

CASE REPORT

How do GTR and GBR Differ? A Periodontitis Case Treated Using an Equine-derived, Enzyme-deantigenic, Collagen-preserving Bone Graft, and Collagen Membranes

Riccardo Tizzoni¹, Marta Tizzoni²

ABSTRACT

Aim: The present case illustrates how a tooth, which had a highly questionable prognosis, was preserved by carrying out a periodontal regeneration surgery.

Background: Treatment of periodontitis involves a careful consideration of all the factors that may allow the achievement of a favorable outcome; among those, the skillful use of guided tissue regeneration (GTR) membranes is of paramount importance.

Case description: A 39-year-old patient presented with a mobile central upper incisor due to severe periodontitis and was treated according to GTR principles using a collagen membrane. A collagen-preserving bone graft was also used, as a scaffold for clot formation and cellular infiltration, which was covered with a second collagen membrane. The patient was contacted for follow-up assessment at 3, 6, 12, and 18 months after surgery. Follow-up radiographs showed that bone regeneration occurred around the involved tooth and very little tooth mobility was observed. The patient's masticatory function, appearance, and comfort were favorable.

Conclusion: The use of two equine collagen membranes with the purpose of creating the best conditions to carry out periodontal regeneration according to GTR principles, in association with an equine, collagen-preserving, enzyme-deantigenic bone graft, allowed sufficient bone regeneration to salvage a tooth that was deemed otherwise lost because of periodontitis.

Clinical significance: In cases of teeth that are severely compromised by periodontitis, the use of collagen membranes according to GTR principles can allow the regeneration of the periodontal tissues; the association with a bone substitute having well-known performance rates, covered with a collagen membrane (guided bone regeneration, GBR) can, in some cases, improve bone regeneration at the defect site.

Keywords: Bone regeneration, Case report, Guided tissue regeneration, Periodontal diseases.

The Journal of Contemporary Dental Practice (2019): 10.5005/jp-journals-10024-2571

BACKGROUND

The etiology of periodontal disease is often multifactorial and effective treatment frequently requires a multidisciplinary approach.^{1,2} Early-onset periodontitis originates, usually in predisposed patients, from uncontrolled plaque accumulation and consequent calculus deposits that lead to chronic gingival inflammation. Occlusion,³ uncontrolled nocturnal forces and endodontic conditions,^{4,5} may all play a crucial role in its onset. Later stages include the loss of the epithelial seal and the creation of a connective tissue pocket, as well as significant gingival recession. Eventually, in uncontrolled cases, the total loss of the periodontal ligament and bone is observed. Consequently, the tooth often becomes mobile, and the prognosis questionable, if not completely unfavorable. At this stage, regenerating the periodontal tissues becomes quite challenging as any negative factor, including the patient characteristics, the suboptimal use of surgical approaches and materials, and a surgeon's insufficient clinical skill and experience may lead to a dramatically negative outcome.⁴ Treatment of periodontitis aims to increase the amount of periodontal connective tissue attachment, with new cementum and new bone formation of the severely compromised tooth and to decrease the pocket depth, causing a minimal or zero increase in gingival recession. Residual periodontal ligament plays a pivotal role in the regeneration of periodontal ligament itself, new cementum and bone, as a source of cells;⁶⁻⁸ accordingly, when treating periodontitis with the aim of regenerating lost

^{1,2}Private practice in Milan, Italy

Corresponding Author: Riccardo Tizzoni, Via San Barnaba 30, Milan, Italy, Phone: +39-025453560, e-mail: dottori.tizzoni@gmail.com

How to cite this article: Tizzoni R, Tizzoni M. How do GTR and GBR Differ? A Periodontitis Case Treated Using an Equine-derived, Enzyme-deantigenic, Collagen-preserving Bone Graft and Collagen Membranes. *J Contemp Dent Pract* 2019;20(5):639-644.

