

Healing of periosteal pedicle graft in the treatment of gingival recession defects: A histological study in rabbits

ABSTRACT

Purpose: Various surgical techniques are described in the literature to manage gingival recession defects but the histopathologic evidence to support the current available techniques, is scarce. Recently periosteal pedicle graft (PPG) has proven to be an effective treatment option to treat gingival recession defects (GRD) with results comparable to subepithelial connective tissue graft (SCTG).

Objective: The present histopathological study was done to evaluate the healing pattern of periosteal pedicle autogenous graft along with coronally advanced flap in the treatment of gingival recession defects.

Materials and Methods: The present study was performed on 10 sites in 5 rabbits. Two sites were selected in each animal and gingival recession defects were surgically created and then treated using periosteal pedicle graft along with coronally advanced flap procedure. Healing pattern was assessed histopathologically at pre-defined intervals till 6 months.

Results: On the 7th day of healing, dilated blood vessels with inflammatory cells were seen, while rudimentary rete-pegs appeared on 14th day. Between 3 months to 6 months, advanced histological repair with connective tissue organization with initiation of junctional epithelium, cementum and bone formation were observed.

Conclusion: Gingival recession defects treated with PPG had evidences of regeneration of cementum, bone and periodontal ligament fibers with new connective tissue attachment.

Keywords: Histology, periosteal pedicle graft, recession, wound healing

INTRODUCTION

Gingival recession is defined as the displacement of the marginal tissue apical to the cement–enamel junction,^[1] which may result in problems such as impaired esthetics, plaque accumulation, root caries, dentin hypersensitivity, and gingivitis.^[2] The etiology of gingival recession defects is multifactorial, and numerous surgical procedures have been described in the literature for the treatment of gingival recession defects^[3,4] with variable success rates.^[3,5]

Periosteal pedicle graft (PPG) was an innovative technique first described in 1999 and has shown promising treatment outcomes when used to manage gingival recession defects.^[6] The technique has shown results, which are comparable with those of the subepithelial connective tissue graft (SCTG) in terms of root coverage. Recent studies have rated PPG better than connective tissue graft owing

to its immense regenerative potential and better patient satisfaction.^[7]

The long-term success of root coverage procedures has often been questioned due to the lack of histopathological

KANWARJIT SINGH ASI, AJAY MAHAJAN, DEEPA RAYAST¹

Department of Periodontology, Himachal Pradesh Government Dental College and Hospital, Shimla, Himachal Pradesh,

¹Department of Dental Surgery, Raipur Institute of Medical Science, Chhattisgarh, India

Address for correspondence: Dr. Deepa Rayast, Department of Dental Surgery, Raipur Institute of Medical Science, Chhattisgarh, India.
E-mail: deeparayast@gmail.com

Received: 22 October 2022, **Revised:** 29 May 2023, **Accepted:** 02 June 2023, **Published:** 24 July 2024

This is an open access journal, and articles are distributed under the terms of the Creative Commons Attribution-NonCommercial-ShareAlike 4.0 License, which allows others to remix, tweak, and build upon the work non-commercially, as long as appropriate credit is given and the new creations are licensed under the identical terms.

For reprints contact: WKHLRPMedknow_reprints@wolterskluwer.com

How to cite this article: Singh Asi K, Mahajan A, Rayast D. Healing of periosteal pedicle graft in the treatment of gingival recession defects: A histological study in rabbits. *Natl J Maxillofac Surg* 2024;15:283-7.

Access this article online

Website:
www.njms.in

DOI:
10.4103/njms.njms_187_22

Quick Response Code



evidence explaining regeneration at the site of treated gingival recession defects.^[5,8] Therefore, this study was conducted to evaluate the healing of PPG when used to treat gingival recession defects.

The study is based on the hypothesis that healing by new attachment and regeneration might happen at the denuded root and PPG interface owing to the inherent regenerative capacity of the periosteum.

MATERIALS AND METHODS

This study was performed at an animal house and approved by the ethical community [Registration no SO2/PO/Re2001/CPCSEA and 18/1/2017]. The surgical team consisted of two periodontal surgeons with more than 10 years' experience and a veterinary surgeon who assisted in the presurgical preparation and postsurgical maintenance of the animals.

