

## Effects of CPP-ACP Paste on the Shear Bond Strength of Orthodontic Brackets

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### ABSTRACT

**Objective:** To evaluate the effect of casein phosphopeptide–amorphous calcium phosphate (CPP-ACP) paste on shear bond strength and debonding failure modes of orthodontic brackets.

**Materials and Methods:** Freshly extracted premolars were randomly divided into four groups (n = 18) as follows: in groups 1 and 3, the enamel was treated with a solution of CPP-ACP dissolved in artificial saliva; groups 2 and 4 served as controls, and the enamel was treated with artificial saliva. After conventional acid etching, in groups 1 and 2, brackets were bonded using a light-cured bonding system (Blugloo); while in groups 3 and 4, brackets were bonded using a conventional bonding system (Unite Bonding Adhesive). Bonded specimens were subjected to thermal cycling for 1000 cycles before debonding procedures. After debonding, teeth and brackets were examined under a stereo-microscope at 10× magnification to determine whether any adhesive remained, in accordance with the adhesive remnant index. The acid-etched enamel surfaces were also observed using scanning electron microscopy after treatment with and without CPP-ACP paste.

**Results:** The shear bond strengths of group 1 were significantly higher than those seen in group 2 ( $P < .01$ ). There was no significant difference in the shear bond strengths of groups 3 and 4 ( $P > .05$ ). Scanning electron microscopic observation showed that the pretreated enamel surface was rougher than that of the control surface after acid etching.

**Conclusion:** The use of CPP-ACP can be considered as an alternative prophylactic application in orthodontic practice since it did not compromise bracket bond strength. (*Angle Orthod.* 2009; 79:945–950.)

**KEY WORDS:** Shear bond strength; Casein phosphopeptide–amorphous calcium phosphate; Enamel; Orthodontic brackets

### INTRODUCTION

Dental caries remains a major public health problem in most communities, although the prevalence of the

disease has decreased since the introduction of fluorides. White-spot decalcification and caries formation under and around orthodontic bands or brackets are still common problems in orthodontics.<sup>1</sup> Recently, a milk protein derivative, casein phosphopeptide–amorphous calcium phosphate (CPP-ACP) complex, has been advocated for caries prevention and enamel remineralization.<sup>2</sup> The role of CPP-ACP has been described as localization of ACP on the tooth surface, which buffers the free calcium and phosphate ions. This helps to maintain a state of supersaturation with respect to the enamel by suppressing demineralization and enhancing remineralization.<sup>3</sup> The literature has shown that CPP-ACP can prevent the tooth structure demineralization caused by acidic solutions such as

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cola and white wine.<sup>4-6</sup> The recommended professional applications in orthodontics for CPP-ACP are white-spot prevention/removal and to provide a topical coating for patients suffering from erosion and caries.<sup>7</sup>

The technique of bonding orthodontic brackets to enamel with acrylic resin was introduced in 1964.<sup>8</sup> The procedure incorporated the use of an acid-etch technique to better adhere the brackets to enamel.<sup>9</sup> CPP-ACP increases the acid resistance of tooth enamel<sup>10</sup> and may affect adhesion by interfering with the acid etching. However, the literature concerning the effect of CPP-ACP in orthodontics is limited.<sup>11,12</sup> Keçik et al<sup>11</sup> found that pretreatment with CPP-ACP significantly increased the shear bond strength of orthodontic brackets. In an earlier experiment, Dunn<sup>12</sup> suggested that orthodontic brackets bonded to teeth with an ACP-containing composite material failed at significantly lower forces than brackets bonded to teeth with conventional resin-based composite orthodontic cement. In experiments not related to brackets, conflicting results were also seen. Moule et al<sup>13</sup> found that the shear bond strength of resin to enamel using a self-etching priming adhesive (Clearfil SE Bond), rather than an all-etch adhesive (Single Bond), may be affected if the enamel is treated with CPP-ACP. In contrast, the study of Adebayo et al<sup>14</sup> suggested that the application of a CPP-ACP-containing remineralizing paste (Tooth Mousse, GC Corp, Tokyo, Japan) did not affect micro-shear bond strength to enamel for either Single Bond or Clearfil SE Bond. Technique sensitivity may explain the conflicting results.

