REVIEW ARTICLE

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# Stem cells in craniofacial and dental tissue engineering

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**Abstract**

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Mesenchymal stem cells (MSC) have been identified in a variety of adult tissues as a population of pluripotential self-renewing cells. Based on their adherence and colony forming properties, a small number of MSC can be isolated from most mesenchymal tissues as well as bone marrow. In the presence of one or more growth factors, these cells commit to lineages that lead to the formation of bone, cartilage, muscle, tendon and adipose tissue; recent studies indicate that stem cells for cementum, dentine and the periodontal ligament also exist. All of these cells can be expanded *in vitro*, and, embedded in a scaffold, inserted into defects to promote healing and tissue replacement. Increased understanding of the molecular mechanism directing lineage specification and morphogenesis is providing a rational approach for the regeneration of craniofacial tissues and oral structures.

**Key words:** bone; craniofacial biology; mesenchymal stem cells; tissue engineering

## Introduction

Enforcement of laws that forbid embryonic stem cell research has energized studies of the use of adult cells to regenerate and reconstruct the craniofacial apparatus. In most cases, this is achieved by transplanting to the diseased site a complex of bioactive molecules, a supportive scaffold and a progenitor cell population (1,2). Ongoing investigations suggest that the progenitor cells are present in mature skeletal and dental tissues. This population of self-renewing stem cells, termed mesenchymal stem cells (MSC), is capable of driving postnatal growth, and orchestrating repair and regeneration.

Surprisingly, unequivocal evidence supporting the existence of stem cells *in vivo* has yet to be demonstrated. However, many studies, especially those that relate to the necessity of stromal cells maintaining hematopoiesis, indicate that MSC exist, and serve a functional role in the adult organism (3). Indeed, all cell-based therapeutic strategies are based on the assumption that in a specific tissue, in response to molecular cues, a small population of self-renewing MSC can reconstitute the parent tissue.

A major focus of contemporary studies in developmental biology has been to delineate the biological cues that drive stem cell proliferation and differentiation (4). Four signaling protein families that govern patterning and morphogenesis have been identified: fibroblast growth factors, hedgehog proteins, bone morphogenetic proteins, and wingless- and int-related proteins (Wnts) (5). Proteins from each of these families are now being evaluated for their utility for stem cell based engineering of craniofacial defects (6,7). Recently, these applications have been extended to address the ravages of endodontic and periodontal disease, as well as serving as adjunct therapy for oral-maxillofacial and alveolar ridge surgery and augmentation and repair of lesions of the temporomandibular joint. Details of the identification and use of stem cells for periodontal and orthognathic surgery are discussed below.

## MSC identification and localization

The painstaking studies of the Russian scientist, Alexander Friedenstein provided much of the basic knowledge of MSC biology (8,9). Over a period of two decades, Friedenstein and co-workers demonstrated that bone marrow derived adherent cells were capable of committing to a number of lineages, including those responsible for osteogenesis. To identify and quantitate tissue MSC levels, the colony forming unit-fibroblast (CFU-F) assay is commonly utilized (10). While of great practical value, it is unlikely that this *in vitro* assay provides an accurate assessment of the number of stem cells that colonize a specific tissue niche. Nevertheless, it is generally agreed that progenitor cell number in adult tissues is very low: for example, in adult bone marrow there is one MSC/$10^4$ –$10^6$ total cells (11). Although there is some dispute about the effect of patient's age on MSC number, it is likely that stem cell quantity decreases with age (12).

Aside from the ability to form CFU, more definitive characteristics of the MSC include: expression of a large number of proteins (antigens) on the cell surface (CD44, CD71, CD90, CD105, CD120a, CD124, CD166 and Flt-3 and Kit ligands) and absence of antigens specific for cells of the hematopoietic lineage (13). In addition, these cells secrete a cassette of cytokines (IL-6, -7, -8, -11, -12, -14, -15, LIF, GM-CSF) (14). These proteins can direct the commitment of the MSC into one of a number of different differentiation pathways. Ultimately, commitment and differentiation is dependent on the biological characteristics of the tissue niche itself. Viewed from this perspective, the current focus on niche and MSC specific marker protein analysis represents a critical arena of stem cell research.

