ORIGINAL ARTICLE

Ridge preservation of extraction sockets with buccal bone deficiency using poly lactide-co-glycolide coated β-tricalcium phosphate bone grafts: An experimental study in dogs

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Abstract

Background: The alveolar ridge undergoes pronounced reduction in height and width following tooth extraction. This study aims to comparatively evaluate the potential for ridge preservation in extraction sockets with buccal bone deficiency of β-tricalcium phosphate coated with poly lactide-co-glycolide (β-TCP/PLGA) and conventional particulate β-TCP.

Methods: In six beagles, maxillary first premolars were extracted after removal of their buccal bone plates. Standardized bone defects (4 [mesiodistal width] $\times$ 4 [buccopalatal width] $\times$ 5 [depth] mm) were created at the sites of extraction sockets and filled with β-TCP/PLGA (test sites) or particulate β-TCP (control sites). Microcomputed tomography, histologic, and histometric evaluations were performed 12 weeks post-surgery.

Results: The test sites exhibited a significantly greater bone volume than the control sites (25.7 ± 2.14 versus 16.0 ± 3.3 mm^3), although no statistically significant difference was detected in bone material density (746.3 ± 23.9 versus 714.5 ± 37.0 g/cm^3, respectively). Relative to the control sites, the test sites exhibited significantly greater alveolar-ridge coronal (2.0 ± 0.4 versus 1.1 ± 0.3 mm) and middle (2.9 ± 0.2 versus 2.1 ± 0.3 mm) horizontal widths and proportions of woven bone ($50.3 \pm 8.1\%$ versus $38.0 \pm 5.2\%$) and bone marrow ($17.7 \pm 6.6\%$ versus $9.7 \pm 4.1\%$) but a significantly lower proportion of connective tissue ($10.7 \pm 4.5\%$ versus $18.3 \pm 5.7\%$).

Conclusion: Within the limitations of this study, the moldable β-TCP/PLGA graft appears to exhibit a greater potential than the conventional particulate β-TCP graft for ridge preservation of extraction sockets with buccal bone deficiency.

KEYWORDS
alveolar ridge augmentation, β-tricalcium phosphate, biocompatible materials, dogs, osteogenesis, tooth extraction

1 | INTRODUCTION

The alveolar ridge undergoes pronounced reduction in its original height and width following tooth extraction.[1,2] Previous studies have demonstrated that these dimensional changes are greater in the buccal bone than in the lingual bone.[2,3] A previous review reported horizontal and vertical bone loss of 29%–63% and 11%–22%, respectively, within 6 months following tooth extraction.[4] This alveolar ridge reduction might have considerable adverse effects on the predictable outcomes of functional and esthetic tooth-restoration therapy, particularly when implant-supported restorations are required. In addition, this resorptive process often results in a more lingually relocated alveolar ridge.[5] In beagle dogs, it has been shown that pronounced dimensional remodeling occurs at extraction sockets with buccal bone deficiency when graft materials are not used.[6,7] Various alveolar ridge preservation techniques following tooth extractions have been developed to date.[8,9] Denissen et al. have demonstrated that the use of dental implants in fresh extraction sockets could prevent alveolar-bone reduction.[10] However, when this technique was investigated in animal and human experiments, buccal bone loss was observed.[11,12] Other ridge preservation techniques advocate the use of grafting biomaterials in extraction sockets.[13,14] Autologous bone grafts have been considered as the gold standard in reconstructive therapy for many years.[15] However, these require additional surgery, which leads to a subsequent increase in the risk of morbidity.[16] Various biomaterials such as allografts,[17,18] xenografts,[19,20] and alloplastic grafts[21,22] have been used for alveolar ridge preservation. Ridge-preservation approaches using allografts and xenografts significantly limit the resorption of hard tissue.[18,20] Among existing alloplastic grafts, β-tricalcium phosphate (β-TCP) is a biocompatible material that exhibits biodegradability, bioactivity, and osteoconductivity.[23] Some clinical studies have demonstrated the potential of β-TCP as a graft material for alveolar ridge preservation following tooth extraction.[24,25] However, it is difficult to maintain the defect configuration by using particulate β-TCP, which requires the use of barrier membranes for containment and stabilization in the early phase of socket healing.[26] A unique biomaterial consisting of β-TCP coated with poly lactide-co-glycolide (β-TCP/PLGA) has been developed, which has the property of solidifying after filling in the bone defect and retaining its shape. In this material, a pure-phase β-TCP particle is coated with a thin layer of biodegradable PLGA. Before application, PLGA is mixed with N-methyl-2-pyrrolidone (NMP), which softens the PLGA and renders the particles easily to moldable. After application, the NMP is flushed out upon contact with body fluids, resulting in the formation of a stable bone graft.[27] This β-TCP/PLGA biomaterial might greatly decrease the use of membranes for stabilization of bone grafts in the defect, resulting in a fast and simple surgical approach. Although this biomaterial appears to be effective for use in human clinical applications, previous reports have only demonstrated the histologic components of bone samples extracted using a trephine drill before dental implant placement.[28,29] Moreover, there are still insufficient preclinical data on comparison of β-TCP/PLGA and conventional bone grafts, including pure β-TCP. Therefore, the present study aims to comparatively evaluate the efficacy of β-TCP/PLGA and conventional particulate β-TCP in preventing linear and volumetric changes of the alveolar ridge in extraction sockets with buccal bone deficiency.

