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Original Research

Impact of Various Cavity Disinfectants on the Fracture Resistance in Tooth Fragment Reattachments- An In-Vitro Study

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ABSTRACT:
Background: Dental crown fractures are one of the most common forms of dental injury encountered in children and adolescents, where the reattachment of tooth fragments is crucial for the best esthetics and function. Tremendous numbers of disinfectants are used in preventing bacterial invasion during this process; however, their action on bonding strength and fracture resistance of reattached fragments remains unknown. **Methods:** An in vitro study was devised to test the four different cavity disinfectants, namely 3% sodium hypochlorite (NaOCl), 20% citric acid, 2% chlorhexidine and glutaraldehyde/methacrylate, on the fracture resistance of reattached incisor fragments. Standardized incisor fragments were prepared by sectioning fifty-five incisor teeth, which were further subdivided based on the disinfectant treatment being applied. The fracture resistance was measured using a universal testing machine. The statistical analysis used ANOVA, followed by Tukey's post hoc test. **Results:** The highest mean fracture resistance was recorded in the citric acid group. Mean fracture resistance was also closely ranked in the glutaraldehyde group. Sodium hypochlorite, on the other hand, significantly decreased fracture resistance compared with others. There were noticeable differences in fracture resistances of treatment groups ($p < 0.05$) wherein citric acid and glutaraldehyde performed significantly superior than sodium hypochlorite and the control group. **Conclusion:** The most efficient disinfectants identified were citric acid and glutaraldehyde for strengthening the fracture resistance of tooth fragment reattachment. Sodium hypochlorite was demonstrated to be detrimental to the bonding strength and must be used with caution in procedures that require strong adhesion to be present.
Keywords: Cavity disinfectants, tooth reattachment, fracture resistance, sodium hypochlorite, citric acid, glutaraldehyde, chlorhexidine

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INTRODUCTION

Crown fractures, especially of the anterior incisors, are among the most common dental injuries. Many are caused by falls, sports injury, or other accidents. In most patients, attachment of the fractured tooth fragment is the most advisable form of treatment because it conserves as much of the natural tooth structure as possible, offers optimal esthetics, and retains original resistance to wear [1]. For most patients, this is a conservative treatment with a different outlook because it would generally outperform alternatives such as the placement of composite resin or a crown in terms of appearance and function [2].

One of the major risks connected with fragment reattachment, however is the possibility for bacterial contamination through the exposed dentin tubules potentially leading to pulpal inflammation or disease [3]. An adjunct to this, therefore is to use cavity disinfectants before the adhesive bonding procedure. Although these are effective for bacterial contamination, such disinfectants interfere with the bonding strength between the tooth fragment and the remaining tooth structure, and subsequently may affect the fracture resistance of the reattached fragment [4].

Chemical disinfectants may alter the organic and inorganic components of dentin, so the structural properties of its substrate, such as microhardness, solubility, and permeability, would be changed [5]. Microhardness testing has been widely used to determine the alterations in the mineral content of dentin as well, which may eventually affect the bonding capacity of its substrate [6-7]. There are various studies recorded in relation to the effect of chemical irrigants on radicular dentin microhardness. According to previous studies, changes in dentin microhardness are linked to corresponding micro-shear bond strength variations [8].

Cavity disinfectants tend to have different mechanisms of action and hence will vary in how they interact with the dentin surface [9]. Sodium hypochlorite, for example, has been commonly used because of its antibacterial effects but causes dentin collagen fiber breakdown, which could reduce bond strength [10]. Conversely, citric acid has antioxidant properties that could potentially nullify the oxidative effect of sodium hypochlorite and stabilize the resin-to-dentin bond [11-13]. Chlorhexidine, as an antimicrobial agent known to inhibit microbial growth, and glutaraldehyde, known for its cross-linking property, can also affect bonding mechanisms [2, 7].

Given these considerations, the aim of our study was to evaluate and compare the effects of four commonly utilized disinfectants, namely NaOCl, citric acid, chlorhexidine, and glutaraldehyde on the fracture resistance of reattached tooth fragments, and ascertain the best disinfectant for optimal bond strength and fracture resistance of the reattached fragments.