## Use of MSC for dental and craniofacial tissue-engineering

Depending on environmental cues, MSC have the ability to differentiate (commit) to pathways that lead to the formation of bone, cartilage, fat, muscle and tendon (15,16). For dental and craniofacial tissue

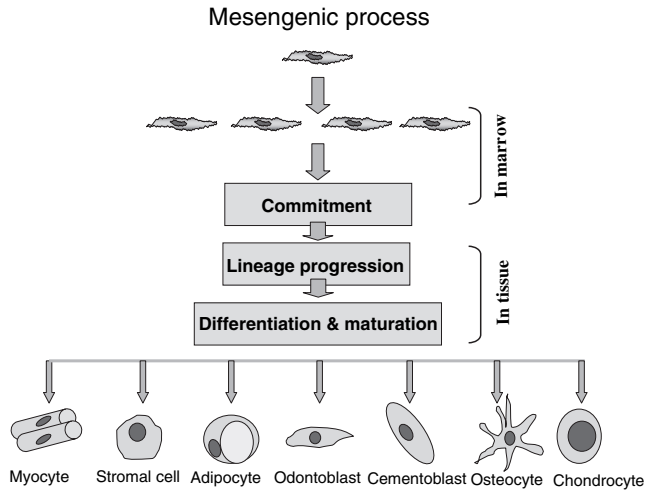


*Fig. 1.* Cartoon showing differentiation of MSC into cells of skeletal and dental tissues. The cartoon shows that MSC can commit to a number of different pathways and assume the phenotype of cells of muscle, bone, cartilage, fat, ligament and cementum. Commitment to a particular lineage is driven by the presence of local morphogenic factors. Lineage-committed cells progress through a number of transitory stages. Once differentiation is initiated, proliferation is down regulated and there is biosynthesis of tissue specific proteins. It is thought that MSC are present in all organs of the body where they serve to maintain tissue homeostasis.

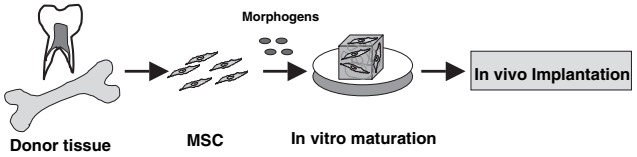


*Fig. 2.* A flow diagram showing the strategy utilized for engineering MSC to regenerate damaged/diseased tissue. MSC are isolated from donor tissue (bone marrow or dental tissues) and cultured on biodegradable scaffolds in the presence of factors (morphogens) that support their differentiation into cells of the target tissue. This cell-scaffold construct is then transplanted into the patient to enhance tissue regeneration.

engineering, stem cells are used to generate osteoblasts, chondrocytes and periodontal ligament cells; more recently there has been interest in engineering odontoblasts and cementocytes (see Fig. 1). To promote osteogenesis, cells can be harvested from a number of autologous sources including bone marrow and fat, without significant donor site morbidity or immunogenic response (17). Although vanishingly small in number, they can be expanded in culture to produce an adequate numbers of cells for tissue engineering strategies (see Fig. 2). One advantage of using committed MSC is that compared with resident differentiated cells (e.g. osteocytes or chondrocytes), which appear to be metabolically quiescent, these committed cells display a high biosynthetic response. A final advantage of using these progenitor cells is that MSC do not display the same surface antigen profile as mature cells. Accordingly, they can be used allogeneically as a therapeutic cell source.

Because of current concerns about HIV and other lethal virus infections, autologous cell transplantation therapy is now desirable. An obvious benefit of cell therapy is that MSC can be harvested directly from the patient, prior to tissue grafting, thereby eliminating worries about infection, and with minimum complications associated with immune rejection of allogenic tissue. Based on all of these considerations, tissue engineering, using the patient's own cells, offers a number of clear advantages over conventional therapy or genetic engineering using viral vectors.

#### Craniofacial applications

Bone marrow derived MSC are now under consideration for the repair of craniofacial bone and even the replacement or regeneration of oral tissues. Commonly, osseous defects are because of post-cancer ablative surgery, trauma, congenital malformations and progressive skeletal disease (18,19). These defects may be treated with autogenous bone grafts and/or alloplastic materials (20,21). Reconstruction of craniofacial and dental defects using MSC avoids many of the limitations of both auto- and allografting techniques (22). Studies using experimental animal models have shown the utility of stem cell based craniofacial regeneration procedures (23,24). From a practical viewpoint, the basis for all of these procedures is that stem cells are seeded onto an appropriate scaffold material. Following proliferation and differentiation, the hybrid is transplanted into the bone defect (Fig. 2). Subsequent evaluation of the transplanted tissue shows that the MSC generate a powerful osteogenic response.

