Evaluation of Freshly Prepared “Arginine-Calcium Carbonate-Fluoride” and “Casein Phosphopeptide-Amorphous Calcium Phosphate-Fluoride” Desensitizing Agents on Crown Retention: An In Vitro Study

Kottem Supraja1, Dileep Nag Vinnakota2, Divi VV Vamsi Krishna3, Srinivas R Pottem4

ABSTRACT

Aim and objective: The prevalence of dentinal hypersensitivity after tooth preparation is high and there is a need to explore the usage of contemporary agents in prosthodontics for this purpose. Therefore, the present study aims to evaluate the retention of fabricated copings on prepared teeth coated with freshly prepared arginine-calcium carbonate-fluoride and casein phosphopeptide (CPP)-amorphous calcium phosphate-fluoride desensitizing agents.

Materials and methods: Forty-five extracted premolar teeth were mounted in autopolymerizing acrylic resin and prepared for complete cast metal copings following the standardized protocol. These preparations were randomly divided into three groups for the application of desensitizing agent: arginine based, CPP based, and control (without any agent). Each group was further subdivided into three and luted using either glass ionomer (GIC), resin modified glass ionomer (RMGIC), or resin cement. All these specimens were subjected to tensile bond strength evaluation using a universal testing machine.

Results: The mean bond strengths (in Newtons) in the control group were 308.62 ± 58.84, 176.89 ± 35.46, and 300.35 ± 27.9 with GIC, RMGIC, and resin types of cement, respectively. On the application of arginine-based desensitizer, the bond strengths decreased to 90.26 ± 10.68, 85.07 ± 18.82, and 236.05 ± 43.62 with GIC, RMGIC, and resin types of cement, respectively. On the other hand, on the application of CPP-based desensitizer, the bond strengths in the same order of luting were 272.32 ± 30.5, 203.47 ± 60.57, and 158.66 ± 25.32.

Conclusion: Arginine-based desensitizer did not influence the retention of crowns with resin cement, whereas CPP-based coat did not affect the retention of crowns luted with GIC as well as RMGIC.

Clinical significance: The present study shows the importance of choosing a desensitizing agent based on the luting cement selected for prosthesis retention.

Keywords: Arginine, Casein phosphopeptide-amorphous calcium phosphate, Dentin desensitizing agents, Prosthesis retention.


INTRODUCTION

In prosthodontics, tooth preparation of vital teeth leads to millions of dentinal tubules exposure.1–4 Hence, short-term as well as prolonged dentin hypersensitivity has been associated with the preparations on vital teeth, sometimes eventually requiring endodontic treatment.3,4 Many theories have been proposed for this, the most plausible one being the hydrodynamic theory by Brannstrom.5–7 According to this theory, the stimulus is transmitted to the pulp by the hydrodynamic movement of fluids in the dentinal tubules. This fluid movement due to desiccation and friction during tooth preparation stimulate the odontoblasts, which elicits a response by nerve fibers as pain.8–11 Additionally, the pressure exerted during the cementation of the crown pushes the unset cement into the tubules causing excessive hydrostatic pressure, leading to further sensitivity and irritation of pulp.12,13 Other factors like changes in temperature, poor provisional restoration, the acidity of cement, bacterial leakage, and removal of the protective smear layer also play a role in increasing the chance of hypersensitivity.14–16 To avoid this consequence, occlusion of potential dentinal tubules is essential to control the flow of fluids in the tubules. To accomplish this, the use of dentin desensitizing agents before crown cementation has been suggested as a useful clinical treatment.17–19 Many substances have been proposed for reducing the sensitivity, among which nonpolymerization desensitizers, polymerizable adhesives, potassium nitrate, copalite varnish, fluoride, calcium hydroxide, LASER treatment, casein, and arginine-based pastes are the important ones.20–22 The effect of all these substances, except arginine and casein, fortified with fluoride, on the retention of crowns cemented with zinc phosphate, glass ionomer, resin-modified glass ionomer cement, or resin cement has been studied in literature with varied results. Thus, the influence

