

Comparative Evaluation of Microleakage Between Nano-Ionomer, Giomer and Resin Modified Glass Ionomer Cement in Class V Cavities- CLSM Study

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ABSTRACT

Introduction: Marginal integrity of adhesive restorative materials provides better sealing ability for enamel and dentin and plays an important role in success of restoration in Class V cavities. Restorative material with good marginal adaptation improves the longevity of restorations.

Aim: Aim of this study was to evaluate microleakage in Class V cavities which were restored with Resin Modified Glass Ionomer Cement (RMGIC), Giomer and Nano-Ionomer.

Materials and Methods: This in-vitro study was performed on 60 human maxillary and mandibular premolars which were extracted for orthodontic reasons. A standard wedge shaped defect was prepared on the buccal surfaces of teeth with the gingival margin placed near Cemento Enamel Junction (CEJ). Teeth were divided into three groups of 20 each and restored with RMGIC, Giomer and Nano-Ionomer and were subjected to thermocycling. Teeth were then immersed in 0.5% Rhodamine

B dye for 48 hours. They were sectioned longitudinally from the middle of cavity into mesial and distal parts. The sections were observed under Confocal Laser Scanning Microscope (CLSM) to evaluate microleakage. Depth of dye penetration was measured in millimeters.

Statistical Analysis: The data was analysed using the Kruskal Wallis test. Pair wise comparison was done with Mann Whitney U Test. A p-value<0.05 is taken as statistically significant.

Results: Nano-Ionomer showed less microleakage which was statistically significant when compared to Giomer (p=0.0050). Statistically no significant difference was found between Nano Ionomer and RMGIC (p=0.3550). There was statistically significant difference between RMGIC and Giomer (p=0.0450).

Conclusion: Nano-Ionomer and RMGIC showed significantly less leakage and better adaptation than Giomer and there was no statistically significant difference between Nano-Ionomer and RMGIC.

Keywords: Beautifil II, Dye penetration, Ketac N 100, Marginal adaptation, Rhodamine B dye

INTRODUCTION

Microleakage is defined as the chemically undetectable passage of bacteria, fluids, molecules or ions between the cavity walls and restorative materials. Clinically microleakage is the major cause for the failure of restorations in Class V cavities, as margins of such restorations are generally located in dentin/cementum [1,2].

Earlier amalgam and gold restorative materials were used to restore Class V cavities but became obsolete mainly because of their esthetic shortcomings. Now-a-days restorative materials like glass ionomers, hybrid ionomers, compomers and composite resins are recommended to restore Class V cavities [3].

Glass Ionomer Cements (GICs) have undergone many modifications since its invention by Wilson and Kent in 1970's [4]. Glass ionomers have several advantages like ability to bond to dental hard tissues, fluoride release. Co-efficient of thermal expansion of glass ionomers which is similar to tooth structure provides good marginal adaptation, less microleakage and good retention of restoration in clinical scenario. Resin Modified Glass Ionomer Cements (RMGIC) were introduced in 1990s to overcome the drawbacks of conventional GIC, by possessing a prolonged working time, improved translucency, faster set and attainment of early strength [4].

Giomers are hybrid between glass ionomers and resin composites. Giomers are directed to have an increased wear resistance, shade conformity, increased radiopacity, improved light diffusion and fluorescence like resin composites along with high fluoride release and rechargability similar to GIC. Giomers uses pre-reacted glass

filler technology where pre-reaction of fluoroaluminosilicate glass fillers with polyacrylic acid forms a stable phase referred as "wet siliceous hydrogel" which is then freeze dried, milled, silane treated and ground to form PRG fillers. Beautifil II uses S-PRG (surface reaction type) where only the surface of the glass filler is attacked by polyacrylic acid and a glass core remains [5].

Recently nano-ionomer was introduced which consists of 69% by weight nano sized fillers like, silane-treated silica and zirconia along with the fluoroaluminosilicate glass. Manufacturer claims that nanofilled RMGIC has increased mechanical characteristics like wear resistance, enhanced surface finish, polishability and precision for shade match characterization [6].

AIM

Aim of the present in-vitro study was to compare and evaluate microleakage in Class V cavities between tooth and restoration interface when teeth were restored with RMGIC, giomer and nano-ionomer.