The aim of this study, therefore, was to compare the shear bond strength (SBS) of orthodontic brackets luted with two different adhesive systems to untreated enamel and enamel topically treated with CPP-ACP. The null hypothesis was that pretreatment with CPP-ACP would have no effect on the SBS of orthodontic brackets to enamel with either bonding system.

## MATERIALS AND METHODS

### Teeth and Brackets

Eighty freshly extracted human premolars, without cracks or erosion, were obtained from the Oral and Maxillofacial Surgery Department of the Ninth People's Hospital, Shanghai Jiaotong University. These teeth had been extracted for orthodontic reasons with the informed consent of the patients. The project was approved by the hospital's bioethics commission. All teeth were cleaned and stored in physiological saline. In no case was a tooth stored for more than 2 weeks after extraction.

For SBS testing, 72 premolars were used. For scanning electron microscopic (SEM) observations, the remaining eight premolars were used. Seventy-two met-

**Table 1.** Composition of Artificial Saliva

| Component*                           | wt%  |
|--------------------------------------|------|
| NaCl                                 | 0.08 |
| KCl                                  | 0.12 |
| MgCl <sub>2</sub> ·6H <sub>2</sub> O | 0.01 |
| K <sub>2</sub> HPO <sub>4</sub>      | 0.03 |
| CaCl <sub>2</sub> ·2H <sub>2</sub> O | 0.01 |
| CMC-Na                               | 0.10 |
| IEW                                  | 99.6 |

\* CMC-Na indicates sodium carboxymethyl cellulose; IEW, ion-exchanged water.

al premolar brackets were used (Standard Edgewise 22/Assortments, REF 790-606-01, Dentaurem Group, Ispringen, Germany).

### Pretreatment and Bonding Procedures

Seventy-two teeth were randomly divided into four groups as follows:

- Group 1: Buccal enamel was treated with CPP-ACP and the brackets were bonded with a light-curing bonding system (Blugloo, Ormco, Orange, Calif);
- Group 2: Buccal enamel was treated with artificial saliva (Table 1) and the brackets were bonded with Blugloo;
- Group 3: Buccal enamel was treated with CPP-ACP and the brackets were bonded with a conventional bonding system (Unite Bonding Adhesive, 3M Unitek, Monrovia, Calif);
- Group 4: Buccal enamel was treated with artificial saliva and the brackets were bonded with Unite Bonding Adhesive.

Before bonding in groups 1 and 3, the buccal enamel of the tooth was treated with a diluted solution (1:10 ratio) of CPP-ACP paste (Tooth Mousse) with artificial saliva for 1 hour per day for 5 days; in groups 2 and 4, only artificial saliva was used.

Eighteen premolars were bonded in each group. Following treatment, each tooth was thoroughly washed in flowing distilled water for 1 hour. At the time of bonding, specimens were initially rinsed with an air-water syringe for 5 seconds; the enamel was then treated with 37% phosphoric acid for 30 seconds, rinsed with an air-water syringe for 30 seconds, and dried until desiccated. The brackets were then bonded using Unite Bonding Adhesive or Blugloo according to the manufacturers' instructions. Each bracket was bonded at the center of the buccal surface of the crown. Briefly, for Blugloo, the primer was applied to the frosty enamel and the back of the bracket base with a microbrush and gently air thinned. The adhesive paste was applied to the base of the bracket, which was then placed on the tooth with firm pressure. Ex-

