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CELL SHEET ENGINEERING AND REGENERATIVE MEDICINE IN PERIODONTAL REGENERATION- A NEW PERSPECTIVE

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KEYWORDS :

INTRODUCTION-

In regenerative medicine, the success and application of tissue engineering technique has shown significant potential in the past 10–15 years, however, cell-based therapies have been clinically used for <40 years.

Multiple treatment options using conventional tissue engineering rationales have yielded clinical products that have their utilization in bone, cartilage and blood vessels.[1] Therefore, these results features the emergence of regenerative therapies using techniques that are superior to simple cell injection.

Periodontitis, a highly prevalent inflammatory infectious disease, commonly induces tissue destruction of the periodontal complex in humans. This disease is initiated by bacterial products such as lipopolysaccharide (LPS), which can stimulate cytokines to signal precursor cells to differentiate and activate osteoclastic cells and/or the periodontal inflammatory process by bacterial biofilm.

Various non-surgical periodontal therapies have been still performed such as scaling, root planning, or anti-infective therapies to eliminate inflammation, periodontitis-causing bacteria, and microbial biofilm. However, the periodontal surgery is critically required for periodontal health and highly expected to regenerate tooth-supportive structures and restore periodontal functions such as pocket reduction procedures and regeneration procedures (guided tissue regeneration; GTR or guided bone regeneration;GBR). In severe cases of periodontitis, dental prosthetics including dental implants or dentures are commonly required after tooth-extraction socket healing and bone regeneration; these approaches use various bioactive molecules, osteoconductive biomaterial fillings, or stem cell therapies to regenerate implant-supportive bone tissues. Therefore, stem cell-based approaches for periodontal regeneration have been studied and translated into clinical settings.

"Cell Sheet Engineering" was developed as an alternative technology for cell transplantation using temperature responsive culture dishes.

METHODS FOR HARVESTING CELL SHEETS

Temperature-responsive culture dishes. Okano et al[2], developed an alternative cell sheet technique for regenerative therapies using temperature-responsive culture dishes. The temperature-responsive polymer poly(N-isopropylacrylamide) (PIPAAm) can undergo a distinct transition from hydrophobic to hydrophilic at its lower critical solution temperature of 32˚C. Thus, PIPAAm was immobilized covalently on ordinary tissue culture polystyrene (TCPS) surfaces at a nanometer-scale thickness, and subsequently cell adhesion and detachment can be controlled by simple temperature changes[3]. On these surfaces, there were no differences in cell adherence, spread or proliferation of various types of cells compared with normal TCPS at 37˚C. Following this, cultured cells can be detached, in the form of a cell sheet, by reducing the incubation temperature, with the conversion of the coated PIPAAm from hydrophobic to hydrophilic. Varieties of cell sheets from different sources were harvested using the temperature-responsive culture dishes and the method of harvesting cell sheets is currently used widely in research. Additionally, overlapping cell sheets can be harvested and multilayered cell sheets can be constructed for tissue repair.

POLYMERIZED HUMAN FIBRIN-COATED DISHES:

Itabashi et al[4] cultured rat cardiomyocytes on polymerized human fibrin-coated dishes, prepared with fibrinogen monomers mixed with thrombin. As the fibrin had been digested by the intrinsic protease, the cell sheet could be readily dissociated intact from the polymerized fibrin layer. Cardiomyocytes cultured on these dishes formed a myocardial cell sheet within 4 days.

Vitamin C treatment:

Wei et al[5] cultured periodontal ligament stem cells treated with vitamin C (Vc) at various concentrations. The cells form cell sheet structures at concentrations of Vc >20 μg/ml, as Vc can increase cell matrix production. However, lower concentrations of Vc (0.0, 5.0 or 10.0 μg/ml) did not result in the formation of sheets.

Periodontal regeneration:

Tsumanuma et al[6] performed cell sheet transplantation using three types of mesenchymal tissue-derived cells (periodontal ligament, alveolar periosteum and bone marrow) to compare differences between cell sources in a canine severe defect (one-wall intrabony defect) model. Three-layered cell sheets from each cell source, supported with woven polyglycolic acid, were transplanted autologously to the denuded root surface. One-wall intrabony defects were filled with a mixture of β-TCP and collagen. Periodontal regeneration with newly formed cementum and well-oriented PDL fibers and alveolar bone regeneration was identified in all the groups 8 weeks after transplantation. The highest amount of bone and periodontal regeneration was revealed in the PDL sheet group.

MSC sheets for bone regeneration:

MSCs are multipotent cells that have positive roles in bone regeneration. In a previous study, MSCs combined with various materials were shown to regenerate bone defects using cell suspension systems.[7] However, the adhesion rate of MSCs is low due to the low surface area to volume ratio of scaffolds. The cell sheet technique can achieve highly efficient cell delivery.[8] Additionally, the adhesion molecules on the cell surface and cell-cell interactions remain intact. Furthermore, osteoblasts attached to the mineralized layered cell sheet may mimic the in vivo deposition of bone matrix.

MSC sheets assembled on demineralized bone grafts or frozen tendon grafts by a wrapping technique were cultured for 3 weeks. When assembled with demineralized bone matrix, the MSC sheet was similar to the in situ periosteum and could differentiate into the osteochondral lineage.