A total of 10 sites in five rabbits (R1, R2, R3, R4, and R5) were used for this study. Two sites were selected in each animal: the left mandibular central incisor and the right mandibular incisor. On the day of surgery, the gingival recession defect was surgically created in all the animals [Figure 1a-c], which was then treated after 30 days using a PPG along with a coronally advanced flap procedure [Figure 1d-f]. To assess the pattern of healing, specimens were taken from animals R1, R2, R3, R4, and R5 after 7 days, 14 days, 1 month, 3 months, and 6 months, respectively, as shown as follows:

RESULTS

On the 7th day, basal cells and prickle cells were seen with plump fibroblasts in the oral epithelium of the flap without rete pegs. The connective tissue in the healing wound consisted of engorged blood vessels, dilated capillaries, and active osteoblasts. Fibroblasts, osteoblasts, supporting vasculature, and laying of osteoid material were seen in the tissue away from the graft [Figure 2a and b].

On the 14th day, cellular organization and initiation of wound maturation by the appearance of rudimentary rete pegs at the interface of epithelium and the connective tissue were clearly evident. Collagen fibers and ground substance were also seen in the connective tissue.

At the end of 1 month, oral epithelium had almost reached normal structure with basal layer, prickle cell layer, and rete pegs that had further extended into the connective tissue. Junctional epithelium appeared for the first time at this stage. The interface between graft and flap showed organized granulation tissue with bundles of matured collagen fibers and fibroblast cells [Figure 3a-d].

After 3 months, the length of epithelium and soft tissue thickness increased to 3 mm, and the sulcular epithelial lining appeared more stratified and organized with proliferation and further elongation of rete pegs [Flow Chart 1]. A progressive increase in the amount of connective tissue fibers was also seen. The surface epithelium along with junctional epithelium appeared to be in close approximation to the tooth surface.

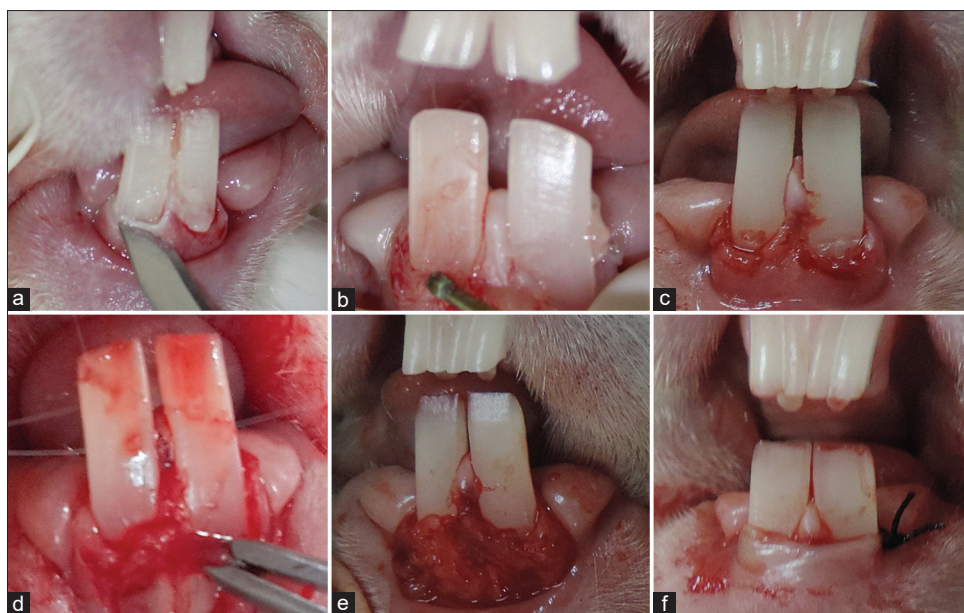


Figure 1: External bevel incision given for the defect creation (a), notch at CEJ (b and c), harvested periosteal pedicle graft (d), periosteal pedicle graft placed over the gingival recession defect (e), and periosteal pedicle covered with coronally advanced flap (f)

A 2 mm thick layer of newly formed cementum and bone was evident [Figure 4].

