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ORIGINAL ARTICLE

Push-out bond strength of three different calcium silicate-based root-end filling materials after ultrasonic retrograde cavity preparation

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Abstract

Objective The aim of this study was to evaluate the bond strength of three calcium silicate-based root-end filling materials.

Materials and methods The root canals of 30 single-rooted teeth were endodontically treated; their root ends were resected and root-end cavities were prepared using ultrasonic tip. The teeth were randomly divided into three groups according to the material: (1) Micro-Mega mineral trioxide aggregate (MM-MTA), (2) Biodentine, and (3) TotalFill root repair material (RRM). Push-out test was performed using universal testing machine, and failure mode was analyzed by stereomicroscope. The data were statistically analyzed using Kruskal-Wallis and Man-Whitney post hoc tests. All p values < 0.05 were considered significant.

Results TotalFill RRM exhibited significantly higher bond strength (12.69 MPa) than Biodentine (9.34 MPa, $p = 0.023$) and MM-MTA (7.89 MPa, $p = 0.002$). The difference between Biodentine and MM-MTA was not significant ($p = 0.447$). Mixed failures were the most noted in all three groups. MM-MTA had more adhesive failures than Biodentine and TotalFill, and no cohesive failures, but without statistical significance ($p = 0.591$).

Conclusion The bond strength was the highest for TotalFill RRM.

Clinical relevance In order to provide a persistent apical seal, root-end filling materials should resist dislodgement under static conditions, during function and operative procedures. TotalFill RRM exhibited higher bond strength to dentin than MM-MTA and Biodentine.

Keywords Biodentine · MM-MTA · Push-out test · Root-end filling materials · TotalFill RRM

Introduction

When conventional root canal treatment or retreatment is associated with post-treatment disease, surgical endodontics may be indicated [1]. In such cases, the recommended surgical procedure involves the resection of the apical 3 mm of the root, followed by retrograde cavity preparation and root-end filling placement [1]. The usage of ultrasonic tips for root-end cavity preparation enables clinician to obtain cleaner, deeper, and more centered cavities favoring the marginal adaptation of the root-end filling materials [2]. This prevents the leakage of microorganisms and their toxins from the root canal system and contributes to periapical healing [3].

Along with the shape and position of the cavity, the quality of apical seal greatly depends on the root-end filling material's properties. An ideal root-end filling material ought to have dimensional stability, radiopacity, proper setting time, antimicrobial activity, biocompatibility, biomimetic properties, and resist dislodging forces and solubility [4, 5]. Much evidence support a calcium silicate cement MTA as the gold-standard material, not only the for root-end filling but also for a series

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of other clinical procedures like pulp capping, pulpotomy, apexogenesis, apexification, root perforations repair, and root canal filling [6]. However, MTA has poor handling properties and long setting time [7]. There are several other calcium silicate-based materials claiming improved handling properties and reduced setting time, including MM-MTA (Micro-Mega, Besançon Cedex, France) and Biodentine (Septodont, Saint Maur-des Fosses, France) which are presented in capsules, and TotalFill root repair material (FKG Dentaire, La Chaux-de-Fonds, Switzerland) which is presented in ready to use form.

In order to provide a persistent apical seal, a root-end filling should adhere well to root canal dentin so that the integrity of the filling material–dentin interface is maintained not only under static conditions but also during function and operative procedures [8]. Push-out test was shown to be an efficient and reliable method for the assessment of the property of a set material to resist dislodgement forces in vitro [9].

The aim of this study was to evaluate the bond strength of MM-MTA, Biodentine, and TotalFill RRM to dentin using push-out test after ultrasonic retrograde cavity preparation and to analyze the failure mode using stereomicroscope.

Material and methods

Preparation of specimens

The research was approved by the Ethical committee of the School of Dental Medicine, University of Zagreb, and patients signed the informed consent allowing the usage of their teeth in the study. Thirty single-rooted extracted human maxillary incisors with straight, fully formed roots, a single canal, and no previous endodontic treatment were used. Any extraneous tissue and calculus were removed using curettes, and the teeth were stored in saline prior to instrumentation. The crowns were sectioned below the cementoenamel junction using a water-cooled diamond drill, leaving 16-mm long roots. The root canals were instrumented with rotating ProTaper instruments (Dentsply Maillefer, Ballaigues, Switzerland) in a standard sequence to the apical F3.

