Effect of modulated photoactivation of bulkfill composite on microleakage in fluorosed and nonfluorosed teeth: A confocal laser scanning microscopy study

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Abstract

Aim: The aim of this study is to compare the microleakage of bulkfill composite activated by modulated photoactivation between fluorosed and nonfluorosed teeth using the confocal laser scanning microscope.

Methodology: One hundred and twenty intact human premolar teeth with Thylstrup and Fejerskov index fluorosis index 0–4 were stored in 0.5% thymol at the room temperature until further use. Standardized Class V preparations of 2 mm depth, 3 mm width, and 2 mm height were prepared on the buccal surface. The cavities were etched with 37% phosphoric acid, rinsed and primed with Tetric N bond, cured for 20 s with Quartz Tungsten Halogen (QTH) variable intensity light-curing unit spectrum-800 operating at 450 mW/cm². Later, bulk fill composite was placed in the cavity and cured. Depending on the curing mode used, all the fluorosed and nonfluorosed teeth were divided into three subgroups each (n = 20) – Conventional light curing, stepped curing, and pulse delayed curing. All samples were stored in distilled water at the room temperature for 24 h and subjected to 500 thermocycles. The prepared teeth were placed in 0.6% rhodamine solution for 48 h; sectioned longitudinally using a hard-tissue microtome and scanned under a confocal laser scanning electron microscope. Data were analyzed using the one-way ANOVA, Wilcoxon signed-rank test, and Kruskal–Wallis test.

Results: Significant differences were observed between fluorosed and nonfluorosed groups. Intragroup comparisons showed significant differences between fluorosed step and conventional subgroups.

Conclusion: Fluorosed teeth had higher microleakage values than nonfluorosed teeth. Pulse-delayed subgroup had the least microleakage to that of conventional and stepped curing subgroups, in both fluorosed and nonfluorosed groups.

Keywords: Bulkfill composite; confocal laser scanning electron microscopy; fluorosis; microleakage

INTRODUCTION

Amalgam restoration was the material of choice for more than 100 years. To overcome drawbacks such as esthetics and corrosion, use of resin-based composites (RBC) is advocated.[1] Despite having good physical properties such as high compressive strength, excellent translucency, and ease of application, the main drawbacks are polymerization shrinkage and stress, resulting in internal micro cracks, bonding agent separation, and marginal microleakage.[2]

Bulkfill RBC introduced has better properties of curing 4–5 increments in single step and allowing better adaptation of cavity walls.[3] Fillers in bulk fill composites are generally increased in size involving a lower filler matrix interphase,
reduced light scattering, and increased transmittance for blue light in depth.\textsuperscript{[4]}

Anticariogenic and positive effects of fluoride on teeth and carious lesions were proved in dentistry. However, excessive intake of fluoride results in dental fluorosis which manifests as varying degrees of discoloration and pitting. Fluorosed enamel is characterized by an outer hypermineralized surface and porosity of subsurface layer which necessitates prolonged etching time to acid etch.\textsuperscript{[5]}

Microleakage is marginal permeability to bacterial, chemical, and molecular invasion at resin dentin interface that is determined by many \textit{in vitro} techniques. Among these methods, confocal laser scanning electron microscopy (CLSM) is a technique used for visualizing subsurface tissue characteristics.\textsuperscript{[6]}

The concept of soft-start polymerization proposes to increase the pregelation time enabling a slower rate of conversion and better flow of resin with decrease in contraction stresses. It may be divided into three separate techniques: Stepped curing, ramped curing, and pulse-delayed curing.\textsuperscript{[7]} Spectrum 800 (Dentsply Sirona, Germany) is a variable intensity halogen curing light device in which the intensity can be adjusted from 300 mW/cm\textsuperscript{2} up to 800 mW/cm\textsuperscript{2}.\textsuperscript{[8]}

This study aims to evaluate the comparative effect of modulated photoactivation on microleakage of bulkfill composite restoration in fluorosed and nonfluorosed teeth using CLSM.

**METHODOLOGY**

Approval for the study was obtained from the institutional review board, and samples were collected from the department of oral and maxillofacial surgery and private dental clinics. Intact one hundred and twenty human permanent premolar teeth without any caries, cracks, fractures, restorations, and fluorosed teeth with Thylstrup and Fejerskov index (TFI) index 0–4 were included in this study. The exclusion criteria were deciduous teeth, teeth with any caries, cracks, fractures, restorations, and fluorosed teeth with Thylstrup–Fejerskov index (TFI) index 5–9. The samples were divided into fluorosed and nonfluorosed groups ($n = 60$), stored in 0.5% thymol at the room temperature until further use.

