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Operative Techniques in
Orthopaedics

Stem Cell Therapy in Cartilage Repair—Culture-Free and Cell Culture–Based Methods

Norimasa Nakamura, MD, PhD,[*] James Hui, MD, MBBS,[†] Kota Koizumi, MD,[*] Yukihiko Yasui, MD,[*] Takashi Nishii, MD, PhD,[*] Dnyanesh Lad, MD,[‡] Georgios Karnatzikos, MD,[‡] and Alberto Gobbi, MD[‡]

In order to overcome potential problems associated with autologous chondrocyte implantation, mesenchymal stem cell-based therapies could be potential alternatives. Conventional stem cell-based therapy accompanies the separation of cells from tissue followed by monolayer culture for the expansion of cell numbers. On the other hand, the cost of cell culture under quality control is high, which could be a potential barrier for industrialization. In order to reduce the cost associated cell culture, culture-free cell-based therapies have been investigated with the use of bone marrow aspirate. In this chapter, we will introduce the three stem cell-based therapies in cartilage repair. The first two procedures are using cell culture methods and the last one with cell-free method. All the three methods have been into the stage of clinical trials and their surgical procedures as well as their preliminary results will be reported.

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KEYWORDS stem cell, cartilage repair, cell cuture, knee, clinical trial

Introduction

It is widely accepted that chondral injuries usually do not heal spontaneously.[1,2] Therefore, a variety of approaches have been tested to improve cartilage healing. Among them, chondrocyte-based therapies have been extensively studied since the successful report of autologous chondrocyte implantation. However, this procedure may have limitations including the sacrifice of undamaged cartilage within the same joint and alterations associated with the in vitro expansion of the cells. Furthermore, because of the degenerative changes in cartilage accompanying aging, the availability of the cells may be limited in elderly individuals.[3]

To overcome such potential problems, cell-based therapy using mesenchymal stem cells (MSCs) could be a promising alternative because of the relative ease of harvest and their strong potential for differentiation into multilineage tissues including cartilage.[4,5]

In this article, we introduce 3 interesting topics related to stem cell–based therapy in cartilage repair. The first 2 topics feature the cell culture–based methods (clinical and translational) and the third topic introduces a culture-free method.

This article has been approved by all the institutional review boards and a written informed consent was obtained from every patient before their inclusion in the study.

Bone Marrow MSC–Based Therapy—From Bench to Bedside

There is little doubt among scientific and medical communities that stem cells, especially MSCs, can be used to treat diversified clinical conditions, from immune modulation to tissue

*Department of Orthopaedics, Osaka University Graduate School of Medicine, Osaka, Japan.
†Department of Orthopaedic Surgery, National University of Singapore, Singapore.
‡OASI Bioresearch Foundation, Milan, Italy.
Address reprint requests to Norimasa Nakamura, MD, PhD, Department of Orthopaedics, Osaka University Graduate School of Medicine, Osaka, Japan. E-mail: Norimasa.nakamura@ohsu.ac.jp

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regeneration. In cartilage repair, there have been many experimental and clinical studies on MSC-based therapy reported with promising results.[6-13] However, unfortunately, the use of stem cells in therapy without proper regulatory and clinical control has been still controversial and, in some way, scandalous.[14-16]

A multidisciplinary orthopaedic-based research group, led by Hui et al, in National University Hospital, Singapore, has systematically developed and translated MSCs for cartilage repair: starting from in vitro research, through animal studies, and finally into clinical trials. First, they have demonstrated that the human bone marrow (BM) is a better source of MSCs than the adipose tissue is.[17] In this study, BM and adipose MSCs were cultured from the same sets of donors. Results showed that although MSCs from both sources were capable of trilineage differentiation, BM MSCs produced significantly more collagen II and s-glycosaminoglycan, suggesting their superior potential for cartilage repair. The next significant progress was demonstrated in small and large animal models, where Lee et al[18] demonstrated that BM MSCs were capable of enhancing cartilage repair.[19]

Nejadnik et al answered an important question of which is superior: autologous BM MSCs vs autologous chondrocyte implantation. In a cohort study, they were able to report comparable efficacy when using BM MSCs as compared with using chondrocytes. The results showed that although both cell types were capable of improving cartilage repair, MSCs were superior in longer-term follow-up, worked just as well in older patients (older than 45 years, which was not replicated in chondrocytes), and required 1 session less of knee surgery, translating into cost savings and potentially lower health risks.[19]

Current techniques of cartilage repair such as autologous chondrocyte implantation require another open surgery after the initial biopsy and harvesting. The use of intra-articular, injectable, cultured, autologous BM MSCs for cartilage repair was investigated and optimized by the research group. Lee et al,[18] using a porcine model, were able to demonstrate the efficacy and viability of using intra-articular injections of MSCs suspended in hyaluronic acid.

