

Research article

In vitro assessment of bracket adhesion post enamel conditioning with a novel etchant paste

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Abstract: Background: 37% phosphoric acid (PA) is the traditional enamel etching technique prior to bracket adhesion, yet it has been implicated in numerous enamel injuries. The purpose of the current study was to create a calcium phosphate (CaP) etching paste in a simplified capsule formula that can underpin clinically adequate bracket bond strength without jeopardizing the integrity of enamel upon the debonding procedure. Materials and Methods: micro-sized hydroxyapatite (HA) powder was mixed with 40% PA solution to prepare experimental acidic CaP paste. Sixty human premolars were assigned into two groups of 30 each. Enamel conditioning was accomplished using 37% PA-gel for control group and CaP paste for experimental group. Each group was further divided into two subgroups regarding the water storage (WS) period (24 h and 30 days). Shear bond strength (SBS) test conducted with examination of debonded surfaces for adhesive remnants and enamel damage using a digital microscope. Results: CaP paste produced significantly lower SBS values than PA ($p < 0.01$), yet sufficient for clinical use. PA etching caused often cracked enamel surfaces with excessive retention of adhesive remnants (mainly ARI scores 2 and 3). Contrarily, enamel treated with the experimental CaP paste exhibited smooth, unblemished surfaces mostly clean of adhesives residues (scores 0 and 1). Conclusion: a newly developed CaP paste in a capsule formula fosters clinically adequate bracket adhesion with a sustained bonding performance, allows a harmless bracket removal with minimal or no adhesive residues on debonded surfaces; thus, it can be introduced as a suitable alternative to PA.

Keywords: Enamel Conditioning, Calcium Phosphate, Bracket Bond Strength.

Introduction

Direct bracket adhesion to tooth enamel represents a milestone in contemporary orthodontics. Bracket adhesion using resin-composite adhesives is typically preceded by alteration of the enamel surface via 37% PA etching. Such treatment was found to provide a robust bracket adhesion as it induces the most retentive surface topography for resin adhesive ⁽¹⁾. Nevertheless, this technique enclosed several potential harms to tooth substrate. Of these, the deep and pronounced demineralization effect which substantially minimizes micromechanical properties of the enamel, besides the potential cracks and tear-outs occurring in the course of the debonding procedure because of the excessive force applied in brackets removal at the end of orthodontic therapy ⁽²⁻⁴⁾. Moreover, the detriment can be aggravated with remaining a lot of adhesives on the debonded surfaces that necessitate removal using traumatic rotary burs that inevitably induce additional enamel scratching ⁽⁵⁾.

Several other acids with different concentrations, e.g. citric, pyruvic, and maleic acids, were investigated for enamel conditioning. It was found that such acids were less effective compared with

PA in terms of providing clinically useful bracket bonding strength ⁽⁶⁾. In the last decades, self-etch primers (SEPs) were suggested as an alternative enamel conditioning technique ⁽⁷⁾. SEPs were advocated to facilitate the fixation of orthodontic brackets to teeth surfaces by reducing bonding steps as they merge etching and priming steps excluding the water-rinsing phase. It was observed that SEPs induce a conservative etching effect with decreased resin infiltration, which more likely minimizes the extent of enamel loss ⁽⁸⁾. However, contradicting data were documented regarding the SBS to enamel, adhesive remnants and the occurrence of enamel damage at bracket debonding ⁽⁹⁾.

Besides these, a novel etchant system was recently evolved, based on mixing two CaP compounds with a PA solution to formulate an acidic CaP paste capable of inducing gentle enamel etching accompanied by CaP precipitation, prior to orthodontic bracket bonding. It was observed that this paste provided bracket bond strength suitable for clinical use, and also induced minimal residual adhesives remained with a low incidence of enamel harms upon bracket debond. Nevertheless, a time-consuming manual mixing, of two types of CaPs, basic (β -TCP) and acidic (MCPM) powders, was required for paste preparation ⁽¹⁰⁾.

Research is ongoing to find out a time-conserving and tooth-friendly enamel conditioning system that can induce orthodontic bracket bonding and debonding with minimal undesirable iatrogenic impacts. From a clinical perspective, the bonding system being effective when providing a tooth-adhesive interface able to survive storage in a degrading medium ⁽¹¹⁾. Therefore, the aim of this study was to develop a CaP-based etchant in a simplified recipe as a mixing capsule formula to be more clinically convenient for enamel conditioning during orthodontic bonding procedure, and investigate its bonding efficiency in terms of shear bond strength, amount of residual adhesive, and enamel surface damage after short- and long-term WS in comparison with the standard etchant material (37% PA).