The smear layer was removed using 2 ml of 17% EDTA (pH 7.7) for 1 min. The final irrigation was carried out with saline; the canals were dried and obturated using gutta-percha size 30 (Dentsply Maillefer, Ballaigues, Switzerland) and endodontic sealer (AH Plus Dentsply, DeTrey, Konstanz, Germany) using a single cone technique. The specimens were stored at 37 °C in a 100% moist environment.

After 1 week, they were sectioned perpendicularly to their long axis, 3 mm short of the apex and 3-mm deep retrograde cavities were prepared using R1D ultrasonic retrotip coupled to an ultrasound device (Piezomed W&H, Bürmoos, Austria, lot 00PE51410). The specimens were randomly divided into three groups ($n = 10$) and the cavities were filled with MM-MTA, Biodentine, or TotalFill RRM according to the manufacturers' instructions. After placement, each root-end filling was compacted with a small plugger and the specimens were stored in saline (0.9%) for 3 months.

Push-out test

The specimens were embedded in acrylic resin (Orthocryl, Dentaurum, Ispringen, Germany). The apical part of each specimen was cut perpendicular to the long axis into 1-mm thick slices with a diamond blade using Isomet 1000 precision saw (Buehler, Düsseldorf, Germany), at a speed of 150–200 rpm. The thickness of each slice was measured using digital caliper (precision level +/− 0.001 mm, Roc International Industry Co., Ltd., Guangdong, China), and the value was recorded. The push-out test was performed in a coronal to apical direction due to the reversed taper during preparation. The bonding surface was calculated using the conical frustum formula:

$$Area = \pi(R1 + R2)\surd(R1\text{–}R2)^2 + h^2$$

with apical radius $R1$ as the larger radius, coronal $R2$ as the smaller radius, and h as the thickness of a slice. A compressive push-out load was applied using a universal testing machine (double-column 3300 series, Instron, Illinois, USA). The slices were centered over a hole in the device, and a compressive load was applied with a 1.0-mm diameter blunt-shaped probe at a speed of 0.5 mm/min until failure. The push-out bond strength expressed in megapascals was calculated by dividing the load at failure by the bonding surface. The slices were observed under a stereomicroscope (×10–×50) to verify the failure mode (adhesive, cohesive, or mixed).

Statistical analysis A priori power analysis was performed to calculate adequate number of samples to be included in the study. From the results of the pilot study, it was estimated that the expected mean values of bond strength would be around 8 MPa for MM-MTA, 9 MPa for Biodentine, and 13 for TotalFill RRM with corresponding standard deviations around 3. These data gave us an estimated effect size f value of 0.72 that we used to calculate minimal necessary number of samples for one-way analysis of variance (ANOVA). For the three groups, with α level of probability of 0.05 and sample power of 90%, we needed 28 samples. Normality of data distribution was checked with Kolmogorov-Smirnov test. Since the distribution of our final data was not normal, we used non-parametric Kruskal-Wallis test instead of ANOVA. The total sample size for Kruskal-Wallis test could be even smaller. Post hoc analysis of differences in bond strength between the three tested materials was done using Mann-Whitney U test. Fisher-Freeman-Halton exact test was used to analyze differences in types of fracture distribution among the three experimental groups at the level of significance $p < 0.05$. IBM SPSS Statistics version 23 was used in all statistical procedures, and power analysis was done with G*Power for Windows (version 3.1.9.2) computer software [10].

Results

Differences in bond strengths (MPa) between MM-MTA, Biodentine, and TotalFill RRM are shown in Table 1, as well as the number of slices, i.e., samples for each of the three experimental groups. The lowest median value was in MM-MTA group 7.89 (6.34–10.48) MPa while the highest was in TotalFill RRM group 12.69 (10.82–16.19) MPa. Biodentine group had bond strength median value of 9.34 (7.69–12.21) MPa. Additional post hoc analysis showed that TotalFill RRM group had significantly higher bond strength than MM-MTA group ($p = 0.002$) and Biodentine group ($p = 0.023$). There was no significant difference between MM-MTA and Biodentine group ($p = 0.447$; Fig. 1).

