Alveolar Ridge Preservation Prior to Implant Placement with Surgical-Grade Calcium Sulfate and Platelet-rich Plasma: A Pilot Study in a Canine Model

Bin Shi, PhD, DDS[1]/Yi Zhou, MD, DDS[2]/Yi Ning Wang, PhD, DDS[3]/Xiang Rong Cheng, BD, DDS[4]

Purpose: *To evaluate the combination of surgical-grade calcium sulfate (SGCS) and platelet-rich plasma (PRP) for alveolar ridge preservation prior to implant placement.* ***Materials and Methods:*** *Five mongrel dogs were used as subjects. Four enlarged mandibular extraction sockets, 2 on each side, were created in each dog. According to a split-mouth design, the 2 anterior sockets received either SGCS/PRP (SGCS/PRP$_{ant}$) or were left unfilled, while the 2 posterior sockets received either SGCS/PRP (SGCS/PRP$_{post}$) or SGCS. Computerized tomographic (CT) scans were conducted at 1 day and 8 weeks postextraction to detect the change in ridge height. Bone scintigraphy was performed at 2, 4, and 6 weeks to investigate new bone formation activity. At 8 weeks, 1 dog was sacrificed for histologic and histomorphometric study. Meanwhile, implants were placed in the remaining 4 dogs. These 4 dogs were sacrificed after 3 months.* ***Results:*** *Less ridge resorption was observed in the anterior SGCS/PRP–filled sites compared to unfilled sites (*P = *.001), while no significant difference was found between the SGCS/PRP$_{post}$ and SGCS groups (*P = *.544). Bone scintigraphy showed that sites filled with SGCS/PRP showed significantly higher count/pixel at 2 (*P = *.028), 4 (*P = *.009), and 6 weeks (*P = *.037) than the unfilled sites. Nevertheless, the SGCS/PRP$_{post}$ group achieved significantly higher values than the SGSC group only at 2 weeks (*P = *.036). Histomorphometrically, the SGCS/PRP$_{ant}$ group showed a significantly higher percentage of bone-implant contact than the unfilled group (*P = *.024), but no significant difference was detected between the SGCS/PRP$_{post}$ and SGCS groups (*P = *.979).* ***Conclusion:*** *Grafting SGCS/PRP in fresh extraction sockets reduced alveolar ridge resorption and promoted the bone formation in this canine model. The addition of PRP to SGCS resulted in the enhancement of bone regeneration in the early phase of healing.* (Pilot Clinical Trial) (More than 50 references) Int J Oral Maxillofac Implants 2007;22:656–665

Key words: *alveolar ridge preservation, calcium sulfate, dental implants, platelet-rich plasma, Tc-99m-MDP*

[1]Assistant Professor and Vice Director, Key Laboratory of Oral Biomedical Engineering of Ministry of Education, School of Stomatology, Wuhan University, Wuhan City, People's Republic of China; Department of Prosthodontics, Hospital of Stomatology, Wuhan University, Wuhan City, People's Republic of China.
[2]PhD Candidate, Key Laboratory of Oral Biomedical Engineering of Ministry of Education, School of Stomatology, Wuhan University, Wuhan City, People's Republic of China.
[3]Professor and Director, Department of Prosthodontics, Hospital of Stomatology, Wuhan University, Wuhan City, People's Republic of China.
[4]Professor, Department of Prosthodontics, Hospital of Stomatology, Wuhan University, Wuhan City, People's Republic of China; Key Laboratory of Oral Biomedical Engineering of Ministry of Education, School of Stomatology, Wuhan University, Wuhan City, People's Republic of China.

Correspondence to: Dr Xiang Rong Cheng, Department of Prosthodontics, Hospital of Stomatology, LuoYu Road 237, Wuhan City, HuBei Province, PR China. Fax: +86 027 87873260. E-mail: xiangrongcheng@hotmail.com.

Bin Shi and Yi Zhou contributed equally to this article.

Tooth extraction is usually followed by partial resorption of the residual alveolar ridge. Approximately 0.34 to 7.7 mm of horizontal ridge reduction and 0.2 to 3.25 mm of vertical ridge reduction occurs in the 6- to 12-month period following extraction, primarily in the initial 3 months.[1,2] Where postextraction bone resorption has occurred, it may be difficult to place an implant in an anatomically appropriate location or to achieve a satisfactory esthetic outcome[3] and long-term functional stability of the implant-supported restoration.[4] Furthermore, greater bone contour changes take place at multiple adjacent extraction sites than at a single extraction site.[2,5,6] Preservation of the alveolar ridge may be needed to optimize the success of implant placement in terms of both esthetics and function.

Bone grafting with synthetic dense hydroxyapatite (HA) has demonstrated clinical effectiveness for long-term ridge preservation.[7–9] The high modulus of elasticity of dense HA is not suitable for placement

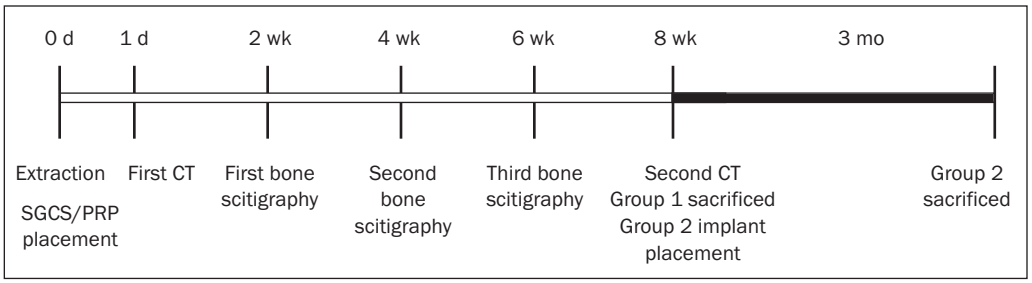

Fig 1 The timeline of the experiment. CT = computerized tomography.

in sites where implants are planned,[10] especially for delayed implantation several weeks postextraction. Resorption speed and ability to promote bone formation, among other factors, must be considered, because the goal of the grafting procedure is to increase the amount of living bone surrounding the implant at the time of insertion.

