Comparative evaluation of a bioactive restorative material with resin modified glass ionomer for calcium-ion release and shear bond strength to dentin of primary teeth—an *in vitro* study

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Objectives: *This study aimed to evaluate the release of calcium ions from a bioactive restorative material and its shear bond strength (SBS) to primary dentin.* ***Study design:*** *Occlusal surface of extracted non-carious primary molars were flattened, onto which 2 × 2 mm cylinders of ACTIVA™ BioActive Restorative (PULPDENT® Corporation, Watertown MA) or Fuji II LC (GC Corporation, Tokyo, Japan) were prepared using a polypropylene straw mould. SBS of the materials to primary dentin was tested using a universal testing machine. The mode of bond failure was assessed using stereomicroscopy. 10 mm × 2 mm disks of each material were prepared and immersed in Milli-Q water for 1, 7, 14 and 21 days. The released calcium ions in the immersion media were quantified using Atomic Absorption Spectroscopy.* ***Results:*** *ACTIVA™ BioActive Restorative showed a mean SBS of 4.29 ± 0.65 MPa to primary dentin and calcium ion release of 0.76 ± 0.12 ppm over 21 days.* ***Conclusion:*** *ACTIVA™ BioActive Restorative showed a significantly higher mean SBS to primary dentin, and significantly higher calcium ion release compared to Fuji II LC.*

Keywords: *BioActive, Calcium ion release, Primary teeth, Shear bond strength*

INTRODUCTION

Fluoride-releasing materials have come to be the mainstay in paediatric restorative dentistry as a result of a shift towards the philosophy of preservative dentistry in the management of dental caries. Glass ionomers, besides displaying fluoride release and recharge abilities, bond chemically to the tooth structure and are extremely biocompatible[1]. Efforts to overcome their brittle nature, early moisture sensitivity, slower development of strength and subpar aesthetics led to the development of resin-modified glass ionomers (RMGIs) which were formulated with the incorporation of light-curable resins such as 2-Hydroxyethyl methacrylate (HEMA), or Bisphenol A glycidyl methacrylate (Bis-GMA) into the conventional glass ionomer matrix[2,3]. However, resin-based restorative materials may cause pulpal irritation and microleakage as a result of polymerisation shrinkage[4].

The philosophy of paediatric restorative dentistry not only involves the preservation of the natural dentition but also the reduction or elimination of tooth demineralisation. In this regard, a variety of materials with additives that reduce demineralisation of the tooth such as fluorides, calcium phosphates, *etc.*, have been investigated. Recently, ACTIVA™ BioActive Restorative (Pulpdent® Corporation, Watertown MA), a

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resin-based glass ionomer matrix without Bis-GMA or BPA-based monomers, was formulated with an ability to release calcium ions. It is claimed to be wear-resistant, fracture-resistant and shows less microleakage. By virtue of its ability to release calcium, phosphate, and fluoride ions, it is marketed as a bioactive material by the manufacturers [5]. *In vitro* studies on ACTIVA™ BioActive Restorative have demonstrated that the material shows fluoride release, superior mechanical characteristics, acceptable marginal integrity and wear resistance [4,6–10].

Bioactivity of any material can be gauged through estimation of its calcium, phosphate, and fluoride ion release [11–14]. From a clinical perspective, the release of calcium and phosphate ions can increase the oral environmental pH, leading to deposition of an apatite-like material [15].

ACTIVA™ BioActive Restorative has been used for posterior restorations in paediatric dentistry [16]. Existing literature indicates its beneficial effects when used in caries-prone areas, due to the release of calcium and phosphate [17]. However, such bioactivity of these materials is directly dependent on the quantum of release of ions which has not been reported widely.

Adhesion between a restorative material and dentin is crucial for the marginal integrity and longevity of a restoration [18]. In general, the bond strength of dental materials to dentin of primary teeth have been reported to be lower than that with permanent teeth. Low shear bond strength of restorative materials causes early loss of restoration under masticatory forces, leading to odontogenic re-infection [19], and it is a cause for concern in children showing high caries risk, who may require multiple interim restorations prior to the placement of full-coverage restorations. In this regard, it is interesting to investigate the presence of calcium and its release from a restorative material on the shear bond strength to the tooth. Although, the shear bond strength of ACTIVA™ BioActive Restorative to the dentin of permanent teeth has been studied [18], there is a lack of information regarding its shear bond strength to primary dentin.

