Platelet-Enriched Fibrin Glue and Platelet-rich Plasma in the Repair of Bone Defects Adjacent to Titanium Dental Implants

Tae-Min You, DDS[1]/Byung-Ho Choi, DDS, PhD[2]/Shi-Jiang Zhu, MD[3]/Jae-Hyung Jung, DDS[1]/Seoung-Ho Lee, DDS, PhD[4]/Jin-Young Huh, DDS[1]/Hyun-Jung Lee, DDS[1]/Jingxu Li, DDS[3]

Purpose: *The aim of this study was to compare the effects of platelet-enriched fibrin glue and platelet-rich plasma (PRP) on the repair of bone defects adjacent to titanium dental implants.* ***Materials and Methods:*** *In 6 mongrel dogs, 3 screw-shaped titanium dental implants per dog were placed into the osteotomy sites in the tibia. Before implantation, a standardized gap (2.0 mm) was created between the implant surface and the surrounding bone walls. Six gaps were left empty (control group), 6 gaps were filled with autogenous particulate bone mixed with PRP (PRP group), and 6 gaps were filled with autogenous particulate bone mixed with platelet-enriched fibrin glue (fibrin glue group).* ***Results:*** *After 6 weeks, the bone-implant contact was 59.7% in the fibrin glue group, 29.2% in the PRP group, and 10.2% in the control defects; this difference was statistically significant (*$P < .05$*).* ***Discussion and Conclusion:*** *Greater bone-implant contact was achieved with platelet-enriched fibrin glue than with PRP. The results indicate that platelet-enriched fibrin glue can induce a stronger peri-implant bone reaction than PRP in the treatment of bone defects adjacent to titanium dental implants.* Int J Oral Maxillofac Implants 2007;22:417–422

Key words: *bone grafting, bone regeneration, dental implants, fibrin glue, platelet-rich plasma*

Bone resorption after tooth extraction reduces the height and width of the alveolar crest, hindering the use of dental implants.[1,2] The placement of a dental implant immediately after tooth extraction has been recommended as a means to minimize bone loss and shorten the time of the prosthetic treatment.[3,4] Immediate implantation into fresh extraction sockets is often associated with a residual bone defect between the implant neck and the residual bone walls. As has been reported,[5,6] large gaps may jeopardize the success of immediate implant procedures. Such gaps may cause cell migration from the connective and epithelial tissue into the gap, which may prevent osseointegration. Various techniques, including the use of barrier membranes and grafting material,[7] have been proposed for the management of these defects. In particular, grafting material mixed with platelet-rich plasma (PRP) has been reported to enhance the osseointegration of dental implants,[8,9] as it contains large numbers of platelets which release significant quantities of growth factors known to promote wound healing.[9,10]

The idea of using platelet-enriched fibrin glue to fill bone defects is attractive because this glue contains many more platelets than PRP.[11] In addition, the high concentrations of fibrinogen in fibrin glue can produce a dense fibrin clot with sufficient adhesive strength to hold particulate bone in a configuration. For these reasons, platelet-enriched fibrin glue was considered for use in bone defects adjacent to titanium dental implants. The aim of this study was to compare the effects of both platelet-enriched fibrin glue and PRP on bone defect repair adjacent to tita-

[1]Graduate Student, Department of Oral & Maxillofacial Surgery, College of Dentistry, Yonsei University, Seoul, South Korea.
[2]Professor, Department of Oral & Maxillofacial Surgery, College of Dentistry, Yonsei University (Brain Korea 21 Project for Medical Sciences), Seoul, South Korea.
[3]Research Assistant, Department of Dentistry, Yonsei University Wonju College of Medicine, Wonju, South Korea.
[4]Associate Professor, Department of Periodontology, Ewha Womans University, Seoul, South Korea.