Calcium sulfate (CS), also known as plaster of paris, has been used to fill bone defects for approximately 100 years. It is a biocompatible, osteoconductive, and completely resorbable biomaterial which can accelerate osseogenesis.[11–13] It was observed histologically that medical-grade CS was completely resorbed within 3 months in human fresh extraction sockets and that CS did not interfere with socket healing.[14] Surgical-grade CS (SGCS) is a pharmaceutically prepared product with an alpha-crystal structure, as opposed to the CS used historically, such as medical-grade CS, which has a beta-crystal structure. SGCS is purer and more uniform than medical-grade CS, and it is designed to be resorbed at a more predictable rate.[12]

Platelet-rich plasma (PRP), a volume of autogenous plasma with concentrated platelets, has been used for enhancing bone regeneration in bone defects.[15,16] It was observed in a recent study that bone cell proliferation increased when CS was used as a carrier for PRP in vitro.[17] An in vivo experiment also found that placement of a mixture of particulate dentin, medical grade CS, and PRP promoted new bone formation in bone defects around implants.[18]

Although SGCS and PRP have been used successfully in various bone defects, no study using this combination in extraction sockets has been reported. It is unclear whether this combination can preserve the alveolar ridge and promote bone formation in fresh extraction sockets. The proposed hypotheses for the current pilot study were as follows:

1. For delayed implant placement, the use of an SGCS/PRP combination in fresh extraction sockets would decrease alveolar ridge resorption and enhance bone formation.
2. The addition of PRP to SGCS would result in better bone regeneration effects than SGCS alone.

MATERIALS AND METHODS

The research protocol was approved by the Ethics Committee for Animal Research, Wuhan University, China. This experimental study was designed as a pilot study employing 5 male mongrel dogs 1.5 to 2 years old and 19 to 21 kg. The animals were maintained on a soft diet throughout the study. Figure 1 shows the timeline of the experiment. Four enlarged mandibular extraction sockets were created in each dog (Fig 2). According to the split-mouth design, the 4 sockets were divided into 2 pairs, anterior and posterior. SGCS/PRP was placed into 1 of the anterior sockets, while the other socket was left unfilled as a control. SGCS or SGCS/PRP was placed in the 2 posterior sockets. Each dog was subjected to a computerized tomographic (CT) scan at 1 day and 8 weeks and to bone scintigraphy at 2, 4, and 6 weeks after surgery. One dog was selected to be sacrificed at 8 weeks for histologic and histomorphometric studies. The other four dogs were subjected to implant placement surgery. Forty-eight custom-made mini-implants were placed in the edentulous sites, with 3 implants in each site. Three months postplacement, the 4 dogs were sacrificed for histologic and histomorphometric studies.

The CS used in this study was a surgical-grade product (Osteoset, Wright Medical Technology, Arlington, TN) supplied in pellet form. Through milling, sifting, and sterilizing, SGCS particles with a diameter of 0.15 to 1.0 mm were prepared. Ti-Mo-Zr-Al alloy rods (Northwest Institute for Non-ferrous Metal Research, Xi'an, ShanXi, China) were machined with high-precision equipment to create parallel-sided mini-implants 2.8 mm in diameter and 5 mm in length with a hemisphere-shaped apex. The surface of implants was treated according to the method described by Wang[19] as follows: *(1)* ultrasonic bathing in pure water (Millipore, Bedford, MA), acetone, and ethanol for 15 minutes each; *(2)* washing 3 times with Millipore pure water; *(3)* passivating in 50% nitric acid for 15 minutes; *(4)* washing 10 times in Millipore pure water; and (5) sterilizing with autoclaving.

In a modification of the method used by Kim et al,[18] 18 mL venous blood was drawn from each ani-

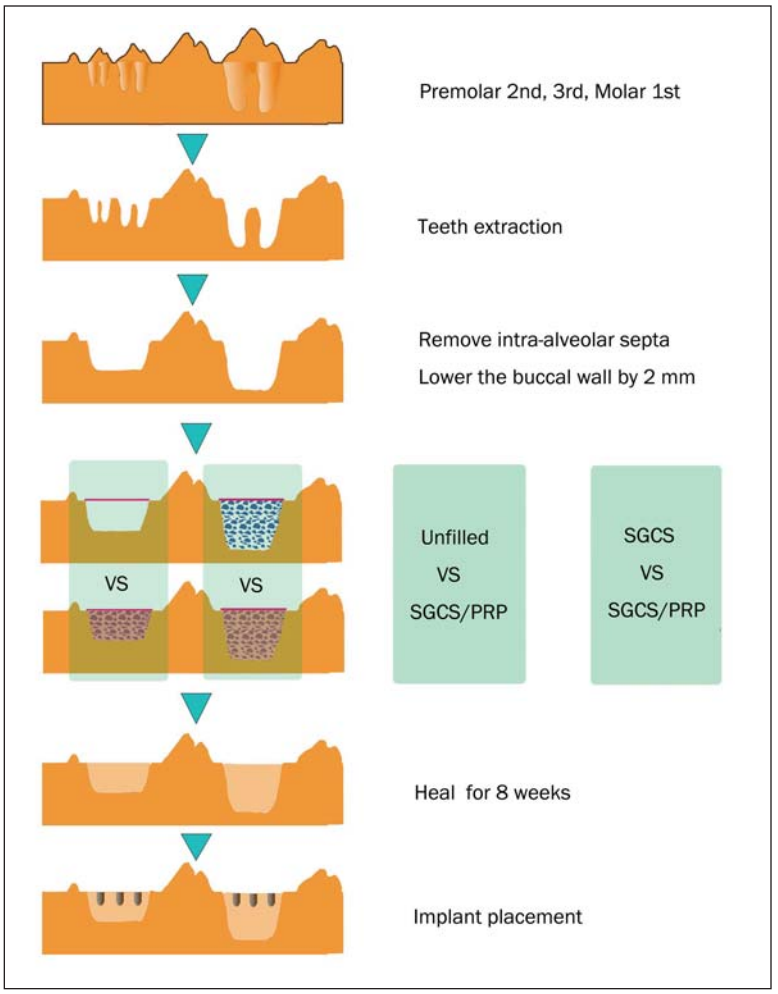

Fig 2 A review of the surgical procedures.

mal using 6-mL sterile vacuum tubes (JinXing Medical, Wuhan, China) containing anticoagulant citrate dextrose-a (ACD-a). The whole blood was centrifuged with a double-spin method at 1,000 rpm (201 g) for 15 minutes and 2,000 rpm (805 g) for 20 minutes at room temperature to prepare PRP. SGCS particles, PRP, 10% calcium chloride, and 300 units of bovine thrombin were then mixed. The concentrations of platelets in the whole blood and PRP were assayed manually and automatically with a blood cell counter.

