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Comparative evaluation of the type of fracture mode of a 7th generation self etch adhesive and an 8th generation universal adhesive on dentin of deciduous teeth following micro-tensile bond strength test. An *in-vitro* scanning electron microscopic study

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Abstract
Introduction: Composite restorations are placed following pretreatment of cavities with an adhesive system. The degree of interface adhesion and chemical stability is critical for clinical success of restoration.
Aim and Objectives: The objectives of this *in vitro* study were to compare and evaluate the fracture mode of 7th generation adhesive and universal adhesive on dentin of deciduous teeth following micro tensile bond strength test under scanning electron microscope.
Methodology: Flat buccal surface of 10 teeth each were divided into 2 groups and treated with GC-G BOND (7th Generation Adhesive) (Group 1); GC-G-PREMIO BOND (Universal adhesive) (Group 2) to develop a composite resin cone. The tensile bond strength was measured using Instron Universal testing machine. Fracture mode was evaluated from each group under SEM.
Results: The greatest mean adhesive fracture was exhibited by 7th Generation adhesive while Universal adhesives exhibited majority of resin cohesive fractures.
Conclusion: Fracture mode of a material is dependent on the adhesive system, substrate depth and adhesive depth interaction. Hence composition of the adhesive and substrate treatment should be considered for good adhesion.

Keywords: tensile bond strength, 8th generation adhesives, 7th generation self-etch adhesives, fracture mode

Introduction

Tooth-coloured restorative materials are widely used in paediatric dentistry for the restoration of carious primary teeth. Of these materials, resin composites have been gaining increasing popularity over the past few decades because of their favourable aesthetic and mechanical properties. Aside from the possible problems encountered during patient management, the aspect ratio of cavities in primary dentition is often quite unfavourable for the use of conventional restorative materials. In addition to these factors are the significant occlusal loads that even young children can inflict on their teeth. Therefore, composite core material should have excellent bond strength to the pulpal floor dentin to maximize the seal and improve resistance to fracture [1].

Primary dentin has a higher tubular density, with a larger diameter in peritubular and intertubular dentin and lower mineral content, turning this substrate more reactive to acid conditioners. Therefore, an increased demineralization occurs in primary dentin [2, 3]. The deeper demineralization of primary dentin jeopardizes the adhesion by the collapse of collagen fibrils, calcium phosphate crystals precipitation, and thus, less penetration of resin monomers occurs into the demineralized dentin [4, 5]. The unprotected collagen fibrils zone formed at the base of the hybrid layer is considered the weakest area within the adhesive interface which is highly susceptible to both hydrolytic and enzymatic long-term deterioration [5, 6].

Advancements in the formulation and application of dentin bonding agents have moved from a multistep bonding process (etching, washing, drying, primer, adhesive) to simplification i.e., self-etch and single bottle system [7]. The seventh-generation bonding systems were introduced between the late 1990s and early 2000s.

The seventh-generation or all in one adhesive combine etchant, primer and adhesive in one bottle. The advantage of this generation was that no mixing was required and the bond strengths reported were found to be fairly consistent [7]. Innovations in nanotech dentistry have led to the development of nano-composites and nano-adhesives which contains nano-sized fillers which are currently marketed as eighth-generation bonding agents. The incorporation of nano-fillers with an average particle size of 12 nm has been claimed to increase the penetration of resin monomers and the hybrid layer thickness, which further improves the mechanical properties of the bonding systems [8].

In vitro tests play a critical role in providing the necessary information regarding the effectiveness of new materials in a short period. The most routinely applied laboratory parameters to evaluate the effectiveness of dentin bonding agents are assessment of fracture modes and evaluation of shear and tensile bond strength tests. While bonding to permanent teeth has been studied abundantly, very few studies have addressed resin bonding to deciduous teeth using the newest nano bonding agents. Thus, the aim of this *in vitro* study was to evaluate the type of fracture mode of a 7th generation self-etch adhesive and 8th generation universal adhesive on dentin of deciduous teeth following micro-tensile bond strength test.

