

Regeneration of Bone in Large Periapical Defect Associated with Maxillary Anterior Teeth after Endodontic Microsurgery Using Sticky Bone and Guided Tissue Regeneration Barrier Membrane – A Report of Two Cases

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Abstract

Endodontic surgery focuses on eliminating periapical pathology and achieving healing through the complete regeneration of bone and periodontal tissue. However, epithelial cells migrate at a rate ten times faster than the cells that form bone and periodontal tissue, making regeneration challenging, especially in large or through-and-through lesions where healing often occurs by repair rather than regeneration. To address this, a barrier membrane is placed over the defect site, and bone graft material, combined with various growth factors, is used to accelerate regenerative healing. The current report discusses two cases regarding the surgical management of large periapical lesions associated with maxillary anterior teeth. The treatment involved the combined use of a newly developed bone graft material called sticky bone and a guided tissue regeneration membrane to act as a barrier against unwanted tissue formation. Healing was evaluated using cone beam computed tomography (CBCT) scans, and patients were examined clinically and radiographically at 3-, 6-, and 12-month post-surgery. The patients remained asymptomatic, and CBCT scans showed a significant reduction in the size of the bony defects and an increase in bone density.

Keywords: Autologous fibrin glue, Barrier membrane, Guided tissue regeneration, Periapical surgery, Sticky bone.

INTRODUCTION

The periapical lesion is a sequel of endodontic infection. In most of the cases, the periapical lesion heals through mechanical debridement and with good chemical irrigation. However, in 10–15% of cases reported failure and may lead to the emergence of uncontrolled symptoms.^[1] Endodontic surgery is indicated for teeth with persistent periradicular pathoses that have not responded to nonsurgical approaches or when nonsurgical retreatment is impractical or unlikely to improve on the previous result.^[2]

Following endodontic surgery, periapical wound healing can be led toward repair or regeneration, depending on various factors such as the type and size of the lesion and the availability of cells from the host and biological factors potentially stimulating the wound healing process in the damaged area.^[3,4] The availability of cell types and the presence or absence of signals necessary to stimulate these cells decide the fate of injured tissues whether it is healed by regeneration

or repair.^[5] The incorporation of growth factors and barrier membranes in endodontic surgery increases the possibility of true regeneration by reconstructing the periapical tissue.^[6] True tissue regeneration is controlled by three things – stem cells, scaffold biomaterials, and growth factors.^[7]

Guided tissue regeneration (GTR) principles are implemented in endodontic surgery by the use of bioactive materials such as bone grafts and membranes, either alone or in combination. Their function is to encourage the growth of key surrounding

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tissues while excluding unwanted cell types such as epithelial cells from the area of regeneration. A new concept of fabricating growth factors enriched bone graft matrix (also known as “sticky bone”) using autologous fibrin glue (AFG) was introduced in 2010 by Sohn *et al.*^[8] Sticky bone provides stabilization of bone graft in the defect and therefore accelerates tissue healing and minimizes bone loss during the healing period.^[8] It contains a much longer, denser, richer fibrin matrix enriched with growth factors than platelet-rich-fibrin (PRF) and has both osteoinductive and osteoconductive character.

Barrier membranes are inert materials that maintain a confined space, which is one of the key biological requirements for bone regeneration.^[9] When a space is formed after periapical surgery, cells from the adjacent tissues will grow into this space to form their parent tissue. The placement of a physical barrier over an osseous defect can prevent the faster proliferating oral epithelium and gingival connective tissue from growing into the bone defect, allowing the cells of the periodontal ligament and endosteum to colonize the blood clot and regenerate the lost tissue.^[10] The present two case reports describe surgical management of large periapical defects associated with maxillary anterior teeth using sticky bone and GTR membrane.

CASE REPORT

Case no. 1

A 30-year-old systemically healthy female patient reported to the Department of Conservative Dentistry and Endodontics of the Dental College and Hospital with a chief complaint of pain in her upper right front teeth region for the past 4 months. She gave a history of trauma in the maxillary anterior region 6–7 years back.

On extraoral examination, there was no abnormality detected. Intraorally, teeth #11 and #12 were found slightly discolored (Figure 1a) and both teeth were tender on percussion. Palatally, there was huge swelling seen (Figure 1b) concerning #21, 22, and 23, but no discharging sinus was found there. Periodontal pocket depth was within normal range. There was Grade I mobility of #11 and #12 but no gingival recession was there. Furthermore, the concerned three teeth demonstrated no response to the cold test and electric pulp test (EPT).