Hence, this study aimed to quantify the release of calcium ion from ACTIVA™ BioActive Restorative and estimate its shear bond strength to primary dentin, compared to a conventional resin modified glass ionomer cement. The null hypothesis of the study was that there is no difference between the bioactive restorative material and resin modified glass ionomer in terms of calcium ion release and shear bond strength to dentin of primary teeth.

MATERIALS AND METHOD

12 non-carious primary molars were included in this study, that were extracted either due to pre-shedding mobility or orthodontic reasons. Carious, hypoplastic or fractured teeth were excluded from the study. To achieve a 5% level of significance at 80% power, a sample size of 6 per group was obtained. The study duration was 4 months, extending from September 2020 to December 2020 following approval by the institutional ethical committee.

For estimation of both shear bond strength to dentin of primary teeth and calcium ion release, two study groups were formed (Table 1), namely:

Group A: ACTIVA™ BioActive Restorative

Group B: Fuji II LC

The extracted primary teeth were cleaned and stored in 0.9% saline, till they were subjected to intervention, as reported in previous studies [20]. The surface enamel was removed using a carborundum disc under running water, until a flat smooth surface of fresh dentin was exposed, to standardise the smear layer formed [21].

The samples were mounted on acrylic blocks of 50 mm height, 20 mm width and 15 mm thickness prepared using autopolymerising acrylic resin (RR Rapid Cure, DPI, India) poured into a custom-fabricated hydrophilic vinyl polysiloxane mould (Reprosil, Dentsply Sirona Pty Ltd.). The flattened occlusal surfaces of the samples were kept exposed. The acrylic blocks were then stored in distilled water at 25 °C, until testing was carried out. The prepared samples were randomly allocated to the two study groups.

Group A: ACTIVA™ BioActive Restorative

The dentin of the specimens was air-dried but not completely desiccated, as recommended by the manufacturer, and was etched using 37% phosphoric acid etchant (Eco-Etch™, Vivadent, Lichtenstein), gently agitated to avoid skipping effect [22,23], using an applicator tip for 15 seconds. Following rinsing of the etchant and air drying, bonding agent (3M ESPE Adper Single Bond) was applied using a micro brush for 20 seconds, gently dried with a blast of air, and light cured using calibrated Montex BlueLEX LED unit (intensity: 1000 mW/cm^2; Output: 9 V, 1.3 A, 5 W; Wavelength: 420–490 nm).

A polypropylene straw of 2 mm internal diameter was used as a mould to build material cylinders on the prepared tooth surfaces for shear bond strength testing. The material was dispensed (premixed) using the auto mix syringe provided by the manufacturer onto the tooth surface to form a cylinder of 2 mm height and 2 mm diameter. The material was then light cured, and the samples were aged in distilled water for 24 hrs prior to the testing of shear bond strength.

Group B: Fuji II LC

The powder and liquid were weighed on an electronic weighing scale, mixed as per the recommended ratio, and dispensed onto the exposed dentinal surface in a single increment of 2 mm, using a polypropylene straw. The material was then light cured for 20 seconds using the light cure unit. The samples were stored in distilled water for 24 hours, prior to the shear bond strength measurement.

Shear Bond Strength Evaluation

The specimens were mounted on to a universal testing machine (Wagner Beam Setup, Bengaluru, India) with the machine crosshead oriented perpendicular to the interface of the cylinder and the tooth surface. Each specimen was subjected to shear bond testing at a crosshead speed of 0.5 mm/min, until there was debonding of cylindrical specimens from the tooth surface. Maximum load recorded during the testing divided

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Table 1: Composition and manufacturer's instructions for the materials used in the study.

Material	Manufacturer (Lot number)	Composition
Group A:	Pulpdent®, Watertown, MA, USA	Blend of diurethane methacrylate and other methacrylates with modified polyacrylic acid
ACTIVA™ BioActive Restorative	(Lot No. 181204)	Silica, amorphous
		Sodium fluoride
Group B: GC Fuji II LC	GC Corporation, Tokyo, Japan (Lot No. 1912251)	Powder: Fluoro-aluminosilicate glass Liquid: Acrylic-maleic acid copolymer HEMA Water Camphoroquinone

by the area of the specimens was considered as shear bond strength and reported in MPa (n = 6).