During surgery, the animals were anesthetized with intramuscularly administered ketamine (2.5 mg/kg, Hengrui Pharmaceutical, Nanjin, Jiangsu, China) and atropine (0.04 mg/kg, Xuzhou Ryen Pharmaceutical, Xuzhou, Jiangsu, China) and a 4% solution of intravenously administered pentobarbital sodium (25 to 30 mg/kg, PentothalA, Hengrui Pharmaceutical). Sulcular incisions were made, with subsequent reflection of full mucoperiosteal flaps. According to the methods described by Verstraete[20] and Gauthier et al,[21] the second and third mandibular premolars and the first mandibular molar were carefully removed. Subsequently, the sockets were prepared by a modified alveolectomy in which the intra-alveolar and interradicular septa were removed and the buccal wall of the socket was reduced by 2 mm. As a result, 4 enlarged extraction sockets with buccal dehiscences were created. According to the split-mouth design, the resulting anterior sockets were either filled with mixture of SGCS and PRP ($SGCS/PRP_{ant}$ group) or left empty (unfilled group) as a control, while the posterior sockets were filled with SGCS and PRP ($SGCS/PRP_{post}$ group) or with SGCS alone (SGCS group). Primary tension-free wound closure was accomplished by advancing the mucoperiosteal flaps using periosteal releasing incisions and suturing with interrupted and mattress sutures.

Antibiotic therapy (ampicillin sodium; 25 mg/kg intramuscularly daily) was administered for 5 days, and the sutures were removed at 15 days postsurgery. Eight weeks after the first operation, 1 dog (group 1) was selected to be sacrificed. At the same time, the other 4 dogs (group 2) underwent the second operation, during which 48 implants were placed in the middle of healed sockets in the buccolingual direction, with 3 implants in each socket. The space between implants and between implants and adjacent teeth was 4 mm. A review of the surgical procedures is shown in Fig 2.

CT scans of the mandible of each dog were obtained at 1 day and at 8 weeks postextraction with a spiral CT device (PQ6000; Picker International, Highland Heights, OH), and the data were transferred to a workstation (Voxel Q; Picker, International). The axial scans were reformatted to produce sections in the coronal plane. On these coronal planes, the distance from the middle point of the tops of the cortical walls to the inferior border was measured. The mean of these measurements was used as the alveolar ridge height value. Then the change of alveolar ridge height was obtained by comparing the 2 alveolar ridge height values of the same dog 1 day and 8 weeks postsurgery.

Bone scintigraphy was applied in all 5 animals at 2, 4, and 6 weeks postextraction to compare the new bone formation activities among different sockets. Each dog was given an intravenous dose of 0.33mCi/kg technetium-99m-methylene diphosphonate (Tc-99m-MDP, China Institute of Atomic Energy, Beijing, China). Three hours later, the animal was anesthetized and fastened to the examining table. A gamma camera fitted with high-resolution parallel collimators (Diacam; Siemens, Erlangen, Germany) was rotated under the mandible of the animal. The head of the dog was positioned so that the inferior margins of the mandible paralleled the detector. To block the radiation emitted from the maxillary tissue, a lead plate was inserted between the maxillary and mandibular teeth. The scintigraphic images of the mandibular bone were obtained using a high-resolution collimator, and the data were processed with a processing system (ICON Siemens, Siemens, Germany). The areas of the 4 sockets and the mandibular symphysis were chosen as regions of interest, and the values were reported as mean counts/pixel (ie, the volume of Tc accumulation per pixel). To compare individual differences and observe the change of bone metabolism activity during the healing period, the uptake volume of Tc-99m-MDP of the 4 sockets was calculated as a ratio: the mean counts/pixel of the individual socket divided by that of the mandibular symphysis.

Sectioned blocks of the mandible from group 1 were fixed in 4% neutral buffered paraformaldehyde, decalcified in 10% neutral buffered EDTA, dehydrated in a graded series of ethanols, and embedded in wax. Five-micron sections were made in a buccolingual plane and stained with hematoxylin-eosin.

The 4 animals in group 2 were sacrificed after the 3-month postimplantation healing period. Tissue blocks with implants were fixed and dehydrated as described. They were subsequently infiltrated and embedded in methylmethacrylate. The bone blocks were axially sectioned into 50-μm-thick sections in the anterior-posterior direction on a slow-speed diamond saw (Leica SP1600; Leica, Milan, Italy), and eventually 3 sections of the center part of an implant were stained with methylene blue.

Histologic analyses were performed using a microscope equipped with an image system (Q-500 MCA; Leica, Germany). Photographs of the specimens were obtained, and histomorphometric analysis was performed using a computer-based NIH image analysis system, Image J (downloaded from http://rsb.info.nih.gov/ij/). All parameters were named and assessed according to the guidelines of the Nomenclature Committee of the American Society of Bone and Mineral Research.[22] Bone volume/tissue volume (BV/TV) was assessed for group 1. The percentage of bone-implant contact (BIC) was assessed for group 2.[23]

Statistical Analysis

Paired *t* tests were used to evaluate differences between groups, and $P < .05$ was considered the level of significance. The statistical analysis was performed with SPSS 11.5 for Windows (SPSS, Chicago, IL).

RESULTS

The average peripheral blood platelet count was 3.17 $\times$ 10^9/L, with the range of 2.08 to 4.45 $\times$ 10^9/L. The average platelet count in PRP was 14.20 $\times$ 10^9/L, with a range of 9.26 to 19.67 $\times$ 10^9/L (Fig 3). The SGCS particles formed a cohesive mass when mixed with PRP, which allowed easy manipulation and packing into the extraction sockets. All animals made a rapid postoperative recovery, and the wound healing was uneventful. At the overview of the control extraction sites, there were obvious depressions at the unfilled extraction sites, while the tops of the treated sockets were flat or slightly protuberant (Fig 4). At the time of euthanization of the animals in group 2, all implants appeared clinically immobile.

