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Evaluating the marginal seal of a bioactive restorative material activa Bioactive and two bulk fill composites in class II restorations: an *in vitro* study

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Abstract
Aims and objectives: To evaluate and compare the microleakage in class II cavities restored with a new bioactive restorative material (ACTIVA bioactive) and two different bulk fill composites (Tetric-N-Ceram bulk fill and Filtek bulk fill).
Materials and methods: 60 freshly extracted mandibular molars were selected and standard class II cavities were made on mesial surface of each tooth. Samples were then randomly divided into 3 groups i.e. Group I- Activa bioactive (n=20), Group II- Tetric-N Ceram (n=20) and Group III- Filtek Bulk fill (n=20) and restored with the respective composite material. After storage and thermocycling, the samples were subjected to dye extraction method for microleakage analysis using a spectrophotometer.
Results: Results demonstrated that the bioactive restorative material Activa Bioactive (Group I) and Tetric-N-Ceram bulk fill (Group II) showed least microleakage followed by Filtek bulk Fill composite (Group III).The difference between Group I and Group II was not statistically significant (p<0.05) whereas the Group I and Group III and Group II and Group III was statistically significant (p<0.05).
Conclusion: Based on the results of the study it can be concluded that Activa bioactive and Tetric - N-Ceram showed least microleakage.

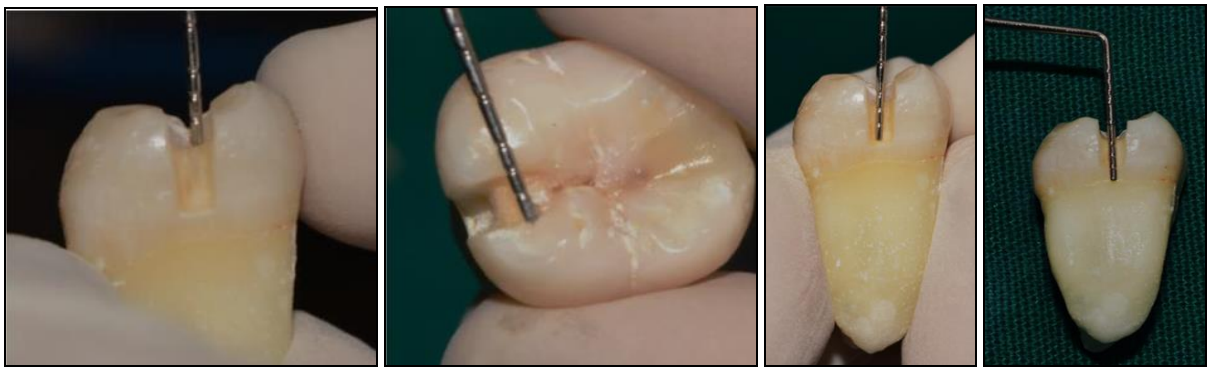
Keywords: Marginal microleakage, Class II restoration, activa bioactive, bulk fill composites

Introduction
The success rate in restoring a class II cavity lesion depends upon the type of dental material used for restoration as well as the operator's skill in performing the procedure [1]. In recent years, the popularity of tooth colored restorative material has led to a rapid increase in the use of composite resins. Composites were introduced in the 1960's and since then have undergone a lot of research and development, starting from the conventional composite resins which allow placement of 2mm increment to the most recent being the bulk fill composites that allow placement of 4-5mm increment at a time thereby making the restorative procedure less time consuming [2, 3]. The clinician's main concern when placing direct posterior resin-based composite restorations is the polymerization shrinkage stress, which would result in marginal gap formation contributing to micro leakage and thus a source of postoperative sensitivity, pulpal inflammation and recurrent caries [1, 4-6].
The incremental layering technique and use of low-modulus intermediate liner material such as flowable composites have been suggested to reduce this shrinkage [7-8]. The most recent attempt to reduce micro leakage is to use Bulk fill composites as they use new resin monomers with novel chemistries (low polymerization shrinkage) to compensate shrinkage stress [9].
Yet another bulk fill restorative material which is also a bioactive material is Activa Bioactive Restorative (Pulpdent Corporation, Watertown). It is the first bioactive composite (with advantages of Glass ionomer cement) with an ionic resin matrix, a shock-absorbing resin component and bioactive fillers. The manufacturer's claim that it is a Dynamic "Smart" Material which is esthetic, chemically bonds to teeth and seals teeth against microleakage [10].
However, the potential for development of internal and marginal discrepancies exists with bulk placement leading to gap formation and microleakage [11].
Therefore the aim of the present invitro study was to evaluate and compare the microleakage of a new bioactive restorative material (Activa Bioactive) and two bulk fill composites

(Tetric N Ceram & Filtek Bulk Fill) at the gingival restoration-dentin interface in class II restorations. The null hypothesis tested was there is no difference in the microleakage values of the 2 bulk fill composites and Activa Bioactive restorative.