Stereomicroscopic Analysis of fractured surfaces

Following SBS testing, the samples were analysed under a stereomicroscope (Labomed CSM2, Amar Industries, India) at 4 × magnification in order to assess mode of bond failure. The mode of bond failure was then categorised as (i) adhesive failure, (ii) cohesive failure or (iii) a mixture of both [18].

Estimation of calcium ion release

A custom-fabricated mould of 2 mm thickness was prepared from hydrophilic vinyl polysiloxane (Reprosil, Dentsply Sirona Pty Ltd). Circular cavities of 10 mm diameter and 2 mm depth were punched out from the mould, in which the test materials were poured, and light cured for 20 seconds. The discs were then immersed in double deionised water (Milli-Q water) at 25 °C.

At regular intervals of time, the discs were taken out and a 30 mL sample of the immersion media was analysed for the presence of Calcium ions released from the test specimens via Atomic Absorption Spectroscopy (AAS) using iCE 3000 Series Atomic Spectrometer (Thermo Fisher Scientific Inc., Waltham, Massachusetts, USA). The amount of calcium ion release was reported in parts per million (ppm) (n = 6).

Statistical Analysis

Data was tabulated on an excel spreadsheet, and the statistical analysis was done using Minitab software, v.19.2020.1 (Minitab, LLC, State College, PA, USA) with level of significance $p < 0.05$. Mann-Whitney U test was performed to determine the difference between shear bond strengths of ACTIVA™ BioActive Restorative and resin modified glass ionomer cement. Calcium ion release values were analysed using Wilcoxon signed ranked test for intragroup comparison between the time intervals, and intergroup comparison was done using Mann-Whitney U test. A p-value < 0.05 illustrated that the null hypothesis can be rejected, implying that the median values of both samples are not equal.

RESULTS

Shear bond strength (SBS) testing

Table 2 shows the mean and median values of macro-shear bond strength of groups A and B. Intergroup comparison using Mann-Whitney U test indicated that group A (ACTIVA™ BioActive Restorative) showed a significantly higher SBS to primary dentin than that of Group B (Fuji II LC). Fig. 1 represents the box-plot graph of shear bond strength of group A and group B.

Stereomicroscopy results

As shown in Table 2, both the groups showed predominantly adhesive failures following debonding from dentin.

Calcium ion release

The mean and standard deviation of group A and group B for calcium ion release are tabulated in Table 3. The normality of the groups of data was tested using Anderson darling's test and the samples were found to be not normal. Pairwise comparisons were done using Wilcoxon signed rank test. For group A, it was observed that there was a significantly higher calcium release between 24 hours and 7 days, and between 14 days and 21 days. The difference between calcium release between 7 days and 14 days was not significant. For group B, there was a consistent release of calcium ion during the study period, but the difference in calcium ion release between 24 hours and 7 days, between 7 days and 14 days and between 14 days and 21 days, was not statistically significant.

Intergroup comparison was done using Mann-Whitney U test. It was observed that there was a significant difference in calcium estimates between group A and group B after 24 hours, 7 days and 21 days. The difference between the calcium ion release between both the groups at 14 days was not statistically significant.

From the box plot graph of calcium ion release at varying time intervals, it can be observed that the mean values of group A were more than the mean values of group B at 24 hours, 7 days, 14 days, and 21 days (Fig. 2).

DISCUSSION

This *in vitro* experimental study was undertaken to evaluate the release of calcium ion and shear bond strength of a bioactive restorative material to the dentin of primary molars.

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Table 2: Intergroup comparison of shear bond strength to primary dentin between groups A and B using Mann-Whitney U test.

Groups	Mean SBS (in MPa) and Standard Deviation (S.D.)	Median	p value (<0.05)	Mode of Bond Failure
A: ACTIVA™ BioActive Restorative	4.29 (0.65)	3.90	0.0122*	Adhesive: 83% Cohesive: - Mixed: 17%
B: Fuji II LC	2.47 (0.32)	2.34		Adhesive: 66.7% Cohesive: 16.6% Mixed: 16.6%