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of arginine and casein combined with fluoride on crown retention forms the significant lacunae in this field. The desensitizing paste based on arginine and casein fortified with fluoride is reported to be an effective treatment modality for dentin hypersensitivity.\textsuperscript{23} However, these commercially available pastes are blended with many other substances to improve the palatability and shelf life of the product. These components are not needed for desensitizing the prepared tooth. Additionally, these might hamper the retention of the crown. Thus, for prosthodontic purposes, only the products attributed to desensitize the dentin are needed. Therefore, in the present study, arginine-calcium carbonate-fluoride and casein phosphopeptide (CPP)-amorphous calcium phosphate (ACP)-fluoride, the principle ingredients theorized as agents that decrease the dentin sensitivity in arginine-based paste (Pro-Argin technology) and GC Tooth Mousse Plus, respectively, are considered. As the effect of these essential substances has not been tested so far, the present in vitro study was planned to determine the impact of the freshly prepared arginine-calcium carbonate-fluoride and CPP-ACP-fluoride, as desensitizing agents, on crown retention.

**Materials and Methods**

The present in vitro study was conducted in Department of Prosthodontics. Freshly extracted, noncarious, nonrestored human premolar teeth were cleaned and disinfected using a 0.5% aqueous chloramine solution for 1 week. Then, all the teeth were stored in a 0.05% thymol solution until used for further experimentation. The roots were notched, and a vent was created in the apical one-third of the root to accommodate a small metal piece to ensure retention of tooth in the jig. The enclosed metal piece was added as an additional retentive aid to resist the displacement of the tooth from the mold. Then, the teeth were mounted in the customized 1-inch square jig using autopolymerizing acrylic resin, such that the cemento-enamel junction (CEJ) was 1 mm above the cold-cure resin. All the teeth were prepared for complete cast metal crowns using round-end tapered diamond bur (TR-19. ISO 200/204) for occlusal reduction of 1.5 mm, and by following a standardized protocol using long round-end tapered crosscut fissure carbide bur (H33LR) connected to ISO A1 milling machine, axial reduction of 1.5 mm was prepared.

All the preparations were standardized with a Chamfer finish line, 6° taper, and the height maintained at 4 mm axial length above the CEJ. Water spray was continuously used to prevent dessication during tooth preparation. A single trained investigator did all the preparations. All the castings were fabricated using cobalt-chromium alloy by additive manufacturing (direct metal laser sintering). The casts were designed with an additional ring attached to the occlusal surface for accommodating the hook of the universal testing machine to hold the coping. All the standard laboratory procedures were followed.

The prepared teeth, along with the castings, were numbered and randomly assigned to three groups, using block randomization, as mentioned below:

- **Group I**: Tooth preparation was applied with arginine-calcium carbonate-fluoride desensitizer, and complete cast copings cemented with either glass ionomer/resin-modified glass ionomer cement/resin cement.
- **Group II**: Tooth preparation was applied with CPP-ACP-fluoride desensitizer and complete cast copings cemented with either glass ionomer/resin-modified glass ionomer cement/resin cement.
- **Group III**: Control, with no application on tooth preparation and complete cast copings, were directly cemented with either glass ionomer/resin-modified glass ionomer cement/resin cement.

**Preparation of Arginine-Calcium Carbonate-Fluoride**

For the preparation of this desensitizer, 2 mg of arginine, 25 mg of calcium carbonate, and 0.002 mg of sodium fluoride powders were blended in a mortar and pestle. This formulated powder was mixed with distilled water to form a smooth paste for application.

**Preparation of Casein Phosphopeptide-Amorphous Calcium Phosphate-Fluoride**

For the preparation of this desensitizer, 1 mg of CPP, 0.1 mg of ACP, and 0.002 mg of sodium fluoride powders were blended in a mortar and pestle. This formulated powder was mixed with distilled water to form a smooth paste for application.