Material and Methods

Experimental CaP etchant paste was prepared as follows: micro-powder of hydroxyapatite (HA) (Sigma-Aldrich CHEMIE GmbH Co., Steinheim, Germany) was used to be mixed with PA solutions of 40% by weight that prepared by diluting PA (85 wt%, CARLO ERBA Reagent, France) with distilled water (DW). Using a 0.4:1 powder to liquid ratio (P: L), the assigned amounts of powder and liquid were gathered into a 2ml Eppendorf tube, that was used as a "mixing capsule" and placed in a dental amalgamator (Lingchen dental Co., Ltd, China). A homogenous workable paste was obtained after 15 s mixing, ready to be applied on the buccal enamel surface. The pH of the resultant paste in addition to 37% PA was assessed using a digital pH meter supplied with a flat-surface pH electrode (Model S450CD, Sensorex, USA) after mixing under ambient lab conditions (22-25° C, 30-40% humidity).

The sample of current study involved sixty human premolars extracted for orthodontic reasons from individuals with an age range of 15-30 years after acquiring ethical approval from the Research Ethical Committee of the College of Dentistry at the University of Baghdad (Ref. no.: 355421/355). After extraction, the soft tissues and blood residuals were cleaned from teeth surfaces using running tap water. Teeth were stored in 1% chloramine-T trihydrate as a bacteriostatic/bactericidal solution for one week, then transmitted into DW for storage. The teeth selected for this study had a sound enamel surface with no cracks or hypoplasia and did not undergo any chemical treatment. To facilitate handling and control of the specimens during SBS test, each premolar tooth was fixed vertically inside a custom-made silicone mould (18×18×15 mm). The analyzing rod of dental surveyor (Addler,

Golden Nimbus LTD, Maharashtra, India) was utilized to align the buccal surface, thereby the bracket base, to be parallel to the debonding force applied during the shear test. Self-cure acrylic resin (Major Repair, Major Prodotti Dentari S.P.A., Italy) was mixed to be poured around the root up to 1mm apical to the cemento-enamel junction.

After complete acrylic curing, the mounted teeth were kept moist at the lab temperature until the bonding time. They were divided into two main groups (n=30) regarding the etchant applied for enamel etching, i.e. commercial 37% PA and developed CaP paste. The etch-and-rinse (EAR) protocol was followed for conditioning enamel surfaces in all study groups. This protocol was implemented as follows: 30 s etchant application, 10 s water rinsing and 10 s oil-free air drying. The surface of each specimen was primarily polished with non-fluoridated pumice at a low speed (10 s), then rinsing with water (10 s) and air-drying (10 s). The etchant material, 37% PA-gel (Proclinc, Madrid, Spain) in the control group and CaP paste in the experimental group, was applied on the middle third of the buccal surface of each specimen. Afterwards, stainless steel premolar brackets (Pinnacle, MBT prescription with 0.022" slot, Orthotechnology, USA) were used for bonding in both groups using Transbond XT primer and Transbond XT adhesive (3M Unitek, Monrovia, California, USA). A thin layer of Transbond XT primer was applied onto the etched enamel surface and spread by air-jet for 3 s. after loading the base with a thin layer of Transbond XT adhesive, the bracket immediately positioned on the specimen surface and firmly pressed (300 g force for 10 s) using a pressure gauge (Dontrix force gauge, DTC Medical Apparatus Co., Hangzhou, China). Light-curing of adhesive was conducted for 10 s on each mesial and distal side of the bracket using light emitting diode (LED) curing light (SDI Radium Plus, 1,500mw/cm² light intensity, Victoria, Australia) according to manufacturer instructions. After bonding, the specimens were incubated inside DW at 37°C for 24 h, after that half of each group (n=15) was tested for shear bond strength (SBS), while the other half continued storage in DW for 30 days with daily DW refreshment.

After the mentioned storage periods, specimens were subjected to SBS test using Universal Testing Machine (WDW-100E, Time Group Inc., Beijing, China) with a 4 KN load cell, at the College of Material Engineering/University of Kufa; the bracket debonded with a crosshead speed of 0.5 mm/min. The surface area of the bracket base (11.46 mm) was input into the computer prior to testing, so that the readings were demonstrated in megapascal (MPa). The debonded bracket besides the specimen were set aside in a labelled container showing the subgroup name for post-debonding surface examination.