**Statistically significant. SBS: shear bond strength.*

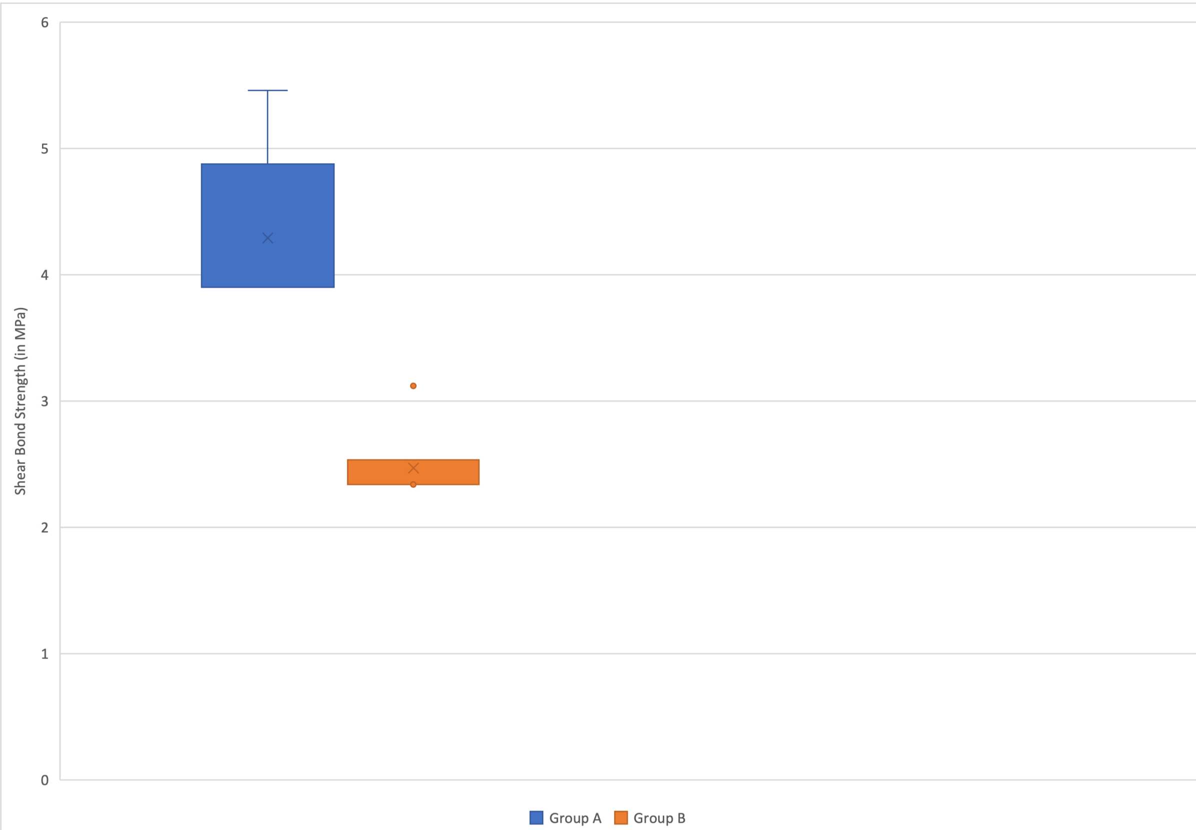

Figure 1: Box plot for comparison of shear bond strength between group A and group B.

In dentistry, restorative materials have been used to restore the form and function of a tooth, either temporarily or permanently. The longevity of a dental restoration relies on its strength and adhesion to the tooth structure. Moreover, the materials should be biocompatible and have acceptable aesthetics comparable to that of the natural tooth [24]. Glass ionomer cements have been indicated in children with high caries risk, due to their property of fluoride release and recharge. However, their brittle nature, along with their subpar aesthetics and relatively inferior mechanical properties, has led to the development of resin modified glass ionomers (RMGIs) [12].

RMGIs contain added light-curable resins in the glass ionomer matrix such as 2-hydroxyethyl methacrylate (HEMA) or Bis-GMA [12]. These are dispensed as powder-liquid systems, which can be mixed manually or via premixed capsules. After mixing the powder and liquid, an acid-base reaction occurs along with the polymerisation initiated by light curing. The uniformity of the polymerisation is ensured by light curing the RMGI in increments of 2 mm [25]. Various studies have demonstrated their high durability, greater bond strengths and release of fluoride [26–29]. However, RMGIs are susceptible to microleakage in both primary and permanent teeth [2,19,25,30–42] and have shown secondary caries formation, which are the primary reasons for their observed low success rates, especially in primary teeth [42]. Their fluoride release is also lesser than that observed in conventional glass ionomers, which is attributed to their polymerised resin matrix preventing ion-exchange with the external environment [43].