**Sample Size Calculation**

With the level of significance set at 0.05, power of 80%, and considering the tensile bond strength as primary outcome measure, a sample size of 45 (5 in each subgroup and 15 in each group) was essential.

**Statistical Analysis**

The data were entered into the Microsoft Excel spreadsheet 2016. The statistical analysis was done by using the SPSS 17.0 version for Windows (Chicago, IL, USA). The normality of the data was tested using the Kolmogorov-Smirnov test. The intergroup comparisons of tensile bond strength among full-coverage restorations luted using glass ionomer (GIC)/resin modified GIC/resin types of cement with/without desensitizing agents were tested using the one-way ANOVA test and the post hoc Bonferroni correction test. The intergroup comparisons of tensile bond strength among full-coverage restorations in control/arginine-based/CPP-based groups luted with various types of cement were tested using a one-way ANOVA test and post-hoc Bonferroni correction test.

**Results**

A total of 45 tooth preparations with complete cast metal crowns were randomly assigned to three groups: control ($n=15$), arginine-based group ($n=15$), and CPP-based group ($n=15$). Five samples in each group were cemented with either glass ionomer, resin-modified GIC, or resin cement. All the specimens were subjected to tensile bond strength evaluation using a universal testing machine.

**Effect of Desensitizing Agents on Tensile Bond Strength (Table 1)**

<table>
<thead>
<tr>
<th>Glass Ionomer Cement</th>
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<tr>
<td>Strength (MPa)</td>
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<td>308.62 ± 58.84</td>
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</table>

The mean tensile bond strength in the control group was 308.62 ± 58.84. On the application of arginine-based paste, the tensile bond strength was 90.26 ± 10.68. Thus, there was a...
statistically significant decrease in retention values \( (p \leq 0.001) \). On the other hand, in specimens with an application of CPP-based paste, the tensile bond strength was \( 272.32 \pm 30.5 \). There was a nonsignificant decrease in retention with CPP-based coat \( (p = 0.49) \).

**Resin-modified Glass Ionomer Cement**

The mean bond strength in the control group was \( 176.89 \pm 35.46 \). On the application of arginine-based paste, the tensile bond strength was \( 85.07 \pm 18.82 \). Thus, there was a statistically significant decrease in retention values \( (p = 0.01) \). On the other hand, in specimens with an application of CPP-based desensitizer, the tensile bond strength was \( 203.47 \pm 60.57 \). There was a nonsignificant increase in retention with CPP-based coat \( (p = 1) \).

**Resin Cement**

The mean bond strength in the control group was \( 300.35 \pm 27.9 \). On the application of arginine-based paste, the tensile bond strength was \( 236.05 \pm 43.62 \). Thus, there was a statistically significant decrease in retention values \( (p = 0.03) \).

On the other hand, in specimens with an application of CPP-based desensitizer, the tensile bond strength was \( 158.66 \pm 25.32 \). There was a significant decrease in retention with CPP-based coat \( (p \leq 0.001) \).

**Comparison of Tensile Bond Strength of the Considered Types of Cement (Table 2)**

**Control Group**

The mean tensile bond strengths of the specimens cemented with glass ionomer, resin-modified GIC, and resin types of cement were \( 308.62 \pm 58.84 \), \( 176.89 \pm 35.46 \), and \( 300.35 \pm 27.9 \), respectively. The bond strength with resins is significantly less compared to GIC and resin cement. On the other hand, there was no difference between GIC and resin cement.

**Arginine-based Desensitizer Group**

The mean tensile bond strengths of the specimens cemented with glass ionomer, resin-modified GIC, and resin types of cement were \( 90.26 \pm 10.68 \), \( 85.07 \pm 18.82 \), and \( 236.05 \pm 43.62 \), respectively. The bond strength with resin cement was significantly more compared to GIC and resin-modified GIC. On the other hand, there was no difference between GIC and resin-modified GIC.