MATERIALS AND METHODS

Study Design

This in vitro study involved extraction of 55 incisors. Each tooth was sectioned to simulate crown fractures, with standardized incisal fragments prepared. Specimens were allocated into five groups (n = 11 group) based on the disinfectant applied to the fracture surface (Figure 1).

- Control Group A: No disinfectant.
- Group B: 3% sodium hypochlorite (NaOCl).
- Group C: 20% citric acid.
- Group D: 2% chlorhexidine.
- Group E: Glutaraldehyde + methacrylate.

Sample Size Determination

The sample size was calculated using the following formula:

$k = n2/n1 = 1$

$$n1 = \frac{(\sigma 1^2 + \sigma 2^2/K)\,(z_{1-\alpha/2} + z_{1-\beta})^2}{\Delta^2}$$

n1= 11

n2 = K∗n1=11

Wherein,

$\Delta = |\mu_2 - \mu_1|$ = absolute difference between two means

σ_1, σ_2 = variance of mean #1 and #2

n_1 = sample size for group #1

n_2 = sample size for group #2

α = probability of type I error (usually 0.05)

β = probability of type II error (usually 0.2)

z = critical Z value for a given α or β

k = ratio of sample size for group #2 to group #1

According to Nihan Gohulol et al's data [8], a minimum sample of 55 teeth was necessary such that any statistically significant difference would be detected.

Figure 1: All disinfectants utilised in our study

Protocol for Applying Disinfectant

The disinfectants were applied according to the protocols described below:

- NaOCl (3%): Fragments and tooth surface received NaOCl for 20 seconds followed by a rinsing of 10 seconds with distilled water and then air-dried.
- Citric Acid (20%): Fragments received citric acid for 10 minutes, rinsed and air-dried
- Chlorhexidine (2%): Fragments received chlorhexidine for 20 seconds, washed, and dried.
- Glutaraldehyde + methacrylate: The disinfectant was applied actively for 30 seconds, then rinsed and dried.

Sample Preparation

The root sections of the specimens were encased in self-curing acrylic resin cylinders, positioned 1 mm from the cemento-enamel junction (Figure 2).

Figure 2: Root sections encased in self-curing acrylic resin cylinder

Following surface treatment, the specimens underwent selective acid etching with 35% phosphoric acid for 15 seconds, followed by rinsing and drying (Figure 3). The universal adhesive was applied, and fragments bonded using a flowable microhybrid composite (Figure 3).

Figure 3: Flowable microhybrid composite

The composite was light cured for 40 seconds, after which the specimens were embedded in self-curing acrylic resin (Figure 4).

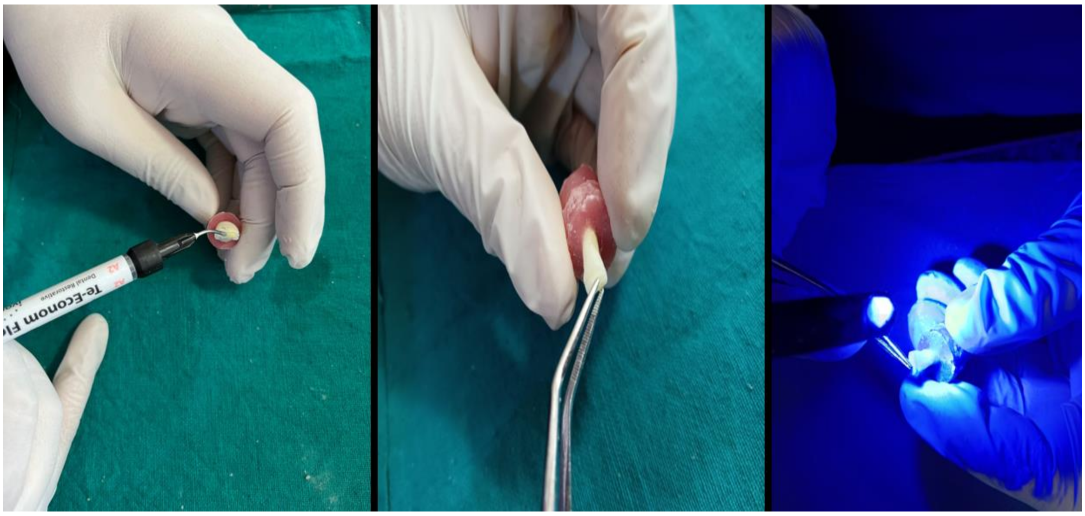

Figure 4: Light curing of specimens

Fracture Resistance Test

A universal testing machine was used to determine the fracture resistance of the reattached fragments. The application was done with a stainless-steel tip loaded perpendicularly to the long axis of the tooth, on its labial surface. The load application was done for a period of 5kN, with a cross-head speed of 1mm/min. The force required to fracture the reattached fragment was recorded.