Materials & Methodology

Materials

1. Teeth- 20 primary molar teeth.

INCLUSION CRITERIA

- 20 primary molar teeth with intact buccal and lingual surfaces.

EXCLUSION CRITERIA

- Teeth with caries on both buccal and lingual surfaces, fractured crown, hypoplastic or hypo mineralized teeth or any kind of developmental anomaly.

2. 7th Generation Adhesive (GC-G-Bond, GC America Inc)
3. 8th Generation Universal Adhesive (GC-G-Premio Bond, GC America Inc)
4. Composite Resin (Tetric N Ceram, Ivoclar Vivadent)
5. Silicone Rubber mould for embedding specimens in acrylic resin.
6. Cold Cure Acrylic Resin
7. Hollow metal spilt Teflon mould

Fig 1: Tooth Specimens

Fig 2: GC-G-BOND (7th Generation Adhesive)

Fig 3: GC-G-PREMIO BOND (8th Generation Adhesive)

Fig 4: TETRIC N CERAM, Ivoclar Vivadent (Composite Resin)

Equipments
1. Universal Testing Machine (UTM) (Instron-3365)
2. Air Rotor Handpiece (NSK S max Pico B2) With Straight Fissure Diamond point
3. Orthodontic (Model) Trimmer (Unident Instruments Pvt. Ltd)
4. Visible Light Cure Unit (Woodpecker Mini- S Light Cure LED)

Methodology
Collection of Samples
20 freshly extracted primary molar teeth were washed and cleaned with periodontal scaler and stored in distilled water until use.

Sample Preparation
Routine prophylactic procedures were carried out using aqueous slurry of pumice on all the teeth before they were treated. Depth orientation holes measuring 1.5 mm weremade in the most prominent part of buccal/lingual surface of each tooth with a straight fissure diamond point. All the samples were then wet ground on a model trimmer to expose a flat dentinal surface. Specimens were then embedded in cold cure acrylic resin within their respective silicone rubber moulds horizontally exposing the flat dentinal surface. The samples were then categorized into 2 groups of 10 specimens each namely, (Group I- GC-G-Bond-7th generation adhesive, Group II - GC-G-Premio Bond-8th generation adhesive). Each group was colour coded for identification *viz*. (Group I-colour coded Pink, Group II - colour coded Blue), following these the respective bonding agents were applied according to manufacturer's instructions.

Tensile Bond Strength Evaluation
A hollow metal split Teflon mould was used to build an inverted composite resin cone on the adhesive treated surface of the specimens. The diameter of the inverted cone-shaped hollow measured 2mm at its lower end, 4mm at its upper end and 5mm in height. Once the mould was held upright and secured on the dentinal surface with the help of adjustment screws, an increment of composite resin of thickness 2 mm was placed inside the mould and condensed. A 26-gauge ligature wire was fabricated in such a way that it was twisted at its one end and looped at the opposite end, following this, the twisted end was placed inside the 2 mm of composite resin, held straight and light-cured for 40 seconds. The second increment of 2mm thickness of composite resin was placed over the first increment and light-cured for 40 seconds. The final increment of 1mm thickness of composite resin was then placed over the second increment and cured for 40 seconds. Following the complete curing, the metal mould was split and removed from the surface of the specimen leaving a composite resin cone of height 5mm along with the twisted wire bonded to 2mm surface area of the dentin. The metal mould was reused for the preparation of the other specimens in the same way. All the specimens were immersed in water for 24 hours at room temperature. The tensile bond strength was then measured using the Universal Testing Machine. The resin

block was clamped to the stable lower jaw of the Universal Testing Machine and the wire was looped onto the movable upper jaw of the same. A cross head speed (CHS) of 1mm /minute was selected and the tensile load was applied to the specimen until the composite resin inverted cone was dislodged from the surface of the tooth. The breaking load values for each specimen were recorded through a computer connected to Universal Testing Machine. The values obtained were in ‘Kilogram (kg)’ and bond strength was calculated using the formula mentioned below, and expressed in ‘Megapascal (MPa)’.

$$\text{Tensile Bond Strength (MPa)} = \frac{\text{Load} \ \underline{r}}{\text{Surface area (mm}^2\text{)}}$$

Surface area = πr^2 (r = radius of bonded material)
The values obtained thereby were subjected to statistical analysis.