MATERIALS AND METHODS

This study was conducted at Sibar Institute of Dental Sciences, Takellapadu, Guntur, Andhra Pradesh, India. This in-vitro study included 60 extracted human mandibular or maxillary premolars. Teeth were divided into three groups of 20 each. Teeth were randomly selected with respect to the inclusion and exclusion criteria and were distributed into three experimental groups. Human premolars extracted for orthodontic reasons were included in

this study. Teeth with previous restorations, visible cracks, decay, fracture, abrasion or structural deformities were excluded from the study.

Teeth were cleaned with ultrasonic scaler one week prior to restoration. Then, all the teeth were disinfected with 0.5% chloramine for 24 hours and stored in distilled water at room temperature. Wedge shaped defects were prepared using 008-diamond bur (Diatech Dental AG), at the buccal surfaces of teeth with air/water spray. The preparation was 4mm in length, 4mm wide mesiodistally, and 2mm deep with an occlusal margin in enamel and a gingival margin in cementum. All the cavity preparations and restorations were performed by the same operator to eliminate inter operator variability. After cavity preparations, the teeth in each group were assigned numbers and were randomly divided into three experimental Groups (I, II, and III). Teeth in Group I were conditioned with Self Conditioner (GC Corp., Tokyo, Japan.) and restored with RMGIC (Fuji Filling LC, GC Corp., Tokyo, Japan.) which was taken as Control Group. Teeth in Group II followed self-etch protocol and restored with GIOMER (Beautifil II, Shofu, Kyoto, Japan.). Teeth in Group III were restored with the Nano-Ionomer (Ketac N 100, 3M ESPE, St. Paul, MN, USA) after the application of primer. Cavities were restored in incremental technique, with a minimum thickness of 1mm to minimize C-factor and polymerization shrinkage. Finishing and polishing of the restorations was performed using an extra-fine diamond point (Mani, Tochigi, Japan) and SofLex disks (3M ESPE, St.Paul, MN,USA).

Microleakage Testing: The specimens were stored in 100% relative humidity at 37°C for 24 hours and then were submitted to 500 thermocycles at 5°C and 55°C with a dwelling time of one minute at each temperature. The apices of all teeth were sealed using composite and all the teeth were covered with two coats of nail varnish except for 1-2mm around the margins of the restorations to limit dye penetration to cavity margins. Teeth were then immersed in 0.5% Rhodamine B dye for 48 hours. After separating the radicular portion, the coronal portions were washed and embedded into acrylic resin. These acrylic blocks were sectioned longitudinally from the middle of cavity (buccolingually) into two mesial and distal parts. Each specimen was observed under Confocal Laser Scanning Microscope (CLSM) to evaluate microleakage. The degree of dye penetration was identified according to the criteria given by Wahab et al., [7].

0=no penetration;

1=penetration to the enamel or cementum aspect of the preparation wall;

2=penetration to the dentin aspect of the preparation wall, but not including the pulpal floor; and

3=penetration including the pulpal floor of the preparation.

STATISTICAL ANALYSIS

The depth of dye penetration along the occlusal and cervical margins towards the pulpal wall was measured in millimeters using UTHSCSA Image-Tool for Windows, v 3.0 software. The data were submitted to statistical analysis using the Kruskal Wallis test. Pair wise comparison among the experimental groups was done with Mann Whitney U Test. A p-value of <0.05 is taken as statistically significant.

RESULTS

Intergroup comparison of three restorative materials with respect to micro leakage showed significant difference ($p = 0.0080$) [Table/ Fig-1]. Group III showed less microleakage which was statistically significant when compared to Group II ($p=0.0050$) [Table/Fig-2]. Statistically no significant difference was found between Group III and Group I ($p=0.3550$) [Table/Fig-2]. There was statistically

Micro-leakage	Group I	%	Group II	%	Group III	%	Total	%
Score 0	10	50.00	8	40.00	14	70.00	32	53.33
Score 1	4	20.00	3	15.00	2	10.00	12	20.00
Score 2	4	20.00	1	5.00	4	20.00	10	16.67
Score 3	2	10.00	8	40.00	0	10.00	6	10.00
Total	20	100.00	20	100.00	20	100.00	60	100.00
Comparison by	H-value = 9.5660							
Kruskal Wallis test	p-value = 0.0080*							

