We are IntechOpen, the world's leading publisher of Open Access books Built by scientists, for scientists

4,800
Open access books available

122,000
International authors and editors

135M
Downloads

Our authors are among the

154
Countries delivered to

TOP 1%
most cited scientists

12.2%
Contributors from top 500 universities

CLARIVATE ANALYTICS BOOK CITATION INDEX INDEXED

WEB OF SCIENCE™

Selection of our books indexed in the Book Citation Index in Web of Science™ Core Collection (BKCI)

Interested in publishing with us?
Contact book.department@intechopen.com

Numbers displayed above are based on latest data collected.
For more information visit www.intechopen.com

Chapter 4

Cartilage

Cartilage regeneration using chondrocyte

Cartilage tissue engineering comprises three factors: cell source, growth factors, and scaffolds. Chondrocytes from other cartilage such as rib cartilage are most commonly used for the formation of cartilage tissue [1]. However, their cell number is limited and it is difficult to construct a tissue of large size. Differentiation of embryonic stem cells toward chondrocytes has been accomplished, but its clinical application is impractical at present from ethical points of view [2, 3]. In contrast, MSCs are promising because they can easily be prepared from patients without invasive surgery. These cells grow rapidly, retaining their capacity to differentiate into chondrocytes under certain conditions [4, 5]. Several growth factors such as TGF-β, BMPs, FGFs, and IGFs are involved in chondrocyte differentiation, proliferation, and maintenance [6]. These molecules are used for cartilage tissue engineering. The application of scaffolds has two advantages in this type of engineering. It enables three-dimensional culture, which is a necessary microenvironment for maturing the chondrocyte phenotype. It serves as an artificial matrix that gradually becomes replaced with native cartilage matrix. Although several methods have been attempted, with consideration of these factors, no cartilage tissue has been engineered that fulfills clinical requirements. There has been accumulating evidence that stimulation of chondrocytes facilitates cartilage matrix formation [7].

For instance, hydrostatic pressure on bovine chondrocytes is known to enhance their matrix synthesis and accumulation [8, 9]. Direct compression on bovine chondrocytes embedded in agarose gel increases glycosaminoglycan and collagen composition [10]. Thus, stress may serve as another important factor in cartilage tissue engineering. In clinical studies and animal models, low-intensity ultrasound (US) promotes fracture repair and increases mechanical strength. US also promotes cartilage healing by increasing glycosaminoglycan synthesis of chondrocytes. As MSCs have the ability to differentiate into chondrocytes, US may promote their differentiation. Here, we evaluated the effects of US on the differentiation of MSCs toward chondrocytes and cartilage matrix formation. When human MSCs cultured in pellets were treated with TGF-β (10 ng/ml), they differentiated into chondrocytes as assessed by alcian blue staining and immunostaining for aggrecan, but nontreated cell pellets did not. Furthermore, when low-intensity US was applied for 20 minutes every day to the TGF-β-treated cell pellets, chondrocyte differentiation was enhanced (Fig. 24).

Biochemically, aggrecan deposition was increased by 2.9- and 8.7-fold by treatment with TGF-β alone, and with both TGF-β and US, respectively. In contrast, cell proliferation and total protein amount appeared unaffected by these treatments. These results indicate that low-intensity US enhances TGF-β-mediated chondrocyte differentiation of MSCs in pellet culture and that application of US may facilitate larger preparations of chondrocytes and the formation of mature cartilage tissue.

MSCs had to be cultured in pellets for differentiation into chondrocytes. Even in the

presence of TGF-β, they did not differentiate when cultured on plates. Induction of chondrocyte differentiation in MSCs in pellets may imply a requirement for cell–cell interaction different from that in plate culture, as confluent cells show little differentiation, and the pellet culture may provide a microenvironment similar to mesenchymal condensation, which normally takes place on initiation of chondrogenesis [11]. It has been demonstrated that chondrocytes maintain differentiation in pellets 12 or in three-dimensional culture coupled with scaffolds such as alginate beads, 34 collagens, and polyglycolic acid [12, 13]. The cells in the pellet may have an appropriate microenvironment for differentiation. Studies on the expression patterns of MSCs cultured in pellets during chondrocyte differentiation demonstrate that these cells exhibit sequential expression of molecules involved in chondrocyte differentiation. The pellet culture system of MSCs will enable us to study US effects at different stages of chondrocyte differentiation.

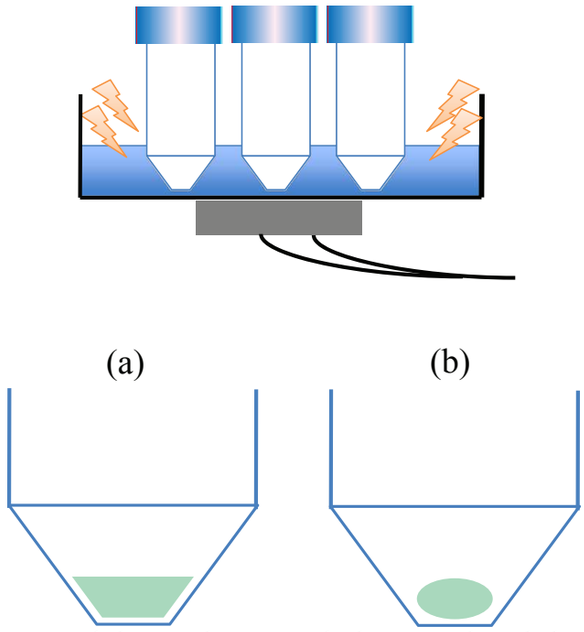

Fig. 24. Schematic drawing of the application of ultrasound and the pellet culture. (a) Cell pellet of hMSCs immediately after centrifugation appears quadrilateral. (b) The cell pellet after 24 hours of incubation appears oval (From Ebisawa et al. 2004).