Sample Preparation for Sem Analysis

After testing the tensile bond strength, specimens were selected from each group. The tooth part of each specimen was wet ground by a metal disc to get a $5mm^2$ tooth specimen without any disturbance to the de-bonded surface. The wire elements were cut from the de-bonded resin cones without any disturbance to the de-bonded surface. Then, a total of 20 pairs of tooth and resin specimens were kept separately in petri dishes with their respective group name and number. After dehydration of tooth and resin specimens in a hot air oven, they were mounted in a sputter coater (auto fine coater) and platinum coating was given on de-bonded surfaces under a small electric field at vacuum. Then specimens were then transferred to SEM and examined at 100x and 500x magnification.

Fig 5: Tooth Sectioning

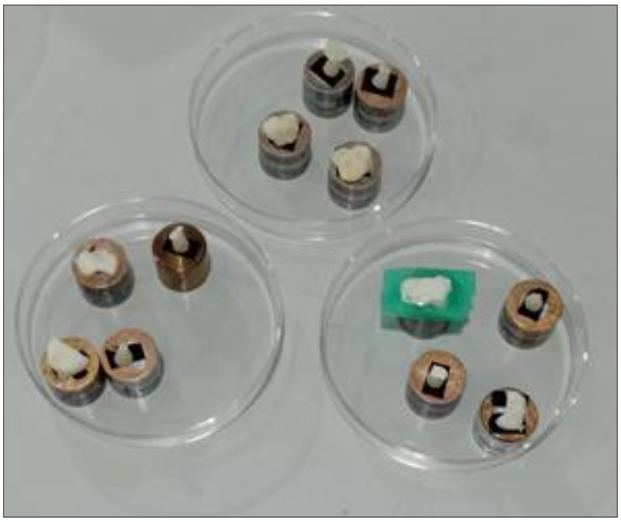


Fig 6: Specimens Mounted On Aluminium Stubs

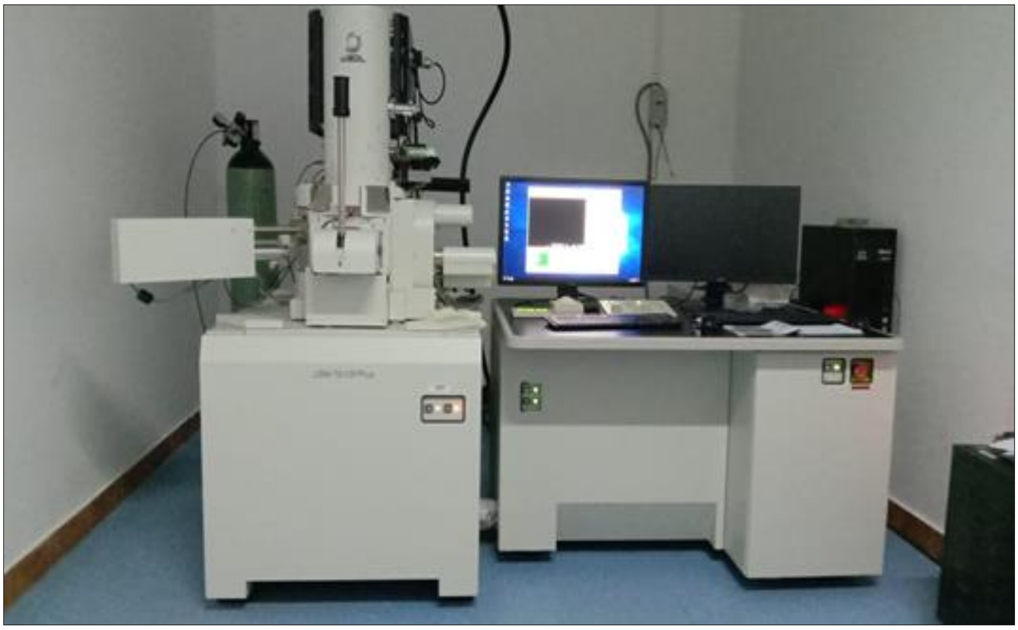


Fig 7: Scanning Electron Microscope Model No: JSM 7610 FPLUS

Fracture modes were designated according to the following criteria (Knobloch, Gailey *et al*. 2007): [9]