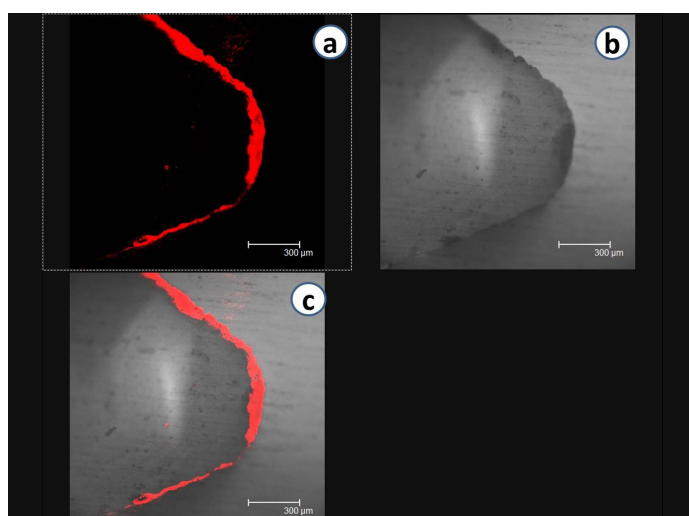
[Table/Fig-1]: GroupWise comparison of three restorative materials with respect to microleakage.

* = p-value is statistically significant.

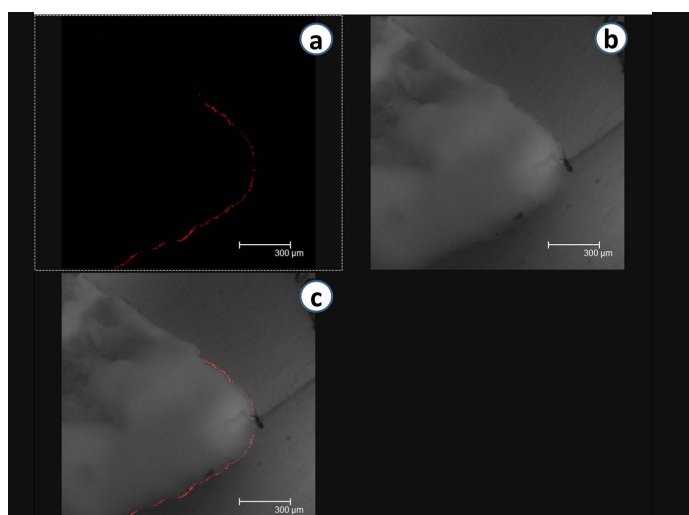
GROUP III vs GROUP II	$p=0.0050^*$
GROUP III vs GROUP I	$p=0.3550$
GROUP II vs GROUP I	$p=0.0450^*$

[Table/Fig-2]: Pair wise comparison of the three restorative materials with respect to microleakage.

* = p-value is statistically significant.



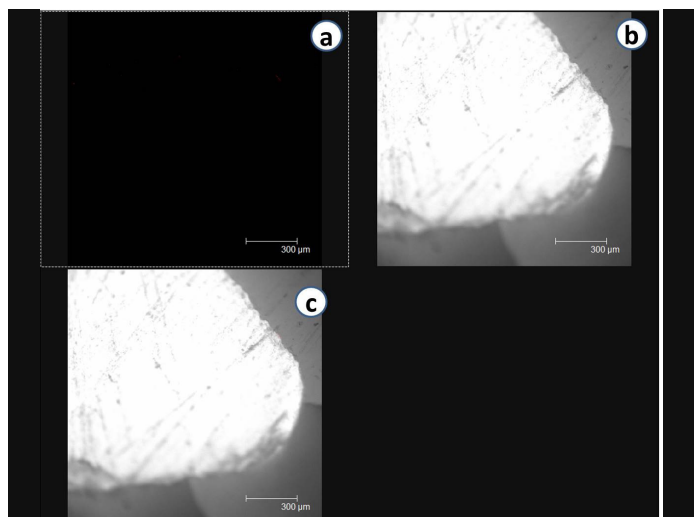
[Table/Fig-3a-c]: CLSM image of Giomer restorative material with presence of fluorescence between cavity wall and restoration.



[Table/Fig-4a-c]: CLSM image of RMGIC restoration with microleakage on gingival margin and good adaptation with cavity wall.

significant difference between Group I and Group II ($p=0.0450$) [Table/Fig-2].

