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# Phosphoric acid treatment enhances adaptation of glass-ionomer cement to bioceramic sealer-conditioned dentin

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## ABSTRACT

**Background:** This study evaluated the interface between fresh eugenol/bioceramic sealer-conditioned coronal dentin and high-viscous glass-ionomer cement (HVGIC), treated with various dentin conditioners (saline, 10% polyacrylic acid, and 37% phosphoric acid).

**Materials and Methods:** Standard endodontic access preparation and instrumentation were done in 21 freshly extracted mandibular molar teeth in this *in vitro* study. Teeth were divided into two interventional groups ($n$ = 9/group), based on the type of sealer (zinc oxide eugenol [ZOE]/bioceramic [BioRoot RCS] sealer) used for obturation. Samples were further subdivided based on the type of dentin-conditioning procedures performed (saline/10% polyacrylic acid/37% phosphoric acid). Post dentin conditioning, the access cavity was sealed with HVGIC. Later, material-dentin interfacial analysis and elemental analysis were done using scanning electron microscopy (SEM) and energy-dispersive X-ray spectroscopy.

**Results:** The interfacial SEM images of HVGIC layered over B-RCS/ZOE sealer-conditioned dentin, treated with saline, showed predominantly adhesive debonding failures, whereas cohesive debonding was observed with polyacrylic and phosphoric acid. In the elemental analysis, the intensity of zirconium (depicting the residue of B-RCS)/zinc (depicting ZOE sealer) was very high on the dentin side treated with saline, in comparison to the dentin treated with polyacrylic and phosphoric acid. Furthermore, the intensity of elements from HVGIC was low on the dentin side of the groups with saline, whereas these elements showed maximum penetration into the dentin when treated with phosphoric acid.

**Conclusion:** Conditioning of the endodontic access cavity using 37% phosphoric acid immediately postobturation resulted in higher penetration of HVGIC into the dentin, in comparison to the other dentin conditioners.

**Key Words:** Dentin, endodontics, glass-ionomer cement, scanning electron microscopy, spectrometry

## INTRODUCTION

The introduction of bioceramics which is a ceramic or metal oxides has created a new epoch in various fields of dentistry. The superior sealing ability,[1] biocompatibility,[2] and antimicrobial action[3] has

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expanded its use in dentistry for procedures such as vital pulp therapy, apexification, root end filling, and root resorption.[4-6]

BioRoot RCS (B-RCS) is a water-based sealer composed of tricalcium silicate and zirconium oxide.[7] It releases two times more calcium ions in comparison to EndoSequence BC and eight times more calcium ions in comparison to mineral trioxide aggregate (MTA) Fillapex and Apexit Plus when placed in contact with Hank's Balanced Salt Solution over a period of 14 days.[7] It also forms a calcium phosphate phase in contact with physiologic solution.[7] The viability of periodontal ligament cells as well as the secretion of growth factors, fibroblast growth factor-2, and osteogenic factor (Bone Morphogenetic Protein (BMP-2)) was found to be superior with B-RCS in comparison to zinc oxide eugenol (ZOE) sealer.[8]

However, MTA has shown to interact with some restorative materials when layered over them resulting in incomplete hydration/increase in porosities.[9] Ionic exchange between MTA/Biodentine and glass-ionomer cement (GIC) does not occur because of their similarity in composition.[9] Furthermore, a physical change with an evident gap has been demonstrated at the interface between these two materials.[9,10] Layering GIC over dentin replacement materials is not advocated due to the weak bond that is formed between GIC and tricalcium silicate cement.[10] Similarly, layering composite over Biodentine after etching can lead to migration of various ions which might affect the properties of the tricalcium silicate cement.[10]

ZOE cement introduced by Sorel in 1958 has various applications in dentistry till date ranging from being used as a provisional restoration to a root canal sealer.[11] However, studies have shown that eugenol inhibits polymerization of dental composites and decreases its bond strength.[12] ZOE interacts with calcium hydroxide to form calcium hydroxide-eugenol set cement, which ultimately results in a weak-binding face and causes ZOE to become brittle and granular.[13] Layering ZOE over MTA causes retardation of cement hydration and increases the porosity of the latter.[14]

High-viscous GIC (HVGIC) is one of the commonly used intracoronal sealing materials/intraorifice barriers.[15] Bonding of GICs to dentin can decrease due to the presence of smear layer or other materials; thus, the removal or modification of smear layer has been advocated with the use of dentin conditioners.[16] Various dentin conditioners such as polyacrylic acid, phosphoric acid, maleic acid, and citric acid have shown to improve the bond strength of GIC to dentin.[16,17] Literature search has shown lack of studies assessing the interface formed between sealer-conditioned coronal dentin and GIC.