Adhesive Fracture: It means that there is <25% of adhesive resin, composite resin and /or dentin on the interface at the dentin side.

Mixed Fracture: It means that in some areas of the interface, there is adhesion type failure and in some areas, there is cohesive failure in adhesive resin, composite resin and/or dentin.

Dentin Cohesive Fracture: It means that there is 75% or >75% dentin on the interface at the composite side.

Resin Cohesive Fracture: It means that there is 75% or >75% composite resin on the interface at the dentin side.

Results

The composite resin–dentin interfaces of each sample of the groups were examined by SEM analysis at different magnifications. When SEM images of fracture types were examined, smear plugs and dentin surface covered with tags were observed in adhesive type fracture. During the examination of cohesive fracture type in composite, it was seen that the dentin surface was covered with resin. Upon examining the images of mix fracture type, smear-coated tubules and composite resin residues were observed on dentin surface.

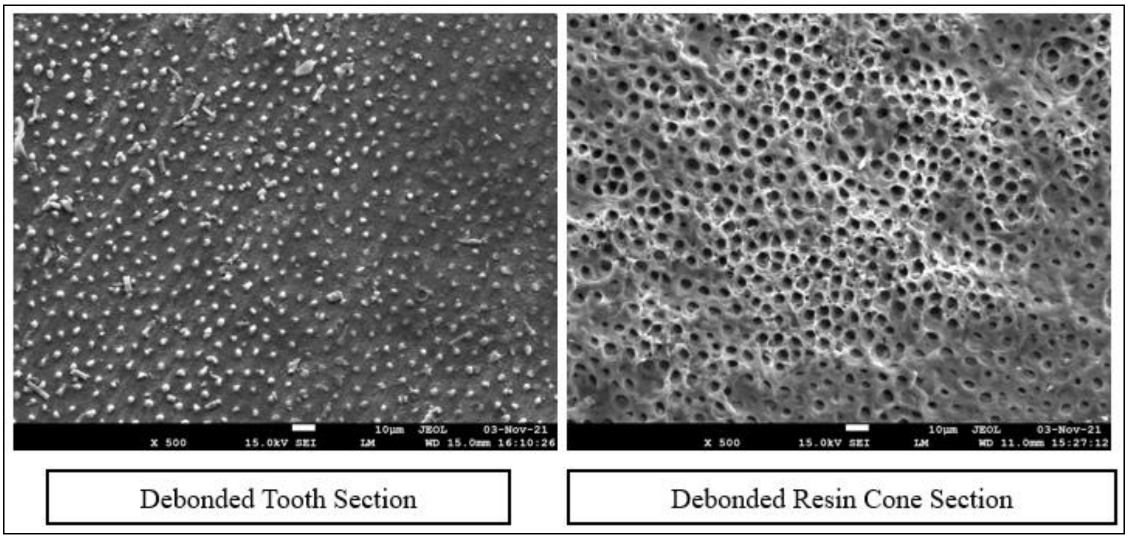


Fig 8: SEM Images of Group I-GC-G-BOND showing Adhesive Fracture

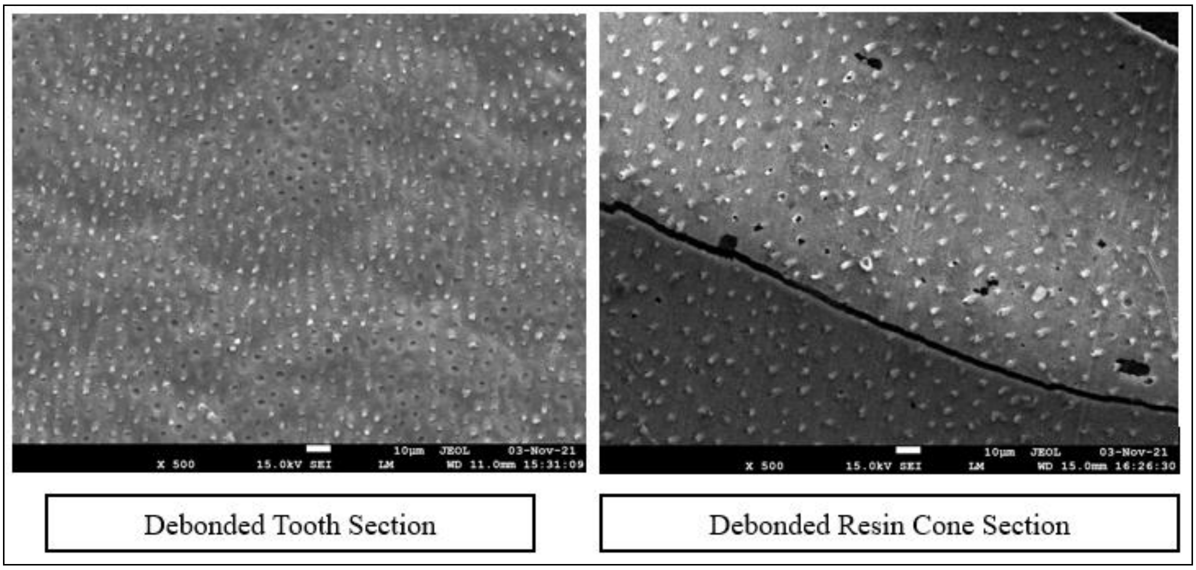


Fig 9: SEM Images of Group II-GC-G PREMIO BOND showing Resin cohesive Fractures

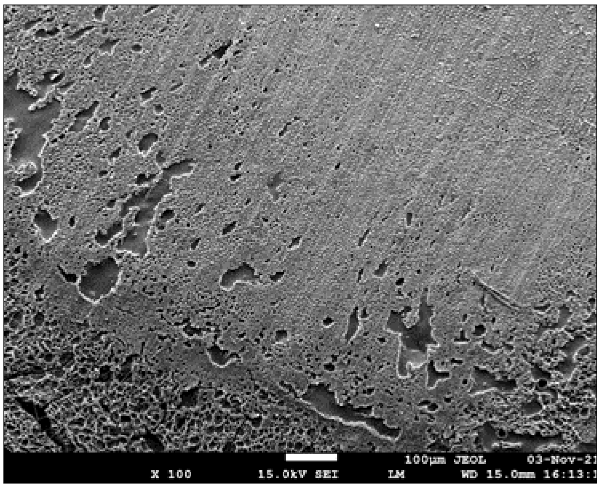


Fig 10: SEM Images of Group I-GC-G BOND showing mixed fracture.

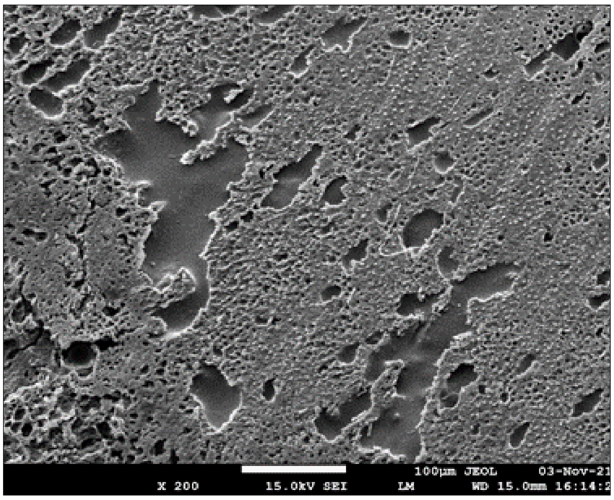


Fig 11: SEM Images of Group II-GC-G PREMIO BOND showing mixed fracture.