Mean values of dye penetration for Group I (1276.23mm), Group II(2655.35mm), Group III (321.23mm) respectively. CLSM images were presented in florescent mode [Table/Fig-3a,4a,5a], reflected mode [Table/Fig-3b,4b,5b], superimposition of both florescent and reflected modes [Table/Fig-3c,4c,5c].



[Table/Fig-5a-c]: CLSM image of Nano Ionomer shows absence of florescence between cavity wall and restoration and well adaptation with cavity wall.

[Table/Fig-3] shows fluorescence between restoration and tooth surface in Group II which indicates a poor adaptation of restorative material and Gap formation. [Table/Fig-4] shows microleakage between restoration and tooth surface in Group I with good adaptation of the material. [Table/Fig-5] shows no microleakage between restoration and tooth surface in Group III which indicates the well adaptation of restorative material.

DISCUSSION

Microleakage between cavity wall and restorative material is one of the main causes of post-operative sensitivity, recurrent caries and pulpal pathosis [1,8]. Restoration of Class V cavities which are usually located in cervical area of the tooth, presents a special challenge to the clinician [9]. The coronal margins of these Class V cavities are in enamel while the gingival margin is usually located in cementum or dentin. Despite several improvements in adhesive systems, the adaptation and bonding of these adhesive systems to cementum and dentin is less predictable. The cyclic flexure of tooth in these cervical areas along with polymerization shrinkage of adhesive material may also lead to loss of marginal adaptation [9-12].

In-vitro evaluation tests are done to predict the clinical performance of the restoration [2]. Various methods for detection of marginal adaptation of restorative material include dye and bacterial leakage studies, chemical and radioactive tracers and Scanning Electron Microscopy (SEM) [12]. Bond failure between the tooth and restoration interface are commonly assessed with microleakage dye penetration tests [1,2].

CLSM in the present study for evaluation of microleakage in Class V cavities is used to obtain thin optical sections below the surfaces of the specimens [8]. Unlike Scanning Electron Microscopy (SEM) and Transmission Electron Microscopy (TEM), CLSM is a non-destructive method, eliminates sectioning and dehydration of specimens and polishing artifacts that exaggerate dye penetration [2,8]. Mount has suggested that CLSM is the least intrusive and destructive method of studying the interface between glass ionomer and tooth structure [8].

In the present study experimental teeth were subjected to thermocycling, similar to other microleakage studies [9-11,13]. Thermocycling is a standard protocol which is employed when evaluating the microleakage of bonded restorative materials under in-vitro conditions, simulating in vivo aging by subjecting these bonded materials to cyclic exposures to hot and cold temperatures [14].

One of the primary requisite for marginal seal is by better adaptation of the restorative material to tooth structure. Adhesion

of RMGIC to tooth structure is by two fold, one is by chemical bonding to enamel and dentin and other by hybridization [15]. Using X-ray photoelectron spectroscopy and Fourier-transformed infrared spectroscopy Fukuda et al., observed chemical bonding between RMGIC and inorganic content of enamel and dentin [16]. CLSM demonstrated hybrid layer formation in dentin with RMGIC [17, 18]. Results of the present study regarding the microleakage of RMGIC restorations were similar with other studies [9-11].

Nano-Ionomer bonds chemically to the tooth structure. Coutinho et al., through a TEM analysis observed tight interface between the nano-ionomer and dentin, without signs of dentin demineralization or hybrid-layer formation when the restorations were pretreated with primer provided by the manufacturer [6]. S Abd El Halim & Zaki under in a SEM study observed an indistinct interface between the margin of the tooth structure and the restoration, suggesting that a chemical bond had formed between the GIC and tooth structure