After 6 months, more amount of mature bone with an increase in the thickness of the sulcular epithelium was evident. A well-organized and matured junctional epithelium of 0.5 mm in length was also attached to the tooth surface at the cemento-enamel junction. The surface epithelium regained its normal shape, thickness, and appearance with well-formed rete pegs. The connective tissue appeared to be well organized with dense matured collagen fibers throughout [Figure 5a-f].

DISCUSSION

Various techniques have been described in the literature to treat the gingival recession defect (GRDs),^[2,7-11] but scientific evidence on the healing pattern of treated GRDs is scarce. The use of periosteum as a pedicle graft in the treatment of gingival recession defects is a recent technique, which

utilizes the immense potential of periosteum to revascularize and regenerate^[2] This study is a pioneer study evaluating the healing pattern of GRDs treated with PPG in experimental animals. The healing pattern of PPG was observed till 6 months in five experimental rabbits. Several studies have used rabbits to evaluate the healing pattern in oral wounds. Similarities between human and rabbit periodontal tissue anatomy and physiology made it possible to assess the healing pattern in treated gingival recession defects.^[9-12]

Histopathological specimen of the 7th day showed that inflammatory cells invaded the graft, length of oral epithelium was 2 mm, and no rete pegs were seen. A few dilated blood vessels with blood clot embedded in the graft were also noted. Guiha *et al.*^[12] and Staffileno and Levy^[13] reported the same results. On the 14th day, the epithelium itself had increased in thickness and rete pegs of epithelium started to appear. Connective tissue revealed a decrease in the number of inflammatory cells and an associated increase in fibroblasts, and a few cementoblasts were also seen, but no signs of new bone formation were observed. In addition, granulation tissue was observed as an interface between graft and flap and blood clot disappeared within the blood vessels. Junctional epithelium had not yet formed. Staffileno and Levy^[13] also reported similar findings in the 14th-day specimen in their histological and clinical study of mucosal transplants in dogs.

After one month of healing, our results revealed that the thickness of the oral epithelium had increased and

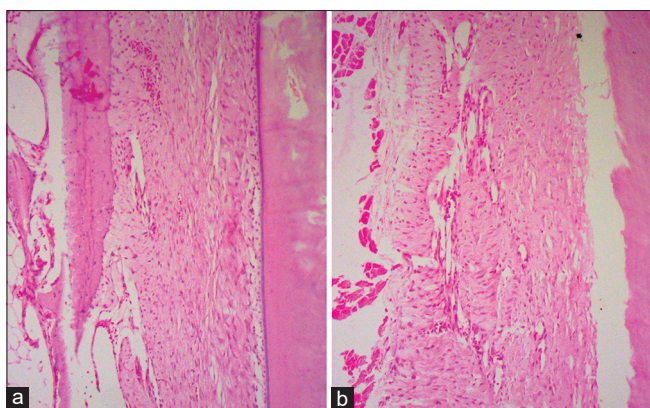


Figure 2: (a and b) 7th-day histopathological analysis of the biopsy specimen showing engorged blood vessels (magnification 10x)

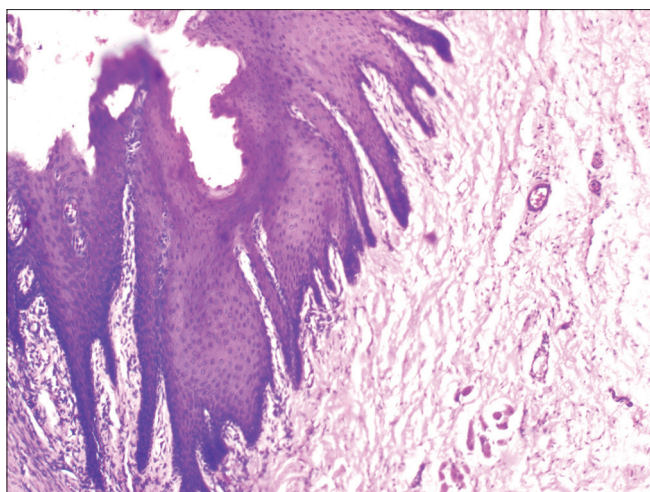


Figure 4: 3-month histopathological analysis of the biopsy specimen showing epithelium, which has regained its normal shape, thickness, and appearance