Standardized Class V preparations were made on the exposed buccal surface with the dimensions of 2 mm depth, 3 mm width, and 2 mm in height with \# 245 bur in air/water cooled high-speed hand piece. The cavity on each tooth was etched for 15 s, primed with Tetric N bond and cured for 20 s with QTH variable intensity light-curing unit spectrum-800 operating at 450 mW/cm\textsuperscript{2}. Later, Tetric N Ceram bulkfill composite was placed in the cavity and cured for 40 s.

Depending on the curing mode used, all the fluorosed and nonfluorosed teeth were divided into two groups of three subgroups each ($n = 20$):

**Nonfluorosed teeth (Group 1):** TFI-0
- Group 1A – Conventional light curing
- Group 1B – Stepped curing
- Group 1C – Pulse-delayed curing.

**Fluorosed teeth (Group 2):** TFI 1–4
- Group 2A – Conventional light curing
- Group 2B – Stepped curing
- Group 2C – Pulse-delayed curing.

Finishing and polishing were done to all teeth with Shofu composite polishing kit. All samples were stored in distilled water at the room temperature for 24 h. They were further subjected to 500 thermocycles with 30 s bath at temperature of 5°C and 55°C and dwell time of 10 s in a resting bath at 24°C. Then, the teeth were covered with nail polish varnish except for one mm around the margins of the restoration to prevent dye penetration in other areas. The prepared teeth were placed in 0.6% rhodamine solution for 48 h; then rinsed under running water to remove excess dye, air dried, and mounted further into acrylic moulds.

The teeth were sectioned longitudinally in a bucco-palatal plane using a hard-tissue microtome and were scanned under a CLSM to measure the extent of microleakage. A 1–6 scoring system was used to describe the severity of dye penetration:
- 1 – No dye penetration
- 2 – Dye penetration within one-half of occlusal or gingival wall
- 3 – Dye penetration extending to the end of occlusal or gingival walls
- 4 – Dye penetration through the gingival or occlusal wall to one-third of the axial wall
- 5 – Dye penetration through the gingival or occlusal wall to two-thirds of the axial wall
- 6 – Dye penetration throughout the axial wall.

Data were recorded [Figure 1a-f] and analyzed using the one-way ANOVA, Wilcoxon signed-rank test, and Kruskal–Wallis test at the 0.05 level of significance ($P < 0.05$).

**RESULTS**

Statistical analysis showed that there were significant differences between fluorosed and nonfluorosed groups. Microleakage mean that rank values were the highest
for conventional light curing subgroups and least for the pulse-delayed light-curing subgroups.

There were statistical significant intragroup differences in the fluorosed group except between fluorosed step subgroup and conventional subgroup. In the nonfluorosed intragroup comparisons, statistical significant differences were observed between the subgroups [Table 1].

Intergroup comparison between fluorosed and nonfluorosed groups showed statistical significant differences between nonfluorosed conventional and fluorosed conventional sub groups and nonfluorosed pulse delayed and fluorosed pulse-delayed subgroups. However, no statistical significant differences were observed between nonfluorosed stepped and fluorosed stepped sub groups [Table 2].

**DISCUSSION**

Dental fluorosis of enamel is caused by successive exposures to high concentrations of fluoride during tooth development, leading to lower mineral content and increased porosity. It not only affects adversely both teeth and bones, but also damages DNA and chromosomes. Esthetic changes in permanent dentition are of greatest concern occurring in children between 20 and 30 months of age who are excessively exposed to fluoride.[9]

Fluorosed enamel is characterized by an outer hypermineralized acid resistant layer and retention of porous enamel in the areas of sub surface. Water and enamel secretory proteins are retained in areas of porous enamel due to the effects of excessive fluoride on ameloblasts which makes bonding to fluorosed enamel challenging. Teeth with high concentrations of fluoride are acid resistant and necessitates extended time for etching.[10]

Thylstrup and Fejerskov[11] in the year 1978 developed TFI index based on 10-point ordinal scale, closely to the histological changes which occur and fluoride content found in the enamel. It is more appropriate for the use in clinical trials or analytical epidemiologic studies, primarily because teeth are dried and fluorosis can be identified in its milder forms. Only teeth with TFI index 1–4 were used in accordance to a study done by Al-Sugair and Akpata.[12]

Marginal integrity is essential to increase the longevity of any restoration.[13] This integrity is compromised, when microleakage occurs resulting from polymerization shrinkage. The most common cause of failure of direct posterior composite restorations is polymerization shrinkage. It is dependent on the various aspects such as the boundary conditions, the material’s formulation, and the amount of material in the polymerization reaction.[2] This occurs because monomer molecules are converted into a polymer network, and therefore, exchanges Vander walls spaces into covalent bond spaces, creating contraction stresses in the resin composite leading to microleakage.[14]
Stress reduction is very important in the 1st s after the start of polymerization, and therefore, retardation in attaining gel point results in a higher viscous point with increased flow subsequently reducing polymerization stresses. The control of such stresses improves the bond strength and marginal seal of composite resin to dentin. Various approaches to compensate polymerization stresses can be by the use of bulkfill composite resins, low viscosity bonding agents enhancing the configuration factor, and modified light-curing modes.