Correspondence to: Prof Byung-Ho Choi, Department of Oral & Maxillofacial Surgery, College of Dentistry, Yonsei University, 134 Shinchon-dong, Seodaemun-gu, 120-752 Seoul, South Korea. Fax: +0082 33 748 2025. E-mail: choibh@yonsei.ac.kr

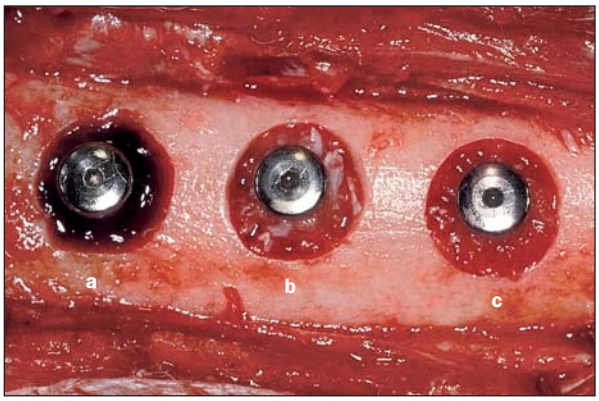


Fig 1 Experimental design. *(a)* The control defect remained empty. *(b)* The cortical defect was filled with autogenous particulate bone mixed with PRP. *(c)* The cortical defect was filled with autogenous particulate bone mixed with platelet-enriched fibrin glue.

nium dental implants in the dog tibia. The experimental model used was designed to simulate immediate implantation into fresh tooth extraction sockets.

MATERIALS AND METHODS

Animal Model

Six adult female mongrel dogs weighing more than 15 kg were used in this experiment. Approval was obtained from our animal care committee of Yonsei Medical Center, Seoul, Korea.

PRP Preparation

The PRP was prepared using a technique described previously.[12] Briefly, 20 mL of autologous blood withdrawn from each dog was initially centrifuged at 2,400 rpm for 10 minutes to separate the PRP and platelet-poor plasma (PPP) portions from the red blood cells. The PRP and PPP were again centrifuged at 3,600 rpm for 15 minutes to separate PRP from PPP. Platelet counts were then done for each dog, yielding a mean PRP platelet count of 1,490,000 (range, 1,020,000 to 2,140,000). The PRP was activated just before application with a 10% calcium chloride solution and 5,000 units of bovine thrombin to form a gel.

Platelet-Enriched Fibrin Glue Preparation

Platelet-enriched fibrin glue was prepared using a previously described technique.[11] Briefly, 20 mL of autologous blood withdrawn from each dog was treated using the aforementioned technique to separate PRP from the blood. The fibrinogen solution was subsequently prepared using the PRP. To 7.5 mL of PRP, 240 µL of transexamic acid and 900 µL of ethanol were added. This mixture was incubated in an ice-water bath for 20 to 30 minutes. The precipitated fibrinogen was separated by centrifugation at 3000 x g for 8 minutes at 0°C to 4°C. After discarding the supernatant, the fibrinogen precipitate was redissolved at 37°C and diluted to 50% with 0.9% sodium chloride (NaCl). A thrombin solution was then prepared using the remaining PRP. Briefly, 2.5 mL of PRP was added to 22.5 mL of citric acid, and the mixture was then centrifuged at 3000 x g for 5 minutes at 4°C. After discarding the supernatant, the precipitate was dissolved in 150 µL calcium chloride ($CaCl_2$; 0.1 mol/L), and the pH was adjusted to 7 by adding 100 µL of sodium hydrogen carbonate ($NaHCO_3$). After clot formation, the thrombin solution was collected and diluted to 10% with 0.05 mol/L $CaCl_2$. The fibrin glue was formed by mixing the fibrinogen and thrombin solutions in a 3:1 (vol/vol) ratio. Thorn et al[11] reported that autologous fibrin glue prepared by this technique contains high concentrations of platelets and fibrinogen.