Experimental model and fracture modes are shown in Fig. 3. Adhesive, mixed and cohesive fractures were equally distributed between MM-MTA, Biodentine and TotalFill RRM groups ($p = 0.591$). Mixed failures were the most observed failure types for all materials. MM-MTA material had more adhesive failures, when compared to the other materials tested, and no cohesive failures (Fig. 2).

Discussion

The present in vitro study evaluated the bond strengths of three different calcium silicate-based root-end filling materials using push-out test.

Push-out tests are widely used to evaluate bond strength of endodontic filling materials and posts to the root dentin [11]. Marques et al. [12] presented a new methodology to evaluate bond strength of root-end filling materials in order to better resemble clinical conditions and suggested taking slices of apical third of the root for testing instead widely used middle root canal filled slices usually taken to test the push-out bond strength of root canal sealers. In our study, we also used the slices cut from the apical third of the root where actual retrograde cavity and filling are clinically positioned. It was not the aim of this study to reproduce conditions identical to in vivo ones since no push-out stresses act on the apical region of the root in the way reproduced in this test. The results of this study in fact help in assessing which material may have better performances in terms of bonding strength to the prepared cavity and therefore resistance to dislocation [9].

The characteristic of calcium silicate-based materials is to precipitate carbonated apatite in the presence of tissue fluids, followed by the formation of interfacial layer and tag like structures in the dentin [7]. A recent study by do Carmo et al. [13] showed that the interfacial layer maturation and bond strength depend on the storage medium in in vitro studies (PBS vs. distilled water). In our study, the samples were left in saline, and not in distilled water, to better mimic conditions in the tissue fluids which retrograde fillings come in contact with during setting in vivo.

The retention to the dentinal wall and physical properties of these materials depend on water/powder ratio, temperature, humidity, the quantity of air trapped in the mixture, and the particle size [14]. Using capsulated and ready to use formulations like the ones used in our study, the variations in water/powder ratio and air trapped in the mixture are reduced to the minimum. The particle size varied between the materials tested. TotalFill RRM which contains the particles of the nanospheric size of 1×10^{-3} μm (maximum) exhibited significantly higher bond strength compared to MM-MTA and Biodentine. Higher bond strength of calcium silicate-based materials with smaller particle size was previously explained by better penetration of those particles into the dentinal tubules [15]. In our study, cavities were not treated with EDTA prior to the cement placement, so the greatest resistance to dislodgement exhibited by TotalFill RRM cannot simply be explained by its easier penetration into dentinal tubules due to the smaller particle size since the tubules were not open in the first place. Moreover, it was reported that irrigation with EDTA has no effect on the push-out bond strength of the calcium silicate cements [16, 17], which also supports the idea that higher bond strength cannot solely be explained by the deeper penetration into dentinal tubules. However, it could be that the smaller particles of the cement favor a better hydration and consequent calcium ion release [14]. That leads to more calcium phosphate precipitates and

Table 1 Bond strength values (MPa) of MM-MTA, Biodentine, and TotalFill RRM: Kruskal-Wallis test

Material	*N*	Bond strength (MPa)					*p*
		Minimum	Maximum	Percentiles			
				25th	50th (median)	75th	
MM-MTA	9	4.48	13.49	6.34	7.89	10.48	0.005
Biodentine	10	3.99	20.94	7.69	9.34	12.21	
TotalFill RRM	15	7.47	17.98	10.82	12.69	16.19	