**Table 2.** Descriptive Statistics (MPa) and Results of Grouped *t* Test Comparing Shear Bond Strength (SBS) of the Groups

| Group | Adhesive               | Pretreatment | n  | SBS (Mean $\pm$ SD) | <i>P</i> |
|-------|------------------------|--------------|----|---------------------|----------|
| 1     | Blugloo                | CPP-ACP      | 18 | 20.89 $\pm$ 4.93    | .038*    |
| 2     | Blugloo                | No CPP-ACP   | 18 | 17.12 $\pm$ 5.57    |          |
| 3     | Unite Bonding Adhesive | CPP-ACP      | 18 | 27.98 $\pm$ 9.16    | .571     |
| 4     | Unite Bonding Adhesive | No CPP-ACP   | 18 | 26.38 $\pm$ 7.58    |          |

\* Indicates significant difference ( $P < .05$ ).

cess adhesive was removed from around the base of the bracket and the adhesive was then light-cured by positioning the light guide of an Ortholux XT lamp (3M Unitek) for 10 seconds on each interproximal side. The power density was verified with a handheld radiometer (Model 100, Demetron Research, Danbury, CT) before each specimen was polymerized. No light curing was needed for Unite Bonding Adhesive, but otherwise, the bonding procedures were the same as for Blugloo. The teeth were then stored in distilled water at 37°C for 24 hours before the testing procedures.

### Shear Bond Strength Test

For SBS testing, the teeth were embedded in acrylic resin with a mounting jig used to align the buccal surface of each tooth so that it was perpendicular to the bottom of the mold. Specimens were then mounted in the jig, which was attached to a universal testing device (EZ20, Lloyd Instruments, Fareham Hampshire, UK), and were secured in the lower jaw of the machine so that the bracket base of the sample paralleled the direction of the shear force. A shear force was applied as close as possible to the tooth/bracket interface by a sharp chisel-shaped rod attached to the end of the testing machine at a crosshead speed of 1 mm per minute until bracket failure, as in previous studies.<sup>15,16</sup> NEXYGEN<sup>Plus</sup> Material Test and Data Analysis Software (Lloyd Instruments, Fareham Hampshire, UK) was used to record the force required to dislodge the bracket.

### Assessment of Adhesive Remnants

After debonding, all samples were examined under 10 $\times$  magnification to assess adhesive remnants on tooth surfaces using the Adhesive Remnant Index (ARI) system.<sup>17</sup> The scoring criteria for evaluation were:

- 1 = All the adhesive, with an impression of the bracket base, remained on the tooth.
- 2 = More than 90% of the adhesive remained on the tooth.
- 3 = More than 10% but less than 90% of the adhesive remained on the tooth.
- 4 = Less than 10% of the adhesive remained on the tooth.
- 5 = No adhesive remained on the tooth.

### SEM Observations

SEM observations were carried out to observe the acid-etched enamel surfaces pretreated with or without CPP-ACP. Eight premolars were used for ultrastructural examination of the etched enamel surfaces by SEM. The crowns were sectioned from the roots with a diamond bur at the buccal cemento-enamel junction, and each crown was cut longitudinally in an occlusogingival direction to obtain two buccal enamel surfaces. Each surface obtained from the same tooth was randomly allocated to one of two experimental groups. The two experimental groups were the CPP-ACP group and the control group, and each group included eight enamel surfaces. In the CPP-ACP group, teeth were pretreated and etched as described for groups 1 and 3. The teeth of the control group were pretreated and etched as mentioned for groups 2 and 4 (no CPP-ACP). After acid etching, samples were prepared for SEM. In brief, the samples were critical point-dried, sputter gold-coated with a sputter coater, and observed and photographed with an SEM (S520, Hitachi, Tokyo, Japan) operated at an accelerating voltage of 20kV. The protocol of using both pretreatments on the same tooth eliminated the variable of possible differences in tooth composition and age among the different groups.

### Statistical Analysis

Statistical analyses were performed using SPSS 11.0 software (SPSS Inc, Chicago, Ill). A grouped *t* test was used to determine whether significant differences existed between the SBS of the groups. The chi-square test was used to examine whether there were differences in the ARI of the groups.  $P < .05$  was considered statistically significant.