Material and methods: Sixty intact, non-carious, unrestored human mandibular first molars, extracted for therapeutic reasons were collected for the study and the teeth were hand scaled and kept in 0.05% thymol solution at 37°C prior to use. The teeth were inspected under a stereomicroscope for any developmental defects and preexisting cracks. 60 standard class II cavities of the following dimensions: Occlusal depth = 2mm, Bucco-lingual width = 2mm and Axial depth = 4mm with the gingival cavosurface margin 1 mm above the CEJ were prepared on the mesial surface of each sample using a 245 bur under copious water cooling, and the cavities were finished with finishing diamond points (Figure 1). The teeth were then randomly divided into the 3 experimental groups:

Group I: Activa Bioactive restorative (n=20)
Group II: Tetric Ceram Bulk fill (n=20)
Group III: Filtek Bulk fill (n=20)



Occlusal depth = 2mm | Bucco-lingual width =2mm | Axial depth = 4mm | All cavities were kept 1mm short of cemento-enamel junction

Fig 1: Dimensions of the class II cavity preparation (original)

Prior to restoration, the teeth were mounted in dental stone block with the adjacent premolar in place to establish contact, after which tofflemire retainer with ivory no.8 band was placed. The wooden wedge was inserted inter proximally in order to tightly seal the cervical margins. A sharp explorer was used to confirm the adaptation of the matrix band to the cervical margin.

Restorative protocol

Group I: Activa Bioactive

The cavities were etched (Detrey conditioner 36, Dentsply) for 5 seconds and were directly restored with Activa Bioactive restorative without application of bonding agent according to manufacturer's instructions. Activa bioactive was placed as a bulk filling material and was cured using the protocol: occlusal curing = 40 s and after removal of the matrix band, the restorations were light-cured from the buccal and lingual aspects for an additional 20 s on each side with a LED light curing unit (Woodpecker® Dental Curing Light LED D) with output irradiance of approximately 850-1000 mW/cm^2 held in contact with the coronal edge of the matrix band.

Group II: Tetric ceram bulk fill and group III: Filtek Bulk fill

The cavities were etched (Detrey conditioner 36, Dentsply) for 15 seconds, bonding agent was applied and cured for 10 seconds (Adper single bond 2, 3M ESPE) and restored with the respective bulk fill composite i.e. Group II-Tetric N Ceram bulk fill and Group III-Filtek bulk fill. Curing protocol followed was same as Group I.

All restorations were polished using the Shofu super snap composite polishing kit (Shofu dental corporation)

The specimens were stored at 100% relative humidity at 37°C for 24 h.

Qualitative microleakage analysis using Dye extraction method

Samples from all the groups were coated with two coats of nail varnish except 2mm on all sides of restoration-dentin interface at the gingival margin and the apices were sealed with cyanoacrylate adhesive (Figure 2). The samples were then dried and immersed in 5 ml of 2% methylene blue dye (S D Fine Chemical Ltd, India) in 15 ml screw-capped bottle for 24 hours (Figure 3). After 24 hours, the samples were washed under running tap water to remove the traces of the dye. The nail varnish was removed using ultrasonic scaler. The teeth were then immersed and stored in freshly prepared 65% nitric acid (S D Fine Chemical Ltd, India) for 72 hours (Figure 4). After 72 hours, the solutions were filtered using a fine grit filter paper in a centrifugal tube and the obtained sample solutions were centrifuged in a high speed microcentrifuge machine at 14,000rpm for 5 minutes. The supernatant solutions thus collected were used to determine absorbency in a UV visible spectrophotometer at 670 nm (Figure 5). The results were recorded as a measure of transmission of light. The data was analyzed statistically by comparison of mean microleakage using S.P.S.S (Statistical Package for Social Sciences version 20) software.

Fig 2: Sample coated with nailvarnish (original)

Fig 3: Sample placed in 2% methylene blue solution (original)