Author and Year	Materials used in the study	Results	Conclusion
Toledano et al., 1999 [11]	Fuji II LC, Vitremer, Dyract	Significant difference found between the three materials with Fuji II LC showing least leakage.	RMGIC showed less or similar microleakage than the polyacid-modified composite resin (Dyract).
Brackett et al., 1998 [10]	Fuji II LC, Vitremer, Dyract	Dye penetration was observed at 20% of restoration margins for all the materials, with greatest for Dyract.	No significant difference found between the three materials.
P.E. Murray et al., 2002 [20]	RMGIC, Zinc Oxide Eugenol (ZnOE), Calcium Hydroxide (Ca(OH) ₂), Composite Resin (CR), Bonded Amalgam (BA), Gutta Percha (GP), Compomer, Silicate, Zinc Phosphate(ZP)	Rank order of preventing microleakage from best to the worst RMGIC (100%), BA (88%), ZnOE (86%), CR (80%), GP (64%), (Ca(OH) ₂ (58%), Compomer (42%), Silicate (36%), ZP (0%).	The most effective restorative materials to prevent bacterial microleakage and pulp injury from inflammatory activity were RMGIC, BA, ZnOE, CR.
Wahab et al., 2003 [7]	Arabesk, Solitaire 2, Silux Plus, Renew, Durafill, Charisma	Thermocycling had a significant effect and the type of restorative material had no effect.	Thermocycling significantly increased the microleakage. The overall microleakage at enamel margins was significantly less than that of the dentinal margins.
Fernanda et al., 2004 [21]	Aelite LS, InTen-S, Filtek Z250, Heliomolar	Significant difference was found between Heliomolar and Aelite LS.	The low shrinkage materials exhibited contraction stress values similar or higher than the hybrid composite.
David et al., 2008 [22]	Admira, Compoglass F, Fuji II LC, Filtek P60	Significant difference was found between four restorative materials.	Admira and Filtek P60 exhibited the best sealing ability.
S. Vinay et al., 2010 [23]	Single Bond Adper Prompt i Bond Clearfil S3 G Bond	Clearfil S3 Showed significantly less leakage.	Clearfil S3 has better sealing ability at both enamel and dentin margins.
Sabine O. Greets et al., 2010 [24]	RMGIC with & without One-Step Self Etch Adhesives.	No nsignificant difference found between bonding RMGIC to dentin with Step Self Etch Adhesives and Polyacrylic acid.	Bonding RMGIC to dentin with Step Self Etch Adhesives would not enhance the performance.
Shinam Pasricha 2011 [25]	Ormocer, Giomer, Composite	Ormocer was best followed by Giomer and Composite.	Bleaching procedure affected cementum margins more than enamel.

Upadhyay S and Arthi Rao 2011 [26]	Conventional GIC, RMGIC, Nano Ionomer.	Significant difference found between RMGIC and Nano Ionomer.	Nano Ionomer showed least microleakage.
Sarath Chandra et al., 2011 [9]	Nano Composite, Nano Ionomer.	Significantly more microleakage on gingival margins. Palatal surfaces showed less microleakage when coated with resin sealant compared to buccal cavities.	Coating of resin sealant did not completely eliminate microleakage, but was effective in reducing microleakage on gingival margin.
Halim and Zaki 2011 [19]	Ketac N 100, Vitremer, PhotacFil Quick	Significant difference observed between groups after immersion for 60 days.	Ketac N 100 showed the least microleakage.
C. Vishnu Rekha et al., 2012 [27]	Fuji IX GP Fuji II LC Compoglass	No significant difference.	Fuji II LC and Compoglass were superior to Fuji IX GP.
Casselli et al., 2013 [28]	Etch & Rinse [Single Bond 2 (SB) Self-Etching Adhesive (Clearfil SE (CL)].	SB presented higher gaps in dentin than CL. CL showed better adaptation in dentin.	Margin location and adhesive system has an important effect on marginal adaptation of Composite Resin.
Amish Diwanji et al., 2014 [29]	Fuji IX Fuji II LC Ketac N 100	Significant difference observed between Fuji II LC and Ketac N 100, Ketac N 100 and Fuji IX.	Fuji IX showed maximum leakage. Ketac N 100 showed least leakage.
Hussein et al., 2014 [30]	Ketac Molar Easy mix, Fuji II LC, Ketac N 100, Filtek Z 250	No significant difference.	All materials showed more microleakage at gingival margins.
Eranot et al., 2014 [31]	Ketac Molar Ketac N 100	High viscosity glass ionomer showed significantly less microleakage compared to the Nano-Ionomer at occlusal margin. No significant differences were found between the groups at gingival margin.	Nano-Ionomer restorations did not perform better than high viscosity glass ionomer in class V cavities in terms of microleakage assessment.
Shiji Dinakaran 2015 [32]	Dyract Fuji II Fuji II LC	Significant difference observed	Dyract was found to have better marginal adaptation to both cementum and enamel margins followed by Fuji II LC and Fuji II
Fereshtesh Shafiei and Mohadese Abouheydi 2015 [33]	Clearfil AP-X Filtek- P 90 Ketac N 100	Significant difference among the materials at enamel margins.	Additional selective enamel etching might improve enamel sealing.