2 | MATERIALS AND METHODS

2.1 | Bone graft substitutes

In this study, particulate β-TCP refers to pure, continuous, interconnected, porous (65%), fully synthetic β-TCP particles with a particle size of 500–1,000 μm.*[27] A scanning electron micrograph showing the particle size of PLGA/β-TCP as well as the PLGA coating can be seen in Supplementary Figure 1 in the online *Journal of Periodontology*. This type of particulate β-TCP was selected for control sites on the basis of previous studies.[30] β-TCP/PLGA† is a synthetic porous composition of pure-phase particulate β-TCP, which exhibits 70% porosity and a diameter of 500–1,000 μm and is covered with a 10-μm-layer of a bioresorbable PLGA polymer. Particles of PLGA-coated β-TCP were mixed with NMP solution.‡ Clinical preparation of β-TCP/PLGA was performed in accordance with the method described in a previous study.[27]

2.2 | Surgical procedure

Six beagle dogs (males, aged 12 months) weighing about 10 kg were included in this study. The study protocol was approved by the Ethical Committee for Animal Research of Tokyo Medical and Dental University, Tokyo, Japan (0170333A). As premedication, medetomidine hydrochloride (0.05 mL/kg)§ was administered intramuscularly. General anesthesia was induced by intravenous injection of sodium thiopental (0.005 mL/kg)¶ under spontaneous breathing. Lidocaine hydrochloride (2% with 1:80,000 epinephrine)# was administered as a local anesthetic. Experimental defects were prepared as described in a previous study.[7] In each

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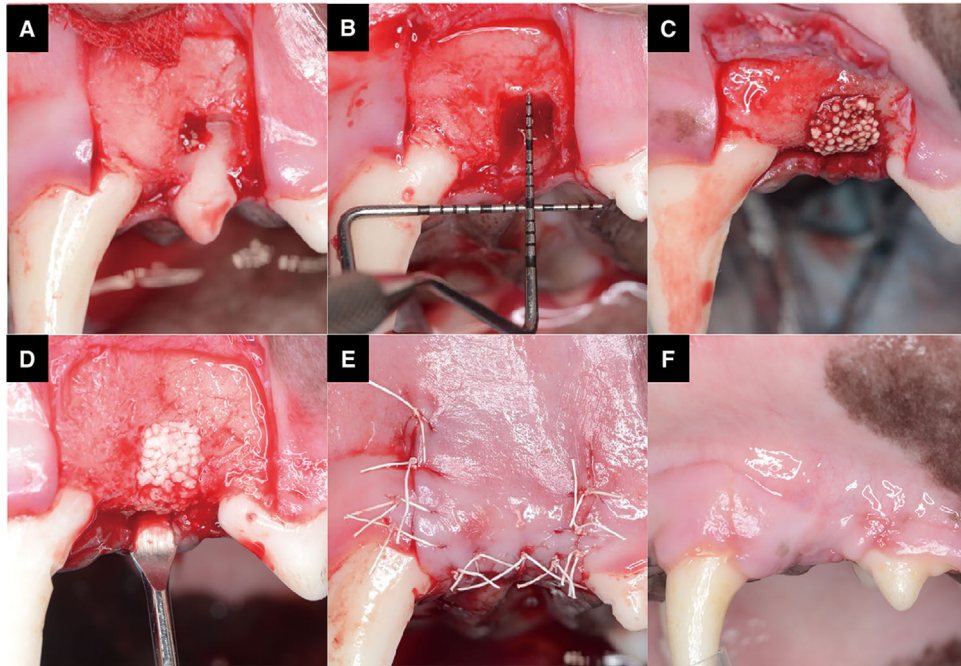* Cerasorb M, granule size 0.5–1 mm; Curasan, Kleinostheim, Germany

† easy-graft CLASSIC, granule size 0.5–1 mm; Degradable Solutions, Schlieren, Switzerland

‡ BioLinker; Degradable Solutions, Schlieren, Switzerland

§ Domitor; Orion, Espoo, Finland

¶ Ravonal; Mitsubishi Tanabe Pharmaceutical, Osaka, Japan

Xylocaine; Fujisawa Pharmaceutical, Osaka, Japan

FIGURE 1 Surgical procedures. **A**) Buccal bone defects were prepared on the first premolars of the maxilla using steel burs. **B**) Tooth extraction was performed carefully, and the bone defects were trimmed to a size of 4 × 4 × 5 mm (length × height × depth). **C**) Bilateral defects were randomly designated as test or control by tossing a coin. In the test group, the fresh extraction sockets with buccal bone defects were filled with a PLGA-coated β-TCP at the level of the adjacent bone. **D**) In contrast, in the control group, particulate β-TCP granules were placed in the extraction sockets. **E**) Coronal advancement of the flap and suturing. **F**) Healing at 12 weeks post-surgery. TCP = tricalcium phosphate; PLGA = poly lactide-co-glycolide

dog, an intracrevicular incision was made from the maxillary canine to the second premolar, and a mucoperiosteal flap was elevated. Buccal bone defects of size 4 × 5 mm (length × height) were created on the buccal side of the maxillary first premolar (P1) using a steel bur (Figure 1A). Then, the P1 was gently extracted, and the granulation tissue in the socket was completely curetted. Then, a fresh bone defect (4 × 4 × 5 mm [width × length × height]) with buccal deficiency was prepared (Figure 1B), and bilateral defects were randomly assigned as test (β-TCP/PLGA; Figure 1C) or control (particulate β-TCP; Figure 1D) sites by tossing a coin. The defects were filled with the bone grafts without using a membrane (Figures 1C and 1D). The flaps were then coronally advanced (because of relief incision in the periosteum) and sutured with interrupted sutures[*] (Figure 1E). Post-surgically, the dogs were administered an antibiotic[†] and analgesic agent,[‡] and a 2% solution of chlorhexidine gluconate[§] was applied to the surgical sites three times a week for 8 weeks. The sutures were removed 2 weeks after operation (Figure 1F). After a healing period of 12 weeks, the dogs were euthanized by sodium thiopental overdose. Subsequently, the maxillae were dissected and trimmed as block sections from the canine to the second premolar and immediately soaked in 10% neutral buffered formaldehyde.[¶]