Abukawa *et al.* used a novel scaffold design, with new fabrication protocol, to generate an autologous tissue-engineered construct. The scaffold was then used to repair a segmental mandibular defect. The tissue engineered construct promoted osteogenesis and enhanced penetration of the bone with blood vessels, thereby accelerating tissue regeneration (25). In an experimental dog model, Yamada *et al.* showed that a mixture of autologous MSC and platelet rich plasma improved bone-implant contact and bone density in a mandibular defect (26).

Development of new scaffold fabrication technologies has facilitated the repair of critical-sized and three-dimensionally complex cranial defects (27). Using a rapid prototyping technology, cell-scaffold constructs have been prepared with a high cell:matrix ratio, permeated by a dense vascular network. Mechanical testing of the reconstructed area revealed partial integration with the surrounding calvarial bone (28). Mechanically, these constructs achieved yield strength of about 85–90% of normal bone (28). Recently, it was shown that the patient's own tissues could be utilized to synthesize a bone-tissue substitute (29). In this study, an extended mandibular discontinuity defect was repaired by ossification of a custom-designed bone transplant implanted within the latissimus dorsi muscle of an adult male patient. After 7 weeks, the implant was then used to repair the mandibular defect. New bone formation was reported and the patient displayed improved mastication (29).

To further enhance the regenerative potential of MSC, genetic engineering technologies have been

utilized (30). Thus, to extend the life span of MSC, and to enhance osteogenesis, cells have been engineered with human telomerase reverse transcriptase (30,31). The telomerase has also been shown to activate osteogenesis by fat-derived MSC (32). In a recent study, the sonic hedgehog gene was transfected into marrow and fat-derived MSC to repair a cranial bone defect (33). Quantitative analysis of the new tissue confirmed that there was a significant increase in bone regeneration by the gene-enhanced cells (33). In summary, cell-derived therapy that is focused on the repair of osseous defects has been enormously successful. Because of the overwhelming success of these animal studies, numerous clinical trails are now in progress to treat human craniofacial defects.

#### Dental pulp applications

Unlike bone, dentin is not remodeled throughout life. However, since there is evidence of limited repair, it was hypothesized that progenitor cells present in the dental pulp differentiate into odontoblasts. Gronthos *et al.* have isolated highly proliferative cells from adult human dental pulp that exhibit a similar immunophenotype to bone marrow derived MSC. Importantly, in culture these cells display high alkaline phosphatase activity and form densely calcified nodule (34). *In vivo* transplantation experiments showed that these cells can form a dentin-like structure. In contrast to bone marrow derived stem cells, the pulp cells do not support the formation of a marrow or adipocytes, elements that are lacking in the dental pulp itself (34). Recently, multipotential stem cells were isolated from exfoliated human deciduous teeth (35). The reparative potential of these cells is new being scrutinized.

#### Cementoblast-like cells applications

Despite profound differences in the organization of bone and cementum, it is not clear if these mineralized tissues are formed by two distinct cell types, or by an osteoblast-like cell that responds to environmental signals that are characteristic of a dental niche. Differentiating between these two possibilities has been difficult, partly because of a lack of specific markers for cementocytes. However, since human (36) and murine (37) cementum-derived cells have been isolated from healthy teeth it may be possible to answer this difficult problem using genomics and proteomic techniques. Very recently, Sato *et al.* have developed a bovine cementoblast progenitor line (38). These cells were transplanted subcutaneously into nude mice on a hydroxyapatite/tricalcium phosphate scaffold. Histological analysis indicated that a bone-like tissue was formed containing cementocyte-like cells in a mineralized matrix. Finally, it is worth noting that periodontal ligament itself may serve as a source of cells for cementum formation (36,39). Seo *et al.* reported isolation of multipotential stem cells from the human periodontal ligament. These cells displayed stem cell characteristics in that they differentiated into cementoblast-like cells or adipocytes (40).