The debonded enamel surfaces were inspected using a digital microscope (Dino-lite pro, AnMo Electronics Co., Taiwan) at x20 magnification to determine signs of crack or tear-out if any, and to evaluate the amount of the adhesive material remaining on the tooth surface to be scored according to the ARI system ⁽¹²⁾.

Statistical analysis

SBS data distribution at both testing times, preliminarily checked by Shapiro-Wilk test, assumed the normality ($P > .05$). Student t-test for independent samples applied to assess the statistical difference between subgroups of 37% PA and experimental CaP pastes at both debonding time points. ARI scores were examined for difference between control and CaP paste subgroups using Mann-Whitney test. The level of significance was set $p < 0.05$ at all statistical tests.

Results

The pH and SBS values of both the control and experimental subgroups at 24 h WS and 30 d WS are shown in table 1 and figure 1. At both testing time points, teeth etched with PA recorded consistently higher SBS values than those etched with the experimental CaP paste. The 30 d WS showed a non-significant influence on SBS of brackets bonded to teeth treated either with 37% PA or acidic CaP paste. Statistically significant differences were also found in the ARI scores between the control and CaP paste subgroups (Table 2). It was obvious that specimens etched with 37% PA tended to retain more amounts of adhesive upon brackets debonding at both testing times. In contrast, debonded teeth in CaP paste subgroups revealed enamel surfaces with minimal or no adhesive residues. In addition, by examining the debonded enamel surfaces using a digital microscope, several specimens of the control subgroups were found to have either cracks or fractures, whereas the specimens of the CaP subgroups showed smooth surfaces with no evidence of enamel injury at both testing time points (figure 2).

Table 1: pH and SBS (MPa) values of the control and experimental etchants after 24 h and 30 d WS.

Etchant	pH	24 h WS	30 d WS
		Mean (SD)	Mean (SD)
CaP paste	1.2	21.9 (7.99) A	18.2 (5.58) A
37% PA	0.6	31.99 (3.83) B	31.86 (8.93) B

Different letters indicate statistically significant differences between control and CaP paste subgroups (t-test).

Table 2: ARI scores after 24 h and 30 d WS for control and experimental subgroups.

Etchant	24 h WS				30 d WS			
	ARI				ARI			
	0	1	2	3	0	1	2	3
CaP paste	9	6	0	0 A	11	4	0	0 A
37% PA	1	7	4	3 B	2	7	2	4 B
	(4 EC)		(2 EF)		(3 EC)		(1 EF)	

EC: enamel crack, EF: enamel fracture. Adhesive remnant index (ARI) scores: (0): No adhesive left on the tooth, (1): Less than half of the adhesive left on the tooth, (2): More than half of the adhesive left on the tooth, (3): All adhesive left on the tooth with a distinct impression of the bracket mesh.

Different letters indicate statistically significant differences between control and CaP paste subgroups (Mann-Whitney test).