2.3 | Radiographic analysis: microcomputed tomography

The specimens were imaged using a microcomputed tomography (micro-CT) system[#] at a voltage of 85 kV, electric current of 116 μA, voxel size of 0.0035 mm/voxel, and slice thickness of 0.1 mm. The scanned data were reconstructed to three- and two-dimensional images using an image-analysis software.[‖]

Following micro-CT analysis, a region of interest (ROI) bearing the same size as the original bone defect (5 × 4 × 4 mm [height × width × length]) was identified, and its position was determined in each, the axial, sagittal, and coronal planes. The horizontal position of the ROI was determined at the center between the maxillary canine and second premolar, and its vertical position was determined on the basis of a notch attached to the canine tooth during surgery. The volume and density of mineralized tissue formed in the ROI were

defined as bone volume (BV, mm^3) and bone material density (BMD, g/cm^3), respectively. Measurement of these radiographic parameters was performed as described in a previous study.[31]

2.4 | Histologic and histomorphometric analyses

The fixed specimens were decalcified using hydrochloric acid,* embedded in paraffin,† and then sliced into 5-μm-thick sections‡ in the buccopalatal plane. The histologic section of the most central portion of the defect, identified using micro-CT images, was selected, stained with hematoxylin and eosin, and then subjected to histologic investigation under a light microscope.§ The width of the alveolar ridge in the coronal (1 mm), middle (3 mm), and apical (5 mm) positions from the top of the palatal plate was measured histologically. The ROI was of the same size as the original bone defect (5 × 4 mm [height × length]). Measurement of these histologic parameters was performed with reference to previous studies.[7,32] Using an image-processing software,¶ bone formation in the ROI—including mineralized bone formation (woven bone, parallel fibered bone, and lamellar bone), bone marrow (intraosseous space mainly occupied by adipocytes), residual β-TCP (grafted β-TCP particles), and connective tissue (mainly collagen fibers but also elastic and reticular fibers)—was measured in each specimen.[22,33]

2.5 | Statistical analysis

Data are shown as mean and standard deviation. Differences between the test and control groups were assessed with the paired *t* test, and a *P* value <0.05 was considered statistically significant.# Sample size was determined by using previous studies[6,7] as a reference. Power calculation was performed by assuming a mean intergroup difference of 1.0 mm in the horizontal width and of 0.5 mm in the standard deviation to attain a power of 0.8 and significance level of 0.05.

3 | RESULTS

3.1 | Clinical observations

There were no clinical signs of infection or wound dehiscence at the test or control sites during the healing period.
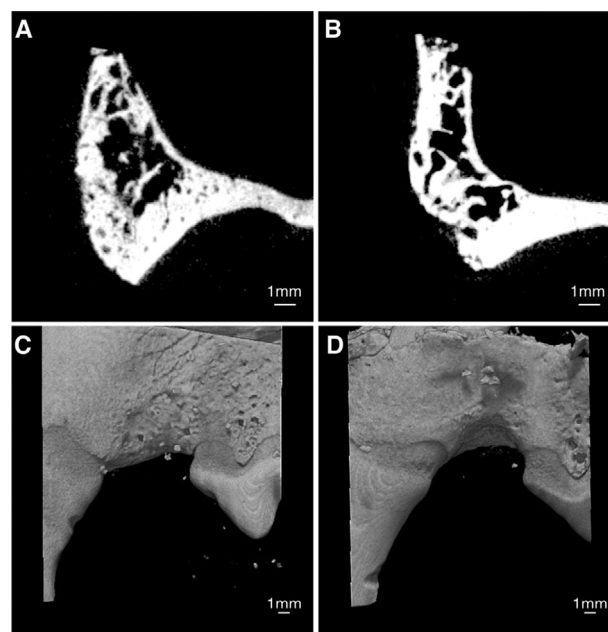
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* K-CX; Falma, Tokyo, Japan

† Tissue-TEKVIP5Jr; Sakura Finetek Japan, Tokyo, Japan

‡ AP 300-3; Micro International, Walldorf, Germany

§ ECLIPSE Ni-U; Nikon, Tokyo, Japan; DP70, Olympus, Tokyo, Japan

¶ Image J v.1.50i; National Institutes of Health, Bethesda, MD

Microsoft Excel v.2011; Microsoft, Redmond, WA

FIGURE 2 Two-dimensional micro-CT buccopalatal images of (**A**) β-TCP/PLGA and (**B**) β-TCP sites. Three-dimensional micro-CT images of (**C**) β-TCP/PLGA, and (**D**) β-TCP sites. CT = computed tomography; TCP = tricalcium phosphate; PLGA = poly lactide-co-glycolide. Scale bars = 1 mm

TABLE 1 Results of measurement of bone volume and mineral density

| Radiographic parameters	β-TCP/PLGA (n = 6)	β-TCP (n = 6)
Bone volume (mm^3)	25.7 ± 2.14*	16.0 ± 3.3
Bone density (mg/cm^3)	746.3 ± 23.9	714.5 ± 37.0

Data are presented as mean ± SD.