#### Periodontal regeneration

The literature on this topic is voluminous and a critical analysis is beyond the scope of this review. However, it is evident that the ligament complex contains stem cells that can commit to a number of pathways (bone, cementum and ligament). Moreover, the cells respond to inductive factors that include members of the TGF-$\beta$ superfamily such as BMP-2 (41–44) BMP-12 (45), BMP-7 (46), TGF-$\beta$ (47), PDGF (48) and b-FGF (49). In an exciting recent study, Kawaguchi *et al.* used autologous bone marrow MSC in combination with atelocollagen to regenerate ligament in an experimental Class III defect in dogs (50). One month after implantation, there was regeneration of cementum, periodontal ligament, and alveolar bone. This study provided firm evidence that MSC embedded in the appropriate environmental niche can be used to regenerate a tissue as complex as the periodontium.

## Conclusions and future directions

Although our understanding of the molecular pathways underlying MSC differentiation is expanding, translation of this knowledge into tissue engineering strategies remains in its infancy. For this reason, research efforts are focused on identifying factors that regulate and control MSC proliferation and commitment. In the context of orofacial tissue engineering, populations of stem cells that form bone, cementum, dentin, and even periodontal ligament have been identified. Within the

next few years, these cells will be used to restore the form and function of the oral cavity using autologous cells, thereby circumventing histocompatability mismatch and transmission of viral disease.