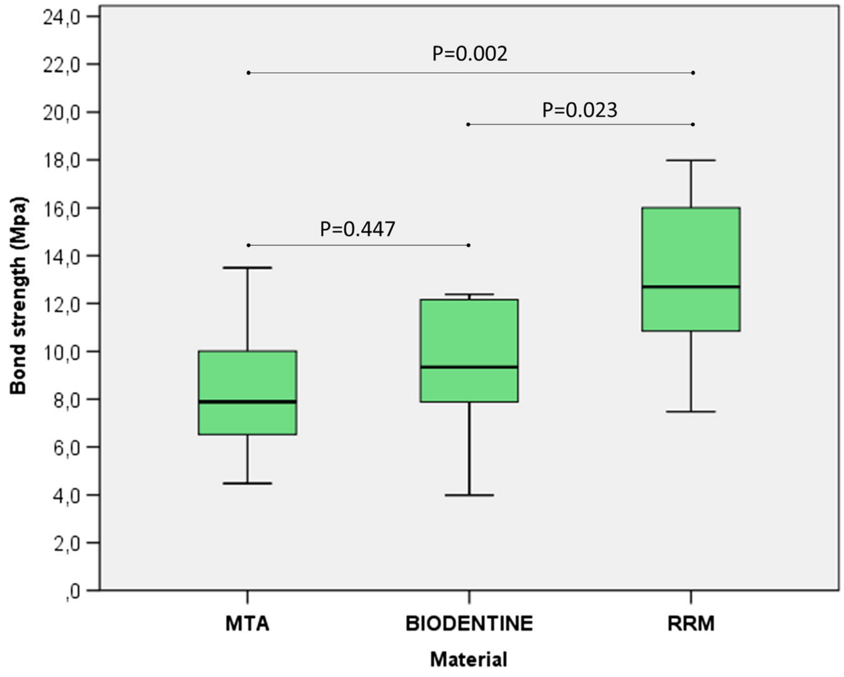

Fig. 1 Post hoc analysis of differences in bond strength between MM-MTA, Biodentine, and TotalFill RRM: Box and Whisker's plot (medians and interquartile ranges) with corresponding Mann-Whitney *U* test

tag-like structures which constitute micromechanical anchorage, thus increasing dislodgement resistance. This is supported by the findings of Han et al. [18] who reported that the depth of calcium and silicon incorporation into dentin was higher for Biodentine which has more homogenous and smaller particles than MTA. Hence, maybe, the higher bond strength of TotalFill RRM in our study could be explained by calcium and silicon uptake by dentin.

Despite belonging to the same group of materials, the materials tested in our study have different compositions which influence setting kinetics and marginal adaptation [7, 18]. Phase composition of MM-MTA powder was reported to be tri-calcium silicate, di-calcium silicate, tri-calcium aluminate, calcium carbonate, calcium sulfate, and bismuth oxide [7]. Biodentine powder contains tri-calcium silicate, calcium carbonate, and zirconium oxide, but no di-calcium silicate [19].

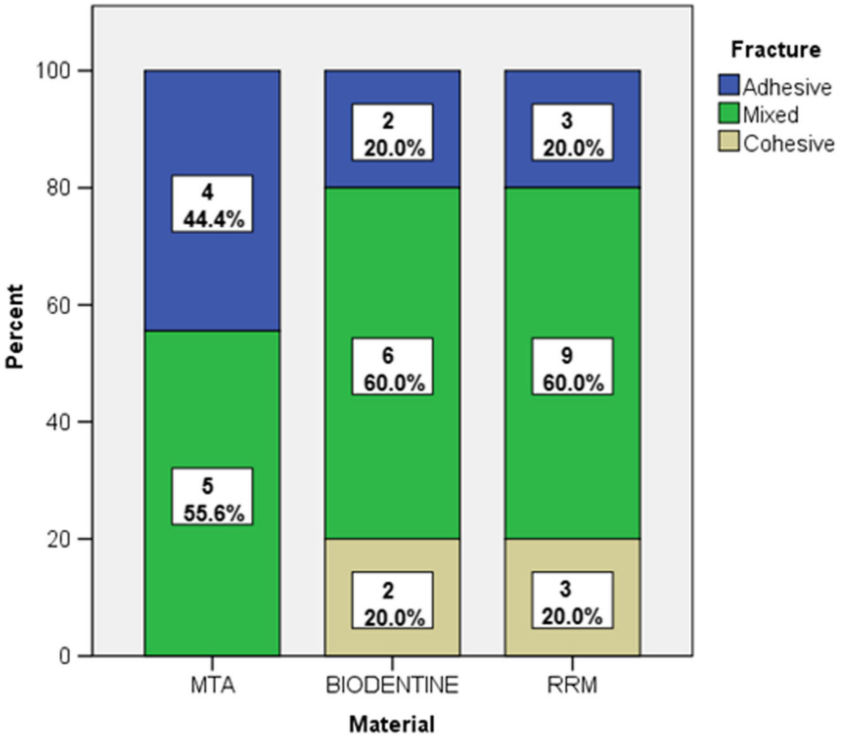

Fig. 2 Differences in failure types distribution between MM-MTA, Biodentine, and TotalFill RRM group: Fisher-Freeman-Halton exact test; $p = 0.591$