*P <0.05, statistically significant difference (Student *t* test; n = 6).

3.2 | Micro-CT findings

Two- and three-dimensional micro-CT images of the alveolar ridge revealed new bone formation and residual granules at the test sites (Figures 2A and 2C). In contrast, the control sites exhibited residual large bone defects and relatively less new bone formation (Figures 2B and 2D). The mean BV at the test sites (25.7 ± 2.14 mm^3) was significantly greater than that at the control sites (16.0 ± 3.3 mm^3). However, there was no significant difference in BMD between the test (746.3 ± 23.9 mg/cm^3) and control (714.5 ± 37.0 mg/cm^3) sites (Table 1).

3.3 | Histologic observations

Histologic analysis of the buccopalatal section revealed the new bone formation observed in the buccal area of the test sites (Figure 3A). At the control sites, the depressed contours of the buccal bone at the coronal and middle areas exhibited
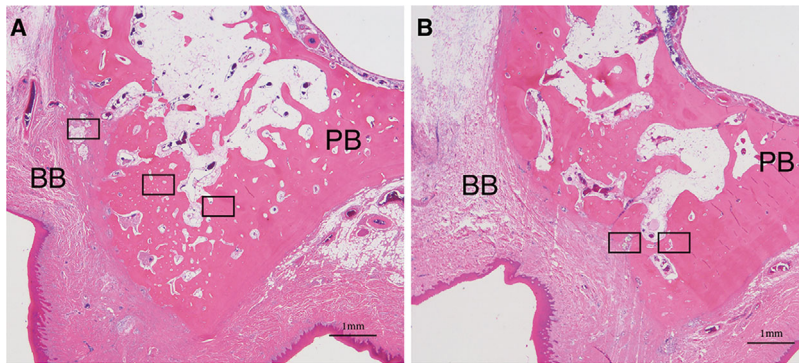
FIGURE 3 Microphotograph of a buccopalatal section. New bone formation continuous with the original bone was observed at the (**A**) test and (**B**) control sites. Although histological findings revealed some residual β-TCP encapsulated in connective tissue at the test sites, the buccal area at the control sites exhibited an evidently greater amount of connective tissue. BB = buccal bone wall; PB = palatal bone wall. Scale bars = 1 mm (original magnification)

limited new bone formation and large connective tissues (Figure 3B). Although, the test sites exhibited some residual β-TCP encapsulated in connective tissue at the apical area, the control sites exhibited an evidently greater amount of connective tissue in the buccal area. Both test and control sites showed continuous bone formation from the original bone (Figures 4A and 4D). Furthermore, mature bone formation represented by the appearance of lamellar bone with woven bone and angiogenesis was observed at the middle portion of the new bone in the test sites (Figures 4B and 4E). Although both test and control sites showed residual β-TCP granules encapsulated in connective tissue (Figures 4C and 4F), the amount of connective tissue was significantly greater in the control sites than in the test sites.

3.4 | Histomorphometric findings

The coronal and middle horizontal widths of new bone formations were significantly greater in the test sites (2.0 ± 0.4 and 2.9 ± 0.2 mm, respectively) than in the control sites (1.1 ± 0.3 and 2.1 ± 0.3 mm, respectively; Table 2). At the

TABLE 2 Results of linear measurement of horizontal width (mm) of the alveolar ridge and histometric results (%)

Parameters	β-TCP/PLGA (n = 6)	β-TCP (n = 6)
Horizontal widths (mm)		
Coronal (1 mm)	2.0 ± 0.4*	1.1 ± 0.3
Middle (3 mm)	2.9 ± 0.2*	2.1 ± 0.3
Apical (5 mm)	3.4 ± 0.5	2.6 ± 1.0
Region of composition (%)		
Mineralized bone	50.3 ± 8.1*	38.0 ± 5.2
Bone marrow	17.7 ± 6.6*	9.7 ± 4.1
Residual β-TCP	2.7 ± 2.0	2.5 ± 3.0
Connective tissue	10.7 ± 4.5	18.3 ± 5.7*

Data are presented as mean $\pm$ SD.

*P <0.05, statistically significant difference (Student t test; n = 6).