Scanning electron microscopy (SEM) helps to visualize the interface formed between the materials and energy-dispersive X-ray spectroscopy (EDX) helps to study the characterization of the interface. Hence, the aim of the present study was to evaluate the interface between fresh eugenol or bioceramic sealer-conditioned coronal dentin and HVGIC, following various dentin-conditioning treatments using SEM and EDX. The null hypothesis ($H_0$) proposed is that there is no difference in the interface formed between fresh eugenol or bioceramic sealer-conditioned coronal dentin and HVGIC following various dentin-conditioning treatments.

## MATERIALS AND METHODS

This *in vitro* study proposal was reviewed and approved by the institutional review board (MADC/IRB-XXX/2018/352). Twenty-one freshly extracted mandibular third molar teeth within the age group of 18–26 years were collected and stored in 0.1% thymol solution. Intact third molars (without caries or restorations) with straight canals were included. Teeth with caries, cracks, curved canals, calcified canals, and resorption were excluded. The teeth were visualized under a stereomicroscope to rule out the presence of any preexisting cracks. Cold-cure epoxy resin (Dental Products of India, Mumbai, India) was used to embed the teeth prior to the procedures. Standard endodontic access cavity was prepared using a 014 round carbide bur and Endo-Z bur (Dentsply Sirona International, York, PA, USA); the root canals were enlarged to size of 25 (6% taper) using ProTaper Gold rotary files (Dentsply Sirona, Ballaigues, Switzerland). Irrigation was performed using 3 mL of 3% sodium hypochlorite (Prime Dental Products Pvt. Ltd., Thane, India), followed by 0.9% physiological saline with a 30-G side vented needle (Max-i-Probe; Dentsply Sirona Tulsa Dental, York, PA, USA) between successive instruments. After canal preparation, the final irrigation was performed using 5 mL of 17% ethylenediaminetetraacetic acid (Prime Dental Products Pvt. Ltd., Thane, India) for 1 min followed

by 3% sodium hypochlorite (Parcan; Septodont, Delhi, India) and 0.9% physiological saline. The canals were dried using paper points. The teeth were randomly divided into three groups. The samples of the control group ($n$ = 3) were not obturated. The samples from the other two groups ($n$ = 9 per group) were obturated either using Tubli-Seal (SybronEndo, Kerr Corporation, Romulus, MI) or B-RCS (Septodont, Saint-Maur-des-Fosses, France), respectively. Based on previous literature, the study was done in triplicate in each group.[10] The sealer was manipulated according to the manufacturer's instructions and coated into the canals using Lentulo spiral and obturated with a corresponding matched taper single cone of gutta-percha (Dentsply Sirona Tulsa Dental, York, PA, USA).

To mimic the clinical scenario, the access cavities were filled with their respective sealers for 3 min and 30 s.[18] The obturated samples were randomly divided into the following subgroups based on the dentin-conditioning procedure.

- ZnS (ZOE, saline); BcS (B-RCS, saline) group – The access cavity was rinsed with saline for 30 s using 30 G with apical opening needle (NaviTip; Ultradent Products, South Jordan, UT, USA) to remove the respective sealers from the samples and blot dried
- ZnPA (ZOE, polyacrylic acid); BcPA (B-RCS, polyacrylic acid) group – After saline rinsing for 30 s, the access cavity was conditioned with 10% polyacrylic acid (GC America, Alsip, IL, USA) for 10 s and again rinsed with saline for 30 s and blot dried
- ZnPH (ZOE, phosphoric acid); BcPH (B-RCS, phosphoric acid) group – After saline rinsing for 30 s, the access cavity was conditioned with 37% phosphoric acid (Prime Dental Products, India) for 15 s and again rinsed with saline for 30 s and blot dried[19]
- Control group – The coronal dentin was conditioned with 10% polyacrylic acid (GC America, Alsip, IL, USA) for 10 s.