### RESULTS

Results of SBS testing for various groups are presented in Table 2. For Blugloo, a statistically significant difference of SBS was observed between the two groups (groups 1 and 2). There was no difference for Unite Bonding Adhesive between groups 3 and 4, although the mean SBS of group 3 was slightly greater than that of group 4.

Table 3 shows the distribution of ARI scores at the



**Table 3.** Frequency Distribution of Adhesive Remnant Index (ARI) Scores of the Groups

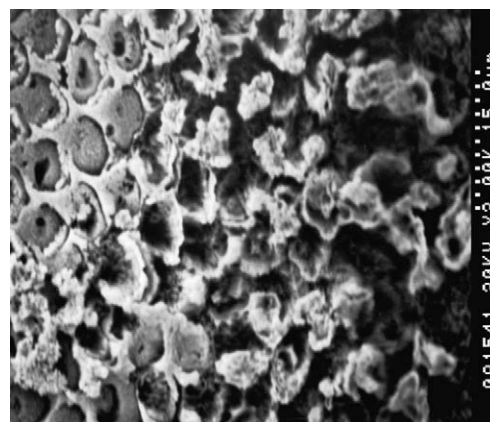
| Group | ARI Scores |   |    |   |   | P    |
|-------|------------|---|----|---|---|------|
|       | 1          | 2 | 3  | 4 | 5 |      |
| 1     | 2          | 5 | 11 | — | — | .406 |
| 2     | 2          | 7 | 9  | — | — |      |
| 3     | 3          | 2 | 6  | 3 | 4 | .482 |
| 4     | 6          | — | 6  | 1 | 5 |      |

debonding sites. The chi-square test revealed no association between both types of pretreatment for both adhesives. Representative SEM images of the etched enamel specimens pretreated with and without CPP-ACP are shown in Figures 1 and 2.

## DISCUSSION

The need for the development of a nontoxic, anticariogenic agent that could supplement the effects of fluoride in an attempt to further lower caries incidence has been highlighted in recent years.<sup>18,19</sup> CPP-ACP is an anticariogenic agent newly derived from milk production. The potential anticariogenicity of CPP-ACP has been demonstrated in various caries models and short-term clinical trials.<sup>20–26</sup> The material appears to be very safe for human use in oral care products, dental professional products, and foodstuffs. The alkaline, stable, and highly soluble CPP-ACP has been trademarked as Recalden and has now been incorporated into sugar-free gum and mints and in dental professional products (Tooth Mousse). In fact, the ability of CPP-ACP in sugar-free chewing gum to remineralize enamel subsurface lesions has been demonstrated in several randomized, double-blind, in situ clinical trials.<sup>10,24,27</sup>

The demineralization of enamel adjacent to orthodontic brackets is a significant clinical problem. White-spot lesions develop as a result of prolonged plaque

**Figure 1.** SEM observation of acid-etched enamel surface after pretreatment with CPP-ACP.**Figure 2.** SEM observation of acid-etched enamel surface after no CPP-ACP pretreatment. This piece of enamel and the enamel shown in Figure 1 were taken from the same tooth.

accumulation on the affected surface, commonly as a result of inadequate oral hygiene. It has been reported that there is a significant increase in the prevalence and severity of enamel demineralization after orthodontic treatment when compared with untreated controls.<sup>28</sup> The use of CPP-ACP to prevent demineralization around orthodontic appliances has recently been advocated.<sup>28,29</sup> However, limited research has focused on the effects of CPP-ACP application on the SBS of orthodontic brackets.<sup>11,12</sup>