[Table/Fig-6]: Results and conclusions of similar previous microleakage studies.

[19].

Results of the present study were similar to results obtained by S Abd El Halim [Table/Fig-6] who observed that nano-ionomer showed least microleakage under in-vitro conditions when compared to RMGIC [19].

Perdigae et al., also observed good marginal adaptation of nano-ionomer than RMGIC, clinically after one year follow up in non carious cervical lesions [34]. Good sealing ability of nano-ionomer could also be related to high filler loading and lower coefficient of thermal expansion which withstands the polymerization contraction stresses [30]. Srirekaha et al., in a three-dimensional finite-element analysis observed that nano-ionomer developed least stresses in the cervical region of tooth with and without occlusal restoration [35].

Giomer uses FL-Bond II adhesive system which belongs to the category of "mild" self-etch primers. Van Meerbeek et al., have suggested that the bonding of self-etch primers to dental hard tissues is by combination of micromechanical and chemical interaction with tooth substrate. They proposed that chemical

bonding of these "mild" self-etch systems may be able to compensate decreased micromechanical interlocking [15]. The FL-Bond system contains 4-AET Acid(4-Acryloxyethyltrimellitic acid), which forms a relatively insoluble calcium (Ca) salt by interacting with the calcium cations of hydroxyapatite to form 4-AETCa, that may improve durability of these adhesive system [5, 36].

In the present in-vitro study microleakage was found to be high at both occlusal and cervical margins of Class V cavities restored with giomer which can be explained by two reasons. Diliperi et al., have observed that "Mild" self etch primers are ineffective in etching of enamel [14]. To overcome this Torii et al., had recommended additional enamel etching with phosphoric acid when a mild self-etch primer is used [37]. In contrary some authors have observed that prior application of phosphoric acid to etch dentin negatively affects dentin bonding of mild self-etch primers. This leads to over-etched dentin and incomplete infiltration of the resin monomer [38]. Thus in present in-vitro study, pre-treatment with phosphoric acid was avoided prior to application of a FL Bond II.

Hakimeh et al., demonstrated that polymerization contraction stresses might be the primary cause of microleakage when the restorations were subjected to themocycling [39]. Reduced marginal adaptation of giomer in the present in-vitro study may also be due to polymerization shrinkage resembling its typical nature like a resin composite. Some authors suggest that the main cause of marginal deterioration of giomer restorations is hygroscopic expansion which is an intrinsic property of this restorative material [5]. The results of the present study were similar to other research [14]. In-vivo studies showed marginal deterioration of giomer restorations [5,36,40].

In summary, the results of our study indicate that nano-ionomer and RMGIC are better than the giomer in terms of marginal adaptation in Class V restorations. In the present in-vitro study restorations in all the groups showed poor sealing ability at dentin or cementum margins compared to the enamel margins, which was similar to other research [9,11,13]. The nano-ionomer has performed better than the control group (RMGIC) and giomer in both the enamel and dentin/cementum margins.

Clinical implications: Based on the results of the present in-vitro dye leakage study, it can be clinically inferred that because of good marginal adaptation of nano-ionomer when compared to RMGIC and giomer, better resists staining around the restorations and prevents post-operative sensitivity, secondary caries and pulpitis in Class V cavities.

LIMITATION

The present study did not compare the tested restorations with self-cure (Conventional) GIC and the teeth were not subjected to any mechanical stress. Hence, future studies should be conducted by comparing these restorations with conventional GIC and by subjecting these restorations to cyclic occlusal loading. The sealing ability of these restorative materials should also be examined through other complex methods like bacterial penetration and with the use of fluid transport model. The present study was done under in-vitro conditions; hence future studies should be focused to be conducted in-vivo conditions to evaluate the clinical behavior of the tested restorative materials.

CONCLUSION

Within the limitations of the present in-vitro study following conclusions can be drawn:

- 1) None of the glass ionomer restorative materials tested in the present study were able to totally prevent the microleakage on the cervical margin of the Class V cavities.
- 2) Nano-Ionomer showed least microleakage and better marginal adaptability when compared to giomer and RMGIC.

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