tubules.

dentin dissolution at the hybrid layer by using less acidic silver nitrate solution [6–8].

Nanoleakage evaluation was performed using SEM in conjunction with EDAX which enables distinct images to be captured together with sensitive and accurate analysis. The concurrent EDAX analysis was carried out in order to identify the existence of metallic silver particles [16]. When SEM was used alone for nanoleakage examination, with secondary and backscattered electron imaging, erroneous interpretations were common. EDAX, in contrast, provides accurate quantitative analysis and distribution for the various existing elements [17].

The results of this study showed that regardless of the mode of curing used in coronal and root dentine, single bond universal in the self-etch technique recorded significantly higher nanoleakage mean values than the total-etch technique. The results are in agreement with [18] who studied nanoleakage within the hybrid layer for etch-and-rinse and self-etch strategies of universal adhesives, and reported that, with regard to the etch-and-rinse approach, the universal adhesives showed improved bond strength in comparison with the self-etch strategy. This may be due to the presence of the smear layer which constitutes a true physical barrier and makes it extremely difficult for

Element	Wt %	At %
C K	45.25	61.99
O K	23.78	24.46
SiK	10.58	06.20
P K	05.37	02.85
AgL	06.48	00.99
CaK	08.54	03.50

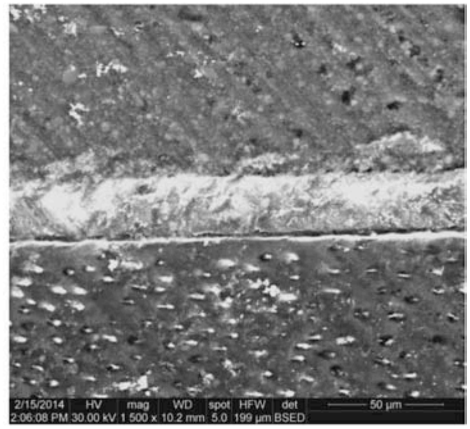

Fig. 4. EDAX element analysis and ESEM photomicrograph of nanoleakage of single bond universal self-etch technique and soft start curing mode in root dentie. The nanoleakage pattern showed continuous heavy silver deposition along the resin dentine interface.

Element	Wt %	At %
O K	35.34	54.80
SiK	13.70	12.10
P K	17.48	14.00
AgL	04.17	00.96
CaK	29.32	18.15

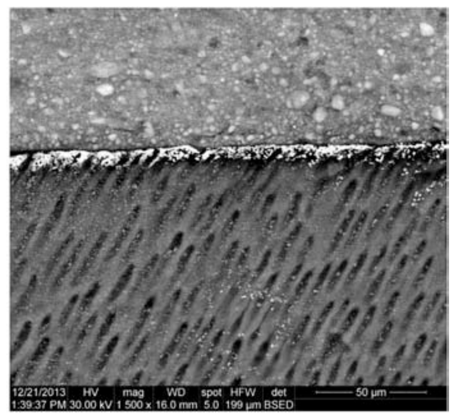

Fig. 5. EDAX element analysis and ESEM photomicrograph for nanoleakage of single bond universal total-etch technique and regular curing mode in coronal dentine. The nanoleakage pattern showed less dense silver deposition along the resin dentine interface than the self etch technique.

Element	Wt %	At %
O K	39.34	58.72
SiK	18.15	15.44
P K	15.51	11.96
AgL	05.88	01.30
CaK	21.12	12.59

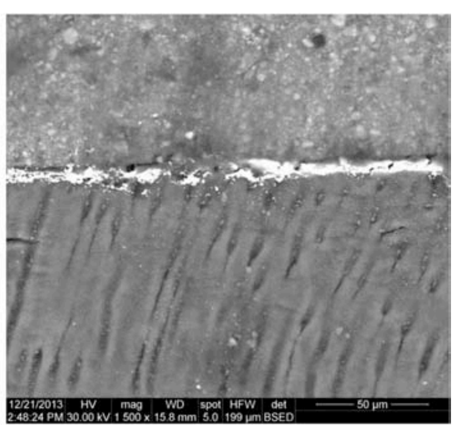

Fig. 6. EDAX element analysis and ESEM photomicrograph for nanoleakage of single bond universal total-etch technique and regular curing mode in root dentine. The nanoleakage pattern showed more dense silver deposition forming a line of varying thickness along the resin dentine interface when compared to the coronal dentine.

the bonding and hybrid layer formation to be fully integrated with the dentine [19]. After preliminary etching with phosphoric acid in etch & rinse approach, the smear layer is removed and superficial dentine is demineralized. This increases impregnation by the adhesive, allowing the creation of a well impregnated hybrid layer [20,21]; and [22].