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## References


1. Sittinger M, Hutmacher DW, Risbud MV. Current strategies for cell delivery in cartilage and bone regeneration. *Curr Opin Biotechnol* 2004;**15**:411–8.
2. Risbud MV, Shapiro IM, Vaccaro AR, Albert TJ. Stem cell regeneration of the nucleus pulposus. *Spine J* 2004;**4**:348S–353S.
3. Majumdar MK, Thiede MA, Haynesworth SE, Bruder SP, Gerson SL. Human marrow-derived mesenchymal stem cells (MSCs) express hematopoietic cytokines and support long-term hematopoiesis when differentiated toward stromal and osteogenic lineages. *J Hematother Stem Cell Res* 2000;**9**:841–8.
4. Thesleff I, Sharpe P. Signalling networks regulating dental development. *Mech Dev* 1997;**67**:111–3.
5. Etheridge SL, Spencer GJ, Heath DJ, Genever PG. Expression profiling and functional analysis of wnt signaling mechanisms in mesenchymal stem cells. *Stem Cells* 2004;**22**:849–60.
6. Nakao K, Itoh M, Tomita Y, Tomooka Y, Tsuji T. FGF-2 potently induces both proliferation and DSP expression in collagen type I gel cultures of adult incisor immature pulp cells. *Biochem Biophys Res Commun* 2004;**325**:1052–9.
7. Moutsatsos IK, Turgeman G, Zhou S, Kurkalli BG, Pelled G, Tzur L et al. Exogenously regulated stem cell-mediated gene therapy for bone regeneration. *Mol Ther* 2001;**3**:449–61.
8. Friedenstein AJ, Deriglasova UF, Kulagina NN, Panasuk AF, Rudakowa SF, Luria EA et al. Precursors for fibroblasts in different populations of hematopoietic cells as detected by the in vitro colony assay method. *Exp Hematol* 1974;**2**:83–92.
9. Luria EA, Panasyuk AF, Friedenstein AY. Fibroblast colony formation from monolayer cultures of blood cells. *Transfusion* 1971;**11**:345–9.
10. Clarke E. Culture of human and mouse mesenchymal cells. *Methods Mol Biol* 2005;**290**:173–86.
11. Kadiyala S, Young RG, Thiede MA, Bruder SP. Culture expanded canine mesenchymal stem cells possess osteochondrogenic potential in vivo and in vitro. *Cell Transplant* 1997;**6**:125–34.
12. Bonyadi M, Waldman SD, Liu D, Aubin JE, Grynpas MD, Stanford WL. Mesenchymal progenitor self-renewal deficiency leads to age-dependent osteoporosis in Sca-1/Ly-6A null mice. *Proc Natl Acad Sci U S A* 2003;**100**:5840–5.
13. Campagnoli C, Roberts IA, Kumar S, Bennett PR, Bellantuono I, Fisk NM. Identification of mesenchymal stem/progenitor cells in human first-trimester fetal blood, liver, and bone marrow. *Blood* 2001;**98**:2396–402.
14. Majumdar MK, Thiede MA, Mosca JD, Moorman M, Gerson SL. Phenotypic and functional comparison of cultures of marrow-derived mesenchymal stem cells (MSCs) and stromal cells. *J Cell Physiol* 1998;**176**:57–66.
15. Gerson SL. Mesenchymal stem cells: no longer second class marrow citizens. *Nat Med* 1999;**5**:262–4.
16. Pittenger MF, Mackay AM, Beck SC, Jaiswal RK, Douglas R, Mosca JD et al. Multilineage potential of adult human mesenchymal stem cells. *Science* 1999;**284**:143–7.
17. Bartholomew A, Sturgeon C, Siatskas M, Ferrer K, McIntosh K, Patil S et al. Mesenchymal stem cells suppress lymphocyte proliferation in vitro and prolong skin graft survival in vivo. *Exp Hematol* 2002;**30**:42–8.
18. Jeffcoat MK. Bone loss in the oral cavity. *J Bone Miner Res* 1993;**8** (Suppl. 2):S467–73.
19. Phillips JH, Forrest CR, Gruss JS. Current concepts in the use of bone grafts in facial fractures. Basic science considerations. *Clin Plast Surg* 1992;**19**:41–58.
20. Jackson IT, Helden G, Marx R. Skull bone grafts in maxillofacial and craniofacial surgery. *J Oral Maxillofac Surg* 1986;**44**:949–55.
21. Oklund SA, Prolo DJ, Gutierrez RV, King SE. Quantitative comparisons of healing in cranial fresh autografts, frozen autografts and processed autografts, and allografts in canine skull defects. *Clin Orthop Relat Res* 1986;**205**:269–91.
22. Bianco P, Robey PG. Stem cells in tissue engineering. *Nature* 2001;**414**:118–21.
23. Krebsbach PH, Mankani MH, Satomura K, Kuznetsov SA, Robey PG. Repair of craniotomy defects using bone marrow stromal cells. *Transplantation* 1998;**66**:1272–8.
24. Quarto R, Mastrogiacomo M, Cancedda R, Kutepov SM, Mukhachev V, Lavroukov A et al. Repair of large bone defects with the use of autologous bone marrow stromal cells. *N Engl J Med* 2001;**344**:385–6.
25. Abukawa H, Shin M, Williams WB, Vacanti JP, Kaban LB, Troulis MJ. Reconstruction of mandibular defects with autologous tissue-engineered bone. *J Oral Maxillofac Surg* 2004;**62**:601–6.
26. Yamada Y, Ueda M, Naiki T, Nagasaka T. Tissue-engineered injectable bone regeneration for osseointegrated dental implants. *Clin Oral Implants Res* 2004;**15**:589–97.
27. Hutmacher DW, Sittinger M, Risbud MV. Scaffold-based tissue engineering: rationale for computer-aided design and solid free-form fabrication systems. *Trends Biotechnol* 2004;**22**:354–62.
28. Schantz JT, Hutmacher DW, Lam CX, Brinkmann M, Wong KM, Lim TC et al. Repair of calvarial defects with customised tissue-engineered bone grafts II. Evaluation of cellular efficiency and efficacy in vivo. *Tissue Eng* 2003;**9**:S127–39.
29. Warnke PH, Springer IN, Wiltfang J, Acil Y, Eufinger H, Wehmoller M et al. Growth and transplantation of a custom vascularised bone graft in a man. *Lancet* 2004;**364**:766–70.
30. Shi S, Gronthos S, Chen S, Reddi A, Counter CM, Robey PG et al. Bone formation by human postnatal bone marrow stromal stem cells is enhanced by telomerase expression. *Nat Biotechnol* 2002;**20**:587–91.
31. Gronthos S, Chen S, Wang CY, Robey PG, Shi S. Telomerase accelerates osteogenesis of bone marrow stromal stem cells by upregulation of CBFA1, osterix, and osteocalcin. *J Bone Miner Res* 2003;**18**:716–22.
32. Jun ES, Lee TH, Cho HH, Suh SY, Jung JS. Expression of telomerase extends longevity and enhances differentiation in human adipose tissue-derived stromal cells. *Cell Physiol Biochem* 2004;**14**:261–8.
33. Edwards PC, Ruggiero S, Fantasia J, Burakoff R, Moorji SM, Paric E et al. Sonic hedgehog gene-enhanced tissue engineering for bone regeneration. *Gene Ther* 2005;**12**:75–86.