All interdental edentulous ridges showed some degree of resorption. The difference between the $SGCS/PRP_{ant}$ (1.39 ± 0.38 mm) and unfilled group (2.77

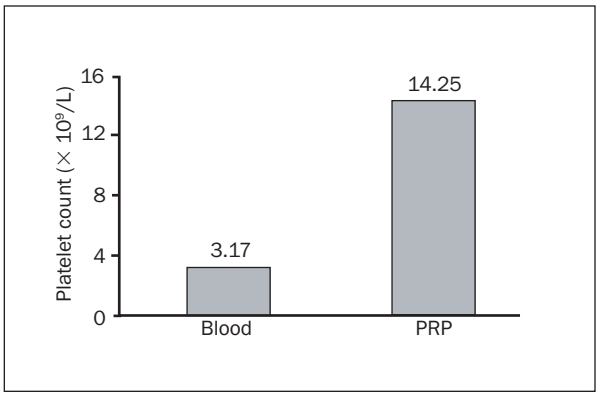

Fig 3 Average platelet counts in peripheral blood and PRP. Platelet counts confirmed that PRP preparation concentrated platelets.

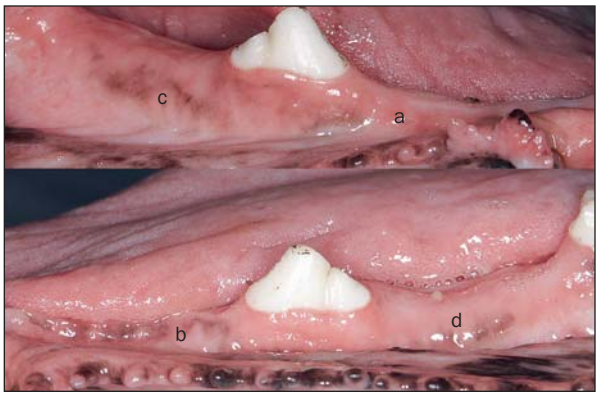

Fig 4 Extraction sockets 8 weeks after surgery. There were obvious depressions at the unfilled extraction sites, while the tops of the treated sockets were flat or slightly protuberant. *(a)* SGCS/PRP_{ant} group, *(b)* unfilled group, *(c)* SGCS/PRP_{post} group, *(d)* SGCS group.

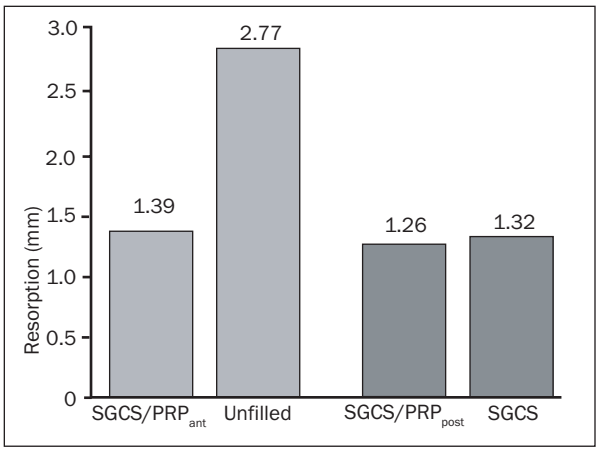

Fig 5 Reduction of alveolar ridges as detected using CT scans.

± 0.30 mm) was significant ($P = .001$), but no significant difference was detected between the SGCS/PRP_{post} (1.26 ± 0.27 mm) and SGCS (1.32 ± 0.34 mm) groups ($P = .544$; Fig 5). Tc-99m-MDP accumulation at the extraction sites was detected 2, 4, and 6 weeks postoperatively (Fig 6). Compared to the unfilled group, the SGCS/PRP_{ant} group showed higher ratio of counts/pixel at 2 ($P = .028$), 4 ($P = .009$), and 6 weeks ($P = .037$) weeks postsurgery. The SGCS/PRP_{post} group showed higher ratio of counts/pixel than the SGCS group at 2 weeks after surgery ($P = .036$), while no marked difference was noted at 4 ($P = .095$) or 6 weeks ($P = .089$) postoperatively (Table 1).

In group 1, the control extraction sockets were filled with newly formed bone after 8 weeks. This bone was mainly composed of woven bone with some lamellar bone. A large number of primary osteons and some secondary osteons were observed. There appeared to be more newly formed bone and bone bridges in the middle and apical areas of the sockets in the SGCS/PRP_{ant} group than in the unfilled group. Compared with the SGCS group, more and thicker bone trabeculae were found in the SGCS/PRP_{post} -treated sockets, especially in the middle and apical parts of the sockets. No remnants of the SGCS particles were observed in any of the treated sockets, indicating that the implanted SGCS was completely resorbed within 8 weeks (Fig 7).

In group 2, the bone-implant interface had mineralized bone matrix in intimate contact with the implant surface in all groups (Fig 8). The bone tissue was characterized by concentric or parallel lamellar bone formation. Under the light microscopy, the difference in BIC between the SGCS/PRP_{ant} and unfilled groups was easy to distinguish, and the implants placed in SGCS/PRP- and SGCS-treated sockets were covered by new bone. However, no apparent difference in BIC or bone density was observed between the SGCS/PRP_{post} and the SGCS groups.

In group 1, the percentage of BV/TV at 8 weeks was 51.88% in the SGCS/PRP_{ant} group compared with 42.32% in the unfilled group and 61.19% in the SGCS/PRP_{post} group compared with 52.61% in the SGCS group. The differences were not statistically significant. In group 2, a statistically higher BIC was detected in the SGCS/PRP_{ant} group than in the unfilled group ($P = .024$). No significant difference was observed between the SGCS/PRP_{post} group and the SGCS group ($P = .979$).