An intraoral periapical radiograph (IOPAR) of the affected teeth showed well-defined large periradicular radiolucency in relation to #11, #12 and #13 (Figure 1c). Cone beam computed tomography (CBCT) evaluation revealed the presence of localized periapical bone loss with loss of facial and palatal cortical plates in relation to teeth #11, #12 and #13 (Figure 1d-f). Based on clinical examination and by CBCT scan and IOPAR evaluation, a diagnosis of pulpal necrosis with symptomatic apical periodontitis in relation to #11, #12, and #13 was made. The patient was explained about the treatment plan and informed consent was taken.

At the first visit, endodontic treatment started with the preparation of access cavities in #11, #12, and #13 under

rubber dam isolation. The canals were cleaned and shaped up to an apical size of #70 K file (Dentsply Maillefer) for tooth #11, size #60 K file for #12, and size #70 K file for #13 using 3% sodium hypochlorite solution (Prime Dental Products Pvt. Ltd.) as irrigant. The canals were dried with paper points and calcium hydroxide (Ca(OH)₂) paste (Ultradent, Uldradent, Jordan) was placed as an intracanal medicament and the access cavities of all three teeth were temporized with zinc-oxide eugenol restoration (Cavition, GC Corporation). The patient was advised to maintain proper oral hygiene.

After 4 weeks of recall visits, resolution of the palatal swelling was noted with normal percussion response in relation to #11, #12, and #13. Intracanal medication was replaced with new Ca(OH)₂ paste in #11, #12, and #13 and the patient was again recalled after 3 months for further evaluation, which revealed no reduction in the size of periapical radiolucency on the IOPAR in relation to the concerned teeth. Hence, surgical intervention was planned. Routine blood examination was done to check whether the patient was surgically fit or not and prophylactic antibiotic was prescribed.

One week before surgery, teeth #11, #12, and #13 were obturated with cold lateral compaction technique using AH plus sealer (Dentsply Maillefer, USA) after checking the apical fit of the mater cones radiographically (Figure 1g). The access cavities were sealed with glass ionomer cement (Fuji II, GC Co., Tokyo, Japan). During surgery, under local anesthesia using 2% lidocaine with 1:80,000 epinephrine (Lignox 2% A, Indoco Remedies Ltd.) a full-thickness mucoperiosteal flap was raised by giving a sulcular incision and two vertical incisions extending from the distal aspect of #13 to distal aspect of #21. Then, bony window preparation was done, the cystic lining was enucleated, and the tissue was sent for histopathological examination. Apical 3 mm of the roots of teeth #11, #12, and #13 were resected and retro cavities were prepared using an E 10 D ultrasonic tip (Guilin Woodpecker, China). The root end cavities of the teeth were filled with mineral trioxide aggregate (MTA) (MTA Angelus, Brazil) (Figure 1h) and were verified by IOPAR.

To prepare sticky bone, 10 mL of venous blood was collected from the patient's antecubital vein and kept in a 10 mL non-coated test tube. Blood was centrifuged immediately without an anticoagulant in a centrifugation machine (Remi R8-C, India) at 2700 RPM for 2 min according to protocol by Sohn *et al.*^[8] After centrifugation, the centrifuge tube encompassed a concoction of two different layers; the uppermost layer contained AFG (Figure 1i) and the bottom layer was composed of red blood corpuscles. The upper AFG was obtained with a syringe and mixed with hydroxyapatite crystals (SYBOGRAF-Plus, Eucare Pharmaceutical Ltd; Chennai) and allowed for 5–10 min for polymerization to produce sticky bone (Figure 1j). The prepared sticky bone was placed in the periapical osseous defect and the GTR membrane (Healiguide, Advanced Biotech Products Pvt. Ltd.,) was used to cover the grafted area (Figure 1k). Then, the flap was repositioned and sutured with 3-0 black silk suture.