Surgical Procedure

All surgical procedures were performed under systemic anesthesia (5 mg/kg ketamine and 2 mg/kg intramuscular xylazine) as well as local anesthesia (2% lidocaine with 1:80,000 epinephrine). The bone surface of the tibia was exposed by an incision made on the internal side of the tibia. Before implantation, corticocancellous bone grafts were harvested from 3 implant recipient sites using a trephine bur of 6.0 mm. Bone defects of 6.1 mm were created at each site by enlarging the upper aspect of the osteotomy site using a round bur. Implants 15 mm in length and 4.1 mm in diameter were then placed through both the defect and the lower cortical bone so that a standardized gap of 2.0 mm was created between the bony walls and the implant neck. In all, 18 implants (Osstem, Seoul, Korea) were placed (3 in each tibia). Subsequently, the obtained corticocancellous bone was ground in a bone mill (Stryker Leibinger, Freiburg, Germany) to a uniform particle size. This particulate bone was then used to fill the gap from which the bone had been obtained. The bone gaps were treated with 1 of the following 3 treatment modalities: *(1)* no treatment (control group), *(2)* grafting with particulate bone mixed with PRP (PRP group), or *(3)* grafting with particulate bone mixed with platelet-enriched fibrin glue (fibrin glue group) (Fig 1). All experimental areas were covered with the soft tissue flap after removal of the periosteum. Cefazolin (Choing Kun Dang, Seoul, South Korea) was administered 1 hour before surgery and once daily for 2 days following surgery.

Sample Preparation

Animals were sacrificed 6 weeks after surgery, and bone blocks with the implants were excised. Resected bone specimens were fixed in 10% buffered formalin and embedded in methylmethacrylate resin. The blocks were cut longitudinally through the middle plane of the implants. Histologic sections (40 μm) were prepared using a cutting-grinding method and were stained with toluidine blue.

Histomorphometry

A morphometric study using an image analysis system (IBAS; Contron, Erching, Germany) was used to quantify the newly formed bone around the implants. The bone-implant contact (BIC), defined as the length of bone surface border in direct contact divided by the implant perimeter (× 100%), was then calculated. BIC was measured at the upper cortical and medullary levels.

Statistical Analysis

The Wilcoxon signed rank test was used to calculate statistical differences between the 2 active treatments. *P* values less than .05 were considered significant.

RESULTS

No postoperative infections or loose implants were observed during the follow-up period.

In the control group, a fibrous membrane with fibers parallel to the implant surface was found in contact with the implant surface, and there was very little direct contact between the bone and the implant in the medullary and transcortical portions of the implant (Fig 2). In the PRP group, the newly formed bone was in contact with the implant surface, and no fibrous membrane was observed. New bone was formed largely at the interface between the implant interface and the upper cortical bone (Fig 3). Compared with the PRP group, the fibrin glue group showed more newly formed trabeculae around the implant in the upper cortical and medullary portions. These trabeculae were thicker than those in the PRP group (Fig 4).

The mean percentages of direct BIC in the 3 groups are shown in Table 1. The quantitative morphometric analysis showed significantly more BIC in the fibrin glue group ($P < .05$) than in the other 2 groups together. The BIC was 10.2% ± 1.9% for the control group, 29.2% ± 7.8% for the PRP group, and 59.7% ± 13.8% for the fibrin glue group.