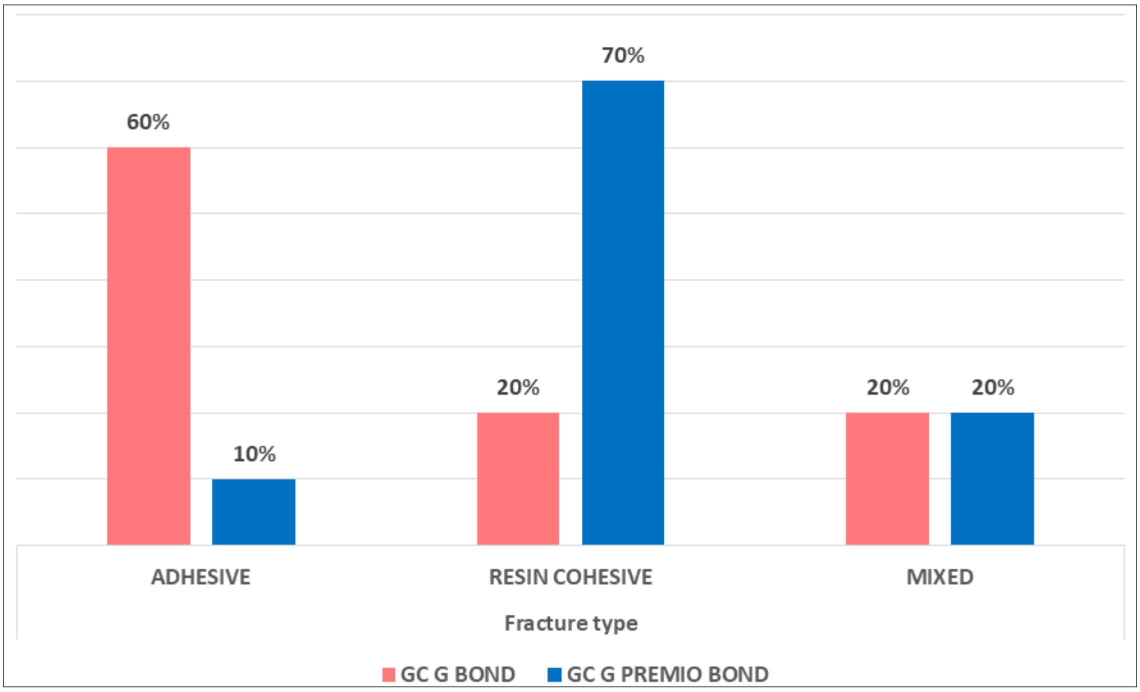
The results of the fracture mode are presented in Table 1. According to the Chi-Square analysis, there was significant difference in the distribution of fracture modes between the two groups (p=0.042) Table 2. According to the SEM analysis, (Group I-GC-G-Bond) showed a maximum of adhesive fractures (60%), while (Group II-GC-G Premio Bond) showed a maximum of resin cohesive fracture (70%). There was an equal distribution of mixed fractures (20%) between the two groups.

Table 1: Distribution of Fracture Modes Between the Groups

		Fracture type * Group Cross tabulation			
			Group I	**Group II**	
			GC G BOND	GC G PREMIO BOND	Total
Fracture type	ADHESIVE	Count	6	1	7
		% within group	60.00%	10.00%	35.00%
	RESIN COHESIVE	Count	2	7	9
		% within group	20.00%	70.00%	45.00%
	MIXED	Count	2	2	4
		% within group	20.00%	20.00%	20.00%
Total		Count	10	10	20
		% within group	100.00%	100.00%	100.00%

Table 2: Results of Chi-Square test showing statistically significant difference between two groups.