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Table 3: Intergroup comparison of mean calcium ion release (in ppm) at different time intervals using Wilcoxon signed ranked test.

Groups	No. of Samples	Mean (ppm) and Standard Deviation			
		24 hours	7 days	14 days	21 days
A: ACTIVA™ BioActive Restorative	6	0.371 (0.053)^{a}	0.463 (0.017)^{b}	0.491 (0.076)^{b}	0.768 (0.127)^{c}
B: Fuji II LC	6	0.198 (0.074)^{a}	0.281 (0.041)^{a}	0.430 (0.179)^{a,b}	0.525 (0.121)^{b,c}
Group A *vs.* Group B ($p < 0.05$)		0.008^{x}	0.005^{y}	0.810	0.020^{z}

The same lower script letters a, b, c indicate no significant difference within the group at different time intervals; x, y, z indicate a significant difference between the groups at different time intervals.

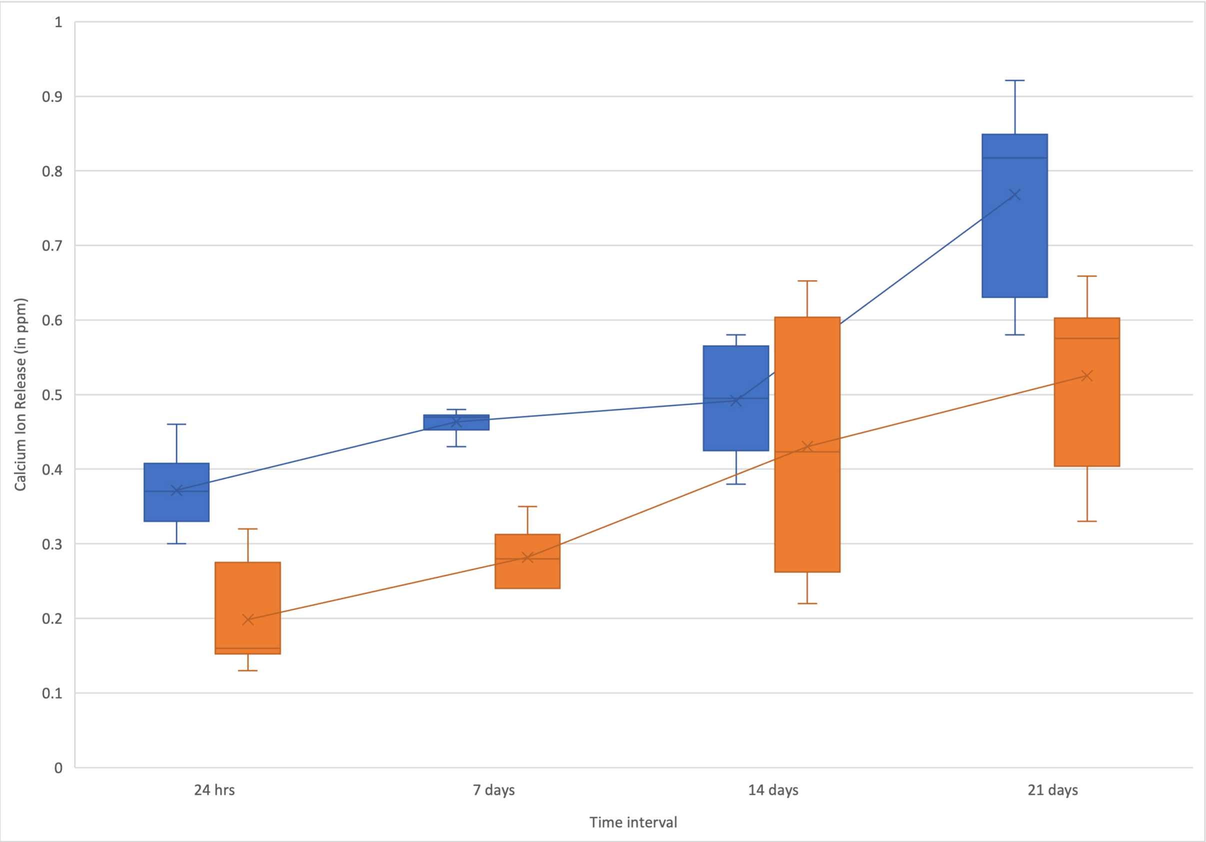

Figure 2: Trend of calcium ion release and box plot for comparison between group A and group B materials at different time intervals.