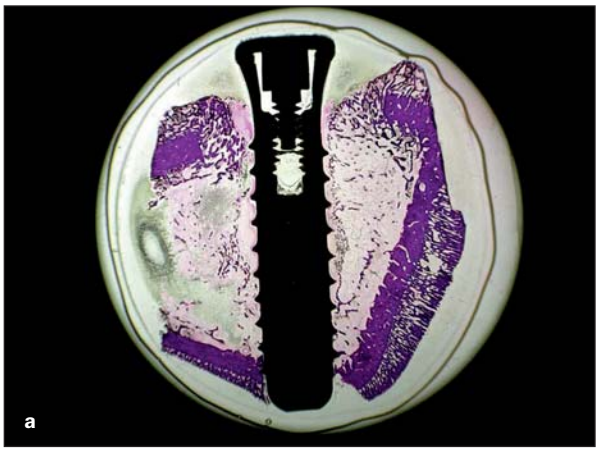
DISCUSSION

In recent years, several substances have been used to improve peri-implant bone response: PRP,[12–15] bone morphogenetic proteins,[16,17] and growth factors.[18–20] The use of PRP is based on the premise that the high quantity of platelets in PRP release significant amounts of growth factors that aid in bone graft maturation.[21] Growth factors released from the platelets have been shown to include platelet-derived growth factor, transforming growth factor-β, platelet-derived epidermal growth factor, platelet-derived angiogenesis factor, insulin-like growth factor 1, and platelet factor 4.[9,10] These factors signal the local mesenchymal and epithelial cells to migrate, divide, and increase collagen and matrix synthesis.[22] It has been suggested that PRP can be used to increase the rate of bone deposition and the quality of bone regeneration when augmenting sites prior to or in conjunction with dental implant placement.[22,23] PRP has also been recommended for use alone or in combination with bone grafts in the treatment of peri-implant defects that result as a consequence of immediate implant placement.[8,9]

The present study showed that when a combination of platelet-enriched fibrin glue and bone grafts was used in the treatment of bone defects adjacent to titanium dental implants, the BIC was significantly higher in the fibrin glue group than in the PRP group. This suggests that the platelet-enriched fibrin glue promotes a stronger peri-implant bone reaction than PRP, most likely due to the properties of the platelet-enriched fibrin glue. Thorn et al[11] reported that the concentration of fibrinogen in platelet-enriched fibrin glue was approximately 12 times that found in PRP and that the concentration of growth factors (as measured by platelet-derived growth factor) was approximately 8 times that in PRP. In the present study, platelet-enriched fibrin glue with high concentrations of platelets and fibrinogen was prepared using the technique described by Thorn et al.[11]

PRP and platelet-enriched fibrin glue mimic the last steps in the coagulation process (the conversion of fibrinogen to fibrin, aided by thrombin and calcium) by helping to cross-link the fibrin into a stable clot.[24] Theoretically, the fibrin clot can keep the platelets on location and serve as an osteoconductor.[25] The platelet-enriched fibrin glue in this study produced a denser fibrin clot than PRP, with more adhesive strength to hold particulate bone in a required configuration due to its high fibrinogen concentration.

Various membrane techniques originally developed for the treatment of bone loss due to periodontal disease have been used to promote bone forma-



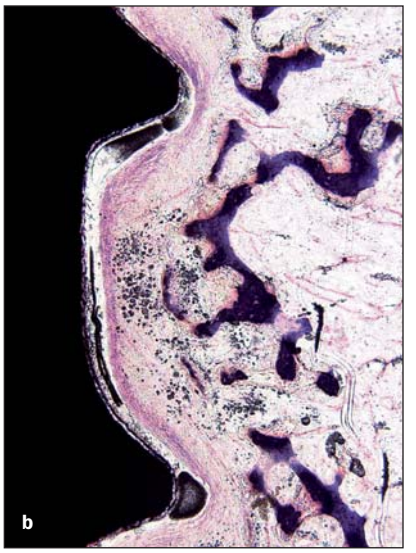
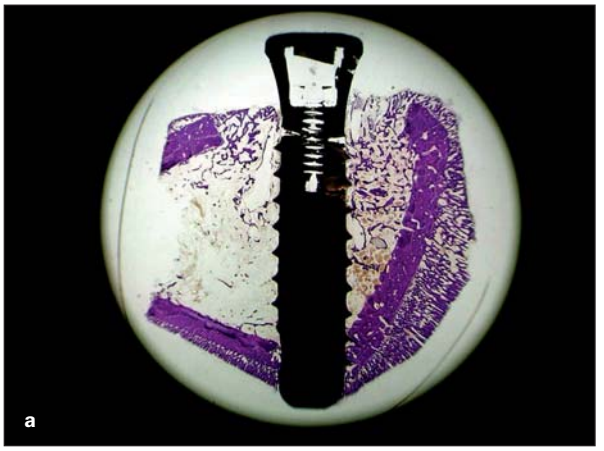


Fig 2 A section from the control group at original magnifications of *(a)* 5× and *(b)* 100× (toluidine blue). A fibrous membrane surrounds the implant surface, and no bone contacts the implant.