After the dentin-conditioning procedure, HVGIC (Fuji IX GP [GC America, Alsip, IL, USA]) was manipulated according to the manufacturer's instructions and condensed into the access cavities using a plugger. The surface of HVGIC was coated with Adper Prompt L-Pop (3M ESPE, MN, USA) for surface protection.[20] All the samples were stored at 37°C and at 100% humidity. Later, the embedded samples were vertically sectioned using hard tissue microtome such that interfaces could be examined in longitudinal sections and polished with progressively finer grits of diamond discs.

### Interfacial analysis

Interfacial and elemental analysis of the material was performed on the floor and wall of the pulp chamber using SEM and EDX (FEI Quanta 400F, Oregon, United States). At each section, three points were analyzed.

A thin conductive coating of gold was spurted to the polished sections prior to examination in the SEM. The SEM used was FEI Quanta 400F, equipped with an energy-dispersive spectrometer, wavelength-dispersive X-ray spectroscopy, and backscattering electron diffraction. The FEI Quanta 400F was operated under high vacuum mode with gaseous backscatter electron detector. Scanning electron micrographs of the material interfaces were captured at ×500. Quantitative elemental analysis of the products was done by EDX. Lines parallel to the interface at increasing incremental distances of 50 µm up to 400 µm were scanned and spectra collected were used to draw atomic ratio plots.

## RESULTS

### Microscopy of uncontaminated materials

The backscattered electron images and the EDX analysis of HVGIC, B-RCS cement, and ZOE cement are shown in Figure 1. Glass particles interspersed in the matrix were observed in the microstructure of HVGIC. The elemental composition of HVGIC was calcium (Ca), silica (Si), aluminum (Al), phosphorus (P), strontium (Sr), and barium (Ba). ZOE was composed of very fine, dense matrix particles with zinc (Zn) as the main component. The microstructure of B-RCS was observed to have a matrix of varying sizes with interspersed filler particles. The major elemental constituents were Si, Al, oxygen (O), and zirconium (Zr).

### Microscopy of interfacial region of high-viscous glass-ionomer cement and nonsealer-conditioned dentin

The interfacial SEM images between HVGIC and dentin not exposed to the sealer (control group) are provided in Figure 2a and b. The microstructure of HVGIC in the control group showed a dense matrix with interspersed filler particles. The HVGIC had detached from the dentin during processing and

showed a cohesive debonding. Numerous microcracks were evident on the surface of the GIC. The EDX analysis of the interface up to 400 µ on the dentin side showed the presence of Si, Al, Ba, and Sr which are the constituents of HVGIC.

### Microscopy of the interfacial region between high-viscous glass-ionomer cement and BioRoot RCS-conditioned dentin

The interfacial SEM images of HVGIC layered over B-RCS-conditioned dentin, treated with saline (BcS group), showed mixed bonding defect, with predominantly adhesive failures [Figure 2c and d]. The HVGIC exhibited microcracking whereas, the interfacial SEM images of HVGIC layered over B-RCS-conditioned dentin, treated with polyacrylic acid (BcPA group) [Figure 2e and f] or phosphoric acid (BcPH group) [Figure 2g and h], showed cohesive debonding. A thick layer of HVGIC was found to be adherent to the surface of dentin.

Microstructure of HVGIC in contact with dentin conditioned with BcPH showed a dense homogenous and uniform matrix with interspersed fillers [Figure 2g and h]. Microcracking of HVGIC was evident. Microstructure of HVGIC in contact with BcPA showed a nonuniform granular matrix which was porous in structure [Figure 2e and f].