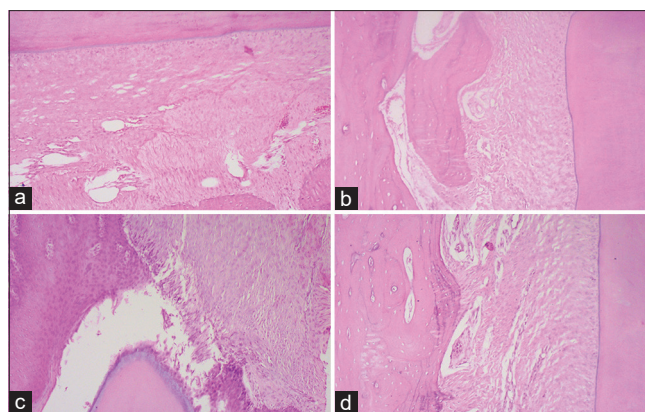
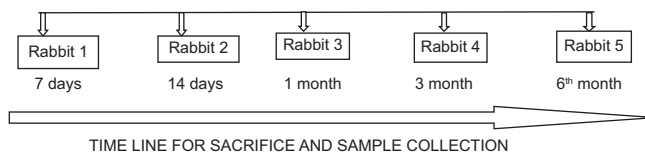


Figure 3: 1-month histopathological analysis of the biopsy specimen showing demarcation zone between graft and periosteum (a), well-organized bundles of collagen fibers (b), increased thickness of surface epithelium (c), and collagen fibers penetrating the graft (d)



Flow Chart 1: Time line for sacrifice and sample collection

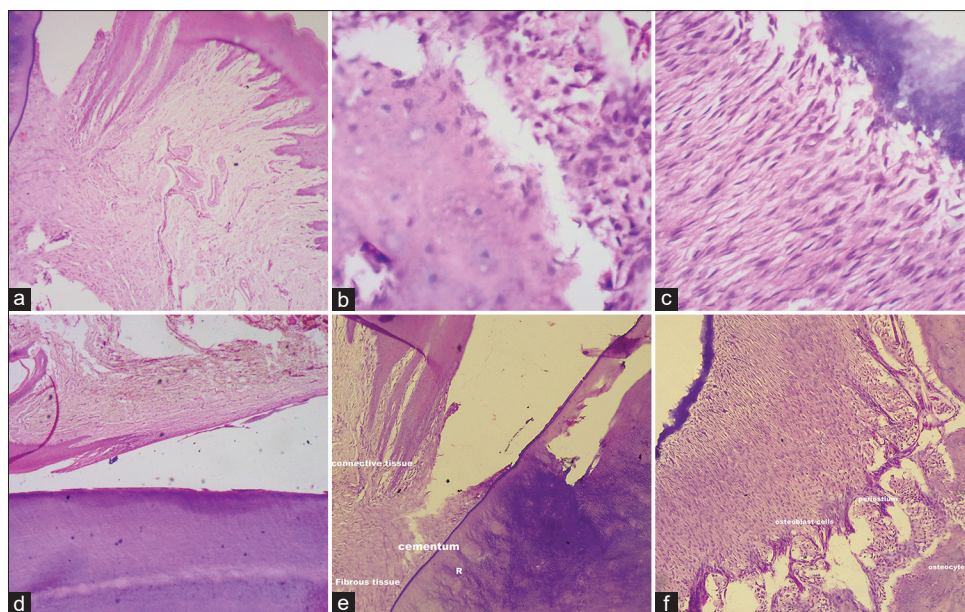


Figure 5: 6-month histopathological analysis of the biopsy specimen showing well-stabilized gingival epithelium (a), osteoblast (b), fibroblast (c), the junctional epithelium (d), and new bone tissue, cementum, and the thick layer of collagen fibers (e and f)

approached the normal structure. Rete pegs had further extended into the connective tissue and the junctional epithelium, notably appeared for the first time at this stage. A fibrous connective tissue attachment from the apical extent of the junctional epithelium was also seen. These findings were also observed in the study conducted by Richardson *et al.*^[14] and Guiha *et al.*^[12] An important finding was new bone formation at the interface of graft and tooth root. This finding was in accordance with Al-Hezaimi *et al.*^[15] in which contralateral gingival recession defects were surgically treated with SCTG + platelet rich plasma (test group) or SCGT (control group) and the evidence of bone formation in the area of the root was noticed after 45 days.