Bulkfill RBC rheology, elasticity, and low polymerization shrinkage stress reduce micro leakage, postoperative sensitivity, and secondary caries. Tetric N Ceram nanohybrid bulk fill RBC has resin matrix composed of Bis-GMA, Bis-EMA, and urethane dimethacrylate monomer, patented light initiator Ivocerin, and advanced composite fillers barium aluminum silicate glass with two different mean particle sizes. Filler content is approximately 61% (volume) and 17% polymer fillers or “isofiller.”

C-factor is defined as the ratio of the bonded surface area to the unbonded or free surface area. The box-shaped cavity design was recommended to reduce polymerization stresses and to resemble most closely the clinical situation resulting in a C-factor of about 3 similar to the study done by Küçüksen and Sönmez.

Nanofilled dentin adhesives (Tetric N Bond, Universal) introduced in year 2015 has been claimed as 8th-generation bonding agent. It is a single component, light-cured “mild-etching” adhesives with a pH of approximately 2.5–3.0 which is based on the combination of monomers of hydrophilic, hydrophobic, and intermediate nature which allows bridging the gap between the hydrophilic tooth substrate and the hydrophobic resin restorative.

Spectrum 800 (Dentsply Sirona, Germany) variable intensity halogen curing light device offers the optimum combination of features, performance, and reliability for the polymerization of all light-curing materials. The intensity can be adjusted from 300 mW/cm² up to 800 mW/cm² by depressing the intensity up/down adjust buttons. For routine-curing needs, the light power intensity should be adjusted to ~550 mW/cm². It also has preset run times of 10 to 60 s programmed in 10 s increments and built in radiometer on base unit.

Microleakage is determined by many in vitro techniques such as staining, bacterial activity, air pressure, chemical agents, markers, neutron activation analysis, radiisotope, ionization, scanning electron microscopy, and CLSM. In this study, CLSM used is a recent three-dimensional nondestructive technique for visualizing subsurface tissue features up to 100 µm in size without sample destruction. Stain spread is avoided which is caused by specimen sectioning and also avoids polishing artifacts that exaggerate dye penetration.

A stepped program emits a low irradiance for 10 s, to contour and shape the restoration in occlusion and then increases immediately to maximum value for the duration of exposure allowing substantial relaxation of polymerization stresses. In pulse-delayed method, a low-energy pulse is given initially of 200 mW/cm² for 3 s followed by a waiting period of 3–5 min for strain relief and final cure obtained by high-intensity exposure of 500 mw/cm².

Pulse-delayed curing subgroup has shown least microleakage and statistical significant differences with the other subgroups. Stepped curing subgroup had an intermediate microleakage due to low irradiance initially but lacking the dark phase between irradiances reducing polymer chain relaxation time. Both pulse delayed and stepped curing modes have more pregel phase allowing polymer chain relaxation and compensating volumetric reduction. Moreover, soft start polymerization reduces the marginal leakage, improves marginal adaptation, and has lower degree of conversion. Another reason for the success of using pulse-delayed curing is the use of bulk-filled resins which have high density of fillers, less organic matrix, and enhanced flow ability, thereby reducing water sorption.

Conventional curing subgroup had highest microleakage due to the reason that continuous high-intensity curing generates stresses leading to bond failure and increase in micro leakage. Furthermore, stresses occurring during postgelation phase are not relieved by material flow and tend to develop at the tissue restoration interface.

The important limitation of the study is that microleakage only reveals a minor aspect of adhesion. Although significant differences were found among the experimental groups, the clinical differences remain questionable. In our study, bonding to fluorosed teeth was significantly lower than to nonfluorosed teeth due to the presence of acid resistant layer and subsurface porosities. Moreover, bonding to dentin and cementum in comparison to enamel is less due to the presence of tubular structure of dentin and more amount of organic collagen phase; hence, resulting in a weaker bond. In addition, thin enamel at cervical margin, abfraction and debonding of restoration at this margin may occur. Hence, the methods to improve bond strength in fluorosed teeth, dentin, and cementum need to be developed.