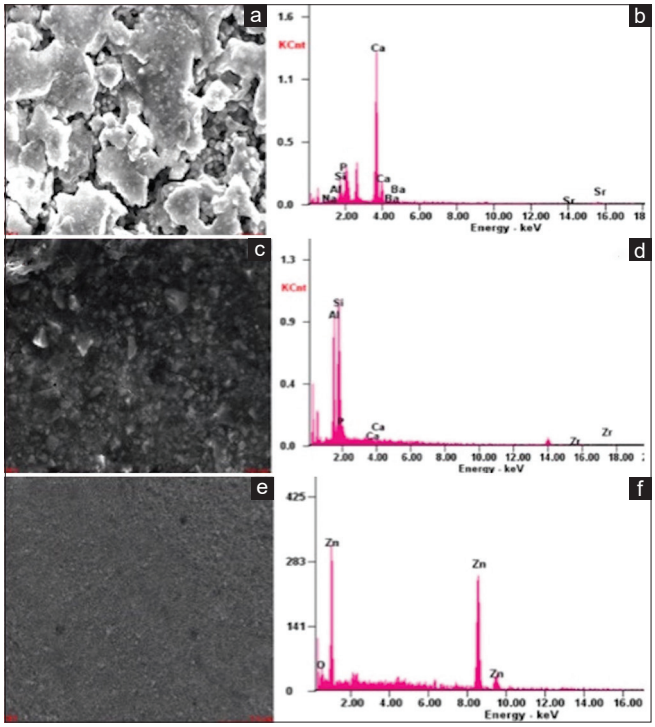


**Figure 1:** Backscatter electron images and EDX analysis of (a and b) HVGIC, (c and d) B-RCS, and (e and f) ZOE (EDX: energy-dispersive X-ray spectroscopy; HVGIC: High-viscous glass-ionomer cement; B-RCS: BioRoot RCS; ZOE: Zinc oxide eugenol).

The overlay of elemental profile of Zr and Sr is depicted in Figure 3. It was observed that the intensity of Zr (depicting the residue of B-RCS) was very high on the dentin side with saline treatment. The intensity of Zr on the dentin side was reduced with polyacrylic acid and was the least with phosphoric acid. The intensity of elements such as Al, Si, Sr, and Ba (depicting HVGIC) was low in the group treated with saline, whereas these elements showed a maximum intensity of penetration up to 400 µm when treated with phosphoric acid.

### Microscopy of the interfacial region between high-viscous glass-ionomer cement and zinc oxide eugenol sealer-conditioned dentin

The interfacial SEM images of HVGIC layered over ZOE-conditioned dentin, treated with saline (ZnS group) showed adhesive debonding failures [Figure 4a and b]. The GIC exhibited microcracking. Dislodgement of the samples was observed in this group during hard tissue microtome of the tooth. On the contrary, the interfacial SEM images of GIC layered over ZOE-conditioned dentin, treated with polyacrylic acid (ZnPA group) or phosphoric acid (ZnPH group), showed cohesive debonding. A thick layer of GIC was found to be adherent to the surface of dentin, indicating that the forces separating the material did not affect the bond strength between the two. Microstructure of GIC in contact with the ZnPA [Figure 4c and d] and ZnPH groups [Figure 4e and f] showed a nonuniform granular matrix which was porous in structure.

The overlay of elemental profile of Zn and Sr is depicted in Figure 5. It was observed that the intensity of Zn (depicting the residue ZOE sealer) was very high on the dentin side with saline treatment. The intensity of Zn on the dentin side was reduced with polyacrylic acid and was the least with phosphoric acid. The intensity of elements such as Al, Si, Sr, and Ba (depicting HVGIC) was low in the group treated with saline, whereas these elements showed a maximum intensity of penetration up to 400 µm when treated with phosphoric acid.

## DISCUSSION

Over the last couple of decades, it has been proven that the outcome of a root canal treatment is directly proportional to the quality of the coronal