In this in vitro study, we examined the effects of CPP-ACP on the SBS of brackets. The CPP-ACP pretreatment was done according to the modified protocols of Moule et al<sup>13</sup> and Oshiro et al.<sup>6</sup> Moule et al<sup>13</sup> treated teeth by applying a small quantity of Tooth Mousse to enamel for 1 hour per day for 5 days. Oshiro et al<sup>6</sup> used a tenfold diluted solution of CPP-ACP paste for 10 minutes. There are two reasons for using artificial saliva as a solvent to dilute the paste in our study. The first is that the product manual for Tooth Mousse emphasizes that saliva will enhance the effectiveness of CPP-ACP, and the longer CPP-ACP and saliva are maintained in the mouth, the more effective the result.<sup>7</sup> The second is that the CPP-ACP paste will be mixed with saliva when it is applied in the mouth. Based on this, application of a tenfold diluted solution of CPP-ACP paste with artificial saliva for 1 hour per day for 5 days may accurately simulate in vivo treatment with GC Tooth Mousse. Our testing was performed in a widely accepted manner first proposed by Fox et al,<sup>30</sup> which permitted comparison with other in vitro bond strength studies.

Dunn<sup>12</sup> found that ACP can cause decreased SBS, while Keçik et al<sup>11</sup> observed that pretreatment with CPP-ACP significantly increased the SBS of orthodontic brackets. The conflicting results may arise from the fact that ACP-containing orthodontic resin cement was

used by Dunn,<sup>12</sup> while Keçik et al<sup>11</sup> performed enamel treatment with CPP-ACP before bonding with Transbond XT, a light-curing adhesive produced by 3M Unitek.

Most published investigations of SBS of orthodontic brackets in the recent period employed light-cured adhesives. However, chemically cured adhesives are still widely used, and it remains worthwhile to investigate the effect of CPP-ACP on the SBS of chemically cured adhesives. We included two adhesive systems in our study. Blugloo, a light-curing adhesive, showed statistically significantly increased SBS after pretreatment with CPP-ACP. This result is similar to that obtained by Keçik et al.<sup>11</sup> Our chemically cured adhesive, Unite Bonding Adhesive, showed no statistically significant difference between the two groups, although the mean SBS of the CPP-ACP group was a bit greater than the mean SBS of the group that was not treated with CPP-ACP.

The chemically cured Unite Bonding Adhesive is dependent upon adequate integration of the primer and the paste to ensure full polymerization. If incomplete polymerization occurs, the potential exists for the bond to be weak and prone to cohesive failure. When the adhesive paste contacts the primer, polymerization occurs first where the paste contact occurs. Rapid polymerization, caused by contact with the primer, may prevent the unpolymerized paste from permeating the etched enamel surface to form more microcomposite tags. The same situation will not occur with light-cured adhesives until light curing is done. In this in vitro study, SEM observations revealed that pretreatment with CPP-ACP resulted in relatively rougher etched enamel than no CPP-ACP pretreatment. A rougher enamel surface results in a greater adhesive area and more resin tags available for bonding. However, after CPP-ACP pretreatment and acid etching, the enamel surfaces were rougher than their counterpart without CPP-ACP pretreatment, and bonding brackets with light-cured adhesive can achieve more resin tags than bonding with chemically cured adhesive. Also, Blugloo may show a more significant increase in SBS (17.12 MPa without treatment versus 20.89 MPa with treatment) after CPP-ACP pretreatment than Unite Bonding Adhesive (from 26.38 MPa to 27.98 MPa).

This study provides some data on the effects of CPP-ACP on the SBS of orthodontic brackets. SBSs in this study ranged from 9 to 42 MPa. This is higher than the 6 to 8 MPa recommended by Reynolds<sup>31</sup> and Whitlock et al<sup>32</sup> as adequate for orthodontic purposes.

The results of this study indicated that SBS of orthodontic brackets is favorably affected when the enamel is pretreated with CPP-ACP. This is true for both light-cured and chemically cured adhesives.

## CONCLUSIONS

- CPP-ACP application may be used safely for caries prophylaxis before orthodontic bracket bonding when either chemically cured or light-cured adhesive is used.
- CPP-ACP application can cause increased SBS of brackets when light-cured adhesive is used. Research on the mechanism causing this increased strength is warranted.

## ACKNOWLEDGMENT

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