In order to overcome these drawbacks, a variety of compositional modifications have been done by adding various additives to RMGIs. ACTIVA™ BioActive Restorative is a resin-modified glass ionomer that is claimed to have bioactive properties and superior mechanical strength than conventional GICs and RMGIs[4].Earlier investigations on ACTIVA™ BioActive Restorative have demonstrated its high flexural strength[5], wear resistance[6], fluoride release[7,10], diametral tensile strength and surface hardness[8,9], and shear bond strength to permanent dentin[11].

Bioactive materials constitute a part of the major advancements in restorative materials, where a paradigm shift is seen from a "passive" restorative material that may restore the form and function of a tooth, to an "active" material that promotes tooth remineralisation and improves marginal integrity of a restoration. Bioactivity of a restorative material is described based on several properties, namely the ability to remineralise the tooth structure, induce hydroxyapatite formation, chemical adhesion to the tooth surface via ion exchange, antibacterial properties and biocompatibility[12]. In order for it to be considered truly bioactive, a restorative material must exhibit hydroxyapatite formation[12]. This can be measured indirectly via detection of calcium, phosphate, and fluoride ion release *in vitro*. Currently, there is limited literature assessing bioactivity of ACTIVA™ BioActive Restorative via ion release[7,10,44].

Atomic Absorption Spectroscopy (AAS) can accurately quantify calcium ions released from a material into an immersion medium. In the present study, the release of calcium ions

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was observed for a 21-day period, as long-term immersion may lead to saturation of the liquid due to continuous passive calcium ion release [45]. The trend of calcium ion release exhibited by ACTIVA™ BioActive Restorative over the course of 21 days in our study, suggests that it releases calcium ions over time and may be considered bioactive. In the present study, the calcium ion release was found to be significantly low for Fuji II LC compared to ACTIVA™ BioActive Restorative. Despite being resin modified glass ionomer cements, the observed differences in the calcium ion release can be attributed to the differences in their composition. ACTIVA™ BioActive Restorative contains a flexible hydrophilic resin matrix based on diurethane dimethacrylate that favours the release of fluoride and phosphate ions, which is pH-dependent. This compositional modification imparts low water solubility and water sorption characteristics to ACTIVA™ BioActive Restorative [46]. Fuji II LC, on the other hand, is based on conventional resin matrix materials such as Bis-GMA that are hydrophobic in nature. Since hydrophobic resin materials are less permeable to water, they tend to release lesser ions.

Under physiological conditions, the ability of a restorative material to induce hydroxyapatite formation relies on its calcium ion release. The released calcium reacts with phosphate ions present in the saliva or in the restorative material itself and can facilitate hard tissue repair [47].The Si-O-Si bonds present in bioglass hydrolyse in the presence of low pH and moisture, leading to a rapid release of fluoride, calcium, silicon, and hydroxyl ions in the oral environment. The resultant alkaline environment created due to hydroxyl ion release allows ion deposition to the tooth structure and inhibition of bacterial growth [12].The results for the estimation of calcium ion release in the present study suggest that ACTIVA™ BioActive Restorative releases higher calcium ion content and exhibits superior shear bond strength to primary dentin compared to Fuji II LC. Due to the in vitro nature of the present study, intraoral conditions were not simulated, and the results must be appropriately inferred. Further studies may be carried out to estimate the quantum of long-term calcium and phosphate ion release.

The longevity of any restoration can be predicted by the strength of adhesion of the restorative material to prepared tooth structure [46]. Shear bond strength (SBS) refers to the ability of two materials to withstand sliding forces applied at their junction. In posterior teeth, high shearing forces are exerted during mastication, which may lead to the restorative material debonding from the prepared tooth surfaces [47]. This becomes clinically relevant especially in class II restorations in primary teeth, where the risk of dislodgement of the restoration is high [45].

SBS testing is beneficial in specimens with large areas of bonding, despite its questionable validity in areas of heterogenous stress distribution [48]. Among the materials evaluated in the present study, ACTIVA™ BioActive Restorative showed higher bond strength compared to Fuji II LC. However, the bond strength values observed in the present study are lower than the reported values of bond strength on permanent teeth [49].