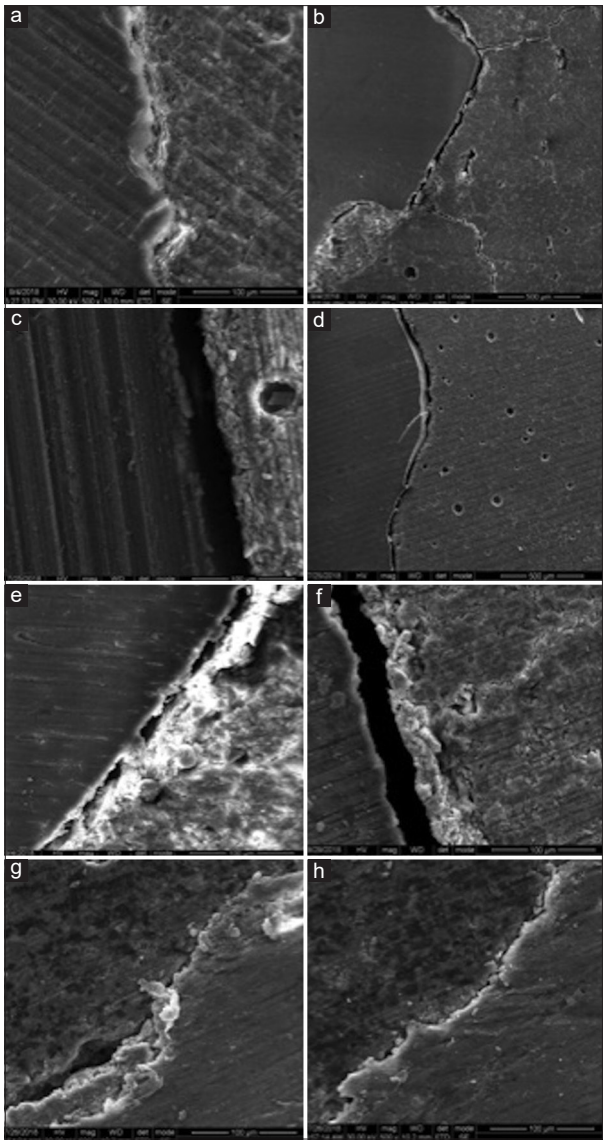


**Figure 2:** Backscattered images at different magnifications of the interfacial region of (a and b) control group, (c and d) BcS group, (e and f) BcPA group, and (g and h) BcPH group.

seal provided.[21-23] Despite an inadequate root canal filling, a good coronal restoration with intact margins reduces bacterial penetration, resulting in endodontic success.[24] The prevalence of apical periodontitis in patients with defective coronal restoration and satisfactory endodontic treatment was proven to be 2.8 times more in comparison to the patients with adequate coronal restoration and endodontic treatment.[25] Restoring the endodontic access cavity with a proper intracoronal seal has shown to reduce coronal microleakage.[26,27] The ability of GIC to adhere chemically to normal and sclerotic dentin provides an advantage for its use as a core material.[28]

ZOE-based sealers are economic, less technique sensitive, provide sufficient working time, and have antibacterial properties, thereby remaining popular till date.[11,29] A recent international survey has highlighted the increased demand and use of calcium silicate-based sealers among general dentists and specialists.[30] This may be attributed to their biocompatibility, bioactive properties, ability to set in the presence of moisture, and excellent antimicrobial properties.[30,31] With the advent of bioceramic sealers such as B-RCS, the single-cone technique/sealer-based obturation (SBO) has gained popularity.[32,33] Thus, despite their high cost, the procedure to use these sealers is more simplified and clinician-friendly[30] while also having high success rates.[32]

The SBO technique primarily relies on the sealer to fill a major portion of the canal and is employed using a gentle pumping motion.[33] The in-and-out motion along with the tapered shape of the gutta-percha cone and root canal may cumulatively result in the flow of excess sealer, not only apically but also coronally into the pulp chamber. This may consequently get coated onto the walls and floor of the pulp chamber, thereby interfering with the sealing ability of the intracoronal core material.

Bargrizan *et al.* reported an interesting observation that there was presence of interfacial gap and no adaptation between ZOE and GIC when used to perform pulpotomy in primary molar teeth.[34] B-RCS has the ability to form hydroxyapatite and produce a chemical bond to the root dentin.[35] Nevertheless, placement of GIC over bioceramic materials has shown to produce wide gaps along with microcracking and porosities in the interfacial region.[9,10] Currently, there are no studies assessing the interface and interaction between B-RCS and GIC. Thus, the present study aimed at assessing the role of various dentin conditioners on the interface between HVGIC and B-RCS/ZOE sealer. It is recommended to use phosphoric acid (34%–37%) or polyacrylic acid (10%–20%) to condition the tooth surface before the placement of GIC.[19] Hence, the dentin conditioners used in the present study were 37% phosphoric acid and 10% polyacrylic acid.

In the present study, dentin conditioning with 37% phosphoric acid produced better interfacial adaptation of HVGIC over ZOE/B-RCS-conditioned dentin in comparison to 10% polyacrylic acid and saline. Thus, the null hypothesis was rejected. In the present study, the presence of Zn K-alpha and Zr K-alpha