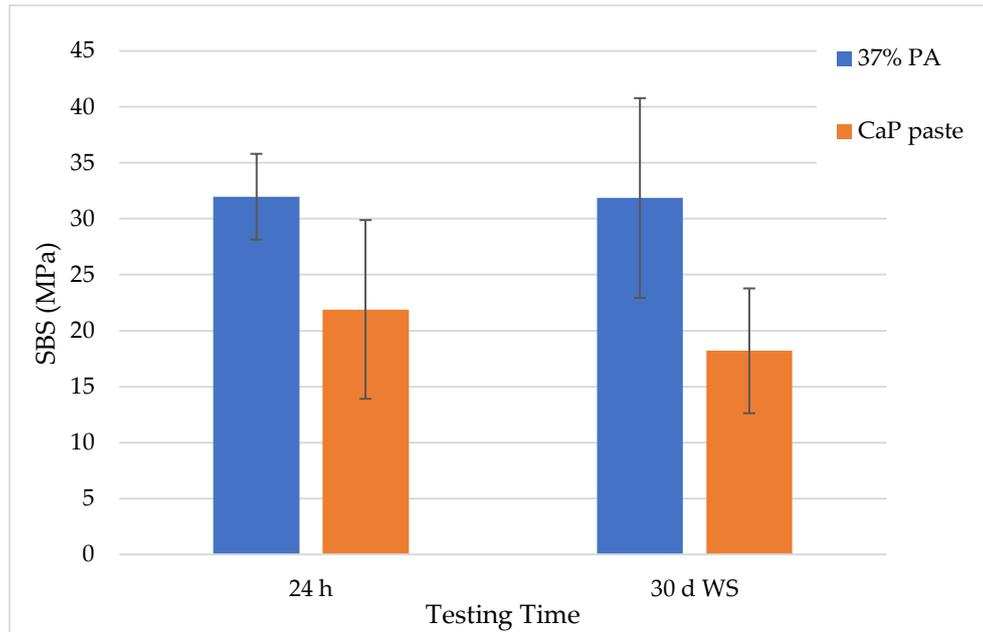


Figure 1: Bar chart illustrating SBS values for all subgroups according to testing time.

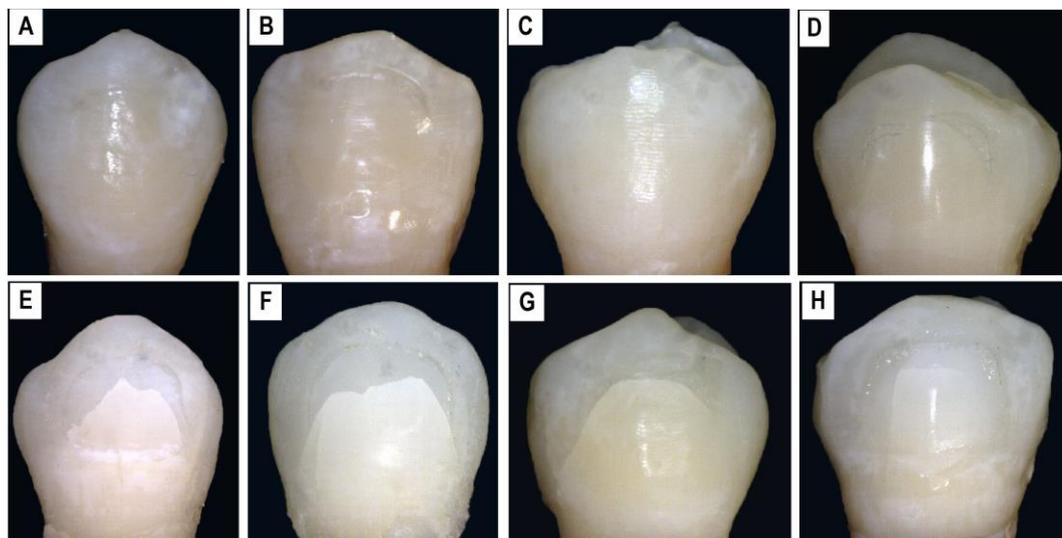


Figure 2: Digital microscope photographs of debonded specimens. A-D: Etched with CaP paste; the enamel surfaces appear smooth and unharmed with minimal or no adhesive remnants. E-H: etched with 37% PA; enamel surfaces have noticeable damage. Bracket debonding was accomplished after 24 h (A, B, E, F) and after 30 d (C, D, G, H).

Discussion

Orthodontic bracket fixation to teeth preceded by 35-40% PA treatment of enamel surface has been undoubtedly an effective approach in clinical practice, yet it has been implicated with a wide range of undesirable iatrogenic impacts on enamel surface. Despite several promising attempts in this regard, enamel conditioning techniques still require to be optimized to establish a secure bracket

adhesion associated with fewer or no adverse effects. Therefore, the current study intended to develop a bioactive etchant material in a simplified capsule formula that can underpin clinically adequate bracket bond strength without jeopardizing the integrity of enamel upon etching and debonding procedures.