These results also are in agreement with [23] who reported that, the etch-and-rinse approaches tested for the single bond universal resulted in immediate bond strength compared to the self-etch mode. These results also support previous findings showing that when phosphoric acid (used to etch peripheral enamel according to the selective enamel etch technique) extends to dentine, the application of a self-etch adhesive results in higher micro-tensile bond strength compared with its application on smear layer-covered dentine [24].

This is also in agreement with several in vitro studies on one-step self-etch adhesives using Transmission Electron Microscopy (TEM) and the Field Emission- SEM (FE-SEM), most of those studies utilized only short-term periods of observation and reported that, although a certain amount of nanoleakage was observed in all the tested groups, it was more pronounced for the self-etch adhesives when compared to the total-etch adhesives [25–30]; and [31]. In particular, the lack of a hydrophobic bonding resin in one-step self-etch adhesive formulations has been demonstrated to reduce bond stability over time, because the bonded interfaces behave as semi-permeable membranes allowing the movement of water across them and expediting hydrolytic degradation

Element	Wt %	At %
C K	28.49	44.49
O K	26.50	31.07
P K	11.35	06.87
AgL	02.60	00.45
CaK	18.14	08.49

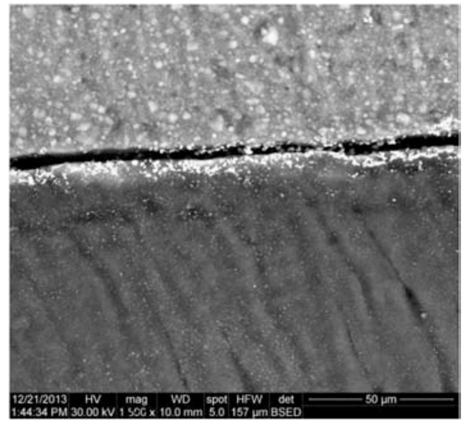

Fig. 7. EDAX element analysis and ESEM photomicrograph for nanoleakage of single bond universal total-etch technique and soft start curing mode in coronal dentine. The nanoleakage pattern showed silver deposition in the form of spots at the resin dentine interface.

Element	Wt %	At %
C K	47.61	64.36
O K	20.35	20.65
P K	07.01	03.68
AgL	03.03	00.46
CaK	10.76	04.36

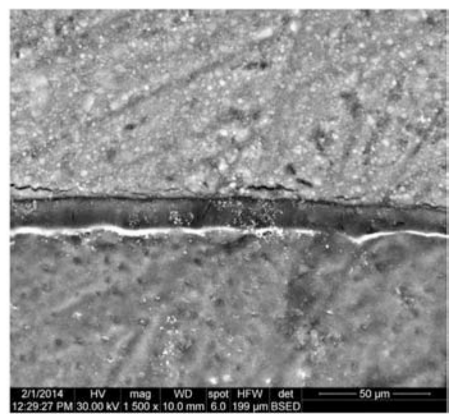

Fig. 8. EDAX element analysis and ESEM photomicrograph of nanoleakage of single bond universal total-etch technique and soft start curing mode in root dentine. The nanoleakage pattern showed thin, uniform, line of silver deposition at the base of the resin dentine interface.

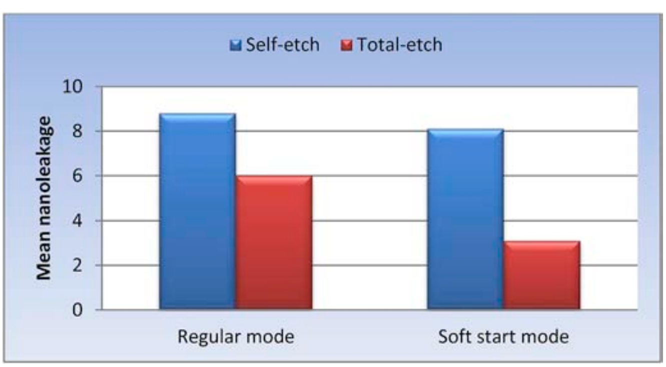

Fig. 9. Nanoleakage% of self-etch and total-etch techniques at coronal dentin with different curing modes.

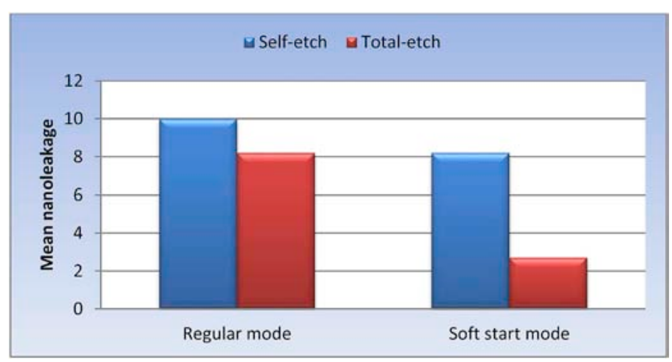

Fig. 10. Nanoleakage % mean values of self-etch and total-etch at root dentin with different curing modes.