	Chi-Square Tests		
	Value	Df	Asymp.Sig. (sided)
Pearson Chi Square	6.349a	2	0.042
Likelihood ratio	6.904	2	0.032
Linear- by- Linear association	2.251	1	0.134
No of valid cases	20		



Graph 1: showing the percentage distribution of fracture modes.

Discussion

Increasing demands for aesthetic restorative treatments have led to the development of adhesive integrated materials such as adhesive systems and composites along with techniques aimed at restoring the natural tooth appearance. Therefore, the objective of an esthetic dental restoration is to obtain morphologic, optical and biologic outcomes miming natural enamel and dentine [10].

A dentin bonding agent can be defined as a thin layer of resin applied between the conditioned dentin and resin matrix of the composite. With progress in technologies, dental adhesives have evolved from no-etch to total-etch (4th and 5th generation) to self-etch (6th, 7th and 8th generation) systems [7].

The seventh-generation bonding systems introduced between 1999 and early 2000s combined all the ingredients required for bonding and was delivered from a single bottle. The advantage of this generation was that no prior mixing was required [11, 12].

Since filled bonding agents produced higher *in vitro* bond strength [10]. The eighth-generation bonding agents containing nano fillers of average particle size of 12 nm were introduced in 2010. These new agents from the self-etch generations had acidic hydrophilic monomers that could be easily used on the etched enamel even after contamination with saliva or moisture thereby improving their versatility in application [13].

Since less chair-side time has an important role in behaviour management in paediatric dentistry, self-etch bonding may be considered the best resin-bonded material for clinical use. In contrast to the etch-and-rinse system, which requires a separate etching protocol, self-etching adhesives reduce the number of steps and application time required [14]. Hence this is invitro study was undertaken to comparatively evaluate the type of fracture mode of a seventh generation self-etch adhesive (GC-G-Bond) and an eighth-generation universal adhesive (GC-G-Premio Bond) on dentin of deciduous teeth following the micro-tensile bond strength test.

In the present study buccal /lingual surface of molars were preferred as flat dentin surface could be prepared which would give wider area of dentin to be treated and bonded to resin [15]. Flat dentin surfaces were created using model trimmer under running water. This preparation was restricted to the superficial dentin just below the dentino-enamel junction. After the application and curing of bonding agents, a split Teflon metal mould shaped in the form of a hollow inverted cone measuring 2mm diameter at lower end, 4mm diameter at the top and 5mm in height was used to build a composite resin cone on the treated tooth surface for evaluation of tensile bond strength. Similarly, a second split Teflon metal mould shaped in the form of a hollow cylinder measuring (2mm diameter and 5mm height) was used build a composite resin cylinder on the the treated tooth surface for evaluation of shear bond strength. This mould was used to restrict the bonding surface area to 2mm diameter on the dentin as it results in fewer defects occurring on the smaller area of bonding thereby exhibiting higher bond strength. During the condensation of composite inside the mould, an incremental technique was used, to decrease the polymerization shrinkage. Twisted wires were placed after placing 2mm thickness of composite resin and cured. The remaining 3mm was restored with composite in two increments and cured separately.

The Micro-tensile Bond Test (µTBS) was introduced in dentistry by Sano *et al.* to measure the bond strength and the modulus of elasticity of the mineralized and demineralized dentin [16]. It is considered as the most valuable test for the evaluation of the adhesion bond strength as the test allows for a more uniform stress distribution than the shear bond strength test, due to axial tensile loading on a reduced interface which reduces the frequency of cohesive fractures in the dentin [17, 18, 19]. Therefore, the current study employed SEM analysis of the fracture mode following a micro tensile bond strength test.

Studies by Sezinando *et al.* and Hanabusa *et al.* suggested that the performance of the multi-mode adhesives not only depends on the adhesive strategy, and tooth substrate but also on the type adhesive used due to the complexity of their chemical composition [20, 21].