Fig 4: Sample placed in 65% nitric acid (original) solution

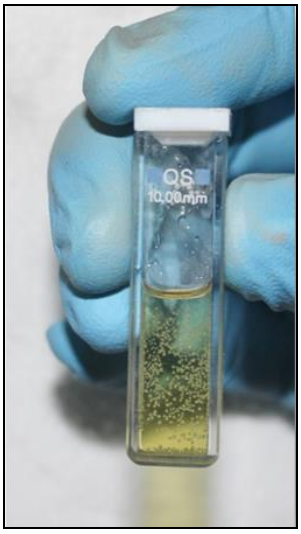
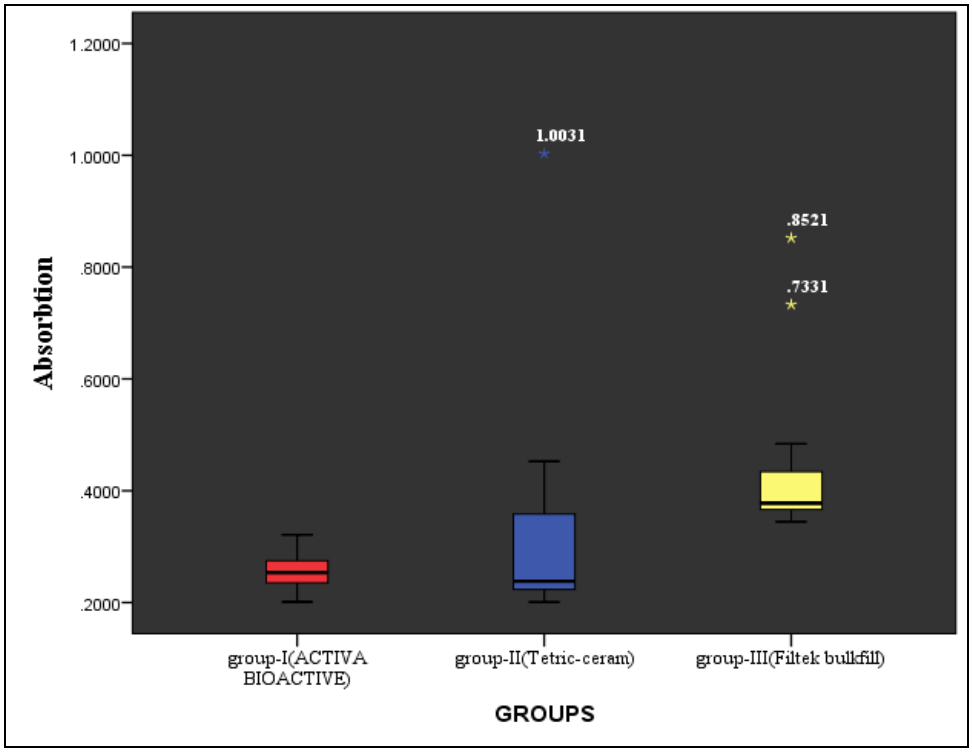


Fig 5: Supernatant collected for spectrophotometric analysis (original)

Results

The variance in the 3 groups studied was not found to be same and data was not normally distributed thus the non-parametric Kruskal Wallis test was used. Mean microleakage values indicate that Activa bioactive restorative showed least microleakage, followed by Tetric N Ceram bulk fill and highest microleakage was shown by Filtek bulk fill. Results suggest that there is a statistical significant difference between the groups ($p \leq 0.05$) (Table 1). Hence, a Pair-wise comparison for the groups was done using the Mann-Whitney U Test. Pair wise comparison of Group I (Activa bioactive restorative) and Group II (Tetric N Ceram bulk fill) showed that there was not a statistically significant difference ($p \geq 0.05$), however comparison of Group II and Group III revealed a statistically significant difference ($p \leq 0.05$) (Table 2). Graph 1 shows a representation of microleakage of the groups.



Graph 1: Graphical representation of microleakage of the groups (original)

Table 1: Intergroup comparison using Kruskal Wallis test (original)

Kruskal wallis test			
Groups		**N**	**Mean rank**
Absorption of dye	Group-I Activa bioactive)	20	21.38
	Group-II (Tetric-N-ceram bulkfill)	20	23.15
	Group-III (Filtek bulkfill)	20	46.98
	Total	60	

Test Statistics	
	Absorption
Chi-Square	26.804
df	2
Asymp. Sig.	.000*

(p<0.05) statistically significant difference

Table 2: Pairwise comparison of Groups using Mann-Whitney Test (original)

Comparison	Group I (Activa bioactive) And Group II (Tetric-N-Ceram bulk fill)	Group I (Activa bioactive) and Group III (Filtek bulk fill)	Group II (Tetric-N-Ceram bulk fill) and Group III (Filtek bulk fill)
Z	-.473	-5.410	-3.503
P	.636	.000*	.000*
Comments	Result not significant at P≤0.05	Result significant at P≤0.05	Result significant at P≤0.05

Discussion

Marginal integrity is essential to increase the longevity of any restoration [11]. Polymerization shrinkage is a major cause of loss of marginal integrity leading to microleakage and thus failure of class II composite restorations. This phenomenon occurs because monomer molecules are converted into a polymer network and therefore exchanges Van der Walls spaces into covalent bond spaces, creating contraction stresses in the resin composite leading to microleakage [13].