After 3 months of healing period, the length of epithelium and soft tissue thickness increased to 3 mm, the sulcular epithelium lining appeared more stratified and organized with proliferation and elongation of rete pegs, and an increase in number of well-organized collagen fiber bundles in connective tissue was seen. A 2 mm thick layer of newly formed cementum was observed coronal to the notch and newly formed bone increased to 1 mm compared with the previous specimen of 1 month. These findings were in agreement with Casati MZ *et al.*^[16] who did a histometric study to assess the healing pattern of gingival recession defects with guided tissue regeneration and coronally positioned flaps. The healing pattern at 6 months was fairly similar to that seen till 3 months except for the fact that an increase in the thickness of the sulcular epithelium, a well-organized and mature junctional epithelium of length 0.5 mm, was noticed. Lewis C. Cummings *et al.*^[17] also reported similar findings in their study.

In addition to the above findings, the authors of this research paper also tried to correlate the clinical studies, which utilized PPG for the treatment of gingival recession defect and the histopathological findings of this study. The majority of the clinical studies attributed the immense success of PPG to the inherent regenerative and vascular properties of the periosteum.^[18,19] Better healing and esthetic results were mainly attributed to the ample vascular supply associated with PPG, due to the release of vascular endothelial growth factor (VEGF) in addition to immediate post-inflammatory vasodilation^[8,12,20] Studies on SCTG healing have revealed that neovascularization in SCTG-treated cases was only due to vasodilation, which follows the surgical trauma.^[12] PPG seems to promote more effective neovascularization compared with SCTG due to the combined effect of VEGF and vasodilatation in cases treated with PPG. Previous clinical studies have suggested that the long-term success of GRDs treated with PPG was supposedly due to the presence of stem cells, osteoblasts, fibroblasts, and their progenitor cells in the layers of periosteum, which contributed to the new attachment between the graft and the denuded root surface.^[21-23] The findings of our study clearly confirmed the presence of these cells during the healing of PPG. The presence of these cells might be responsible for the firm reattachment of the connective tissue fibers with the denuded root surface. On further analysis, it was interesting to observe that the cells with the potential to regenerate cementum and periodontal ligament (i.e., cementoblasts and fibroblasts) were the first to populate the root surface followed by osteoblast and their progenitor cells, which were immediately behind the fibroblasts and populate the

defect. So, the presumption that PPG places proper cells in the proper position for the regeneration of gingival recession defects was objectively confirmed at the end of this study.^[24]

To summarize, the present study could act as a baseline study, which explains the histopathological mechanism behind the immense clinical success of PPG.

Limitations

Though our manuscript replicated the PPG technique in animals, it is worth mentioning that an actual histological study in humans taking into account etiological factors such as trauma from occlusion will produce stronger evidence.

Financial support and sponsorship

Nil.

Conflicts of interest

There are no conflicts of interest.