The addition of calcium carbonate to the Biodentine's powder (acting as nucleation sites) and $CaCl_2$ to its liquid accelerates the setting reaction [20, 21]. Also, the fast setting of Biodentine of only 12 min [7] could be attributed to the fact that it is essentially composed of tri-calcium silicate. It was reported that the proportion of di-calcium silicate and tri-calcium silicate significantly influences the setting kinetics because di-calcium silicate gets hydrated slowly [22, 23]. Moreover, the absence of di-calcium silicate from its components makes the powder of Biodentine more homogenous, thus enabling better marginal adaptation and higher resistance to dislodgement [7]. This could explain why the bond strength exhibited by Biodentine was somewhat higher than that of MM-MTA, but not significantly. This is in concordance with some previous studies which reported that Biodentine showed higher bond strength to dentin than MTA [24, 25].

However, in our study, the bond strengths of both MM-MTA and Biodentine, when compared to TotalFill RRM, were significantly lower. Unlike MM-MTA and Biodentine, TotalFill RRM, also known as EndoSequence® RRM in the USA and Canada (ERRM; Brasseler USA, Savannah, GA, USA), contains phosphate salts in addition to hydraulic calcium silicates [26]. It namely consists of calcium silicates, zirconium oxide, tantalum peroxide, calcium phosphate monobasic, and fillers [27]. During cement setting, tri-calcium silicate hydrates to produce calcium silicate hydrate (C-S-H) gel and calcium hydroxide; further, calcium phosphate monobasic reacts with $Ca(OH)_2$ to precipitate hydroxyapatite in situ within C-S-H. Bearing in mind the definition of bioactivity in vivo as a specific biological response at the interface of the material resulting in the formation of a bond between living tissues and the material [28], this could explain why the bond of TotalFill RRM to dentin was stronger. Furthermore, it was reported that the materials with accelerated setting release significantly less Ca^{2+} and that such precipitated crystals have significantly lower Ca/P ratios [29]. Since the setting time of TotalFill RRM is relatively long compared to MM-MTA and Biodentine, probably, more calcium ions are available in TotalFill RRM, and crystals precipitate more readily. However, we must point out that this experiment was conducted in vitro and that the bond strength would, in clinical conditions, be influenced by the contamination of the cement with blood during the setting reaction. It was, in fact, reported that contamination with phosphate-buffered-solution during setting significantly reduces the expansion of MTA [30]. In that context, faster setting would reduce the negative impact of contamination, and it is fair to assume that Biodentine would perform relatively better in vivo conditions due to its significantly faster setting time of 12 min, as compared to MM-MTA and TotalFill RRM which are claimed to be at 20 min and 2 h, respectively [7].

Bond strength of the root-end filling material to dentin can also be influenced by the type of ultrasonic tip used to prepare retrograde cavities [31]. In a study by Vivan et al. [31] the CVD T0F-2 ultrasonic tip, compared to Trinity diamond tip and Satelec S1290 L tip, favored higher bond strength to dentin of two types of MTA and zinc oxide eugenol cement. In our study, we used the same ultrasonic unit and same type of ultrasonic tip (R1D) to prepare retrograde cavities, so the differences in bond strength could only be attributed to the materials we used for root-end filling.

All specimens were inspected after ultrasonic preparation and, additionally, after sectioning and prior to push-out test with the stereomicroscope. Few specimens that showed dentin fracture lines were found only after sectioning and were thus