References

1. Brittberg M, Lindahl A, Nilsson A, Ohlsson C, Isakson O, Peterson L. Treatment of deep cartilage defects in the knee with autologous chondrocyte transplantation. N Engl J Med. 331: 889,1994
2. Kramer J, Hegert C, Guan K, Wobus AM, Müller PK, Rohwedel J. Embryonic stem cell-derived chondrogenic differentiation *in vitro*: activation by BMP-2 and BMP-4. Mech Dev. 92: 193,2000
3. Paul G, Li JY, Brundin P. Stem cells: Hype or hope? Drug Discov Today. 7: 295,2002
4. Johnstone B, Hering TM, Caplan AI, Goldberg VM, Yoo JU. *In vitro* chondrogenesis of bone marrow-derived mesenchymal progenitor cells. Exp Cell Res. 10: 238,1998

5. Yoo JU, Barthel TS, Nishimura K, Solchaga L, Caplan AI, Goldberg VM, Johnstone B. The chondrogenic potential of human bone-marrow-derived mesenchymal progenitor cells. J Bone Joint Surg Am. 80: 1745,1998
6. Raddi AH. Symbiosis of biotechnology and biomaterials: applications in tissue engineering of bone and cartilage. J Cell Biochem. 56: 192,1994
7. Darling EM, Athanasiou KA. Articular cartilage bioreactors and bioprocesses. Tissue Eng. 9: 9,2003
8. Mizuno S, Tateishi T, Ushida T, Glowacki J. Hydrostatic fluid pressure enhances matrix synthesis and accumulation by bovine chondrocytes in three-dimensional culture. J Cell Physiol. 193: 319,2002
9. Ikenoue T, Trindade MC, Lee MS, Lin EY, Schurman DJ, Goodman SB, Smith RL. Mechanoregulation of human articular chondrocyte aggrecan and type II collagen expression by intermittent hydrostatic pressure *in vitro*. J Ortho Res. 21: 110,2003
10. Buschmann MD, Gluzband YA, Grodzinsky AJ, Hunziker EB. Mechanical compression modulates matrix biosynthesis in chondrocyte/agarose culture. J Cell Sci. 108: 1497,1995
11. Thorogood PV, Hinchliffe JR. An analysis of the condensation process during chondrogenesis in the embryonic chick hind limb. J Embryol Exp Morphol. 33: 581,1975
12. Guo JF, Jourdian GW, MacCallum DK. Culture and growth characteristics of chondrocytes encapsulated in alginate beads. Connect Tissue Res. 19: 277,1989
13. Wakitani S, Kimura T, Hirooka A, Ochi T, Yoneda M, Yasui N, Owaki H, Ono K. Repair of rabbit articular surfaces with allograft chondrocytes embedded in collagen gel. J Bone Joint Surg Br. 71: 74,1989

(Ebisawa K, Hata K, Okada K, Kimata K, Torii S, Watanabe H, Ueda M)

IntechOpen

Applied Tissue Engineering

Edited by

ISBN 978-953-307-689-8
Hard cover, 76 pages
Publisher InTech
Published online 08, June, 2011
Published in print edition June, 2011

Tissue engineering, which aims at regenerating new tissues, as well as substituting lost organs by making use of autogenic or allogenic cells in combination with biomaterials, is an emerging biomedical engineering field. There are several driving forces that presently make tissue engineering very challenging and important: 1) the limitations in biological functions of current artificial tissues and organs made from man-made materials alone, 2) the shortage of donor tissue and organs for organs transplantation, 3) recent remarkable advances in regeneration mechanisms made by molecular biologists, as well as 4) achievements in modern biotechnology for large-scale tissue culture and growth factor production.

This book was edited by collecting all the achievement performed in the laboratory of oral and maxillofacial surgery and it brings together the specific experiences of the scientific community in these experiences of our scientific community in this field as well as the clinical experiences of the most renowned experts in the fields from all over Nagoya University. The editors are especially proud of bringing together the leading biologists and material scientists together with dentist, plastic surgeons, cardiovascular surgery and doctors of all specialties from all department of the medical school of Nagoya University. Taken together, this unique collection of world-wide expert achievement and experiences represents the current spectrum of possibilities in tissue engineered substitution.

InTech Europe
University Campus STeP Ri
Slavka Krautzeka 83/A
51000 Rijeka, Croatia
Phone: +385 (51) 770 447
Fax: +385 (51) 686 166
www.intechopen.com

InTech China
Unit 405, Office Block, Hotel Equatorial Shanghai
No.65, Yan An Road (West), Shanghai, 200040, China
中国上海市延安西路65号上海国际贵都大饭店办公楼405单元
Phone: +86-21-62489820
Fax: +86-21-62489821

IntechOpen