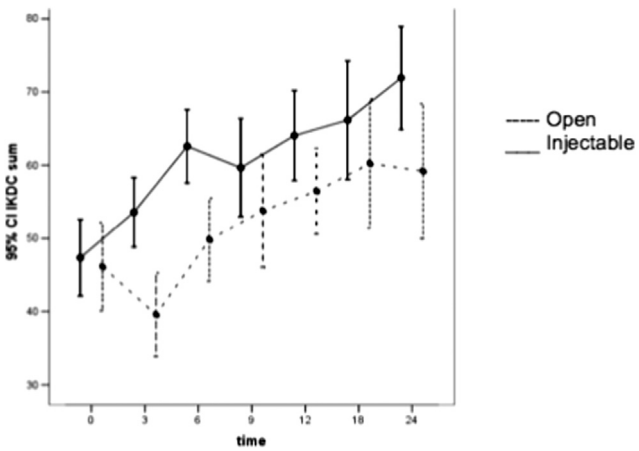

Figure 1 A graph showing the International Knee Documentation Committee (IKDC) sum score: injectable vs open technique.

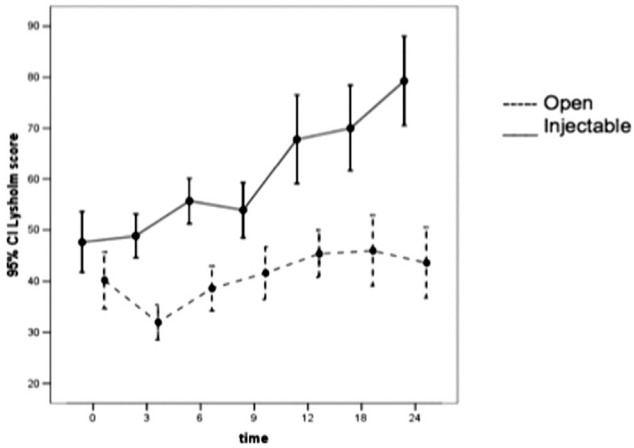

Figure 2 A graph showing the Lysholm score: injectable vs open technique.

In view of concrete consistent evidence from in vitro research to large animal studies, clinical trials are designed and carried out to evaluate the clinical efficacy of using intra-articular, autologous, cultured BM MSCs for cartilage repair. The autologous BM MSCs used are processed in a clean room environment (current good manufacturing practice cell processing facility) and cultured for 3 weeks. From participating subjects, 60 mL of BM and 60 mL of whole blood are collected during the index surgery. The index surgery includes International Cartilage Repair Society classification of the lesion and microfracture techniques as described by Steadman et al.[20,21] After red blood cell removal, the cells are seeded into culture flasks with complete medium change every 2-3 days. When adherent MSCs reach >30% confluency, they are trypsinized and reseeded into the same number of flasks (p0 to p1), usually within 9-12 days. Medium changes continue until MSCs are >80% confluent (usually between 21-28 days after BM collection). Stringent release criteria include no microbial contamination, confluency >80%, normal MSC morphology, and more than 75% viability. The cells are trypsinized and collected, washed, resuspended in autologous serum, and given as intra-articular injections with local anesthesia along with hyaluronic acid.

Clinical trials conducted by Lee et al[22] compared the novel injectable method described earlier with the open technique by which MSCs were implanted beneath a sutured periosteal patch over the defect. In a prospective comparative study comparing both methods with 35 patients in each arm, patients were followed up for 2 years using clinical scoring systems. After 2 years, there were no clinically significant adverse events reported. There was significant improvement in mean International Knee Documentation Committee, Lysholm, SF-36 physical component, and visual analog pain scores in both the groups (Figs. 1 and 2). The injectable group demonstrated better improvement in pain scores and SF-36 scores, but these were statistically insignificant.[22]

Through systematic and careful research and development, MSCs can be safely applied to patients for cartilage repair.