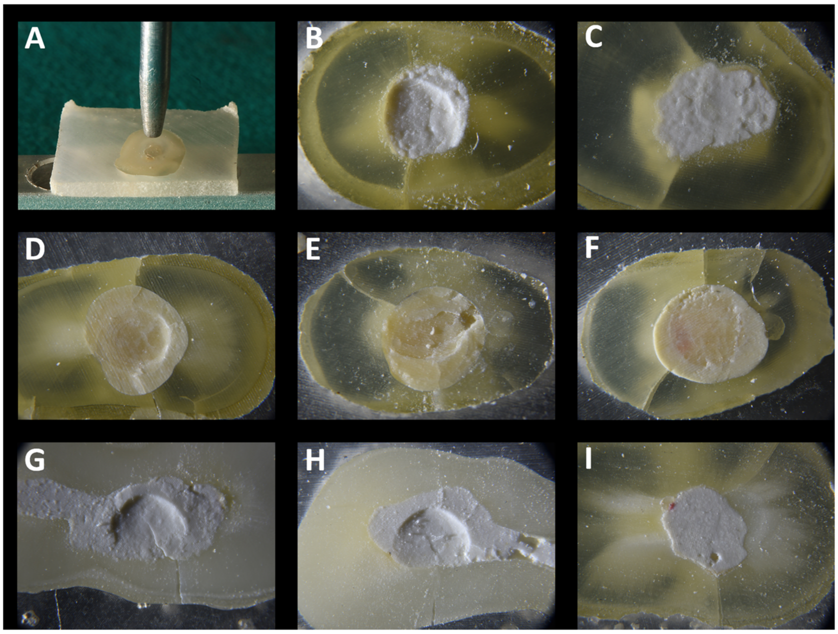

Fig. 3 Representative failure modes for each tested material as follows: **a** experimental setup of the push-out test, the 1-mm-thick slice of tooth embedded in resin was positioned horizontally with the retrograde filling centered under the blunt probe; **b** MTA, mixed failure mode; **c** MTA, adhesive; **d** Biodentine, cohesive; **e** Biodentine, mixed; **f** Biodentine, adhesive; **g** RRM, cohesive; **h** RRM, mixed; **i** RRM, adhesive

discarded. It was thus surprising to find fracture lines in dentin of all specimens radiating from the canal (Fig. 3). First explanation could be that the impact of the probe resulted in lateral stresses during the push-out test, due to lateral dissipation of forces by the Hertzian cone crack system, which eventually compressed dentinal tissue against the epoxy material of the slab. Actually, in this case we would not expect to find dentin fracture lines in specimens characterized by pure adhesive failure since in that case no cone crack, only frictional forces at the interface between the retrograde material and dentinal tissues were produced. It may be speculated that these frictional forces may have dissipated inside dentin producing the fractures visible in all specimens. This might be the result of an early mineral deposit formation at the interface (do Carmo et al. 2017) and may give an indirect proof of the quality of interconnection between the material and the dentinal tissues. Further studies may address this interesting possibility.

The failure types of the MM-MTA were different from those seen for the Biodentine and TotalFill RRM. The bond failures observed in MM-MTA group were either adhesive type or mixed type of failures. This finding is in accordance with some previous studies [15, 25, 32, 33]. Different failure types of MM-MTA compared to Biodentine and TotalFill RRM may be explained by smaller and uniform size of particles in Biodentine and TotalFill RRM, enabling better marginal adaptation and penetration into dentin and resulting finally in better adhesion. Biodentine samples presented predominantly cohesive mode of failure in a recent study, and after SEM analysis, it was reported that Biodentine had particles that appeared firmly attached to the underlying surface [15]. The higher percentage of cohesive failures in Biodentine samples than in our study could be attributed to different setting time of the materials which was in the mentioned study only 4 days [15].

All three materials had mixed type of failures predominantly, which means the presence of cohesive and adhesive failures at the same time. Tested materials showed some weaknesses not only in material itself (cohesive failures) but also in the bond with the radicular dentin (adhesive failures). However, mixed type of failure still indicates that the adhesive interface between radicular dentin and all three retrograde filling materials, which were investigated in the present study, were preserved, at least in part. However, TotalFill RRM exhibited significantly higher bond strength in comparison to Biodentine and MM-MTA. Taking into consideration the same type of failure for all the materials and if we extrapolate the results of our in vitro study to clinical conditions, TotalFill RRM could perform better than the other two materials because of the stronger bond to dentin.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval This article does not contain any studies with human participants or animals performed by any of the authors. The article contains in vitro studies on human teeth where the extraction was indicated. The research was approved by the Ethical Committee of the School of Dental Medicine, University of Zagreb.

Informed consent The participants signed the informed consent allowing the usage of their teeth in the study.

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