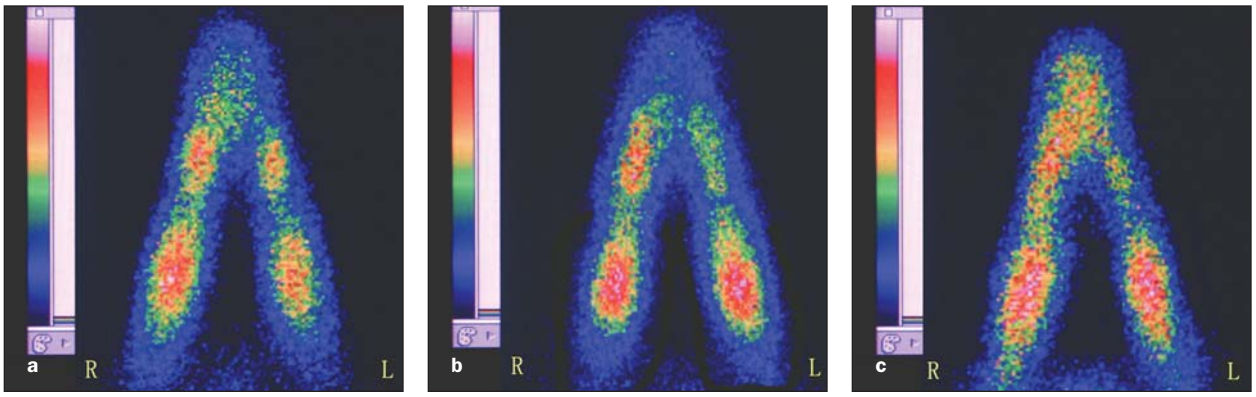

Fig 6 Bone scintigraphy images of the same dog at *(a)* 2 weeks, *(b)* 4 weeks, and *(c)* 6 weeks postsurgery. The pink/red end of the color reference bar indicates the greatest level of Tc-99m-MDP accumulation.

Table 1 Mean Ratios (± SD) of Counts/Pixel at 2, 4, and 6 Weeks Postsurgery

	Anterior			Posterior		
	SGCS/PRP$_{ant}$ (n = 5)	Unfilled (n = 5)	*P*	SGCS/PRP$_{post}$ (n = 5)	SGCS (n = 5)	*P*
2 wk	3.04 ± 0.74	1.51 ± 0.72	.028*	3.44 ± 1.09	1.91 ± 0.56	.036*
4 wk	4.33 ± 1.02	2.26 ± 0.85	.009*	4.93 ± 1.39	4.09 ± 1.23	.095
6 wk	3.52 ± 1.12	1.42 ± 0.72	.037*	3.72 ± 1.32	3.12 ± 1.39	.089

*Statistically significant.

DISCUSSION

Although there has been increasing interest in immediate implant placement,[24] it has been reported that such placement may be adversely affected by the presence of infection,[25–27] lack of soft tissue closure,[28] and defects between the bone and implants. Therefore, delayed implant placement is still one of the main options for clinicians. It is recognized, however, that residual ridge resorption following the tooth extraction is unavoidable, especially in cases where there are multiple adjacent extraction sites. Greater bone contour changes take place in such situations than at single extraction sites.[2,5,6]

In addition to guided bone regeneration, fresh extraction socket grafting has been investigated for alveolar ridge preservation. To avoid the need for a second surgical site, the efficacy of bone substitutes such as allografts, xenografts, and synthetic graft materials has been investigated.[10] The present study was conducted to test a synthetic graft material, SGCS, in combination with PRP for used with delayed implantation 8 weeks after tooth extraction in a canine model.

The CT scans indicated that the reduction of alveolar bone height following tooth extraction was decreased in the SGCS/PRP–treated sockets in comparison to the sockets allowed to heal naturally. The fact that no difference in wound healing was observed between sites treated with SGCS/PRP and those treated with SGCS alone indicates that the alveolar ridge preservation demonstrated may have been largely because of the SGCS rather than the PRP.

The reported resorption rate of common CS has ranged from 2 to 4 weeks in dog alveolar sites.[29–31] Cardaropoli and colleagues[32] carried out a dynamic histologic observation of socket healing to ascertain the change of new bone volume in the extraction sockets in a canine model. They found that mineralized bone was first seen on day 1, and that mineralized bone volume in sockets peaked on day 30, occupying 88% of the socket volume. The value was 23% at 60 days and 37% at 90 days. In the present study,