Data were analyzed by the use of SPSS Statistical Package for the Social Sciences (Version 25). The ANOVA and Tukey's post hoc tests were done to find the statistically significant differences between the groups. In this study, the cut-off for establishing statistical significance was a p-value of <0.05.

RESULTS

Citric acid produced the highest average fracture resistance (98.27 ± 0.14 N), whereas glutaraldehyde occupied the second position with 97.34 ± 0.29 N (Table 1, Figure 5). Both disinfectants showed an improvement in the bonding strength and fracture resistance of reattached tooth fragments over all the other groups. The near-equal fracture resistance between citric acid and glutaraldehyde treatments suggests that both agents are very effective in maintaining the integrity of the structure of the reattached fragments.

Table 1: Comparative evaluation of fracture resistance of reattached tooth fragments

Medicaments	N	Mean	St. Deviation	95% Confidence Interval for Mean	
				Lower Bound	Upper Bound
Glutaraldehyde	12	97.3425	.28753	97.1598	97.5252
Citric acid	12	98.2700	.14403	98.1785	98.3615
NaoCl	12	44.7975	.31352	44.5983	44.9967
Chlorhexidine	12	81.6050	.24441	81.4497	81.7603
Control	12	64.0333	.21060	63.8995	64.1671
'F' statistic	102632.841				
P value	0.000*				

(*=Significant; NS = Not Significant)

The effect of chlorhexidine treatment was moderate in raising the resistance at fracture, taking a mean value of 81.61 ± 0.24 N. Compared to the control group, it presented better performance but was lower than that achieved with citric acid and glutaraldehyde. This gives an indication that although positives on bond strength results by chlorhexidine's antimicrobial properties, its efficacy to increase fracture resistance was not high.

The lowest fracture resistance was in the NaOCl-treated group with a mean value of 44.80 ± 0.31 N. This result is in line with previous researches that revealed sodium hypochlorite impairs the adhesive bond strength since it degraded collagen. The samples treated by NaOCl were even weaker than the rest of groups and thus NaOCl could not be used for any kind of procedure that demands more level of fracture resistance.

The control group, treated with no disinfectants had a mean fracture resistance of 64.03 ± 0.21 N. Although higher than that of the NaOCL group, it was lower than that of the chlorhexidine, glutaraldehyde, and citric acid groups; this may suggest that while application of disinfectants improves bond strength in general, sodium hypochlorite has an adverse effect on bond strength.

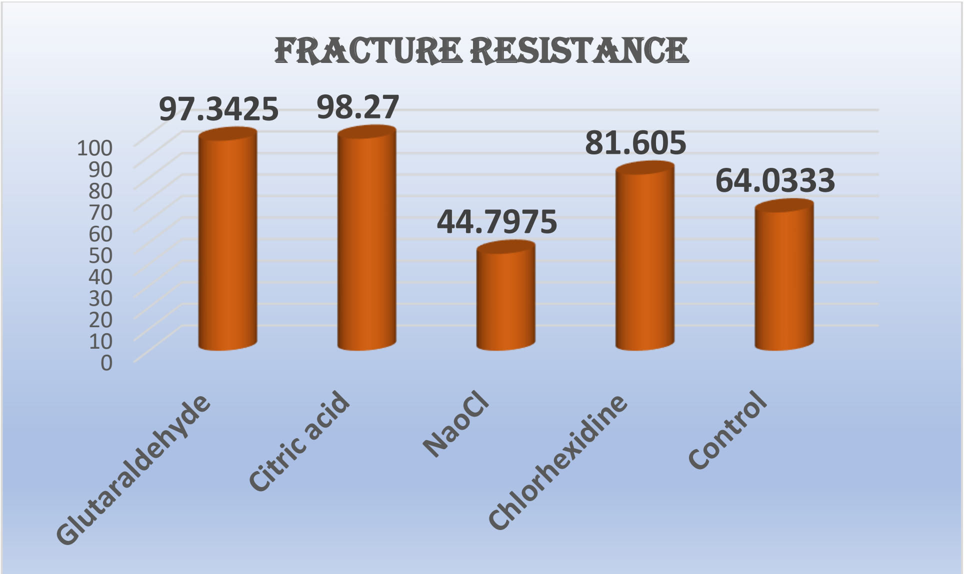

Figure5: Comparative evaluation of fracture resistance of reattached tooth fragments