The magnitude of the stress induced during polymerization shrinkage also depends upon other factors, such as the configuration factor (C-factor) of the cavity and also the effect of light-curing mode [14].

In our study, the cavities were of the same dimensions, the same C-factor, and the light-curing mode was the same for all the restoration specimens.

The recent introduction of ''bulk-filled'' restorative materials has reignited the debate of ''bulk vs. incrementally'' placed composites as the effect of shrinkage stress may be more pronounced with bulk fill since the entire mass polymerizes at one time rather than in small increments [1].

Therefore, in this study, we used 2 bulk fill composites i.e. Tetric N Ceram bulk fill and Filtek bulk fill composite and a bioactive bulk fill material Activa Bioactive.

Activa Bioactive is a bioactive restorative material composed of a patented bioactive ionic resin, patented rubberized resin and bioactive glass ionomer. It combines the esthetics, strength and resilience of composites with bioactive properties and fluoride release that are superior to glass ionomers and contains no Bisphenol A, No Bis-GMA and no BPA derivatives. It releases and recharges calcium, phosphate and fluoride. Also, it chemically bonds to the tooth via ionization reaction and forms a strong resin-hydroxyapatite complex thereby sealing the tooth against bacterial microleakage [10].

Tetric N Ceram Bulk Fill represents the medium viscosity type bulk fill. The curing depth of 4 mm is achieved mainly due to the patented photo-initiator, Ivocerin, which is far more reactive than conventional initiators [15]. Shrinkage stress in Tetric N-Ceram Bulk Fill is however kept to a minimum. Special patented filler which is partially functionalised by silanes, acts as a unique shrinkage stress reliever.

Filtek bulk fill composite posterior restorative resin allows placement of 5 mm increment at a time and is composed of a nanofiller technology and two innovative methacrylate monomers which act to lower polymerization stress without compromising wear. The 2 novel methacrylate monomers are (AUDMA) that decreases the number of reactive groups in the resin and addition-fragmentation monomers (AFM) that reacts with the developing polymer, thereby forming cross links between polymer chains and it has a third reactive site that cleaves through fragmentation process.

The study utilizes a dye extraction method to quantify the amount of microleakage instead of dye penetration because dye extraction method is a quantitative assessment unlike dye penetration which is more of a qualitative assessment. Also, depth of penetration of the dye is not uniform around the margins of a restoration and gives randomly chosen results, raising doubts about their reliability Additionally it is advisable to use dye extraction methods instead of fluid filtration since dye extraction gave the same results as fluid filtration but saved much laboratory time [16].

In the present study, the results were recorded as a measure of absorbance of light. According to the Beer-Lamberts law, the absorbency of the solution is directly proportional to the concentration of absorbing species in the solution and path length. Thus, for a fixed path length, UV visible spectroscopy can be used to determine the concentration of the absorber in the solution. Hence it can be interpreted that absorbance of the solution is directly related to the amount of microleakage. Thus, a UV spectrophotometer is used to calibrate the amount of microleakage [17].

In the present study, there was significant difference in microleakage among the three groups. Activa bioactive and Tetric N Ceram bulk fill showed least microleakage. Pair-wise comparison for the groups using Mann-Whitney U test showed that there was not a statistically significant difference among Group I (Activa Bioactive) and Group II (Tetric N Ceram bulk fill) ($p \geq 0.05$), however comparison of Group II (Tetric N Ceram bulk fill) and Group III (Filtek bulk fill) revealed a statistically significant difference ($p \leq 0.05$).The results of present study are in agreement with that of Cannova *et al*. who demonstrated that the marginal seal of Activa Bioactive when used without bonding agent was at par with the leading composites Filtek™ Supreme Ultra (3M, ESPE/Conventional), SonicFill™(Kerr/Bulk Fill), TetricEvoCeram® (Ivoclar/Bulk Fill) [18].

The superior sealing of Activa bioactive could be attributed to the ionization reaction that helps form hydroxyapatite bond to the tooth structure, while the enhanced seal obtained in Tetric N Ceram bulk fill could be due to the patented photointiator ivocerin and the shrinkage stress relievers present in the formulation.

Further studies with larger sample size and class II cavities extending below the cemento-enamel junction need to be tested to evaluate the potential of the newer bulk fill restorative materials.

Conclusion

Within the limitations of the present study, it can be concluded that in Class II restorations, microleakage is observed regardless of the Bulk Fill composite used. There is

a statistically significant difference among the groups. Activa bioactive and Tetric N Ceram bulk fill composite restorations showed least microleakage, with no statistically significant difference among these two groups. Filtek bulk fill composite resin showed highest microleakage.

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