Source of support: Nil

Conflict of interest: None

tissues, the preservation of the periodontal ligament is of foremost importance. The gingival connective tissue, in fact, cannot form new connective tissue attachments; indeed, it may instead induce root resorption.⁹ Similarly, bone regeneration—when occurring in direct contact with the root—cannot form new attachment and may indeed induce root ankylosis,¹⁰ or sometimes, root resorption. From a biological standpoint, the latter case is due to the necessary osteoclastic activity on the bone, as part of bone remodeling, which, at the same time, acts negatively on the root when this is deprived of the periodontal ligament. Periodontal treatments intended to achieve tissue regeneration usually induce epithelial downgrowth instead; this, in turn, prevents the formation of a new connective tissue attachment, as periodontal ligament cells cannot repopulate the root surface.¹¹ However, the coverage of the root surface by an epithelial layer prevents root ankylosis and resorption that would otherwise be induced by bone and gingival connective tissue. In

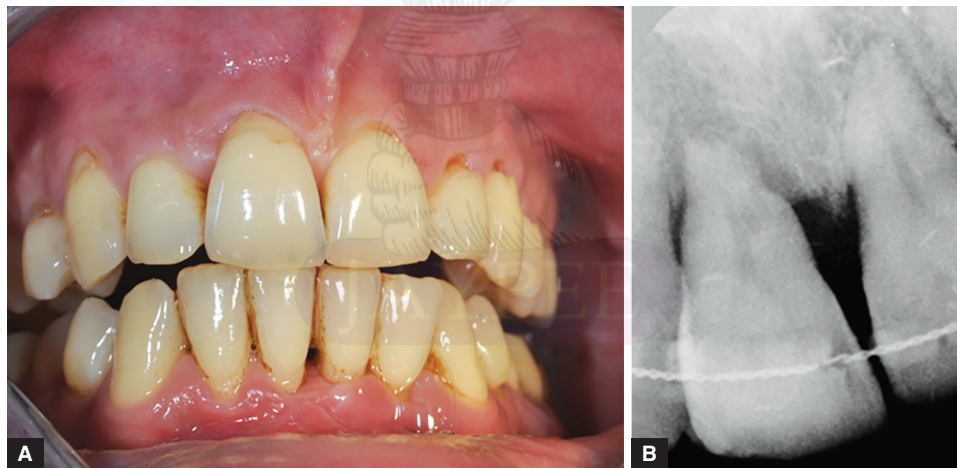
summary, the regeneration of the periodontium involves several cell types including gingival epithelial cells, periodontal ligament cells, including cementoblasts, and bone cells that may populate the tridimensional space surrounding the affected tooth within their well-defined spatial boundaries and, consequently, in a certain time order. Guided tissue regeneration (GTR) techniques aim to achieve periodontium regeneration—that is, to regenerate bone with new cementum formation and a functionally oriented periodontal ligament—using barrier membranes. Appropriate use of barriers may allow any periodontal cells still present to arrive before all the other involved cell types and repopulate the area surrounding the tooth root, preventing bone cells, especially if concomitant bone grafting has been carried out (guided bone regeneration, GBR), and gingival epithelial cells and gingival connective tissue cells from repopulating the area.¹² The present case illustrates how a tooth, which had a highly questionable prognosis, was preserved by carrying out a periodontal regeneration surgery using collagen GTR membranes and an enzyme-treated equine-derived collagen-preserving bone graft, which was covered with a second collagen membrane, to provide supporting bone (GBR) and as a scaffold for clot formation and cellular infiltration.

CASE DESCRIPTION

The patient was a heavy smoker (15–20 cigarettes/day), a 39-year-old man with a noncontributory medical history reporting a nocturnal episode of pain affecting the upper central right incisor (tooth 1.1). A comprehensive periodontal examination was accomplished. Clinical examination (Fig. 1A) showed that the tooth had grade 3 mobility,¹³ grade 2 gingival and bleeding indexes,¹⁴ grade 2 calculus

index,¹⁵ and grade 2 visible plaque index¹⁴ as shown in Table 1. Gingival recession was 2 mm. Radiographic examination revealed a significant periodontal bone loss affecting tooth 1.1 as well as initial root resorption (Fig. 1B). The tooth was negative to the dental pulp test. The patient was informed of the severity of his condition and the high probability of failure of any attempt to maintain the mobile element. The first treatment plan proposed involved extracting tooth 1.1 and replacing it with an implant-supported single crown. A prosthetic rehabilitation by means of a conventional bridge, with a pontic element, was also suggested. The patient firmly refused these options and, after discussions with him about his possibly unrealistic expectations, he was alternatively proposed a procedure that included splinting the mobile tooth, elevating a periodontal flap, and performing regenerative surgery involving the placement of barrier membranes as well as a xenogeneic bone graft, with the aim of preventing definitive tooth loss and rehabilitating the patient while also achieving the best possible aesthetic outcome. The patient gave his informed consent.