Another factor that needs consideration is the presence of voids in the final bulk fill restoration which may impair strength, initiate cracks, and hence, further studies should be modified to evaluate eliminating voids.
CONCLUSION

Within the limitations of the study, fluorosed teeth had higher microleakage values than the nonfluorosed teeth. Among the subgroups, conventional curing subgroup had the highest microleakage followed by the stepped curing subgroup and pulse delayed subgroup had the least microleakage. Soft-start polymerization modes enabled a slower conversion rate and better resin flow with decreased contraction stresses. The real effect of long-term improvement in marginal integrity and decrease in microleakage must be supported with further clinical in vivo studies and other future researches. Definitive results of stress reduction with improved marginal adaptation must be concluded with the use of pulse-delayed exposure method.

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Conflicts of interest
There are no conflicts of interest.

REFERENCES

1. Deliperi S, Bardwell DN. An alternative method to reduce polymerization shrinkage in direct posterior composite restorations. J Am Dent Assoc 2002;133:1387-98.
2. Swapna MU, Koshy S, Kumar A, Nanjappa N, Benjamin S, Nainan MT. Comparing marginal microleakage of three bulk fill composites in class II cavities using confocal microscope: An in vitro study. J Conserv Dent 2015;18:409-15.
3. Ilie N, Bucuta S, Draenert M. Bulk fill resin based composites: An in vitro assessment of their mechanical performance. Oper Dent 2013;38:618-625.
4. Chesterman J, Jowett A, Gallacher A, Nixon P. Bulk-fill resin-based composite restorative materials: A review. Br Dent J 2017;222:337-44.
5. Küçükeşmen C, Sönmez H. Microleakage of Class-v composite restorations with different bonding systems on fluorosed teeth. Eur J Dent 2008;2:48-58.
6. Usha HL, Kumari A, Mehta D. Comparing microleakage and layering methods of silorane-based resin composite in Class V cavities using confocal microscopy: An in vitro study. J Conserv Dent 2011;14:164-8.
7. Chan DC, Browning WD, Frazier KB, Brackett MG. Clinical evaluation of the soft-start (pulse-delay) polymerization technique in Class I and II composite restorations. Oper Dent 2008;33:265-71.
8. Roy KK, Kumar KP, John G, Sooraparaju SG, Nujella SK, Sowmya K. A comparative evaluation of effect of modern-curing lights and curing modes on conventional and novel-resin monomers. J Conserv Dent 2018;21:68-73.
9. Marure FS, Mahamuni A, Ambekar AS, Kangane S, Joshi Y, Khandapure C. Orthodontic bracket bonding challenge for fluorosed teeth. J Int Oral Health 2016;8:476-80.
10. Bakhadher W, Talic N, Korayem M, Ahmira AA, Bamusa B, Bajafar S. Effects of dental fluorosis on bond strength of orthodontic bracket, a review of in vitro studies. Acta Sci Dent Sci 2019;3:78-82.
11. Shyam R, Manjunath BC, Kumar A, Narang R, Ghanghas M, Singh S. Indices for measuring dental fluorosis: A review. Int J Curr Pharm Res 2016;3:1982-6.
12. Al-Sugair MH, Akpata ES. Effect of fluorosis on etching of human enamel. J Oral Rehabil 1999;26:521-8.
13. Majety KK, Pujar M. In vitro evaluation of microleakage of Class II packable composite resin restorations using flowable composite and resin modified glass ionomers as intermediate layers. J Conserv Dent 2011;14:414-7.
14. Radhika M, Sajjan GS, Kumaraswamy BN, Mittal N. Effect of different placement techniques on marginal microleakage of deep Class II cavities restored with two composite resin formulations. J Conserv Dent 2010;13:9-15.
15. Carvalho RM, Pereira JC, Yoshiyama M, Pashley DH. A review of polymerization contraction: The influence of stress development versus stress relief, Oper Dent 1996;21:17-24.
16. Ernst CR, Brand N, Frommator U, Rippin G, Willershausen B. Reduction of polymerization shrinkage stress and marginal microleakage using soft-start polymerization. J Esthet Restor Dent 2003;15:93-103.
17. Malhotra N, Kundabala M, Shaashirashmi A. Strategies to overcome polymerization shrinkage – Materials and techniques. A review. Dent Update 2010;37:115-25.
18. Sachdeva B, Dua P, Mangla R, Kaur H, Rana S, Butal A. Bonding efficacy of 5th, 6th, 7th & 8th generation bonding agents on primary teeth. Int J Dent Med Sci 2018;17:61-6.
19. Enggist L, Tetico N Bond Universal[Brochure], Schaan, Liechtenstein, Ivoclar Vivadent AG; 2015.
20. Al-Habdan AA. Review of microleakage evaluation tools. J Int Oral Health 2017;9:141-5.
21. Malhotra N, Mala K. Light curing considerations for resin based composite materials: A review part I. Compend 2010;31:498-505.
22. Suh BI. Controlling and understanding the polymerization shrinkage-induced stresses in light-cured composites. Compend Contin Educ Dent Suppl 1999; 25:S34 41.