The results of the current study showed that the seventh generation self-etch adhesive (GC-G-BOND) exhibited a maximum of adhesive fractures followed by an equal number of resin cohesive and mixed fractures whereas the eighth-generation universal adhesive (GC-G-PREMIO BOND) exhibited a maximum of resin cohesive fractures followed by mixed fractures and least number of adhesive fractures. Similar results were reported by Knobloch *et al.* in his study where both one step & two step self-etch adhesives exhibited adhesive failure at resin-dentin interface [9]. On the contrary, Stalin *et al.* concluded that the use of self-etch adhesives on primary dentin resulted in mixed fracture over adhesives fractures, where failure was observed within adhesive layer with some part of cohesive failure in dentin [22].

Armstrong *et al.*, Osorio *et al*. and Toledano *et al*., reported in their studies that there is a higher adhesive fracture rate in the systems with low bonding strength and there is a higher risk of mix and cohesive fractures with high bonding strength, implying that the bonding strength and the fracture type can be related [23, 24, 25]. Though a corelation between the bond strength and fracture mode of adhesives were not made in the current study, the eighth generation universal adhesive (Group II-GC-G-PREMIO BOND) used in this study exhibited a higher tensile bond strength and greater number of cohesive fractures when compared to one step self-etch adhesives (Group I-GC-G-BOND) which exhibited much lower tensile bond strength values and greater number of adhesive fractures.

A probable explanation for this result may be due to the fact that despite the similarities between both the adhesive systems, the composition of the 8th generation universal adhesives differs from that of the 7th generation adhesives by the inclusion of monomers that are capable of producing both chemical and micromechanical adhesion to the dental substrates [26, 27].

Methacryloyloxydecyl Dihydrogen phosphate (MDP) is a functional monomer found in 8th generation adhesives. MDP being a hydrophilic monomer with mild-etching properties allows the 8th generation adhesives to be used with any etching techniques. Yoshida *et al.* observed that an effective chemical interaction occurs between MDP and hydroxyapatite, forming a stable nano-layer which could form a stronger phase at the adhesive interface, thereby increasing the mechanical strength of the adhesive interface [28]. Therefore, this stable MDP-Ca salt deposition along with nano-layering may explain the high bond stability and less number of adhesive fractures observed with universal adhesives.

In order to achieve an effective bond strength in dentin, the adhesive system should produce an intermingled layer of resin monomers and organic portion (collagen fibres) of dentin, known as hybridization zone. The quality of hybrid layer may be vary depending upon the pH of etchant, ability of the resin monomer to flow into the demineralized dentin besides the chemo-physiological and morphological characteristics of the primary dentin.

An acidic medium is needed to dissolve the smear layer and the smear plugs, and open the way for adhesive impregnation into the dentin; on the other hand, a very strong acidic medium might remove too much calcium,

consequently decreasing its bonding ability [29]. Perdigao *et al.* referred that the ideal pH for bonding agent containing 10-MDP should be mild (pH ≈ 2), which allows greater formation of stable calcium salts, improving dentin bonding [30]. One step self-etch adhesives used in the current study presents an ultra-mild pH (pH = 3.5). A plausible reason for more number of adhesive fractures being observed with these systems could be due to the acidity level of the bonding agent not being able to sufficiently to demineralize the smear layer to allow the adhesive penetration into the dentin. Yet another reason for the higher incidence of adhesive fracture with these systems could be presence of water as an adhesive solvent. Studies by Landuyt *et al.* and Hashimoto *et al.* report the permeability of the adhesive layer resulting in hydrolytic breakdown of the adhesive interface as one of the primary causes of bond failure seen with these adhesives systems [31, 32].
The type and amount of solvents, the filler content and percentage of monomers along with the diluents used in the mixture can influence the bond strength of an adhesive. These are some of the factors that could affect the bond strength significantly which needs further investigation.

Conclusion
Within the limitations of the study, the following conclusions can be made:

- While 7th generation self-etch adhesive exhibited majority of adhesive fractures, 8th generation Universal adhesive exhibited resin cohesive fractures followed by least number adhesives fractures.
- Hence, 8th generation universal adhesives being nano–filled and less technique sensitive can be considered as superior bonding agents with respect to their use in paediatric dental practice.

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