apical position, there was no significant difference in the horizontal width between the test and control sites. With regard to histologic measurements acquired at the ROI, the proportions of mineralized bone area and bone marrow were
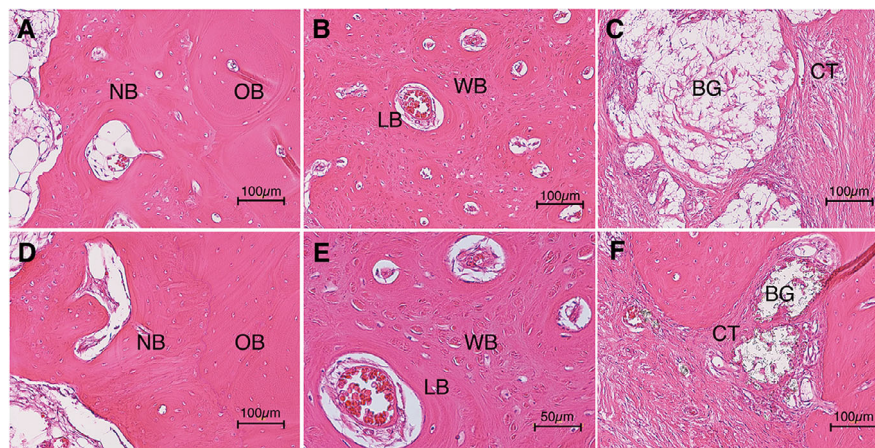
FIGURE 4 Higher magnification of the boxed areas of the boundary between the original and newly formed bones (**A** and **D**) and the boxed areas adjacent to the middle of the buccal bone (**B** and **E**) in Figures 3A and 3B. Mature bone formation represented by the appearance of LB with WB and angiogenesis was observed at the middle of the new bone (**B** and **E**). Although residual BG encapsulated in CT were observed both in the test and control sites (**C** and **F**), the amount of connective tissue was significantly greater in the control group than in the test group. NB = new formed bone; OB = original bone; LB = lamellar bone; WB = woven bone; BG = β-tricalcium phosphate granules; CT = connective tissue. (**A–D**, and **F**) original magnification × 20; scale bars = 100 μm; (**E**) original magnification × 40; scale bars = 50 μm

significantly greater at the test sites (50.3% ± 8.1% and 17.7% ± 6.6%, respectively) than at the control sites (38.0% ± 5.2% and 9.7% ± 4.1%, respectively). There were no statistically significant intergroup differences in residual β-TCP. However, the amount of connective tissue at the control sites (18.3% ± 5.7%) was significantly greater than at the test sites (10.7% ± 4.5%).

4 | DISCUSSION

In the present study, the utility of β-TCP/PLGA for alveolar ridge preservation in extraction sockets with buccal bone defect is compared to that of conventional particulate β-TCP. To the best of our knowledge, no animal histologic or histometric research for comparative evaluation of the effectiveness of β-TCP/PLGA and particulate β-TCP for alveolar ridge preservation in extraction sockets with buccal bone deficiency has been conducted to date. These injectable, moldable, and in-situ-hardening β-TCP particles have been shown to improve clinical handling and form a stable body in the extraction socket, thus serving as solid scaffolds for ridge preservation. Furthermore, stabilization and immobilization of particulate β-TCP at the graft site might help prevent micromovement between the original bone and implanted material, which is known to promote ingrowth of fibrous tissue and obstruct new bone formation at defect sites.[34] PLGA is a biodegradable scaffold commonly used for tissue healing; it degrades by hydrolysis within a few weeks and is known to be biocompatible.[35] On the other hand, it has been speculated that the PLGA coating might inhibit bone regeneration at graft sites.[36] A previous study had reported that grafted PLGA could cause inflammatory responses by virtue of accumulation of lactic acid and glycolic acid produced by hydrolysis.[37] Furthermore, a previous study that used β-TCP/PLGA grafts in a large calvarial-defect model reported inflammatory reaction and limited new bone formation.[38] Although it has not been elucidated why the implanted PLGA causes inflammation at the graft site, some studies have reported no inflammatory response around β-TCP/PLGA grafts in rat calvarial[30] and sheep sinus models.[36] In the present study, no significant inflammatory response was observed around the β-TCP/PLGA grafts, and histologic findings revealed mature bone formation at the test sites. The European Chemicals Agency (ECHA) has amended the risk assessment for mixtures containing NMP at concentrations ≥0.3% and classified them as having reproductive toxicity.[39] Although repeated oral exposure to NMP has been reported to induce a systemic effect that causes a decrease in body weight and food consumption, NMP exhibits low acute toxicity with oral exposure.[40] NMP and its resorption products are excreted primarily through urine in 1 to 3 days.[41] Because of its high metabolic rate, a single administration of NMP seems to

have little cellular toxicity. Moreover, after the application of β-TCP/PLGA to defect sites, NMP is flushed out upon contact with body fluids, which suggests that NMP might have a short-term cytotoxic effect. Although our histological and micro-CT findings demonstrated enhanced bone regeneration at the β-TCP/PLGA sites, we were not able to verity the biosafety of this graft because of the long observation period required. Further research is required to elucidate the effect of PLGA and its byproducts on bone formation.

Conversely, other studies have reported that NMP possesses various bioactivities favoring osteoblastic differentiation and bone regeneration.[42,43] A previous in vitro study reported that NMP acts as an enhancer of bone morphogenetic protein in vitro.[42] It also inhibits osteoclast differentiation and attenuates bone resorption.[44] Therefore, NMP might potentially be useful for prevention of dimensional changes in the alveolar ridge after tooth extraction. Micro-CT findings in the present study showed that the BV at the test sites (25.7 ± 2.14 mm^3) was significantly greater than that at the control sites (17.2 ± 1.33 mm^3). However, there was no significant difference in BMD between the test (746.3 ± 23.9 mg/cm^3) and control (714.5 ± 37.0 mg/cm^3) sites. These positive outcomes suggest that the aggregated and hardened β-TCP/PLGA maintains its shape and secures the regenerative space. Furthermore, the hardened β-TCP/PLGA graft also enlarged the osteoconductive area. Although the PLGA around β-TCP has been reported to be resorbed in ≈ 3 to 4 weeks,[27,45] in the present study, the β-TCP/PLGA grafts were resorbed and replaced with new bone at rates similar to those of particulate β-TCP. Histologic analysis revealed that the new bone formation was found to be continuous with the original bone at both the test and control sites in this study. Some residual β-TCP was encapsulated in connective tissue at the test sites. A previous study using a rat calvaria-defect model[30] reported large remnants of β-TCP/PLGA and β-TCP grafts at 6 weeks post-surgery. Although the present study involved a different animal and experimental site, the grafts were gradually resorbed and replaced with newly formed and mature bone, including osteocyte-like cells, at 12 weeks post-surgery. In the β-TCP/PLGA group, some particles were encapsulated in connective tissue because of delayed dissolution and resorption of PLGA.