Splinting was carried out from the right to the left cuspid. This procedure was done at the first visit (Fig. 2), and the patient underwent full mouth oral hygiene and meticulous sulcular debridement. One month later, the patient went through periodontal surgery. As he showed distress, he was subjected to procedural sedation as follows: after administering 4 mg diazepam (Valium, Roche, Milano, Italy), the patient was administered oxygen (O₂) for 5 minutes and then a nitrous oxide (N₂O)/oxygen mixture with a N₂O increasing concentration up to a 40:60 N₂O:O₂ ratio. For antibiotic prophylaxis, 2 g of amoxicillin/clavulanic acid (Augmentin, Glaxo-SmithKline, Verona, Italy) were administered



Figs 1A and B: (A) The right central incisor shows mobility and unfavorable gingival and bleeding indexes. (B) The radiograph shows significant periodontal bone loss and initial root resorption

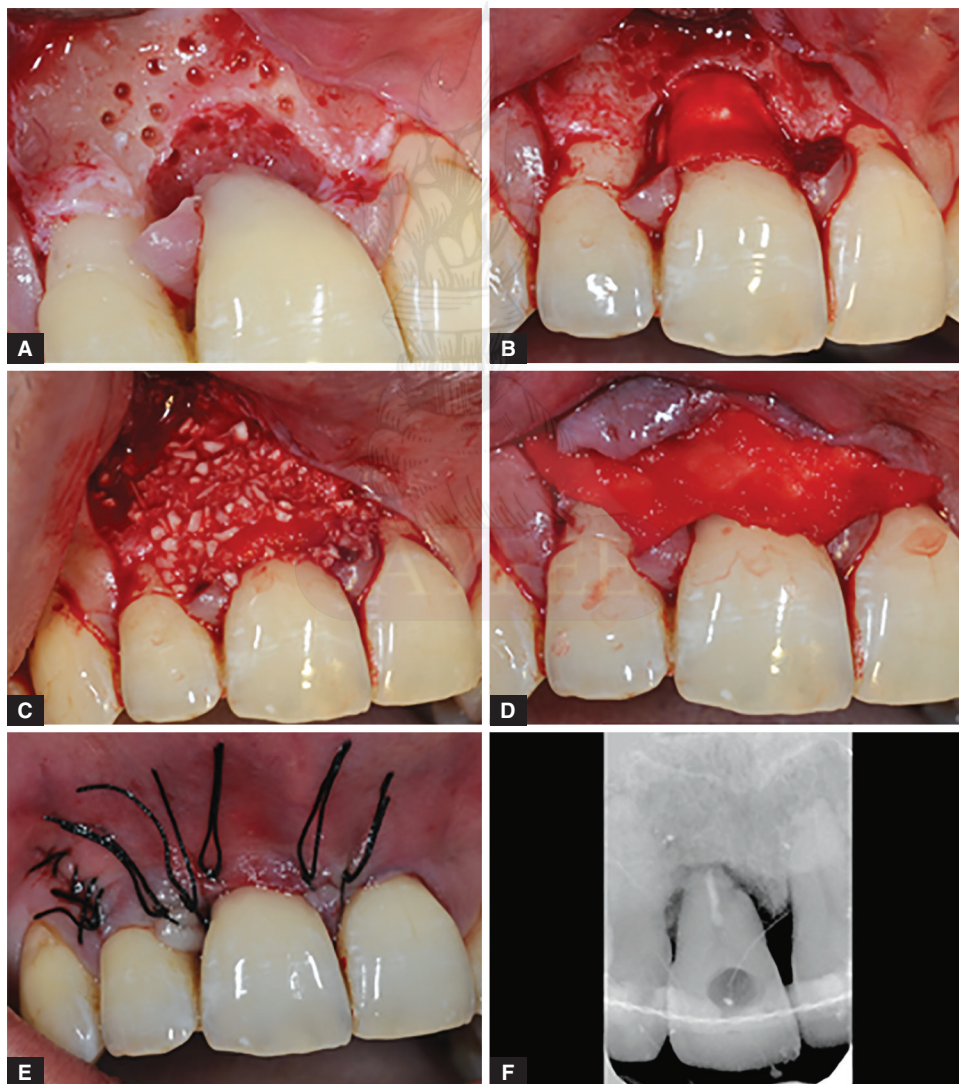
Table 1: Periodontal indexes. Mobility: tooth mobility grade; GI: gingival index; CI, calculus index; VP, visible plaque index; CAL (buccal/palatal), clinical attachment level calculated at three probing sites at the buccal and palatal side, respectively