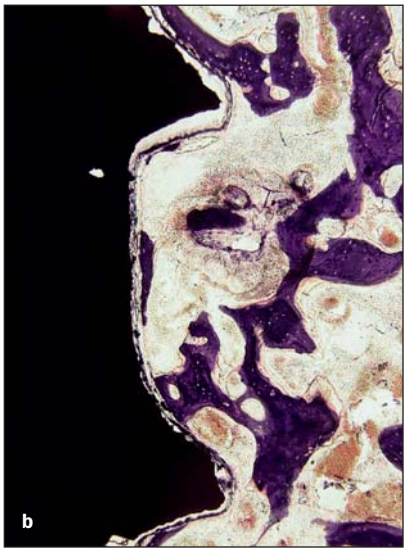
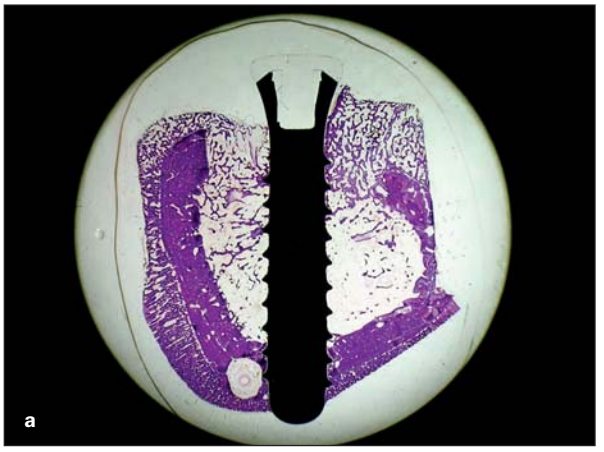


Fig 3 A section from the PRP group at original magnifications of *(a)* 5× and *(b)* 100× (toluidine blue). The newly formed bone contacts the implant surface; no fibrous membrane is seen.



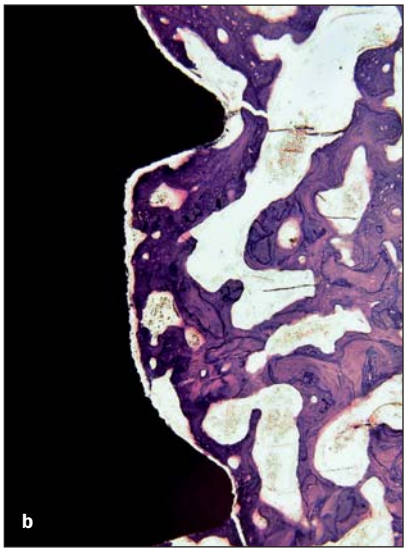


Fig 4 A section from the fibrin glue group at original magnifications of *(a)* 5× and *(b)* 100× (toluidine blue). The new trabeculae are thicker and better formed than in the PRP group.

Table 1 Percentage of BIC in the Examined Dog Tibiae

Dog number	Control group	PRP group	Fibrin glue group
1	8.1	29.6	33.9
2	10.3	30.2	59.0
3	12.7	34.4	62.8
4	10.1	40.1	67.5
5	8.1	21.7	60.5
6	11.7	19.3	74.2
Mean ± SD	10.2 ± 1.9	29.2 ± 7.8	59.7 ± 13.8

tion in the presence of large gaps (> 2 mm) between the bony walls and the implant neck.[5] However, inconsistent results have been achieved. Postoperative infections, membrane exposure to the oral cavity, and partial necrosis of the adjacent bone have been reported.[26] In this study, nongrafted gaps that did not have a barrier membrane showed a fibrous membrane surrounding the implant surface, whereas gaps filled with a bone graft mixed with either PRP or platelet-enriched fibrin glue showed bone in contact with implant surface. Based on these results, it is possible that either platelet-enriched fibrin glue or PRP could be used as an alternative to membranes to prevent connective and epithelial cells from migrating into the gaps.

CONCLUSION

Based on data presented in this study, it can be concluded that platelet-enriched fibrin glue can induce a stronger peri-implant bone reaction than PRP in the treatment of bone defects adjacent to titanium dental implants.

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