When assembled with a frozen tendon graft, the MSC sheet was incorporated within the tissue sheath (peritenon) around the tendon and adopted the characteristic spindle-shaped morphology of tenocyte-like cells. This study further verified the potential of in situ-specific differentiation of MSC sheet cells.

the high morbidity associated with artificial joint loosening, promoting bonding behavior and increasing bone ingrowth between bone and implant is important for the stability of the joint. A novel MSC sheet-wrapped implant can promote bone bonding at the bone-implant interface by providing cells and ECM.

In the study of Zhou et al[9], multilayered rabbit BMSC sheets were assembled with two types of implant (surface-modified titanium and zirconia) to construct an MSC sheet-implant complex. After culturing in osteogenic medium for 2 weeks, two types of cell sheet-implant complex were transplanted under the skin of SCID mice for 8 weeks. For the MSC sheet-titanium implant complex, new bone tissue that formed around the implants followed a predominantly endochondral pathway, exhibiting histological markers of native bone. Intramembranous ossification appeared to occur on the surface of the

zirconia implant, as observed with typical osteocytes embedded in a dense matrix and accompanied by microvessels and marrow cavities in the MSC sheet-zirconia implant complex. Similarly, Yu et al[10] harvested MSC sheets and wrapped them around titanium implants to construct complexes, followed by culturing in an osteogenic medium. The effect of an MSC sheet-implant complex in the right tibia of a diabetic rat model was investigated. The MSC sheet-implant complex showed a significantly higher bone volume ratio and trabecular thickness ($P<0.05$) and significantly less trabecular separation ($P<0.05$) compared to the titanium implants in the diabetic rats 4 and 8 weeks later. Additionally, the amount of new bone tissue forming around the MSC-implant complexes was higher in comparison to with the titanium implants.

Cell sheet engineering techniques in conjunction with routine implant materials can generate MSC-implants possessing osteogenic and vascularization abilities. Therefore, the MSC-implant complexes provide a novel tissue engineering approach that promotes osseous healing and may be useful in the treatment of patients with diabetes

CONCLUSION:

The applications of cell sheet engineering for regenerative medicine should prove useful as a fundamental, generalized technique. Various types of cells particularly MSC have been examined and most of them improved the functions of recipients, suggesting that cell sheet engineering can be an alternative tool for the therapy of tissue engineering.

REFERENCES:

1. CHEN G, QI Y, NIU L, DI T, ZHONG J, FANG T, YAN W. Application of the cell sheet technique in tissue engineering. Biomedical Reports 2015 3: 749-757.
2. Okano T, Yamada N, Sakai H and Sakurai Y: A novel recovery system for cultured cells using plasma-treated polystyrene dishes grafted with poly(N-isopropylacrylamide). J Biomed Mater Res 27: 1243-1251, 1993.
3. Okano T, Yamada N, Okuhara M, Sakai H and Sakurai Y: Mechanism of cell detachment from temperature-modulated, hydrophilic-hydrophobic polymer surfaces. Biomaterials 16: 297-303, 1995.
4. Itabashi Y, Miyoshi S, Kawaguchi H, Yuasa S, Tanimoto K,,Furuta A, Shimizu T, Okano T, Fukuda K and Ogawa S: A new method for manufacturing cardiac cell sheets using fibrin-coated dishes and its electrophysiological studies by optical mapping. Artif Organs 29: 95-103, 2005.
5. Wei F, Qu C, Song T, Ding G, Fan Z, Liu D, Liu Y, Zhang C, Shi S and Wang S: Vitamin C treatment promotes mesenchymal stem cell sheet formation and tissue regeneration by elevating telomerase activity. J Cell Physiol 227: 3216-3224, 2012.
6. Tsumanuma Y, Iwata T, Washio K, Yoshida T, Yamada A, Takagi R, Ohno T, Lin K, Yamato M, Ishikawa I, et al: Comparison of different tissue-derived stem cell sheets for periodontal regeneration in a canine 1-wall defect model. Biomaterials 32: 5819-5825, 2011.
7. Dumas A, Moreau MF, Ghérardi RK, Baslé MF and Chappard D: Bone grafts cultured with bone marrow stromal cells for the repair of critical bone defects: An experimental study in mice. J Biomed Mater Res A 90: 1218-1229, 2009.
8. Yang J, Yamato M, Kohno C, Nishimoto A, Sekine H, Fukai F and Okano T: Cell sheet engineering: Recreating tissues without biodegradable scaffolds. Biomaterials 26: 6415-6422, 2005.
9. Zhou W, Han C, Song Y, Yan X, Li D, Chai Z, Feng Z, Dong Y, Li L, Xie X, et al: The performance of bone marrow mesenchymal stem cell - implant complexes prepared by cell sheet engineering techniques. Biomaterials 31: 3212-3221, 2010.
10. Yu M, Zhou W, Song Y, Yu F, Li D, Na S, Zou G, Zhai M and Xie C: Development of mesenchymal stem cell-implant complexes by cultured cells sheet enhances osseointegration in type 2 diabetic rat model. Bone 49: 387-394, 2011.