Index	Presurgical	6-month control	12-month control	18-month control
Mobility	3	1	1	1
GI	2	1	1	1
CI	2	0	0	0
VP	2	1	1	1
CAL (buccal)	6;7;6	4;4;3	4;4;3	4;4;3
CAL (palatal)	3;3;3	2;2;2	2;2;2	2;2;2
Full mouth bleeding score (%)	74	22	27	29
Full mouth plaque score (%)	83	30	39	40



Fig. 2: Splinting was carried out during the first visit

1 hour before surgery and then 1 g every 12 hours for 8 days following the surgery. The patient also rinsed for 2 minutes with a chlorhexidine 0.20% mouth rinse (Corsodyl, Glaxo-SmithKline, Verona, Italy) and received 100 mg of a nonsteroidal-anti-inflammatory drug (Oki, Dompè, Milan, Italy). Local anesthetic was administered by means of infiltration into the oral mucosa with 2% lidocaine with epinephrine 1:50,000 (Dentsply Italia, Milano, Italy). Periodontal regenerative surgery is shown in Figure 3. An intrasulcular incision was created, extending partially to the left central incisor and to the right lateral incisor, preserving all the papillae; a releasing incision was then created on the distal aspect of the lateral incisor to allow flap elevation and subsequent regenerative surgery. The tooth root as well as the tooth apex and the area underneath the papillae were then debrided of granulation tissue using manual periodontal curettes and ultrasonic scalers. While debriding, the soft tissue was not detached from the coronal portion of the root in the interproximal and palatal aspects. The bone was debrided and bone holes were then created using a



Figs 3A to F: Steps of the periodontal regenerative surgery. After carrying out root debridement and planing, as well as alveolar bone preparation (A), a first collagen membrane is placed to protect the tooth root and possibly favoring periodontal ligament regeneration (B). Bone grafting is then carried out (C) and the graft is protected using a second collagen membrane (D). Finally, the flap is sutured (E) and a control radiograph (F) is taken

diamond round bur mounted on a high-speed rotating handpiece to allow bleeding and favor bone regeneration. Before continuing the grafting surgery, a conventional root canal therapy was carried out and a retrofilling material positioning with no apicoectomy was also performed. As the root canal was both short and wide, dentin/enamel adhesives (Bisco, Schaumburg, USA) were used to seal it and a bonding resin (Bisco, Schaumburg, USA) was added to complete the filling at the crown portion, while a polymer-reinforced zinc oxide–eugenol composition restorative material (IRM, Dentsply Italia, Milano, Italy) was used to seal the apical portion. The root apex and the retrofilling material were finished and polished and the surgical area was rinsed using a 20% chlorhexidine-based solution (Curaden Healthcare, Saronno, Italy). A first equine collagen membrane (Biocollagen, Bioteck, Vicenza, Italy) was then placed over the tooth root with the aim of preventing direct contact between the bone graft (and, after regeneration had occurred, bone tissue) and the root, while possibly favoring—instead—the migration of periodontal ligament cells on the root surface with consequent regeneration of the periodontal ligament itself, new cementum and bone. The bone defect was then grafted with a 1:1 cancellous–cortical mixture of enzyme-deantigenic equine 0.5 to 1.0 mm bone granules (Osteoplant Osteoxenon, Bioteck, Vicenza, Italy) for bone regeneration and to act as a scaffold, for clot formation and cellular infiltration, and the grafted site was then covered with a second equine collagen membrane (Biocollagen, Bioteck, Vicenza, Italy).