Statistical comparison using ANOVA and Tukey's post hoc tests revealed significant differences at $p < 0.05$ (Table 2). The citric acid and glutaraldehyde groups significantly outperformed the control and NaOCl groups by big margins. From the comparison of the pairwise difference in Table 2, it is quite obvious that an average difference of 53.47 units was given by citric acid compared to NaOCl, chlorhexidine, and the control group. Similar comparison portrayed the mean difference of fracture resistance as the highest value between glutaraldehyde and NaOCl since it exhibited 52.55 units and when compared to the control group exhibited 33.31 units.

Table 2: Within group comparison for fracture resistance of reattached tooth fragments

Pairs	Mean Difference	Std. Error	Significance
Glutaraldehyde versus Citric acid	-.92750*	.10096	.000*
Glutaraldehyde versus NaoCl	52.54500*	.10096	.000*
Glutaraldehyde versus Chlorhexidine	15.73750*	.10096	.000*
Glutaraldehyde versus control	33.30917*	.10096	.000*
Citric acid versus NaoCl	53.47250*	.10096	.000*
Citric acid versus Chlorhexidine	16.66500*	.10096	.000*
Citric acid versus Control	34.23667*	.10096	.000*
NaoCl versus Chlorhexidine	-36.80750*	.10096	.000*
NaoCl versus Control	-19.23583*	.10096	.000*
Chlorhexidine versus Control	17.57167*	.10096	.000*

(*=Significant; NS = Not Significant)

Table 2 compares the mean differences in fracture resistance of different disinfectants used on reattached tooth fragments. Citric acid showed a minimal lower fracture resistance compared to glutaraldehyde with approximately about 0.93 units; this was statistically significant. Glutaraldehyde provided a more pronounced increase in fracture resistance compared to NaOCl as shown, which was 52.55 units greater, and compared with chlorhexidine, glutaraldehyde showed higher fracture resistance by 15.74 units as well. The fracture resistance of glutaraldehyde was also much higher than that of the control group by 33.31 units more than the latter.

Citric acid revealed high fracture resistance against the comparative NaOCl by 53.47 units. Similarly, when compared to chlorhexidine, citric acid had shown a high fracture resistance of 16.67 units. Citric acid also has shown greater fracture resistance than that of the control group with a difference of 34.24 units.

Chlorhexidine showed a higher fracture resistance as compared to NaOCl, with a difference of 36.81 units, and the fracture resistance was also significantly more than the control with a difference of 19.24 units. The fracture resistance of chlorhexidine had a very significant difference against the control group by 17.57 units.

DISCUSSION

Our findings showed a significantly high difference in fracture resistance in reattached tooth fragments following disinfectant treatment. From all the tested agents, citric acid and glutaraldehyde produced the highest mean values in fracture resistance, being significantly different and outperforming sodium hypochlorite as well as the control group. This means that citric acid and glutaraldehyde do enhance bonding between the dentin and adhesive, which gives stronger reattachment of the fragments.

This seems consistent with existing literature that points out potential adverse effects of sodium hypochlorite on the resin-dentin interface. Sodium hypochlorite degrades the dentin collagen network, thus producing a less receptive surface for adhesive bonding [7]. Further, it interferes with polymerization through its action as an oxidizing agent and, thereby produces a less than ideal hybrid layer, essential for effective bonding. These factors would most likely explain why the fracture resistance was significantly lower for the NaOCl group [12].

Chlorhexidine had also demonstrated significant improvements in fracture resistance over the control and NaOCl groups but its performance was lower as compared with citric acid and glutaraldehyde [13]. Chlorhexidine has been shown to be effective in inhibiting bacterial growth and thus, has been studied extensively because of its role in preserving bond strength by inhibiting MMP activity. While it improved bond strength in this study, its overall effectiveness in enhancing fracture resistance was not much significant compared to the other disinfectants [14].