With regard to histometric measurements in the present study, the horizontal width of the newly formed bone and the amounts of mineralized bone and bone marrow within the ROI were significantly greater at the test sites than at the control sites. On the basis of these results, it appeared that β-TCP/PLGA is effective for alveolar ridge preservation after tooth extraction with buccal bone deficiency, and mature bone formation was observed at areas distant from the original bone at 12 weeks post-surgery.

In another animal study, the effectiveness of β-TCP/PLGA for ridge preservation in four-wall sockets was compared

with that of a no-graft treatment in a swine model at 12 weeks post-surgery.[46] The authors found no significant difference in change in ridge width at 12 weeks post-surgery between β-TCP/PLGA and no-graft sites, although the β-TCP/PLGA sites showed a smaller decrease in mean horizontal alveolar ridge width than the latter (7.69% and 8.86%, respectively). Furthermore, histomorphometric findings revealed pronounced bone regeneration at both treatment sites. This lack of a statistically significant difference between the two groups might be attributed to the high regenerative potential of the four-wall extraction sockets. In contrast, in the present study, we used an extraction socket with buccal bone deficiency, which might be more sensitive to any difference between treatment methods employed after ridge preservation.

In our previous study, the efficiency of tunnel β-TCP blocks for ridge preservation in extraction sockets with buccal bone deficiency was compared with that in the absence of grafts at 8 weeks post-surgery.[7] The tunnel β-TCP block exhibited a greater volume of new bone formation and horizontal width at the coronal site. Hardened bone grafts such as block-type grafts have a greater potential for bone regeneration than particulate bone grafts.[47] β-TCP/PLGA is easily moldable and hardens rapidly. The merit of block-type grafts is also indicated in β-TCP/PLGA grafts. However, a substantial amount of β-TCP block remained at the graft site at 8 weeks post-surgery in our previous study, which presented the possibility of reduction in horizontal width with resorption and replacement of the graft over a longer observation period. Furthermore, in our previous study, histological findings revealed a greater volume of immature bone formation at both test and control sites at 8 weeks post-surgery. The present findings showed greater amounts of bone marrow and smaller amounts of residual β-TCP at the test sites than at the control sites. This suggests that the present study was able to demonstrate mature bone formation at 12 weeks post-surgery.

Jensen et al.[48] demonstrated that β-TCP granules are resorbed almost completely within 8 weeks after grafting. In the present study, histologic findings showed that residual β-TCP particles were almost completely resorbed in the mineralized bone area. However, previous studies have reported that most morphologic changes occur at 3 months post-surgery.[3,49] Schropp et al.[3] reported that loss of the crestal bone height and width of the alveolar ridge occurs mainly during the first 3 months after single or multiple tooth extraction and that the extent of loss remains almost unchanged from 3 to 12 months post-surgery. Furthermore, a recent review comparatively evaluated the effect of ridge preservation through socket filling following tooth extraction and that of tooth extraction alone on preserving the alveolar ridge dimensions after a minimum healing period of 12 weeks.[50] For a preclinical study, a 3-month observation period is appropriate for confirming the effect of alveolar ridge preservation.

The present study employed particulate β-TCP without a barrier membrane as a control group to verify the self-hardening potential of β-TCP/PLGA. Because of the lack of sufficient defect sites, we were not able to include other treatment groups such as barrier membranes with or without particulate β-TCP. The use of membranes with particulate grafts is common in clinical practice and has been proven to be effective for ridge preservation after tooth extraction.[26] Furthermore, the efficacy of this treatment in preventing midbuccal and midlingual ridge loss has also been confirmed in a recent review.[50] Additional studies are needed to evaluate the efficacy of β-TCP/PLGA relative to that of particulate β-TCP with a barrier membrane.

5 | CONCLUSIONS

Within the limitations of this study, the moldable β-TCP/PLGA graft appears to be more effective than a conventional particulate β-TCP graft for ridge preservation and minimization of linear and volumetric changes after tooth extraction in sockets with buccal bone deficiency.

REFERENCES

1. Pietrokovski J, Massler M. Alveolar ridge resorption following tooth extraction. *J Prosthet Dent*. 1967;17:21-27.

2. Araújo MG, Lindhe J. Dimensional ridge alterations following tooth extraction. An experimental study in the dog. *J Clin Periodontol*. 2005;32:212-218.

3. Schropp L, Wenzel A, Kostopoulos L, Karring T. Bone healing and soft tissue contour changes following single-tooth extraction: a clinical and radiographic 12-month prospective study. *Int J Periodontics Restorative Dent*. 2003;23:313-323.

4. Tan WL, Wong TL, Wong MC, Lang NP. A systematic review of post-extractional alveolar hard and soft tissue dimensional changes in humans. *Clin Oral Implants Res*. 2012;23(Suppl 5):1-21.