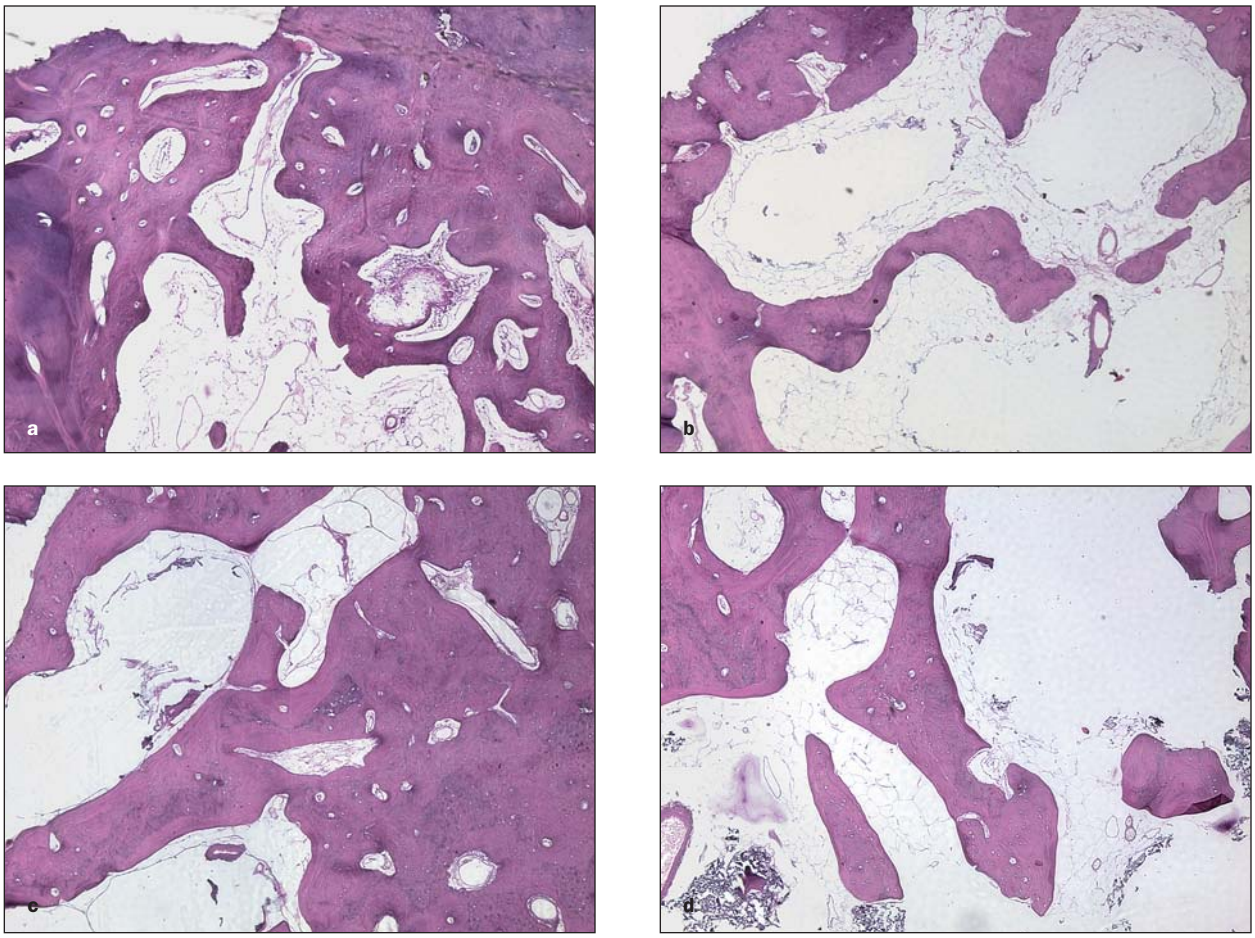

Fig 7 Histologic appearance of group 1 at 8 weeks after tooth extraction. *(a)* SGCS/PRP_{ant}, *(b)* unfilled, *(c)* SGCS/PRP_{post}, *(d)* SGCS (hematoxylin-eosin, original magnification ×10).

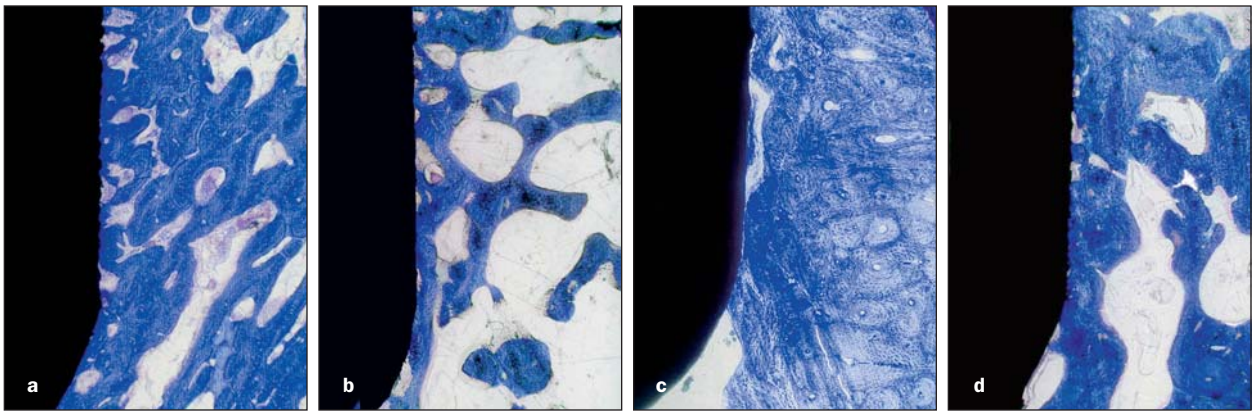

Fig 8 Longitudinal sections through implants placed in *(a)* SGCS/PRP_{ant}, *(b)* unfilled, *(c)* SGCS/PRP_{post}, and *(d)* SGCS 3 months after implantation (methylene blue, original magnification ×4).

SGCS placed with or without PRP was completely resorbed within 8 weeks postoperatively. The implantation of SGCS or SGCS/PRP did not appear to hamper bone formation in fresh sockets, and the rate of new bone formation was close to that of SGCS resorption. The healing of dog extraction sockets is faster than that of human one. For example, a 3-week-old human extraction wound was found to be histologically equivalent to a 9- or 10-day-old wound in dogs, and a 3-month-old human extraction wound was found to be equivalent to an 8-week-old counterpart in dogs.[33] Therefore, the resorption time of less than 8 weeks shown in the present dog model may be a good reflection of human bone healing in the first 3 months postsurgery.[22] It therefore may be reasonable to believe that SGCS could be resorbed completely in 3 months in human sockets, which is the routine period that a patient has to wait for delayed implant placement after tooth extraction.

The presence of vital bone with a sustained ability to remodel is essential to maintain osseointegration over time.[34] To evaluate the combined effects of SGCS and PRP on the enhancement of wound healing and osseogenesis in extraction sockets, Tc-99m-MDP was used in this study to trace bone formation in the healing sockets. Although the specific binding site of Tc-99m-MDP remains unknown, it is clear that Tc-99m-MDP is associated with areas of osteoblast activity, and bone growth as well as vascularization.[35,36] Therefore, it has been used frequently to examine the effects of grafts on the bone healing in vivo.[37,38] In the present study, it was demonstrated by a semiquantified analysis of bone scintigraphy that the region treated with SGCS/PRP had a higher Tc-99m accumulation than the natural healing site throughout the 6-week period. This result indicates SGCS/PRP may promote bone formation. Furthermore, the newly formed bone observed 8 weeks postextraction in the unfilled and SGCS-treated sockets showed histologically similar bone maturity. As the accumulation of Tc-99m-MDP is partly because of vascularization of the graft, the higher bone formation activity at the SGCS/PRP–treated sockets may attribute to enhancement of the vascularization at the extraction sockets. Strocchi and colleagues[39] investigated the microvessel density of bone defects treated with CS compared with defects grafted with autogenous bone. It was found that CS-treated sites had higher microvessel density than those treated with autogenous bone after 4 weeks of healing.

The use of PRP in extraction sockets was first reported by Anitua,[40] who found no negative effect when PRP was used in fresh sockets. Then an in vitro study showed that PRP carried with CS increased bone cell proliferation.[17] In this study, with the aid of bone scintigraphic semi-quantified evaluation, it was found that PRP promoted the bone regeneration in the early healing phase when combined with SGCS. These results are consistent with a study by Wiltfang et al,[41] who noted that PRP was able to enhance bone healing significantly at 2 weeks when applied in combination with autogenous bone, but not at 4 or 12 weeks. In another study, PRP was instilled into the host sites before the implants were placed, and the animals were sacrificed at 3, 6, and 12 weeks.[23] The histomorphometric evaluation showed significantly higher BIC at 3 and 6 weeks after PRP application but not at 12 weeks.