Gonulol et al. [8] were interested in the effect of different cavity disinfection protocols on fracture strength of reattached tooth fragments. They found that ascorbic acid treatment after sodium hypochlorite treatment led to improvement of bond strength to dentin. On the other hand, this is contrary to what has been seen in our study, wherein NaOCl treatment had the lowest fracture resistance. In their study, they also used ascorbic acid, which could neutralize the detrimental effect that NaOCl may have on bond strength.

Recently, Haralur et al. [14] evaluated the effect of chemical disinfectant solutions on the µSBS and microhardness of total-etch and self-etch resin-infiltrated human dentin. According to their results, 2% chlorhexidine did not affect µSBS but resulted in marginal reduction of dentin microhardness compared with the control group. The result obtained is comparable with our study where chlorhexidine treatment presents moderate improvement in fracture resistance compared with the control group.

Barakat et al. [15] compared the effect of NaOCl in both temperatures and concentrations on the compressive strength of root dentin. They found that irrigation with NaOCl with both concentrations and temperatures during root canal preparation affected the compressive strength of root dentin. This was contradictory to our study; here, NaOCl treatment led to the lowest fracture resistance.

Shafiei et al. [16] examined whether pre-treatment of access cavity with sealing with resin before applying the canal irrigant would increase the resistance of the endodontically treated anterior teeth to fracture. They observed that NaOCl/EDTA irrigation resulted in an adverse effect on the fracture resistance, which accords with our findings concerning NaOCl treatment. They also stated that the pre-sealing of access cavity with flowable composites leads to fracture resistance significantly better than with conventional methods.

Chemical disinfection treatments could, therefore, strongly alter the microhardness and thus the mechanical and structural properties of dentin. Such alterations might relate to variations in relative proportions of organic to inorganic components in the dentin matrix. Microhardness measurements indicate alterations in modifications of mineral content that subsequently affect the bonding capacity of the surface of treated dentin. Other studies have reported the harmful effect of NaOCl and EDTA solutions for the microhardness of radicular dentin [16-17]. NaOCl was also found to dissolve major organic constituent of dentin, Type I collagen, resulting in reduced modulus of elasticity and flexural strength of the dentin. Moreover, NaOCl dissolves inorganic constituents including magnesium and phosphate ions while preserving carbonate concentration of dentin [18].

Significant alteration of Ca/P ratio following NaOCl treatment was also documented by Doğan et al. [19]. The alteration of both mineral and organic components thus contributes to the reduction in dentin microhardness. EDTA, having very strong chelating properties, solubilizes the calcified components, which results in a softening effect and decreases in dentin microhardness [15]. On the other hand, the treatment groups with CHX showed marginal microhardness reduction only. Reports by earlier studies have shown that structural properties depend on the contact time and concentration of CHX [17].

Naenni et al. [20] also reported the absence of smear layer removal and tissue dissolution capacity with CHX. The remaining smear layer may thus potentially act as a barrier by limiting the contact time between the irrigant and the surface of dentin. Nonetheless, CHX has been known at higher concentrations (2%), on the other hand, to reduce the Ca/P ratio as well as the microhardness [21]. The marginal reduction of dentin microhardness in the povidone-iodine treatment could be attributed to the inability of necrotic tissues as well as chemical remnants to get solubilized after surface treatment.

Limitations

Being an in-vitro study, we could not simulate real life clinical scenarios where saliva, blood, and oral flora may affect bonding performance and fracture resistance. More importantly, the long-term bond strength effects of these disinfectants were not tested. Resin-dentin bond strength could alter with time due to changes in the ultrastructure and micromorphology of the hybrid layer, especially under thermal and mechanical loading in the oral cavity. Lastly, the sample size was small, although statistically powered; such a small sample size requires additional research to be confirmed using larger sample sizes. This investigation was limited to the immediate fracture resistance and did not consider other significant clinical outcomes, like microleakage, that could possibly affect the outcome of the procedure.

CONCLUSION

The results indicate that citric acid and glutaraldehyde effectively improve fracture resistance in reattached dental fragments. Conversely, NaOCl markedly diminishes bond strength and should be eschewed in treatments necessitating strong reattachment of tooth fragments. Additional research is advised to investigate the prolonged effects of these disinfectants in clinical contexts.

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