The flap was closed using a non-resorbable suture (Monomyd 4-0/5-0 Polyamide Monofilament Suture, Butterfly, Cavenago, Italy). The sedative was then adjusted to 100% O₂ and the patient was monitored closely until complete recovery. Before release, the patient was also provided with a custom rigid bite splint, placed over his lower arch, to protect the upper elements from mechanical trauma. The postsurgical control radiograph (Fig. 3f) showed that a small portion of the root apex had been lost through root planing and polishing after retrofilling. Sutures were removed after 1 week, and the patient returned for follow-up assessment at 3, 6, 12, and 18 months thereafter.

During follow-up observations, staged radiographs showed that partial bone regeneration had occurred around the involved tooth (Fig. 4). Before surgery, and even when splinted, this tooth



Fig. 4: X-ray comparisons: presurgery (left) and at 18-month follow-up (right). A partial periodontal bone regeneration occurred. The effects of intra-operative root-planing and polishing are noticeable



Fig. 5: At the 18-month follow-up, the aesthetic result is favorable and the tooth is no more mobile

could be still rotated on its vestibular and palatal aspects; from the 6-month follow-up control onward, no rotation along any axis could be observed at all. Neither clinical nor radiographic symptoms of root resorption or ankylosis were observed at any control visit. The patient's masticatory function, appearance, and comfort were favorable (Fig. 5).

DISCUSSION

The present report describes a surgical intervention aimed to salvage and restore the function and aesthetics of a tooth with a questionable prognosis because of periodontitis. Periodontal regeneration was carried out according to the principles of GTR. The tooth root had to be planed over most of its surface, except for its most coronal portion, and no bundle bone was present; these were regarded as two conditions highly favoring the probability of observing root resorption or tooth ankylosis because of the consequent reduced (and possibly lost) periodontal ligament regenerative potential. It is known that cells from the periodontal ligament are those promoting periodontal tissues' regeneration and new connective tissue attachment formation,^{9,16} while those from gingival connective tissue cannot originate any connective tissue attachment and possibly induce root resorption instead.⁹ Specifically, alveolar bone, when developing during the embryological phase, originates from the dental follicle.¹⁷ The ectomesenchymal cells of the dental follicle differentiate into osteoblasts which lay down the bone matrix, called the osteoid tissue. Dental follicle cells are stem cells of childhood that can be found only in the growing tooth germ before they actually erupt in the oral cavity. It is thought that dental follicle cells can differentiate into osteoblasts, periodontal ligament fibroblasts, and cementoblasts to produce the tooth supporting tissues, known as periodontium.¹⁷

Mesenchymal stem cells are connective tissue cells of adulthood that derive from the mesoderm. These are undifferentiated cells susceptible of self-regeneration, proliferation, and cell differentiation.^{18,19} They can also migrate by means of blood transportation. Adult stem cells are pluripotent, and they are found in most tissues. They have also been isolated from deciduous teeth, from apical papilla and from the periodontal ligament.^{20–22}

The periodontal ligament is the fibrous connective tissue structure, with neural and vascular components, that joins the cementum covering root to the alveolar bone.

A small subpopulation of mesenchymal stem cells is held in the periodontal ligament and can provide physiological cells turnover and possibly regeneration of periodontal tissues' structure and function, if needed. These cells can differentiate into tooth cementoblasts, fibroblasts, and osteoblasts to create cementum and periodontal ligament-like tissues and alveolar bone.²²