Figure 1: Procedural steps in Case 1: (a) pre-operative clinical view showing discolored front tooth, (b) palatal swelling observed, (c) pre-operative intraoral periapical radiograph, (d) pre-operative cone beam computed tomography scan in axial section, (e) pre-operative cone beam computed tomography scan in coronal section, (f) pre-operative cone beam computed tomography scan in sagittal section, (g) post-obturation radiograph, (h) apicoectomised roots, (i) preparation of platelet-rich fibrin, (j) sticky bone graft placed in the osseous defect, (k) guided tissue regeneration membrane placed

The patient was recalled at 3, 6, 9, and 12 months for clinical and radiographic evaluation (Figure 2). At 12-month follow-up, the patient was asymptomatic. IOPAR showed evidence of a considerable amount of bone deposition. CBCT evaluation revealed that there were a gradual increase in bone density Hounsfield unit (HU) and a decrease in the radiolucent area. It was seen that the percent increase in bone density was 129.42, 156.12, and 188.15 HU at 3, 6, and 12 months, respectively, and at the same time interval, the decrease in the radiolucent area was seen to be 83.48, 84.38, and 88.89% respectively (Table 1).

Case no. 2

A 35-year-old female patient reported to the Department of Conservative Dentistry and Endodontics with the chief complaint of pain in the upper right front teeth region for the past 2 months. She gave a history of trauma around 5 years ago. IOPAR and CBCT scan revealed large, well-circumscribed, corticated periapical radiolucency in relation to teeth #11 and #12 (Figure 3a-d). EPT confirmed that the teeth were non-vital and were tender on percussion.

Endodontic treatment was started in 11 and 12. After completion of proper cleaning and shaping and repeated placement of Ca(OH)_2 paste (UltradentTM XS, Uldradent) as an intracanal medicament for up to 3 months, there was no reduction in the size of periapical radiolucency seen in relation to 11 and 12. Hence, periapical surgery was planned for the treatment of the defect. One week before, surgery obturation was completed in 11 and 12. Surgical procedures were

followed as mentioned in case 1. Apicectomy was carried out (Figure 3e); the sticky bone was prepared by the method used in Case 1 and placed inside the bony cavity (Figure 3f), and GTR membrane (Healiguide, Advanced Biotech Products Pvt., Ltd.) was placed over the defect side (Figure 3g).

The patient was reviewed clinically and radiographically at 3-, 6-, 9-, and 12-month period during which there were no symptoms of pain, discomfort, or inflammation. CBCT evaluation revealed that a percent increase in bone density at was 105.75, 180.37, and 231.74 HU, and at the same time interval, the decrease in the radiolucent area was 83.19, 89.4, and 92.39%, respectively (Table 2 and Figure 4).

DISCUSSION

The main goal of surgical endodontics is the healing of periapical tissue by regeneration rather than repair. The locally available osteoblasts, periodontal ligament cells, and cementoblasts might be capable of restoring damaged periapical tissue.^[11] However, in the case of large periapical lesions, it may heal by repair as the migration speed of connective tissue cells is much higher than the bone-forming cells. Hence, inadequate bone healing may occur by ingrowth of connective tissue cells into the bony defect which interrupts in osteogenesis process. Andreason and Rud proposed that if the size of the osseous defect is large, regeneration of the wound will not occur and the defect will heal by fibrous connective tissue.^[11] To prevent this kind of scenario and soft tissue overgrowth, bone grafts and barrier membranes can be used.