The effect of PRP on bone regeneration has demonstrated variability in the literature.[42,43] The reasons for these conflicting reports may be listed as follows. First, methodology is crucial in order to achieve a blood product with undamaged platelets. Jensen et al[44] tested the influence of PRP on the mechanical fixation of implants placed in conjunction with frozen bone allograft found no effect of PRP. However, EDTA was used as the anticoagulant for the PRP. EDTA is not a recommended anticoagulant for PRP preparation because of its ability to fragment platelets.[43] The PRP used in this experiment was prepared strictly according to the guidelines of Marx,[45] including the use of ACD-a as the anticoagulant and the use of freshly prepared PRP with a platelet concentration 5-fold greater than baseline. Second, PRP may execute its effects only in sites where sufficient osteogenic cells are present. In vitro studies showed that PRP could promote the proliferation and differentiation of bone marrow stromal cells and maintain the function of the differentiated osteoblast.[46–48] When PRP was implanted with autologous mesenchymal stem cells (MSCs) into canine mandible defects, bone density and BIC tended to be slightly higher in PRP/MSC-filled defects (79.4% and 58.6%) than in PRP-filled sites (68.2% and 44.2%).[49] In other studies, PRP was shown to significantly increase BIC when implants reached deeper into the cancellous bone of the mandible,[50] in contrast to implants only involving mandibular cortical bone.[23,51,52] Third, the conflicting results achieved with PRP may be due to the different grafts that have been combined with PRP. In an animal experiment, PRP was mixed with autogenous bone, tricalcium phosphate, anorganic bovine bone, or collagenous sponge, respectively, to treat the critical-size defects. A significant effect on bone regeneration was found in the autogenous group compared with other groups.[41] Thus it seems that PRP, prepared correctly, can be effective when it is combined with certain biomaterials and placed into the bone defects at certain sites.

CONCLUSION

In a canine model, in comparison to untreated control sites, grafting the combination of SGCS and PRP in fresh extraction sockets reduced the resorption of the alveolar ridge and promoted bone formation in extraction sockets. The addition of PRP to SGCS resulted in the enhancement of bone regeneration only in the early phase of healing.

ACKNOWLEDGMENT

This work was supported by HuBei Provincial Key Technologies R&D Program (2004AA304B07), People's Republic of China.

REFERENCES

1. Chen ST, Wilson TG Jr, Hammerle CHF. Immediate or early placement of implants following tooth extraction: Review of biologic basis, clinical procedures, and outcomes. Int J Oral Maxillofac Implants 2004;19(suppl):12–25.
2. 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.
3. Belser UC, Buser D, Hess D, Schmid B, Bernard JP, Lang NP. Aesthetic implant restorations in partially edentulous patients: A critical appraisal. Periodontol 2000 1998;17:132–150.
4. Buser D, Dula K, Hirt HP, Schenk RK. Lateral ridge augmentation using autografts and barrier membranes: A clinical study with 40 partially edentulous patients. J Oral Maxillofac Surg 1996;54:420–432.
5. Lam RV. Contour changes of the alveolar processes following extraction. J Prosthet Dent 1960;10:25–32.
6. Johnson K. A study of the dimensional changes occurring in the maxilla following tooth extraction. Aust Dent J 1969;14: 241–244.
7. Kentros GA, Filler SJ, Rothstein SS. Six months evaluation of particulate Durapatite in extraction sockets for the preservation of the alveolar ridge. Implantologist 1985;3:53–62.
8. Bell DJ Jr. Particles versus solid forms of hydroxylapatite as a treatment modality to preserve residual alveolar ridges. J Prosthet Dent 1986;56:322–326.
9. Nemcovsky CE, Serfaty V. Alveolar ridge preservation following extraction of maxillary anterior teeth. Report on 23 consecutive cases. J Periodontol 1996;67:390–395.
10. Bartee BK. Extraction site reconstruction for alveolar ridge preservation. Part 1: Rationale and materials selection. J Oral Implantol 2001;27:187–193.
11. Walsh WR, Morberg P, Yu Y, et al. Response of a calcium sulfate bone graft substitute in a confined cancellous defect. Clin Orthop Relat Res 2003;406:228–236.
12. Kelly CM, Wilkins RM, Gitelis S, et al. The use of a surgical grade calcium sulfate as a bone graft substitute: Results of a multicenter trial. Clin Orthop Relat Res 2001;382:42–50.
13. Borrelli J, Prickett WD, Ricci WM. Treatment of nonunions and osseous defects with bone graft and calcium sulfate. Clin Orthop Relat Res 2003;411:245–254.
14. Guarnieri R, Pecora G, Fini M, et al. Medical grade calcium sulfate hemihydrate in healing of human extraction sockets: Clinical and histological observations at months. J Periodontol 2004;75:902–908.
15. Sánchez AR, Sheridan PJ, Kupp LI. Is Platelet-rich plasma the perfect enhancement factor? A current review. Int J Oral Maxillofac Implants 2003;18:93–103.
16. Marx RE. Platelet-rich plasma: Evidence to support its use. J Oral Maxillofac Surg 2004;62:489–496.
17. Intini G, Andreana S, Margarone JE III, Bush PJ, Dziak R. Engineering a bioactive matrix by modifications of calcium sulfate. Tissue Eng 2002;8:997–1008.
18. Kim SG, Chung CH, Kim YK, Park JC, Lim SC. Use of particulate dentin-plaster of paris combination with/without platelet-rich plasma in the treatment of bone defects around implants. Int J Oral Maxillofac Implants 2002;17:86–94.
19. Wang JW. The Study of Techniques of Anodic Oxidation and Hydroxyapatite Coating on the Surface of Pure Titanium [thesis; in Chinese]. Wuhan, China: Hubei Medical University, 2000: 39–40.
20. Verstraete FJ. Instrumentation and technique of permanent teeth in the dog. J S Afr Vet Assoc 1983;54:231–238.
21. Gauthier O, Boix D, Grimandi G, et al. A new injectable calcium phosphate biomaterial for immediate bone filling of extraction sockets: A preliminary study in dogs. J Periodontol 1999; 70:375–383.
22. Parfitt AM, Drezner MK, Glorieux FH, et al. Bone histomorphometry: Standardization of nomenclature, symbols, and units. Report of the ASBMR Histomorphometry Nomenclature Committee. J Bone Miner Res 1987;2:595–610.
23. Zechner W, Tangl S, Tepper G, et al. Influence of platelet-rich plasma on osseous healing of dental implants: A histologic and histomorphometric study in minipigs. Int J Oral Maxillfac Implants 2003;18:15–22.
24. Schwartz-Arad D, Chaushu G. The ways and wherefores of immediate placement of implants into fresh extraction sites: A literature review. J Periodontol 1997;68:915–923.
25. Rosenquist B, Grenthe B. Immediate placement of implants into extraction sockets: Implant survival. Int J Oral Maxillofac Implants 1996;11:205–209.
26. Polizzi G, Grunder U, Goene R, et al. Immediate and delayed implant placement into extraction sockets: A 5-year report. Clin Implant Dent Relat Res 2000;2:93–99.
27. Grunder U, Polizzi G, Goene R, et al. A 3-year prospective multicenter follow-up report on the immediate and delayed-immediate placement of implants. Int J Oral Maxillofac Implants 1999;14:210–216.
28. Wilson TG Jr, Weber HP. Classification of and therapy for areas of deficient bony housing prior to dental implant placement. Int J Periodontics Restorative Dent 1993;13:451–459.
29. Radentz WH, Collings CK. The implantation of plaster of paris in the alveolar process of the dog. J Periodontol 1965;36: 650–657.
30. Calhoun NR, Neiders ME, Greene GW. Effects of plaster-of-paris implants in surgical defects of mandibular alveolar processes of dogs. J Oral Surg 1967;25:122–128.
31. Petlier LF, Orn D. The effect of the addition of plaster of paris to autogenous and homogenous bone grafts in dogs. Surg Forum 1958;8:571–574.
32. 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.
33. Amler MH, Johnson PL, Salman I. Histological and histochemical investigation of human alveolar socket healing in undisturbed extraction wounds. J Am Dent Assoc 1960;61:32–44.