GBR and GTR procedures are carried out adapting the surgical technique to this orchestrated scenario involving different cells playing different roles at different times.²³ In the present case, a first collagen membrane was, therefore, used to cover the entire root to attempt favoring the proliferation of any residual periodontal ligament cells to colonize the root to regenerate periodontal ligament fibers, new cementum, and new alveolar bone. Further, as the residual bone was not enough to provide effective support, a GBR procedure was also attempted. The bone graft was used as a scaffold for clot stabilization and cellular infiltration, according to the principles of GTR when associated with bone grafting.²⁴ Accordingly, the tooth root needed to be isolated to attempt to prevent root ankylosis. Even the processes involved in new bone regeneration cannot lead to new attachment formation. In fact, when block sections of regenerated bone including dental elements were analyzed, a long junctional epithelium between the root surface and the newly formed bone could be observed, even after grafting autogenous bone; areas of newly formed cementum and functionally oriented and inserted periodontal ligament fibers could be observed only at the very base of the defect, as expected, given its proximity to the periodontal ligament.^{25,26} Nevertheless, when bone grafting is carried out, tooth resorption and ankylosis are seldom observed at the tooth–bone interface; this fact may be explained by either coronal migration of periodontal ligament cells with the formation of a new attachment apparatus and/or apical migration of the junctional epithelium, which acts as a protective barrier preventing root resorption and ankylosis. Concerning the case reported in the present study, no conclusions can be drawn about a possible complete *restitutio ad integrum* of the lost periodontal tissue, as no histological examinations could be conducted.

The last radiograph, recorded at the 18-month follow-up, shows that GBR of part of the alveolar process bone was successful and the root has not undergone resorption. This indicates that GTR probably occurred, specifically affecting the periodontal ligament, therefore, preventing the root from gaining direct contact with the bone tissue, especially in its apical portion. Furthermore, no signs of ankylosis were detectable. If, as an alternative, the associated above-described procedures had led to root ankylosis or root resorption as possible consequences of a direct bone-to-tooth contact (i.e., without interposition of the periodontal ligament), we would have considered the clinical result as a pure GBR and not a GTR.

Accordingly, the treated tooth gained mechanical stability over time, and, at present, it is functional and has been stable over a period of more than 18 months while its probing depth has improved dramatically. The gingival recession has reduced to a level fully meeting acceptable aesthetic requirements and the patient is fully satisfied.

CONCLUSION

The use of an equine collagen membrane with the purpose of creating the best conditions to carry out periodontal regeneration according to GTR principles, in association with an

equine, collagen-preserving, enzyme-treated bone graft covered with a second collagen membrane (GBR procedure) in order to allow sufficient bone regeneration, permitted to salvage a tooth that was deemed otherwise lost because of periodontitis.

CLINICAL SIGNIFICANCE

Basilar principles of GTR should always be applied when treating teeth that are severely compromised by periodontitis using regenerative techniques. The use of collagen membranes according to these principles, in association with a bone substitute having well-known performance rates, might be invaluable to carry out regeneration of the periodontal ligament, beyond that of bone tissue only.