[6–8,32]; and [33]. The adhesive permeability of one-step adhesives has also been correlated with suboptimal curing of the hydrophilic acidic resin monomers blended into their formulations, which was associated with phase separation phenomena [34]; and [35]. This holds true specifically that other studies performed by Refs. [36–38]; and [39] considered the type of adhesive system as influential in the leakage of composite restorations as different nanoleakage patterns were observed with different adhesive systems.

However [40], and [41] found no statistically significant difference in nanoleakage between dentin bonding agents and this may be due to the difference in methodology as they applied thermocycling protocol and the difference in the adhesive material as they used two step self etch system. The results are also not constant with [23] who found that nanoleakage expression was lower when Single bond Universal was applied in the self-etch mode compared to the etch-and-rinse this is may be due to difference in the storage period as they made their investigations after 6 month and 1 year of storage in artificial saliva while the storage protocol was not applied in this study. This is also may be due to presence of 10-MDP in Single bond Universal which results in additional chemical bonding which may contribute more to the stability and longevity of the bond rather than its immediate bond strength [42]; and [43].

The results of this study showed that, regular continuous curing mode showed statistically significantly higher nanoleakage % mean

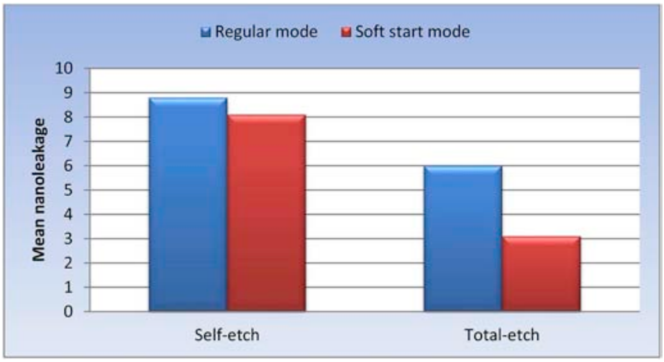

Fig. 11. Nanoleakage % mean values of the two curing modes at coronal dentin.

Fig. 12. Nanoleakage % mean values of the two curing modes at root dentin.

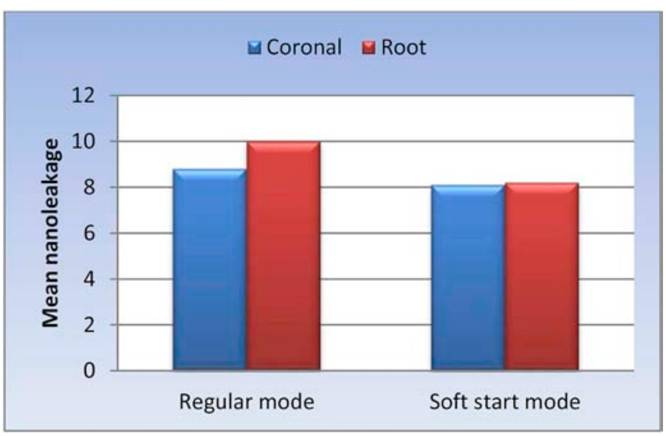

Fig. 13. Nanoleakage% mean values at coronal and root dentin with self-etch technique using different curing modes.

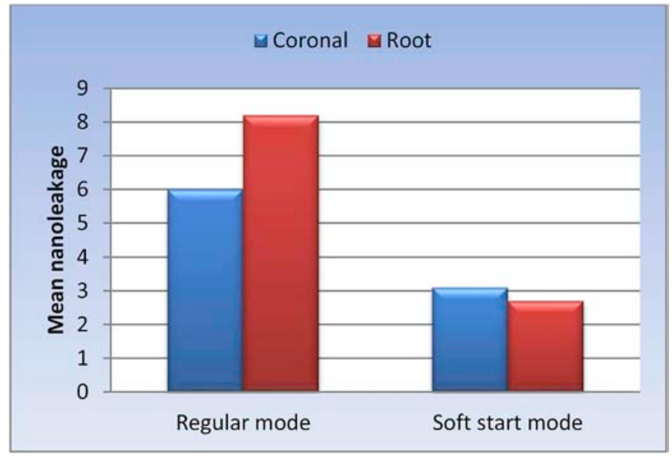

Fig. 14. Nanoleakage% mean values at coronal and root dentin with total-etch technique using different modes of curing.