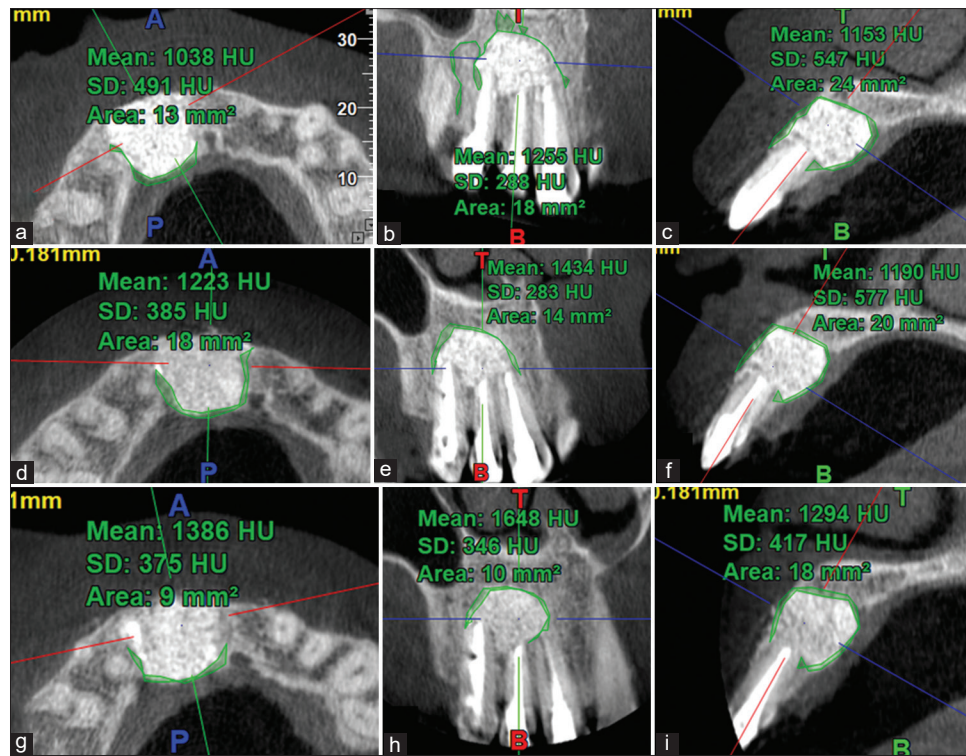


Figure 2: Post-operative cone beam computed tomography evaluation of bone density and lesion size in Case 1 (a) 3 months in axial section, (b) 3 months in coronal section, (c) 3 months in sagittal section, (d) 6 months in axial section, (e) 6 months in coronal section, (f) 6 months in sagittal section, (g) 12 months in axial section, (h) 12 months in coronal section, (i) 12 months in sagittal section

Table 1: CBCT evaluation values showing bone density and lesion size pre- and post-operatively at 3, 6, and 12 months, respectively, in Case 1

Time period	Parameters	Axial	Coronal	Sagittal	Average value	% Gain in BD	% Reduction in LS
Pre-operative	BD	474	529	499	500.66		
	LS	115	80	138	111		
3 rd month	BD	1038	1255	1153	1148.66	129.42	83.48
	LS	13	18	24	18.33		
6 th month	BD	1223	1434	1190	1282.33	156.12	84.38
	LS	18	14	20	17.33		
12 th month	BD	1386	1648	1294	1442.66	188.15	88.89
	LS	9	10	18	12.33		

CBCT: Cone beam computed tomography, BD: Bone density (in Hounsfield's Units); LS: Lesion Size (in mm²)

The body's healing potential can be maximized by the local application of growth factors and host-modulating agents, which further promote periapical tissue regeneration and healing. Platelets are known to release high quantities of growth factors such as platelet-derived growth factor (PDGF), transforming growth factor- β 1 (TGF- β 1) and β 2 (TGF- β 2), fibroblast growth factor, vascular endothelial growth factor (VEGF), and insulin-like growth factor, which stimulate cell proliferation, matrix remodeling, and angiogenesis.^[12] Several techniques to collect platelet aggregate have been utilized to accelerate tissue healing in the dental and medical fields.^[13-17] Fibrin-rich gel is known to release slowly 3 growth factors such as TGF, PDGF, and VEGF and accelerates new bone formation when it is mixed with bone graft.^[18,19] Compared to platelet-rich plasma (PRP) and plasma-rich in growth

factors (PRGF), AFG is simple to make and does not require any synthetic or biomaterials, such as bovine thrombin and calcium chloride, to make gel condition. Hence, it is free from the risk of cross-contamination.^[20] Sticky bone is prepared from AFG by mixing it with bone graft. This sticky bone does not migrate even when shaking it due to a strongly interlinked fibrin network, so the bone loss of the defect during the healing period is minimized.