34. Gronthos S, Mankani M, Brahim J, Robey PG, Shi S. Postnatal human dental pulp stem cells (DPSCs) in vitro and in vivo. *Proc Natl Acad Sci U S A* 2000;**97**:13625–30.
35. Miura M, Gronthos S, Zhao M, Lu B, Fisher LW, Robey PG et al. SHED: stem cells from human exfoliated deciduous teeth. *Proc Natl Acad Sci U S A* 2003;**100**:5807–12.
36. Grzesik WJ, Kuzentsov SA, Uzawa K, Mankani M, Robey PG, Yamauchi M. Normal human cementum-derived cells: isolation, clonal expansion, and in vitro and in vivo characterization. *J Bone Miner Res* 1998;**13**:1547–54.
37. D'Errico JA, Ouyang H, Berry JE, MacNeil RL, Strayhorn C, Imperiale MJ et al. Immortalized cementoblasts and periodontal ligament cells in culture. *Bone* 1999;**25**:39–47.
38. Saito M, Handa K, Kiyono T, Hattori S, Yokoi T, Tsubakimoto T et al. Immortalization of cementoblast progenitor cells with Bmi-1 and TERT. *J Bone Miner Res* 2005;**20**:50–7.
39. Grzesik WJ, Cheng H, Oh JS, Kuznetsov SA, Mankani MH, Uzawa K et al. Cementum-forming cells are phenotypically distinct from bone-forming cells. *J Bone Miner Res* 2000;**15**:52–9.
40. Seo BM, Miura M, Gronthos S, Bartold PM, Batouli S, Brahim J et al. Investigation of multipotent postnatal stem cells from human periodontal ligament. *Lancet* 2004;**364**:149–55.
41. Kinoshita A, Oda S, Takahashi K, Yokota S, Ishikawa I. Periodontal regeneration by application of recombinant human bone morphogenetic protein-2 to horizontal circumferential defects created by experimental periodontitis in beagle dogs. *J Periodontol* 1997;**68**:103–9.
42. Wikesjo UM, Guglielmoni P, Promsudthi A, Cho KS, Trombelli L, Selvig KA et al. Periodontal repair in dogs: effect of rhBMP-2 concentration on regeneration of alveolar bone and periodontal attachment. *J Clin Periodontol* 1999;**26**:392–400.
43. Wikesjo UM, Xiropaidis AV, Thomson RC, Cook AD, Selvig KA, Hardwick WR. Periodontal repair in dogs: rhBMP-2 significantly enhances bone formation under provisions for guided tissue regeneration. *J Clin Periodontol* 2003;**30**:705–14.
44. Selvig KA, Sorensen RG, Wozney JM, Wikesjo UM. Bone repair following recombinant human bone morphogenetic protein-2 stimulated periodontal regeneration. *J Periodontol* 2002;**73**: 1020–9.
45. Sorensen RG, Polimeni G, Kinoshita A, Wozney JM, Wikesjo UM. Effect of recombinant human bone morphogenetic protein-12 (rhBMP-12) on regeneration of periodontal attachment following tooth replantation in dogs. *J Clin Periodontol* 2004;**31**:654–61.
46. Giannobile WV, Ryan S, Shih MS, Su DL, Kaplan PL, Chan TC. Recombinant human osteogenic protein-1 (OP-1) stimulates periodontal wound healing in class III furcation defects. *J Periodontol* 1998;**69**:129–37.
47. Wikesjo UM, Razi SS, Sigurdsson TJ, Tatakis DN, Lee MB, Ongpipattanakul B, Nguyen T, Hardwick R. Periodontal repair in dogs: effect of recombinant human transforming growth factor-beta1 on guided tissue regeneration. *J Clin Periodontol* 1998;**25**:475–81.
48. Nevins M, Camelo M, Nevins ML, Schenk RK, Lynch SE. Periodontal regeneration in humans using recombinant human platelet-derived growth factor-BB (rhPDGF-BB) and allogenic bone. *J Periodontol* 2003;**74**:1282–92.
49. Murakami S, Takayama S, Kitamura M, Shimabukuro Y, Yanagi K, Ikezawa K et al. Recombinant human basic fibroblast growth factor (bFGF) stimulates periodontal regeneration in class II furcation defects created in beagle dogs. *J Periodontal Res* 2003;**38**:97–103.
50. Kawaguchi H, Hirachi A, Hasegawa N, Iwata T, Hamaguchi H, Shiba H et al. Enhancement of periodontal tissue regeneration by transplantation of bone marrow mesenchymal stem cells. *J Periodontol* 2004;**75**:1281–7.