value than the ramp soft start mode in coronal and root dentine with total etch technique. These results are in agreement with [44] who demonstrated that the ramp technique reduced gap formation between the resin composite and dentin, compared to the standard curing mode [45].) also reported that the ramping technique significantly reduced the maximum polymerization shrinkage force and the rate of force development, compared to the standard curing mode. This may be due to the relieve of Polymerization shrinkage stress by flow. A slower curing process may allow more time for flow to occur. The initial low-light intensity could facilitate a certain degree of shrinkage stress relaxation before the system reaches the vitrification stage and while in the pre-gel phase. Laboratory research showed that soft-start polymerization partially relieves shrinkage stress and achieves improved marginal integrity of the restoration [46]. On the other hand; some researchers have showed that the stress relief advantage of soft-start curing might be in fact a result of reduction in the degree of conversion of the resin [47]; and [48]. The purpose of this system is to reduce the speed of conversion and increase the gel phase resulting in viscous flow and better adaptation of composites to the cavity walls [44]; and [49].

Results also showed that, in coronal dentine with self etch technique there was no statistical significant difference between the curing modes. These results are in agreement with [50]; and [51] who reported no significant improvement in cavity adaptation with different modes of curing. These results are also in harmony with [52] who studied the effect of different curing modes on microleakage of composite using self etch adhesives and found no significant differences in the microleakage among the light-curing modes used. This is may be due to material factor. It has been demonstrated that the filler contents, elastic modulus, photo-initiators, matrix resins and adhesive systems greatly affect marginal integrity [29,53]; and [51].

Recent studies revealed that adhesive permeability well correlates with the presence of un-reacted monomers within the hybrid layer. In particular one-step self-etch adhesives revealed the highest permeability associated with the lowest degree of cure (DC) when compared to the unsimplified multi-step systems [34,54]; and [6–8]. This may reduce bond longevity due to elution of un-reacted monomers. The final consequence of this process is the formation of a porous hybridoid structure along the adhesive interface with reduced sealing ability. Also reduced DC of the simplified adhesive films reduces their bonding effectiveness [55]; and [54].

In this study comparison between coronal and root dentine revealed that, there was no statistically significant difference between nanoleakage mean values at coronal and root dentine in self-etch mode regardless of the mode of curing used. However, with total etch technique and regular curing mode root dentine showed a statistically significantly higher nanoleakage mean values than coronal dentine. These results are in agreement with [56]; and [57] who demonstrated that bond strength of total-etch adhesives may be affected by the different locations of dentin, while a self-etch system produces consistent bonding irrespective of dentin location or tubule orientation. They explained that the higher bond strength values attained by the self-etch systems they used might be related to the exposition of a larger mineralized surface area ready for bonding with the10-MDP molecule, thus creating a stronger chemical bond.

Similar findings were observed by Ref. [21] who explained this by the fact that, bonding to parallel-cut dentin implies that the numbers of resin tags, which have been postulated to contribute to bond strength are reduced, so a decrease in bond strengths in the case of the total-etch bonding system is expected. This also could be explained by the findings of [57] who observed that the average number of tubules present in deep coronal (44,243 tubules/mm2) and (42,360 tubules/mm2) in the cervical thirds of radicular dentin. It is well known that, if a lower number of dentinal tubules are present, or if the collapse of the collagenous fibrilar network occurs, preventing monomer diffusion, bond strength values can fall by 90% of their maximal level [58].

However, little information is available on the influence of different dentin morphologies and adhesive techniques on bond strength. Efficacy of current adhesive systems is regularly evaluated by their ability to bond to coronal dentin; nevertheless developments in periodontology significantly increased the demand for restoration of root dentin defects and further investigations are needed.

The polymerization shrinkage stresses are critical for the microscopic integrity of adhesive bond to dentin [59]. Polymerization shrinkage stress can be relieved by flow. A slower curing process may allow more time for flow to occur [60,61]; and [45]. Numerous approaches have been proposed to minimize the shrinkage stress by manipulation of curing protocols and placement techniques [62]. Among these approaches, soft-start curing (which can be either in a step or a ramp mode) and pulse curing have attracted extensive investigations [63]; and [48]. Therefore, in this study the effect of soft-start curing protocol on nanoleakage was investigated. To minimize also the deleterious effect of shrinkage stresses on the marginal integrity of the composite restorations, the incremental filling technique was used in this study. Researchers have shown that increment should be no larger than 2 mm to provide uniform and maximum polymerization [64]; and [65].

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.fdj.2018.04.001.

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