In these two cases, we used sticky bone as the graft material due to its numerous advantages. Sticky bone is moldable, allowing it to adapt well to various shapes of bony defects, and it prevents both micro and macro movement of the grafted bone. The fibrin network within sticky bone entraps platelets and leukocytes, releasing growth factors that accelerate bone regeneration and soft tissue healing. Unlike PRP or PRGF,

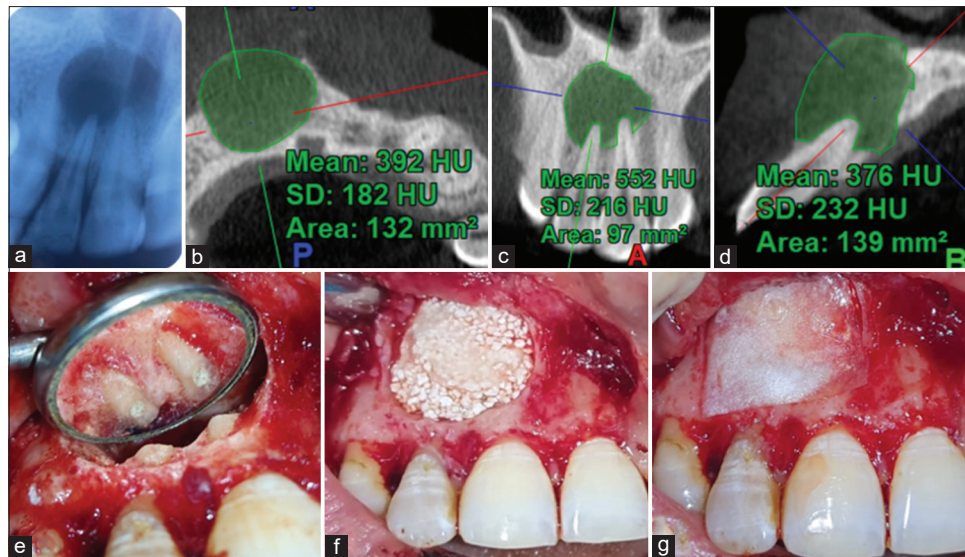


Figure 3: Procedural steps in case 2: (a) pre-operative intraoral periapical radiograph, (b) pre-operative cone beam computed tomography scan in axial section, (c) pre-operative cone beam computed tomography scan in coronal section, (d) pre-operative cone beam computed tomography scan in sagittal section, (e) apicoectomised roots, (f) sticky bone graft placed in the osseous defect, (g) guided tissue regeneration membrane placed

Table 2: CBCT evaluation values showing bone density and lesion size pre- and post-operatively at 3, 6, and 12 months, respectively, in Case 2

Time period	Parameters	Axial	Coronal	Sagittal	Average value	% Gain in BD	% Reduction in LS
Pre-op	BD	392	552	376	440		
	LS	132	97	139	122.66		
3 rd month	BD	831	1162	723	905.33	105.75	83.19
	LS	23	24	15	20.66		
6 th month	BD	1381	1273	1047	1233.66	180.37	89.4
	LS	11	17	11	13		
12 th month	BD	1537	1377	1465	1459.66	231.74	92.39
	LS	8	12	8	9.33		

CBCT: Cone beam computed tomography, BD: Bone density (in Hounsfield's Units); LS: Lesion size (in mm²)

sticky bone does not require any biochemical additives. In addition, the fibrin interconnection in sticky bone minimizes soft tissue ingrowth into the graft, ensuring better integration and stability.^[8] The “sticky bone” is a homogenous product that contains important elements for bone formation. It contains the mineral scaffold for bone cells necessary for bone formation. It also contains growth factors necessary for the stimulation of differentiation or migration of cells.^[8]

GTR techniques have been widely used for bone and periodontal tissue regeneration. In endodontic surgery, GTR has been applied using different bone substitute materials and/or different barrier membranes. The concept of GTR was introduced first by the Lindhe group (Nyman *et al.* 1982).^[21] The principles of GTR are based on the concept that epithelial cells, which migrate approximately 10 times faster than other periodontal cell types, are excluded from the wound space long enough to allow other cell types (such as osteoblasts) with regenerative potential to populate the area. This prevents epithelial downgrowth and enables regeneration. This can be obtained using various barrier membranes with or without bone grafts.^[21]

The objectives of the application of GTR barrier membrane in endodontic surgery resemble those in periodontology and implantology – (i) facilitate tissue regeneration by creating an optimum environment (stable and protected wound) and (ii) exclude non-desired fast proliferating cells from interfering with tissue regeneration.^[21]