Appropriate use of dentin conditioners is a crucial step in achieving good bonding. The use of an etchant and bonding agent further improves a material's bond strength to dentin, attributed to smear layer removal and unplugging of the dentinal tubules. This results in a partially demineralised dentin with an increased surface area for bonding. The resultant microporosities following conditioning and chemical interactions of the carboxyl groups of the conditioner with calcium from the hydroxyapatite around exposed dentinal collagen, contribute to the stronger bond strength between a glass ionomer and dentin [50].It must be borne in mind that dentin conditioners remove smear layer faster in primary teeth than permanent teeth. Surface treatment with a stronger conditioner, such as phosphoric acid can lead to loss of calcium ions from the bonding surface and a resultant weaker bond between the two materials [51]. The low SBS obtained for ACTIVA™ BioActive Restorative in the present study can be attributed to the above-mentioned fact, even though 37% phosphoric acid was used for dentin conditioning as recommended by manufacturers. Shortening the conditioning time or the use of weaker acid solutions for conditioning primary teeth has thus been advocated [50]. Further investigations may be carried out to assess the effect of lowering etching time on the bond strength of ACTIVA™ BioActive Restorative to primary tooth dentin.

Alkhudhairy *et al.* [18] reported higher macro-SBS values for ACTIVA™ BioActive Restorative (18.45 $\pm$ 1.34 MPa) to permanent dentin. In the present study, the mean macro-SBS for ACTIVA™ BioActive Restorative to primary dentin was found to be less (4.29 $\pm$ 0.65 MPa). This can be attributed to the structural differences and mineral concentration gradients between primary and permanent dentin. Dentin of primary teeth has straight dentinal tubules, whereas permanent dentin has "s-shaped" dentinal tubules. This leads to a lesser surface area for bonding of the restorative material to dentin of primary teeth [52]. Courson *et al.* [53] stated that calcium and phosphorous concentrations in intertubular and peritubular dentin of deciduous teeth are lesser than in permanent teeth, which may affect the bond strength to primary dentin. The high density and larger diameters of dentinal tubules, and higher number of micro canals in the dentin of primary teeth than in permanent teeth may cause interference with the adhesion of the restorative material, leading to lower bond strength values [50]. Primary teeth are also more demineralised than permanent teeth [52]. The lower inorganic content in primary teeth along with lesser tubular density may contribute to their decreased chemical and micromechanical adhesion to a restorative material [25]. The mean value of macro-SBS for Fuji II LC to primary dentin in this study was also lower than the values reported in other studies [50,54,56]. Since the difference between the mean macro-SBS to primary dentin of ACTIVA™ BioActive Restorative and Fuji II LC was statistically significant, the null hypothesis of the study was rejected.

The assessment of bond failure can give an indication of the nature of bonding between the restorative material and dentin. Adhesive failures refer to disruption of bonds between the molecules or atoms of two different types of materials, while cohesive failures refer to a disruption of bonds between molecules or atoms of the same species [47]. Adhesive failures at the interface between a restorative material and dentin are

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characterised by open dentinal tubules, while cohesive failures demonstrate an intact hybrid layer [48]. In the present study ACTIVA™ BioActive Restorative demonstrated predominantly adhesive failures. This is in agreement with the study by Alkhudhairy *et al.* [18] where conventional etch and rinse technique used with a universal bonding agent resulted in predominantly adhesive failures with permanent dentin. Fuji II LC showed predominantly adhesive failures with primary dentin as well, which is in agreement with Pacifici *et al.* [50] but in contrast to Abdelmegid and co-authors [56], who obtained predominantly mixed bond failures.

Further studies on the effects of etching time and etchant concentration on the shear bond strength of ACTIVA™ BioActive Restorative to primary dentin can be pursued. The ability of ACTIVA™ BioActive Restorative to bond with affected dentin may also be investigated, as it may bond better with the exposed collagen of the demineralised dentin. Moreover, as the calcium ion release from restorative materials is known to initiate remineralisation of the tooth, the effect of such induced remineralisation on bond strength can also be evaluated.

CONCLUSION

The results of the present study indicate that ACTIVA^{TM} BioActive Restorative releases significantly higher calcium ion over a 21-day period and also exhibits significantly higher shear bond strength to the dentin of primary teeth compared to Fuji II LC. Based on these findings, ACTIVA™ BioActive Restorative may be considered a fair alternative for posterior restorations in primary teeth in children at a high caries risk.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

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