REFERENCES

1. Bhat M, Roberts-Thomson K, et al. Clustering of risk indicators for periodontal disease: a population-based study. *Community Dent Health* 2015 Sep;32(3):158–162.
2. Devi P, Pradeep AR. Classification of periodontal diseases: the dilemma continues. *N Y State Dent J* 2009 Jun–Jul;75(4):30–34.
3. Meynardi F, Pasqualini ME, et al. Correlation between dysfunctional occlusion and periodontal bacterial profile. *J Biol Regul Homeost Agents* 2016 Apr–Jun;30(2 Suppl 1):115–121.
4. Cortellini P, Tonetti MS. Focus on intrabony defects: guided tissue regeneration. *Periodontol* 2000 2000 Feb;22:104–132.
5. Pretzl B, Eickholz P, et al. Endodontic status and retention of molars in periodontally treated patients: results after 10 or more years of supportive periodontal therapy. *J Clin Periodontol* 2016 Dec;43(12):1116–1123. DOI: 10.1111/jcpe.12621.
6. Gottlow J, Nyman S, et al. New attachment formation as the result of controlled tissue regeneration. *J Clin Periodontol* 1984 Sep;11(8):494–503.
7. de Jong T, Bakker AD, et al. The intricate anatomy of the periodontal ligament and its development: lessons for periodontal regeneration. *J Periodontol Res* 2017 Dec;52(6):965–974. DOI: 10.1111/jre.12477.
8. Yamamoto T, Ugawa Y, et al. Modulation of microenvironment for controlling the fate of periodontal ligament cells: the role of Rho/ROCK signaling and cytoskeletal dynamics. *J Cell Commun Signal*. 2018 Mar;12(1):369–378. DOI: 10.1007/s12079-017-0425-3.
9. Nyman S, Karring T, et al. Healing following implantation of periodontitis-affected roots into gingival connective tissue. *J Clin Periodontol* 1980 Oct;7(5):394–401.
10. Karring T, Nyman S, et al. Potentials for root resorption during periodontal wound healing. *J Clin Periodontol* 1984 Jan;11(1):41–52.
11. Caton J, Zander HA. Osseous repair of an infrabony pocket without new attachment of connective tissue. *J Clin Periodontol* 1976 Feb;3(1):54–58.
12. AlGhamdi AS, Ciancio SG. Guided tissue regeneration membranes for periodontal regeneration—a literature review. *J Int Acad Periodontol* 2009 Jul;11(3):226–231.
13. Miller SC. *Textbook of Periodontia*. 3rd ed. Philadelphia, PA: Blakiston; 1950.
14. Löe H. The Gingival Index, the Plaque Index and the Retention Index Systems. *J Periodontol* 1967 Nov–Dec;38(6):Suppl:610–616.
15. Ramfjord SP. The Periodontal Disease Index (PDI). *J Periodontol* 1967 Nov–Dec;38(6):Suppl:602–610.
16. López NJ, Belvederessi M. Healing following implantation of healthy roots, with and without periodontal ligament tissue, in the oral mucosa. *J Periodontol* 1983 May;54(5):283–290. DOI: 10.1902/jop.1983.54.5.283.
17. Chen FM, Sun HH, et al. Stem cell-delivery therapeutics for periodontal tissue regeneration. *Biomaterials* 2012 Sep;33(27):6320–6344.
18. Dominici M, Le Blanc K, et al. Minimal criteria for defining multipotent mesenchymal stromal cells. The International Society for Cellular Therapy position statement. *Cytotherapy* 2006;8(4):315–317.

19. Morrison SJ, Kimble J. Asymmetric and symmetric stem-cell divisions in development and cancer. *Nature* 2006 Jun 29;441(7097):1068–1074. DOI: 10.1038/nature04956.
20. Miura M, Gronthos S, et al. SHED: stem cells from human exfoliated deciduous teeth. *Proc Natl Acad Sci U S A* 2003 May 13;100(10):5807–5812. DOI: 10.1073/pnas.0937635100.
21. Sonoyama W, Liu Y, et al. Characterization of the apical papilla and its residing stem cells from human immature permanent teeth: a pilot study. *J Endod* 2008 Feb;34(2):166–171. DOI: 10.1016/j.joen.2007.11.021.
22. Seo BM, Miura M, et al. Investigation of multipotent postnatal stem cells from human periodontal ligament. *Lancet* 2004 Jul 10–16;364(9429):149–155. DOI: 10.1016/S0140-6736(04)16627-0.
23. Hernández-Monjaraz B, Santiago-Osorio E, et al. Mesenchymal Stem Cells of Dental Origin for Inducing Tissue Regeneration in Periodontitis: A Mini-Review. *Int J Mol Sci* 2018 Mar 22;19(4):1–17. DOI: 10.3390/ijms19040944.
24. Pellegrini G, Pagni G, et al. Surgical Approaches Based on Biological Objectives: GTR vs GBR Techniques. *Int J Dent* 2013;2013:521547. DOI: 10.1155/2013/521547.
25. Moskow BS, Karsh F, et al. Histological assessment of autogenous bone graft. A case report and critical evaluation. *J Periodontol* 1979 Jun;50(6):291–300. DOI: 10.1902/jop.1979.50.6.291.
26. Listgarten MA, Rosenberg MM. Histological study of repair following new attachment procedures in human periodontal lesions. *J Periodontol* 1979 Jul;50(7):333–344. DOI: 10.1902/jop.1979.50.7.333.