**Casein Phosphopeptide-based Desensitizer Group**

The mean tensile bond strengths of the specimens cemented with glass ionomer, resin-modified GIC, and resin types of cement were \( 272.32 \pm 30.50 \), \( 203.47 \pm 60.57 \), and \( 158.66 \pm 25.32 \), respectively. The bond strength with resin cement was significantly less compared to GIC and resin-modified GIC. On the other hand, there was no difference between GIC and resin-modified GIC.

**Discussion**

In prosthodontics, there is a high prevalence of hypersensitivity after cementation of fixed partial dentures. This has been ascribed to the preparation of vital abutments leading to the opening of dentinal tubules. It has also been attributed to the luting cement; both GIC and resins are reported to cause hypersensitivity. The low initial setting pH of GIC and the marginal defect caused by polymerization shrinkage of resin types of cement have been
projected as major reasons for this issue following cementation. Many other factors like an increase in the permeability due to dissolution of the smear layer, hydraulic pressure in dentinal tubules produced by cementation, dehydration of tooth, post-cementation microleakage, as well as bacterial leakage have also been projected as etiologic factors, irrespective of the type of cement. 4,11,25

The sensitivity problem increases as tooth preparation is closer to the pulp, due to various insults such as thermal, chemical, or osmotic stimuli, which also lead to pulpal damage. Thus, there is a need to protect the pulp by covering with biologically compatible materials. So, the selection of the desensitizing agent plays a major role as it should not only decrease the sensitivity but also should protect the pulp. The current desensitizing agents belong to either anti-inflammation, nerve fiber depolarization, protein precipitation, or therapeutic occlusion categories.26 As hydrodynamic theory is a widely accepted explanation for hypersensitivity, products that act as occluding agents are marketed as most appropriate.5,10 However, the resin sealers currently available and used as a desensitizing agent on the preparations cannot have a direct protective role on the pulp. These can only temporarily form a protective seal. On the other hand, the natural ingredients used in the present study, additionally, are proved to help in remineralization of dentin by stimulating sclerotic reparative dentine and dentine bridge formations. 5 Therefore, arginine-calcium carbonate-fluoride and CPP-ACP-fluoride complexes were selected for the current study. The first one is based on the role of natural saliva12 and another on the role of natural milk protein in theoretically reducing dentinal hypersensitivity.

The amino acid arginine is first isolated from the Lupin seedling extract in 1886 by Swiss chemist Ernst Schulz. 27 Further, the combination of arginine with calcium carbonate was investigated for its ability to occlude the tubules and reducing the pain from hypersensitivity. A commercial company adopted this as Pro-Argin technology. Studies done on this technology have proposed that arginine and calcium carbonate work together to accelerate the mechanism of occlusion by depositing dentin-like material containing calcium and phosphate. CPP stabilizes ACP-forming nanocomplexes to remineralize the dentin by maintaining a supersaturation state of calcium and phosphate. CPP casein phosphopeptide; #, ANOVA test; ^, post hoc test; SD, standard deviation; CI, confidence interval; NS, nonsignificant; *, level of significance 0.01; **, level of significance 0.001.