Considerably variable SBS values have been described in the literature for nominally the same enamel etching protocol and adhesive system; this can be attributed to the multiplicity in test configurations among in vitro experiments ⁽¹³⁾. Many parameters, such as etching protocol, adhesive material, debonding time and bracket base shape, have been defined as variables affecting the in vitro evaluation of orthodontic brackets bond strength ⁽¹⁴⁾. Thus, the EAR technique using 37% PA reported a wide spectrum of bond strength values ranged from 9 to 35 MPa ⁽¹⁵⁾. The mean SBS values of all subgroups in the present study came in accordance with the above-reported range. Enamel etching with 37% PA-gel yielded significantly higher SBS values, at both testing times, than etching with the experimental CaP paste. This finding comes in accordance with the fact that the 37% PA etchant produces the highest bond strengths compared to any other etchant used for enamel conditioning ^(16,17). Indeed, the mechanism of bonding to enamel is essentially based on the micro-mechanical interlocking of resin monomers into created micro-pores through the differential removal of minerals from the dental substrate as a result of surface pre-treatment with an acidic etchant material ⁽¹⁸⁾. Thus, the resin infiltration morphology plays a major role in determining the resultant resin-enamel bond strength.

The pattern of inter-crystallite resin infiltration inside the prisms and the deep penetration into the inter-prismatic spaces was associated with high bond strength, whilst a shallow etch depth with inter-crystallite resin infiltrations and lack of inter-prismatic resin tags were accounting for reduced bond strength. The former was frequently correlated to high PA concentrations, whereas the latter was observed when reduced concentrations were used ⁽¹⁹⁾. This is also supported by previous research stating that the resultant morphology and depth of resin infiltration depend on the etching potential and the pH value of the etchant materials (4). Enamel treatment with 37% PA-gel of an acidic pH value (0.6) leads to extensive micro-pores creation post a very short application time (15-30 s) because the pH remained probably unchanged even with the dissolution of HA from the enamel surface leading to a strong etching effect and a recognized inter-prismatic demineralization, thereby thick resin tags are established yielding high bond strengths ⁽²⁰⁾. This finding is supported by a previous study that recorded higher bond strength values with a more acidic meta-PA (pH 0.5), similar to conventional 37% PA ⁽²¹⁾. It was also concluded that the SBS is closely related to the pH of the etchant ⁽²²⁾. On the other hand, enamel etching with CaP pastes of higher pH (1.2) yielded significantly lower SBS values compared with the control (37% PA). Pre-mixing of HA micro-powder with 40% PA imposed an effective buffering effect leading to a rise in the pH of resultant CaP paste. When this paste with less acidity was applied onto the tooth surface, the demineralization process occurred to a limited extent producing a milder etching effect; thereby resin penetration was diminished leading to a drop in resultant SBS. However, the SBS values obtained with the CaP pastes subgroups exceeded the suggested threshold (6–10 MPa) in the vast majority of the literature as admissible for clinical performance ^(23,24).

The change in bond strength following 24 h has received limited attentiveness, where most in vitro experiments were confined to the evaluation of SBS at 24 h presuming that this reflects long-term bonding efficiency. The resultant enamel-adhesive interface actually undergoes several challenges in

the oral environment, which can impose adverse effects on the bonding longevity of an orthodontic adhesive system ⁽²⁵⁾. It was reported that one of the major factors encountered in vivo environment is the bond strength deterioration due to water ageing that is thought to be a result of interface components hydrolysis and water infiltration, which can also weaken the mechanical properties of the adhesive polymer matrix ^(26,27). Therefore, aging in water was the most frequently used artificial aging protocol to examine the durability of created enamel-adhesive joint ⁽²⁸⁾. The testing of bracket adhesion in specimens treated with either the CaP paste or PA revealed a non-significant reduction in SBS outcomes after 30 d WS. This can be attributed to that the created enamel-adhesive joint using the etchant paste or 37% PA was sufficiently appropriate to endure the long-term water ageing process. This agrees with the findings of previous studies, which found that the durable bonding interface established by the EAR technique could tolerate prolonged WS periods, extended for 30 d to 2 years, without significant alteration in bond strengths ^(11,28,29).