The success rate of endodontic surgery depends on lesion size. Even in nonsurgical root canal treatment, the size of the lesion is a major factor in determining the rate of the healing process.^[11,22] Usage of GTR in large lesions (>10 mm), according to a series of clinical studies, significantly improved clinical outcomes regarding periapical area regeneration in comparison with control groups where there was no use of GTR techniques.^[23] Nonetheless, a clinical study mentioned by Corbella *et al.* showed that cases with large lesions (>10 mm) did not benefit from using a GTR procedure. However, it may be recommended to place a bone substitute into the bony crypt to support and prevent the collapse of buccally and lingually placed non-rigid membranes.^[21]

Most of the reports published in the literature have used periapical radiographs to evaluate bony changes associated with

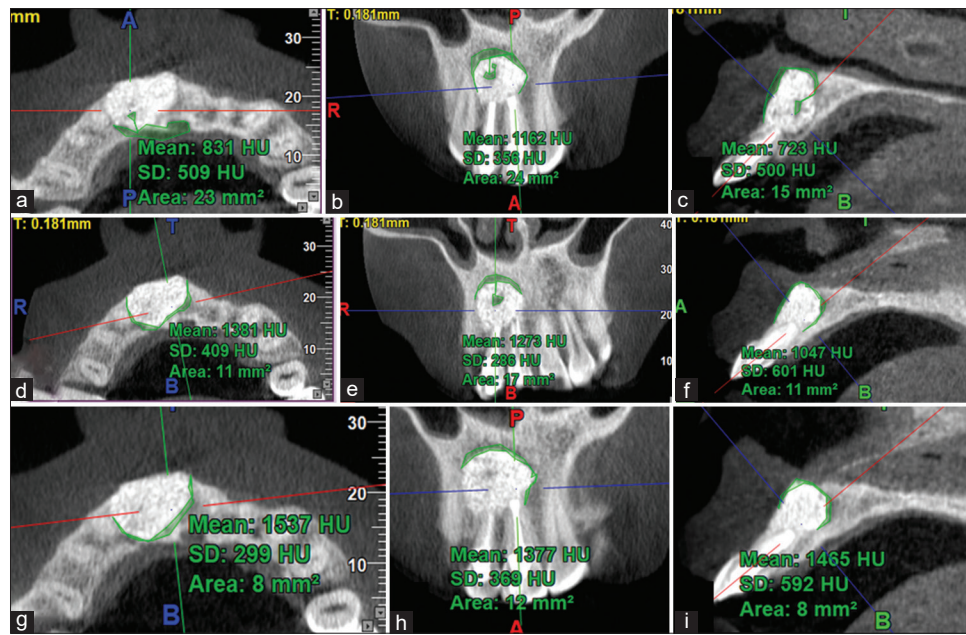


Figure 4: Post-operative cone beam computed tomography evaluation of bone density and lesion size in Case 2: (a) 3 months in axial section, (b) 3 months in coronal section, (c) 3 months in sagittal section, (d) 6 months in axial section, (e) 6 months in coronal section, (f) 6 months in sagittal section, (g) 12 months in axial section, (h) 12 months in coronal section, (i) 12 months in sagittal section

the involved tooth except Rachana *et al.* who have used CT.^[24] Furthermore, Rajput *et al.* used CBCT for initial evaluation.^[25] However, in the present two cases along with IOPA, CBCT has been used not only for initial evaluation but also for post-operative follow-up. At 12 months, radiographic assessment criteria were used to access pre- and post-operative IOPA and CBCT that satisfied the criteria of complete healing. Thus, the result of these two case reports suggests that the combination of sticky bone and GTR barrier membrane promotes bone healing and bone growth in periapical bone defects (Tables 1 and 2).

Although the ability of bone grafts or/or membranes to boost bone regeneration is well documented, periodontal ligament and cementum regeneration remain unpredictable. This indicates that GTR techniques might not result in complete regeneration of periapical tissues after endodontic surgery.^[26] To fully understand tissue regeneration, more information is needed, including details of the various molecular and biological processes involved in the formation of each tissue in the periapical area. Clinicians must know all the factors that negatively influence the outcome of sticky bone and GTR and strict case selection should be implemented regarding this.

CONCLUSION

The present two case reports have shown that the use of sticky bone and GTR membrane may lead to sufficient bony healing of large periapical lesions after endodontic surgery. However, histological studies are more appropriate to confirm bone regeneration. Further long-term follow-up is required to validate the obtained results.

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