### Table 2: Tensile bond strength of specimens luted with various cements

<table>
<thead>
<tr>
<th></th>
<th>Glass ionomer (n = 5)</th>
<th>Resin modified glass ionomer (n = 5)</th>
<th>Resin cement (n = 5)</th>
<th>p value&lt;sup&gt;a&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control group (n = 15)</td>
<td>308.62 ± 58.84 (235.56–381.68), Range: 240.34–377.69</td>
<td>176.89 ± 35.46 (132.87–220.92), Range: 120.66–213.86</td>
<td>300.35 ± 27.90 (265.72–334.99), Range: 270.76–338.45</td>
<td>0.001**</td>
</tr>
<tr>
<td>Intergroup comparisons—p value&lt;sup&gt;a&lt;/sup&gt;</td>
<td></td>
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<td></td>
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<tr>
<td>Glass ionomer vs resin-modified GIC</td>
<td>0.001**</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Glass ionomer vs resin cement</td>
<td>1NS</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Resin-modified GIC vs resin cement</td>
<td>0.002*</td>
<td></td>
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<tr>
<td>Arginine-based lacquer (n = 15)</td>
<td>90.26 ± 10.68 (77.0–103.52), Range: 74.55–103.98</td>
<td>85.07 ± 18.82 (61.7–108.43), Range: 56.9–103.1</td>
<td>236.05 ± 43.62 (181.89–290.21), Range: 199.24–309.02</td>
<td>&lt;0.001**</td>
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<tr>
<td>Intergroup comparisons—p value&lt;sup&gt;a&lt;/sup&gt;</td>
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<tr>
<td>Glass ionomer vs resin-modified GIC</td>
<td>1NS</td>
<td></td>
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<tr>
<td>Glass ionomer vs resin cement</td>
<td>&lt;0.001**</td>
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<tr>
<td>Resin-modified GIC vs resin cement</td>
<td>&lt;0.001**</td>
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<tr>
<td>CPP-based lacquer (n = 15)</td>
<td>272.32 ± 30.50 (234.46–310.19), Range: 240.34–313.92</td>
<td>203.47 ± 60.57 (128.27–278.68), Range: 125.57–280.57</td>
<td>158.66 ± 25.32 (127.23–190.09), Range: 122.63–182.47</td>
<td>0.004*</td>
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<tr>
<td>Intergroup comparisons—p value&lt;sup&gt;a&lt;/sup&gt;</td>
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<tr>
<td>Glass ionomer vs resin-modified GIC</td>
<td>0.07NS</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Glass ionomer vs resin cement</td>
<td>0.003*</td>
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<tr>
<td>Resin-modified GIC vs resin cement</td>
<td>0.35NS</td>
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and phosphate ions favoring mineralization. ACP is capable of rapid conversion into hydroxyapatite crystals under physiological conditions, which obstructs the dentinal tubules. Another point in the present study is the addition of the fluoride component, which also acts as a desensitizing agent by occluding the dentinal tubules and reducing the discomfort.

The study is first of its kind to know the effect of the principal ingredients in the commercially available pastes theorized to have a desensitizing action, on the retention of full-coverage restorations. These primary ingredients alone add to the clinical advantage of eliminating the influence of unnecessary components added to the commercially available desensitizing pastes. Studies on these materials have a significant role in enhancing the scope of a prosthodontist.

The forces required to dislodge the crowns, as observed in the present study, without any application of desensitizing agents, were relatively medium, ranging from 177 N to 309 N. The values are less compared to other reported studies done to determine the retentive force of crowns cemented using GIC/resin modified glass ionomer (RMGIC)/resin cement. This difference can be attributed to the inclusion of premolars in the current study, whereas extracted molars were considered in other studies. Due to changes in the average axial area of crown preparation, the amount of force required to dislodge the crowns might have changed. When all the luting types of cement were compared, the maximum retention observed was with GIC followed by resin cement and RMGIC. However, there was no statistical difference between GIC and resin cement. This observation is not in accordance with the previous study. It is theorized that higher tensile forces are expected to unseat the restorations cemented with luting agents having high compressive strength like resin cement compared to those with low strength like GIC. As resin cement has more compressive strength (250 MPa) compared to GIC (225 MPa), it is expected that resin cement requires higher tensile forces to dislodge full-coverage restorations. However, contrary findings were observed in the present study, which can be attributed to smooth flow and minimum film thickness noted with GIC compared to resin cement. It is reported in the literature that the film thickness with GIC is 7.24–20.5 μm compared to 31 μm to 40–45 μm with resin cement leading to poor marginal sealing. Thus, there might be poor marginal sealing with resin cement. Additionally, the bond strength of resin cement is multifactorial that depends on the type of dentin (coronal/apical), the preparation depth (superficial/close to the pulp), quality of dentin, and the age of the tooth.