In the current study, the examination of the debonded enamel surfaces demonstrated that the etchant type had an impact on the failure mode, with the site of bond failure differing between the control and experimental etchant material. In control subgroups, the failure was mainly an adhesive failure at the bracket/adhesive interface, with the majority of residue adhesive remained on the tooth surface (ARI scores 2 and 3), indicating a stronger bond between the adhesive and enamel. It was observed that when the enamel was etched using 37% PA, where a deep and pronounced interaction occurs with the enamel surface creating the most retentive morphology for adhesive material, hence upon bracket debonding, much of adhesive remains on the tooth surface ⁽²⁰⁾. Moreover, brackets bonded to PA-etched enamel surfaces entail a high debonding force which associated with high incidence of enamel hurts, e.g. cracks and/or tear-out ^(30,31). It was reported that the risk for enamel damage increased with a higher bracket bond strength ⁽⁴⁾. The outcomes of the present study advocate this view, as shown in figure 2 E-H, where bracket removal in 37% PA subgroups was associated with varying forms of enamel damage so that several teeth exhibited enamel cracks and tear-outs. In contrast, the experimental pastes revealed a frequent failure at the enamel/adhesive interface recording significantly lower ARI scores (mostly 0 and 1). Changing the composition and the pH of experimental etchant pastes achieved in the current study probably had a considerable role in the conservative interaction with enamel, hence facilitating adhesive failure at the enamel/adhesive interface, more clinically convenient, which makes the debonding and subsequent polishing much easier ⁽¹⁰⁾. Moreover, the teeth treated with developed etchant paste notably revealed smooth, unblemished enamel surfaces after bracket debonding; this might have resulted from the milder etching effect and diminished resin penetration into the etched enamel substrate that has frequently been associated with a reduced risk of enamel damage upon bracket removal ⁽⁸⁾.

With the absence of the effect of other intraoral factors such as chewing forces besides temperature and pH fluctuations commonly encountered in the normal oral environment, the findings of this study should be extrapolated with caution; the evaluation of bonding performance of the developed CaP paste via a clinical trial is thus required.

Conclusion

This in vitro study presented a newly developed CaP paste in a mixing capsule formula, which fosters clinically adequate bracket adhesion with a sustained bonding performance, and allows a harmless bracket removal with minimal or no adhesive residues on debonded surfaces; thus, it can be introduced as a suitable alternative to conventional 37% PA.

Conflict of interest: None

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**العنوان: تقييم مختبري للتصاق الحاصرات بعد تكييف المينا باستخدام عينة حفر جديد
الباحثون: حيدر عبد المنعم كاظم , سانجوكتا ديب , علي اسماعيل ابراهيم
المستخلص**

الخلفية: حمض الفوسفوريك (PA) 37٪ التقليدي هو المعيار الذهبي لحفر المينا قبل التصاق الحاصرات ، ومع ذلك فقد تورط في العديد من إصابات المينا. الهدف من هذه الدراسة هو تطوير معجون فوسفات الكالسيوم (CaP) بصيغة كبسولة مبسطة يمكن أن تدعم قوة رابطة الحاصرات المناسبة سريريًا دون المساس بسلامة المينا عند عملية إزالة الأقواس. المواد وطرق العمل: تم خلط مسحوق هيدروكسيباتيت (HA) مايكروية الحجم مع 40٪ محلول PA لتحضير معجون CaP الحمضي التجريبي. تم تقسيم ستين ضاحكًا بشريًا سليمًا بشكل عشوائي إلى مجموعتين رئيسيتين (ن=30). تم إجراء تكييف المينا باستخدام 37٪ جل PA للمجموعة الضابطة ومعجون CaP للمجموعة التجريبية. تم تقسيم كل مجموعة إلى مجموعتين فرعيتين فيما يتعلق بفترة تخزين المياه (WS) (24 ساعة و 30 يومًا). تم إجراء اختبار قوة رابطة القص (SBS) مع فحص الأسطح بعد نزع الحاصرات بحثًا عن بقايا اللاصق وتلف المينا باستخدام مجهر رقمي. النتائج: أنتج معجون CaP المطور قيم SBS أقل بكثير من 37٪ (p < 0.01) ، لكنه كاف للاستخدام السريري. تسبب حفر PA في كثير من الأحيان في تشقق أسطح المينا مع الاحتفاظ المفرط ببقايا المادة اللاصقة (بشكل أساسي درجات 2 و 3 من ARI). في المقابل ، أظهر المينا المعالجة بمعجون CaP التجريبي أسطحًا ناعمة لا تشويها شائبة نظيفة في الغالب من بقايا المواد اللاصقة (الدرجات 0 و 1). الاستنتاج: معجون CaP المطور حديثًا في تركيبة كبسولة خلط يعزز التصاق حاصرات مناسب سريريًا مع أداء ارتباط مستدام ، ويسمح بإزالة القوس غير المؤذي مع الحد الأدنى من بقايا اللاصق أو عدم وجودها على الأسطح المنزوعة الحاصرات؛ وبالتالي ، يمكن تقديمه كبديل مناسب لـ 37٪ PA التقليدي.