5. Pinho MN, Roriz VL. Titanium membranes in prevention of alveolar collapse after tooth extraction. *Implant Dent*. 2006;15:53-61.

6. Inomata K, Marukawa E, Takahashi Y, Omura K. The effect of covering materials with an open wound in alveolar ridge augmentation using beta-tricalcium phosphate: an experimental study in the dog. *Int J Oral Maxillofac Implants*. 2012;27:1413-1421.

7. Ikawa T, Akizuki T, Matsuura T, et al. Ridge preservation after tooth extraction with buccal bone plate deficiency using tunnel structured beta-tricalcium phosphate blocks: a 2-month histologic pilot study in beagle dogs. *J Periodontol*. 2016;87:175-183.

8. Darby I, Chen ST, Buser D. Ridge preservation techniques for implant therapy. *Int J Oral Maxillofac Implants*. 2009;24(Suppl): 260-271.

9. Vignoletti F, Matesanz P, Rodrigo D, Figuero E, Martin C, Sanz M. Surgical protocols for ridge preservation after tooth extraction. A systematic review. *Clin Oral Implants Res*. 2012;23(Suppl 5):22-38.

10. Denissen H, Montanari C, Martinetti R, van Lingen A, van den Hooff A. Alveolar bone response to submerged bisphosphonate-complexed hydroxyapatite implants. *J Periodontol*. 2000;71:279-286.

11. Araújo MG, Sukekava F, Wennström JL, Lindhe J. Ridge alterations following implant placement in fresh extraction sockets: an experimental study in the dog. *J Clin Periodontol*. 2005;32:645-652.

12. Sanz M, Cecchinato D, Ferrus J, Pjetursson EB, Lang NP, Lindhe J. A prospective, randomized-controlled clinical trial to evaluate bone preservation using implants with different geometry placed into extraction sockets in the maxilla. *Clin Oral Implants Res*. 2010;21:13-21.

13. Lekovic V, Kenney EB, Weinlaender M, et al. A bone regenerative approach to alveolar ridge maintenance following tooth extraction. Report of 10 cases. *J Periodontol*. 1997;68:563-570.

14. Chan HL, Lin GH, Fu JH, Wang HL. Alterations in bone quality after socket preservation with grafting materials: a systematic review. *Int J Oral Maxillofac Implants*. 2013;28:710-720.

15. Hjørting-Hansen E. Bone grafting to the jaws with special reference to reconstructive preprosthetic surgery. A historical review. *Mund Kiefer Gesichtschir*. 2002;6:6-14 (Article in German).

16. Damien CJ, Parsons JR. Bone graft and bone graft substitutes: a review of current technology and applications. *J Appl Biomater*. 1991;2:187-208.

17. Becker W, Becker BE, Caffesse R. A comparison of demineralized freeze-dried bone and autologous bone to induce bone formation in human extraction sockets. *J Periodontol*. 1994;65:1128-1133.

18. Iasella JM, Greenwell H, Miller RL, et al. Ridge preservation with freeze-dried bone allograft and a collagen membrane compared to extraction alone for implant site development: a clinical and histologic study in humans. *J Periodontol*. 2003;74:990-999.

19. Nevins M, Camelo M, De Paoli S, et al. A study of the fate of the buccal wall of extraction sockets of teeth with prominent roots. *Int J Periodontics Restorative Dent*. 2006;26:19-29.

20. Barone A, Aldini NN, Fini M, Giardino R, Calvo Guirado JL, Covani U. Xenograft versus extraction alone for ridge preservation after tooth removal: a clinical and histomorphometric study. *J Periodontol*. 2008;79:1370-1377.

21. Mangano C, Piattelli A, Perrotti V, Iezzi G. Dense hydroxyapatite inserted into postextraction sockets: a histologic and histomorphometric 20-year case report. *J Periodontol*. 2008;79:929-933.

22. Araujo MG, Liljenberg B, Lindhe J. Beta-Tricalcium phosphate in the early phase of socket healing: an experimental study in the dog. *Clin Oral Implants Res*. 2010;21:445-454.

23. LeGeros RZ. Calcium phosphate-based osteoinductive materials. *Chem Rev*. 2008;108:4742-4753.

24. Artzi Z, Weinreb M, Givol N, et al. Biomaterial resorption rate and healing site morphology of inorganic bovine bone and beta-tricalcium phosphate in the canine: a 24-month longitudinal histologic study and morphometric analysis. *Int J Oral Maxillofac Implants*. 2004;19:357-368.

25. Brkovic BM, Prasad HS, Konandreas G, et al. Simple preservation of a maxillary extraction socket using beta-tricalcium phosphate with type I collagen: preliminary clinical and histomorphometric observations. *J Can Dent Assoc*. 2008;74:523-528.

26. Melcher AH. On the repair potential of periodontal tissues. *J Periodontol*. 1976;47:256-260.

27. Ruffieux K, Kohli M, Benner K. Principles/material science/ handling. In: Ruffieux K, Kohli M, eds. *Bone Augmentation*. 1st ed. Schlieren, Switzerland: DS Dental; 2010:36.

28. Leventis MD, Fairbairn P, Kakar A, et al. Minimally invasive alveolar ridge preservation utilizing an in situ hardening beta-tricalcium phosphate bone substitute: a multicenter case series. *Int J Dent*. 2016;2016:5406736.