The present study highlights that there is a significant impact of the type of dentin treatment on the forces required to remove crowns luted with various types of cement. There is a distinct effect of dentin treatment on cement and cement on dentin treatment, which emphasizes the importance of choosing luting cement and according to the desensitizing agent. GIC relies on both mechanical retention to surface irregularities and formation of ionic bonding to the tooth structure, whereas RMGIC additionally depends on chemical bond. On the other hand, resin cement depends on mechanical interlocking into surface irregularities and chemical bonding to dentin bonding agents and teeth. Thus, there is a need to choose the desensitizing agent based on the requirement of the luting cement. When using a resin cement for luting, the use of arginine-based desensitizer is advisable. On the other hand, when considering RMGIC or GIC for cementation, CPP-based coat is advisable.

The positive aspect of this study is the normal distribution of recorded values in all subgroups. The deviation in the values is also low. This low deviation can be attributed to the precise standardized crown preparations, which were done in a reproducible way. Though there is a lot of ambiguity in the standardization of tooth preparations, in the present study, the axial surfaces of teeth were prepared by rotating diamond bur on a surveyor base. The preparations were done using a milling machine with a movable tridimensional arm equipped with an electrical micromotor with adjustable speed for which a bur was attached to maintain a uniform preparation on the selected premolars. All the premolars chosen in the present study were maxillary teeth to maintain similarity in the anatomy of the teeth.

Another factor that has an impact on the retention of the crowns is the preparation angle. An ideal realistic preparation taper of 6° was selected in the present study. The taper is in contrast with the preparation angles considered in the previous studies, which were in the range of 10–20°. The authors substantiated this for the nullification of the strong influence of preparation angle, which masks the weaker effects of the desensitizing agent on crown retention. Thus, the convergence angle was increased to decrease the impact of taper and crown retention and increase the impact of luting cement. However, these tapers were not considered in this present study as the experienced dentists do not practice these, and the clinical significance of the study will be decreased. Hence, an ideal recommended convergence of 6° taper was considered for all the groups. Similar to this view, 6–10° taper was selected in one study and unusual taper of 4.8° in another.

All the agents selected in the present study are easy to apply, painless, and nonirritating to the oral structures. Another advantage of pretreating the dentin with desensitizer is the reduction in the contamination with provisional cement. Desensitizers are also accepted as a blanket treatment in prosthodontics as they minimize the bacterial contamination of pulp. Hence, further studies on the effect of these desensitizing pastes on the smear layer of the prepared tooth need to be investigated using scanning electron microscopic evaluations. Even the components formed in the dentinal tubules on the application of desensitizing pastes need to be analyzed using an energy dispersive X-ray microanalysis. These in vitro studies should be followed by randomized clinical trials to determine the effectiveness of these agents in decreasing the sensitivity experienced by the participants, as the subjective effect can only be specified in a clinical situation.

The major limitation of the present study is the consideration of ideal realistic preparation taper of 6°, which questions the generalizability of the study findings to those done by the inexperienced dentists.

**Conclusion**

Based on the findings observed in the present study, the following conclusions can be drawn:

- The arginine- and CPP-based desensitizing pastes had a significant impact on the retention of complete cast metal crowns.
- There was a significant influence of the type of luting cement when desensitizing agents were applied to the tooth preparations.
Freshly Prepared Desensitizing Agents and Crown Retention

- When the crowns were luted with GIC, CPP-based desensitizer did not affect the retention, whereas arginine-based one significantly decreased the retention.
- When the crowns were luted with RMGIC, CPP-based desensitizer improved the retention, whereas arginine-based one significantly decreased the retention.
- When the crowns were luted with resin cement, both the pastes significantly decreased the retention, but values in the arginine-based desensitizer group were close to control.
- Arginine-based desensitizer was compatible with resin cement, whereas the CPP-based coat was compatible with GIC as well as RMGIC. Thus, the selection of desensitizing pastes should be based on the desired luting cement.

**Clinical Significance**

The present study shows the importance of choosing a desensitizing agent based on the luting cement selected for prosthesis retention.

**References**