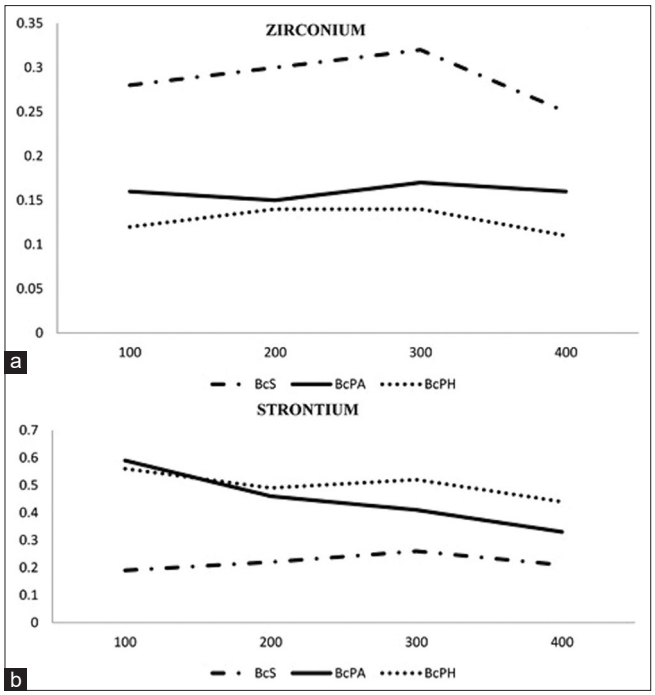


**Figure 3:** Elemental profile of (a) Zr and (b) Sr in the dentin side up to 400 μm from the interface in BcS, BcPA, and BcPH (Zr: Zirconium; Sr: Strontium).

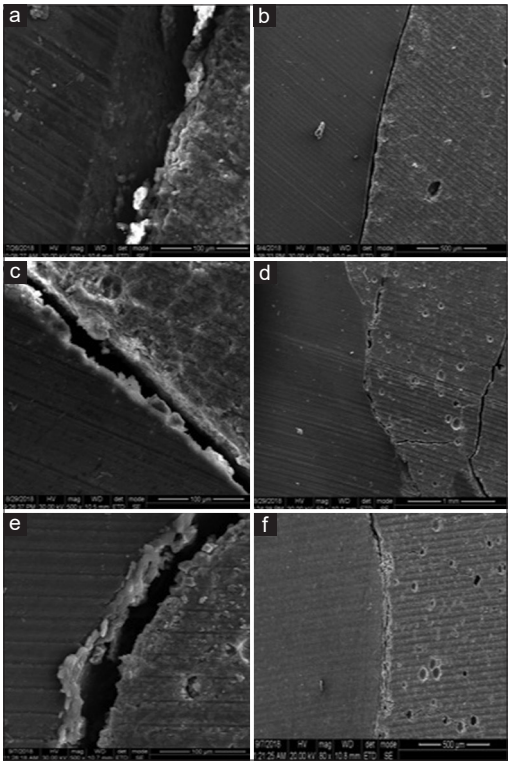


**Figure 4:** Backscattered images at different magnifications of the interfacial region of (a and b) ZnS group, (c and d) ZnPA group, and (e and f) ZnPH group.

elements in the spectroscopic (EDX) graph indicated the residue of ZOE sealer and B-RCS, respectively. The elements Al K-alpha, Si K-alpha, P K-alpha, and Ca K-alpha overlapped between B-RCS and HVGIC. Hence, the K-alpha peak of Sr was chosen to denote and quantify HVGIC.

The spectral analysis showed a distinct peak of Zn K-alpha, Zr K-alpha, and Sr K-alpha at 8.6 keV, 15.7 keV, and 14.16 keV, respectively. The molecular weight percentage (Wt%) of Zn K-alpha on the dentin side was observed to be 5.69, 1.91, and 1.52 with respect to the samples conditioned with saline, polyacrylic acid, and phosphoric acid. Similarly, the molecular Wt% of Zr K-alpha on the dentin side was 1.92 (BcS), 1.06 (BcPA), and 0.22 (BcPH). This indicates that treating the dentin with saline was ineffective to remove the ZOE/B-RCS residue from the walls and floors of the access cavities in comparison to polyacrylic and phosphoric acid groups.

The molecular Wt% of Sr K-alpha on the dentin side was 1.30, 5.26, and 7.11 in the ZnS, ZnPA, and ZnPH groups and 1.47, 4.56, and 6.23 in the BcS, BcPA, and BcPH groups, respectively. This implies that there was a poor penetration of HVGIC into the dentin in the samples treated with saline due to its inability to remove the sealer residue. Furthermore, this could have led to adhesive debonding failures in these groups. Further, hard tissue microtome of the tooth resulted in dislodgement of one sample in the ZnS group even before the SEM/EDX analysis. A probable reason could be that eugenol is a phenol-based compound which is insoluble in water.[36] Since it is hydrophobic, it could have formed an immiscible layer over the exposed dentin surface,[36] which might have interfered with the adhesion of HVGIC to the dentin.