29. Leventis MD, Fairbairn P, Horowitz RA. Extraction site preservation using an in-situ hardening alloplastic bone graft substitute. *Compend Contin Educ Dent*. 2014;35:11-13.

30. Bizenjima T, Takeuchi T, Seshima F, Saito A. Effect of poly (lactide-co-glycolide) (PLGA)-coated beta-tricalcium phosphate on the healing of rat calvarial bone defects: a comparative study with pure-phase beta-tricalcium phosphate. *Clin Oral Implants Res*. 2016;27:1360-1367.

31. Nooh N, Abdullah WA, El-Awady Grawish M, et al. Evaluation of bone regenerative capacity following distraction osteogenesis of goat mandibles using two different bone cutting techniques. *J Craniomaxillofac Surg*. 2014;42:255-261.

32. Araujo M, Linder E, Wennstrom J, Lindhe J. The influence of Bio-Oss Collagen on healing of an extraction socket: an experimental study in the dog. *Int J Periodontics Restorative Dent*. 2008;28:123-135.

33. Cardaropoli G, Araujo M, Lindhe J. Dynamics of bone tissue formation in tooth extraction sites. An experimental study in dogs. *J Clin Periodontol*. 2003;30:809-818.

34. Wang HL, Boyapati L. PASS" principles for predictable bone regeneration. *Implant Dent*. 2006;15:8-17.

35. Patrick CW Jr., Chauvin PB, Hobley J, Reece GP. Preadipocyte seeded PLGA scaffolds for adipose tissue engineering. *Tissue Eng*. 1999;5:139-151.

36. Wildburger A, Bubalo V, Magyar M, et al. Sinus floor augmentation comparing an in situ hardening biphasic calcium phosphate (hydroxyapatite/β-tricalcium phosphate) bone graft substitute with a particulate biphasic calcium phosphate (hydroxyapatite/β-tricalcium phosphate) bone graft substitute: an experimental study in sheep. *Tissue Eng Part C Methods*. 2017;23:404-411.

37. Zhu H, Yang F, Tang B, et al. Mesenchymal stem cells attenuated PLGA-induced inflammatory responses by inhibiting host DC maturation and function. *Biomaterials*. 2015;53:688-698.

38. Bojar W, Kucharska M, Ciach T, Kopersk L, Jastrzebski Z, Szałwiński M. Bone regeneration potential of the new chitosan-based alloplastic biomaterial. *J Biomater Appl*. 2014;28: 1060-1068.

39. European Chemicals Agency. Background document to the opinion on the Annex XV dossier proposing restrictions on 1-methyl-2-pyrrolidone (NMP). Available at: https://echa.europa.eu/documents/10162/a3e8fb17-d794-493c-a5ac-8cce41def0c7. Accessed October 9, 2014.

40. Organisation for Economic Co-operation and Development. 1-methyl-2-pyrrolidone. SIDS initial assessment report for SIAM 24. Available at: https://hpvchemicals.oecd.org/ui/handler.axd?id=84daa4ac-feb7-4b5a-9839-206d17914e42. Accessed April 19–20, 2007.

41. Bader MWR, Blaszkewicz M, van Thriel C. Human experimental exposure study on the uptake and urinary elimination of N-methyl-2-pyrrolidone (NMP) during simulated workplace conditions. *Arch Toxicol*. 2007;81:335-346.

42. Miguel BS, Ghayor C, Ehrbar M, et al. N-methyl pyrrolidone as a potent bone morphogenetic protein enhancer for bone tissue regeneration. *Tissue Eng Part A*. 2009;15:2955-2963.

43. Gjoksi B, Ghayor C, Siegenthaler B, Ruangsawasdi N, Zenobi-Wong M, Weber FE. The epigenetically active small chemical N-methyl pyrrolidone (NMP) prevents estrogen depletion induced osteoporosis. *Bone*. 2015;78:114-121.

44. Ghayor C, Correro RM, Lange K, Karfeld-Sulzer LS, Grätz KW, Weber FE. Inhibition of osteoclast differentiation and bone resorption by N-methylpyrrolidone. *J Biol Chem*. 2011;286:24458-24466.

45. Friedrich D. Die Behandlung von grossen parodontalen defekten mit einem β-TCP composite. *Dent Implantol*. 2009;13:258-262.

46. Leventis M, Agrogiannis G, Fairbairn P, et al. Evaluation of an in situ hardening β-tricalcium phosphate graft material for alveolar ridge preservation. A histomorphometric animal study in pigs. *Dent J (Basel)*. 2018;6:27.

47. Yoo HS, Bae JH, Kim SE, et al. The effect of bisphasic calcium phosphate block bone graft materials with polysaccharides on bone regeneration. *Materials (Basel)*. 2017;10:17.

48. Jensen SS, Broggini N, Hjørting-Hansen E, Schenk R, Buser D. Bone healing and graft resorption of autograft, anorganic bovine bone and beta-tricalcium phosphate. A histologic and histomorphometric study in the mandibles of minipigs. *Clin Oral Implants Res*. 2006;17:237-243.

49. Aimetti M, Romano F, Griga FB, Godio L. Clinical and histologic healing of human extraction sockets filled with calcium sulfate. *Int J Oral Maxillofac Implants*. 2009;24:902-909.

50. Avila-Ortiz G, Elangovan S, Kramer KW, Blanchette D, Dawson DV. Effect of alveolar ridge preservation after tooth extraction: a systematic review and meta-analysis. *J Dent Res*. 2014;93: 950-958.

SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of the article.