34. Garretto LP, Chen J, Parr JA, Roberts WE. Remodeling dynamics of bone supporting rigidly fixed titanium implants: A histomorphometric comparison in four species including humans. Implant Dent 1995;4:235–243.
35. Schuemichen C, Krause T, Umbach G, Wolff T. Localization of 99mTc-diphosphonates in newly formed bone matrix as a measure of bone lesion detectability. Nuklearmedizin 1988; 27:8–11.
36. Schwartz Z, Shani J, Soskolne WA, Touma H, Amir D, Sela J. Uptake and biodistribution of technetium-99m-MD32P during rat tibial bone repair. J Nuclear Med 1993;34:104–108.
37. Hervás I, Floria LM, Bello P, et al. Microvascularized fibular graft for mandibular reconstruction detection of viability by bone scintigraphy and SPECT. Clin Nuclear Med 2001;26:225–229.
38. McCracken M, Zinn K, Lemons JE, Thompson JA, Feldman D. Radioimaging of implants in rats using Tc-99m-MDP. Clin Oral Implants Res 2001;12:372–378.
39. Strocchi RS, Orsini G, Iezzi G, et al. Bone regeneration with calcium sulfate: Evidence for increased angiogenesis in rabbits. J Oral Implantol 2002;28:273–278.
40. Anitua E. Plasma rich in growth factors: Preliminary results of use in the preparation of future sites for implants. Int J Oral Maxillofac Implants 1999;14:529–535.
41. Wiltfang J, Kloss FR, Kessler P, et al. Effects of platelet-rich plasma on bone healing in combination with autogenous bone and bone substitutes in critical-size defects. An animal experiment. Clin Oral Implants Res 2004;15:187–193.
42. Zechner W, Tangl S, Tepper G, et al. Influence of platelet-rich plasma on osseous healing of dental implants: A histologic and histomorphometric study in minipigs. Int J Oral Maxillofac Implants 2003;18:15–22.
43. Freymiller EG, Aghaloo TL. Platelet-rich plasma: Ready or not? J Oral Maxillofac Surg 2004;62:484–488.
44. Jensen TB, Rahbek O, Overgaard S, Søalle K. Platelet rich plasma and fresh frozen bone allograft as enhancement of implant fixation: An experimental study in dogs. J Orthop Res 2004;22:653–658.
45. Marx RE. Discussion. J Oral Maxillofac Surg 2000, 58(3):300.
46. Marx RE. Platelet-rich plasma (PRP): What is PRP and what is not PRP? Implant Dent 2001;10:225–228.
47. Arpornmaeklong P, Kochel M, Depprich R, Kubler NR, Wurzler KK. Influence of platelet-rich plasma (PRP) on osteogenic differentiation of rat bone marrow stromal cells. An in vitro study. Int J Oral Maxillofac Surg 2004;33:60–70.
48. Gruber R, Varga F, Fischer MB, Watzek G. Platelets stimulate proliferation of bone cells: Involvement of platelet-derived growth factor, microparticles and membranes. Clin Oral Implants Res 2002;13:529–535.
49. Gruber R, Karreth F, Frommlet F, et al. Effects of platelet lysates on select bone cell functions. Clin Oral Implants Res 2004;15: 581–588.
50. Yamada Y, Ueda M, Naiki T, Nagasaka T. Tissue-engineered injectable bone regeneration for osseointegrated dental implants. Clin Oral Implants Res 2004;15;589–597.
51. Fuerst G, Gruber R, Tangl S, Sanroman F, Watzek G. Effects of fibrin sealant protein concentrate with and without platelet-released growth factors on bony healing of cortical mandibular defects. An experimental study in minipigs. Clin Oral Implants Res 2003;14:1–7.
52. Fuerst G, Gruber R, Tangl S, Sanroman F, Watzek G. Enhanced bone-to-implant contact by platelet-released growth factors in mandibular cortical bone: A histomorphometric study in minipigs. Int J Oral Maxillofac Implants 2003;18:685–690.