Dentin conditioning with phosphoric acid was more effective than polyacrylic acid in the removal of root canal sealers from the access cavities. Lower molecular weight of phosphoric acid (97.995 g/mol)[37] in comparison to polyacrylic acid would facilitate easy penetration and conditioning of peritubular and intertubular dentin, thus removing smear layer along with smear plugs.[38] Furthermore, polyacrylic acid has a weak acidic nature in comparison to phosphoric acid.[39]

An interesting observation was the presence of microporosities on the HVGIC end of the interface in the groups conditioned with polyacrylic and phosphoric acid. The presence of any residual

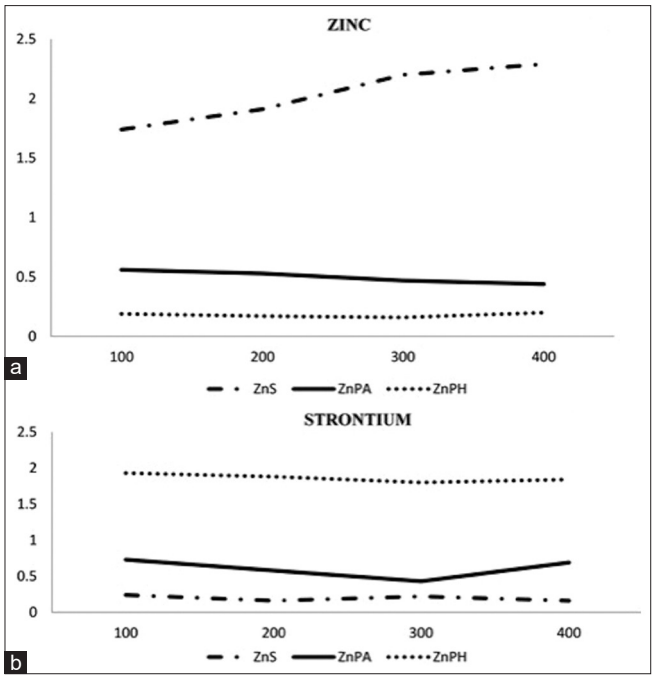


**Figure 5:** Elemental profile of (a) Zn and (b) Sr in the dentin side up to 400 μm from the interface in ZnS, ZnPA, and ZnPH (Zn: Zinc; Sr: Strontium).

polyacrylic or phosphoric acid after dentin conditioning could have depleted the water of hydration from HVGIC due to the hygroscopic nature of the acids,[40] thus causing microporosities. Extensive microporosity was reported by Namazikhah when MTA was exposed to acidic pH.[41]

Although cohesive debonding was seen in these groups, the presence of microporosities could hamper the bonding of HVGIC and its properties. The bond strength of GIC when layered over tricalcium silicates such as Biodentine or Theracal directly has been reported to be low.[10,42] The similarity in composition of tricalcium silicate cement and GIC leads to low ionic exchange which might affect the bond strength.[42]

The assessment of interface and interaction using SEM-EDX analysis has shown to be precise and reliable.[9] EDX analysis is regarded as qualitative analysis. The software considers the similar elements present on either side of the interface as a single entity (homogeneity).[9] In the current study, only the different elements across the interface were accounted for, thus the errors were not significant.

However, this study has certain limitations. SEM can assess only the surface of the interface, as opposed to transmission electron microscopy or confocal laser scanning microscopy which can provide a deeper analysis of the ultrastructure including the depth of penetration of elements. The cracks produced on the surface of HVGIC and the debonding of samples could be due to the high vacuum drying of SEM analysis. Future studies assessing bond strength between various restorative materials and bioceramic materials could provide more details on the nature of the bond formed between the materials.

## CONCLUSION

Conditioning of the endodontic access cavity using 37% phosphoric acid immediately postobturation resulted in better removal of the residual root canal sealer and higher penetration of HVGIC into the dentin, in comparison to the other dentin conditioners.

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### Conflicts of interest
The authors of this manuscript declare that they have no conflicts of interest, real or perceived, financial or non-financial in this article.

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