REFERENCES

- American Academy of Periodontology. Glossary of Periodontal terms. 3rd ed. Chicago: American Academy of Periodontology; 1992.
- Mahajan A. Periosteal pedicle graft for the treatment of gingival recession defects: A novel technique. *Aust Dent J* 2009;54:250-4.
- Chambrone L, Tatakis DN. Periodontal soft tissue root coverage procedures: A systematic review from the AAP regeneration workshop. *J Periodontol* 2015;86:S8-S11.
- Langer B, Langer L. Subepithelial connective tissue graft technique for root coverage. *J Periodontol* 1985;56:715-20.
- Hofmänner P, Alessandri R, Laugisch O, Aroca S, Salvi GE, Stavropoulos A, *et al.* Predictability of surgical techniques used for coverage of multiple adjacent gingival recessions—A systematic review. *Quintessence Int* 2012;43:545-54.
- Lekovic V, Kenney EB, Carranza FA, Martignoni M. The use of autogenous periosteal grafts as barriers for the treatment of Class II furcation involvements in lower molars. *J Periodontol* 1991;62:775-80.
- Prakash P, Rath SK, Mukherjee M. Clinical efficacy of periosteal pedicle graft with subepithelial connective tissue graft in gingival recession coverage. *J Indian Soc Periodontol* 2019;23:442-7.
- Mahajan A. A review of periosteal pedicle graft technique for the management of gingival recession defects. *J Adv Surg Res* 2018;2:10-4.
- Kantarci A, Hasturk H, Van Dyke TE. Animal models for periodontal regeneration and peri-implant responses. *Periodontol* 2000 2015;68:66-82.
- Rinastiti M, Harijadi, Santoso AL, Sosroseno W. Histological evaluation of rabbit gingival wound healing transplanted with human amniotic membrane. *Int J Oral Maxillofac Surg* 2006;35:247-51.
- Oortgiesen DA, Meijer GJ, Bronckers AL, Walboomers XF, Jansen JA. Fenestration defects in the rabbit jaw: An inadequate model for studying periodontal regeneration. *Tissue Eng Part C Methods* 2010;16:133-40.
- Guiha R, el Khodeiry S, Mota L, Caffesse R. Histological evaluation of healing and revascularization of the sub epithelial connective tissue graft. *J Periodontol* 2001;72:470-8.
- Staffileno H, Levy S. Histologic and clinical study of mucosal (Gingival) transplants in dogs. *J Periodontol* 1969;40:311-9.
- Richardson CR, Maynard JG. Acellular dermal graft: A human histologic case report. *Int J Periodontics Restorative Dent* 2002;22:21-9.
- Al-Hezaimi K, Rudek I, Al-Hamdan KS, Javed F, Iezzi G, Piattelli A, *et al.* Efficacy of acellular dermal matrix and coronally advanced flaps for the treatment of induced gingival recession defects: A histomorphometric study in dogs. *J Periodontol* 2013;84:1172-9.
- Casati MZ, Sallum EA, Caffesse RG, Nociti FH, Sallum AW, Pereira SL. Guided tissue regeneration with a bioabsorbable polylactic acid membrane in gingival recessions. A histometric study in dogs. *J Periodontol* 2000;71:238-48.
- Cummings LC, Kaldahl WB, Allen EP. Histologic evaluation of autogenous connective tissue and acellular dermal matrix grafts in humans. *J Periodontol* 2005;76:178-86.
- Mahajan A, Asi KS. Comparison of effectiveness of the novel periosteal pedicle graft technique with coronally advanced flap for the treatment of long-span unesthetic multiple gingival recession defects. *Clin Adv Periodontics* 2018;8:77-83.
- Shah MP, Patel AP, Shah KM. Periosteal pedicle graft: A novel root coverage approach. *J Indian Soc Periodontol* 2015;19:99-102.
- Bourke HE, Sandison A, Hughes SP, Reichert IL. Vascular endothelial growth factor (VEGF) in human periosteum normal expression and response to fracture. *J Bone Joint* 2003;85:4.
- Squier CA, Ghoneim S, Kremenak CR. Ultrastructure of the periosteum from membrane bone. *J Anat* 1990;171:233-9.
- Provenza DV, Seibel W. *Oral Histology: Inheritance and Development*. Lea and Febiger; 1986.
- Godavarthi L, Murthy KR, Pavankumar S. A comparison of acellular dermal matrix allograft and periosteal pedicle graft covered by coronally advanced flap in the treatment of gingival recession: 1-year follow-up study. *Int J Periodontics Restorative Dent* 2016;36:67-75.
- Dandu SR, Murthy KR. Multiple gingival recession defects treated with coronally advanced flap and either the VISTA technique enhanced with GEM 21S or periosteal pedicle graft: A 9-month clinical study. *Int J Periodontics Restorative Dent* 2016;36:231-7.