Chapter 4

Cell Therapy and Tissue Engineering for Cartilage Repair

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Additional information is available at the end of the chapter

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Abstract

The integrity of the articular cartilage is necessary for the proper functioning of the diarthrodial joint. The self-repair capacity of this tissue is very limited and, currently, there is no effective treatment capable of restoring it. The degradation of the articular cartilage leads to osteoarthritis (OA), a leading cause of pain and disability mainly among older people.

Different cell treatments have been developed with the aim of forming a repair tissue with the characteristics of native articular cartilage, including cellular therapy and tissue engineering. Cell therapy-based approaches include bone marrow-stimulating techniques, implants of periosteum and perichondrium, ostechondral grafting and implantation of chondrogenic cells as chondrocytes, mesenchymal stem cells or induced pluripotent stem cells. In tissue engineering-based approaches cell-free scaffolds capable of recruiting endogenous cells or chondrogenic cell-loaded scaffolds may be used.

However, despite the numerous treatments available nowadays, no technique has been able to consistently regenerate native articular cartilage in clinical trials. Although many cell therapy and tissue engineering studies have shown promising results and clinical improvement, these treatments generate a fibrocartilaginous tissue different from native articular cartilage. More research is needed to improve cell-based approaches and prove its efficacy

Keywords: regenerative medicine, chondrogenic cells, mesenchymal stem cells (MSCs), induced pluripotent stem cells (iPS), scaffolds

1. Introduction

The integrity of the structure of the articular cartilage is necessary for the proper functioning of the diarthrodial joint. Articular hyaline cartilage provides a resistant, smooth, and lubricated surface, which avoids friction between bones. Thus, hyaline cartilage absorbs and minimizes the pressures produced in the movement of the joint, allows bones to glide over one another with minimal friction, and facilitates the coupling between articular surfaces. Due to its elasticity, articular cartilage absorbs an important part of the compression force, reducing the load supported by the underlying bone structure [1–3].

Traditionally, osteoarthritis (OA) was defined as a degenerative joint disease, characterized by the alteration in the integrity of the articular cartilage [1]. Nowadays, it is known that although the degradation of articular cartilage is the central event in the pathogenesis of OA, synovial tissue and subchondral bone also participate in the onset and development of this disease [4]. The degree of compromise of these components of the joint leads not only to variability between the clinical profiles of patients, but also between different joints of the same patient [5]. On this basis, the Osteoarthritis Research Society International (OARSI) has defined OA as a heterogeneous disorder of movable joints, manifested as genetic, metabolic, and inflammatory changes in the joint, as well as anatomic and/or physiological conditions that may lead to the symptoms associated with the disease. OA is characterized by cell stress and extracellular matrix degradation initiated by micro- and macro-injury that activates maladaptive repair responses including pro-inflammatory pathways of innate immunity [6]. OA is one of the most common chronic health conditions and a leading cause of pain and disability among adults [2, 7]. OA is one of the most prevalent diseases in older people and its incidence, which increases with age, is expected to rise along with the median age of the population [3, 8].

The self-repair capacity of articular cartilage is very limited as it is an avascular and aneural tissue. Due to this absence of vascularity, progenitor cells present in blood and marrow cannot enter into the damaged region to influence or contribute to the reparative process [9, 10]. In addition, because of aneurality, chondral lesions are not detected, and thus patients are not medically treated until more severe lesions are formed [11, 12].

Currently, there is no effective treatment capable of restoring the physiological properties of the osteochondral unit (**Figure 1A**) [13, 14] and the prosthetic replacement is necessary at the final clinical stage (**Figure 1B**) [6]. Different cell treatments have been developed with the aim of forming a repair tissue with structural, biochemical, and functional characteristics equivalent to those of native articular cartilage (**Figure 2**). Scientists have sought several different ways to repair articular cartilage after traumatic damage, which can lead to secondary OA or degeneration of the cartilage [13, 15–17].

It is necessary to highlight that "repair" refers to the restoration of a damaged articular surface with the formation of a neocartilage tissue, which resembles to the native cartilage and "regeneration" refers to the formation of a tissue indistinguishable from the native articular cartilage [16]. Cellular therapy (using cells) and tissue engineering (combining cells, scaffolds, and bioactive factors) have emerged as alternative clinical approaches. However, despite the numerous treatments available nowadays, no technique has been able to consistently regenerate normal hyaline cartilage in clinical trials [3, 18]. Long-term follow-up studies are expected to be performed in the coming years to confirm safety and effectiveness of these new approaches [3].

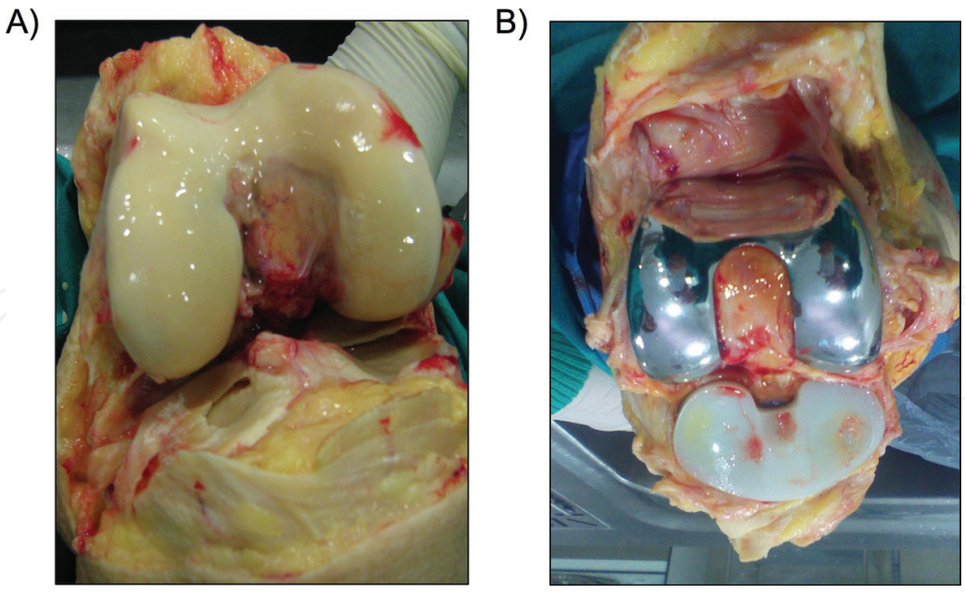

Figure 1. Images showing (A) healthy knee joint and (B) prosthetic joint replacement.

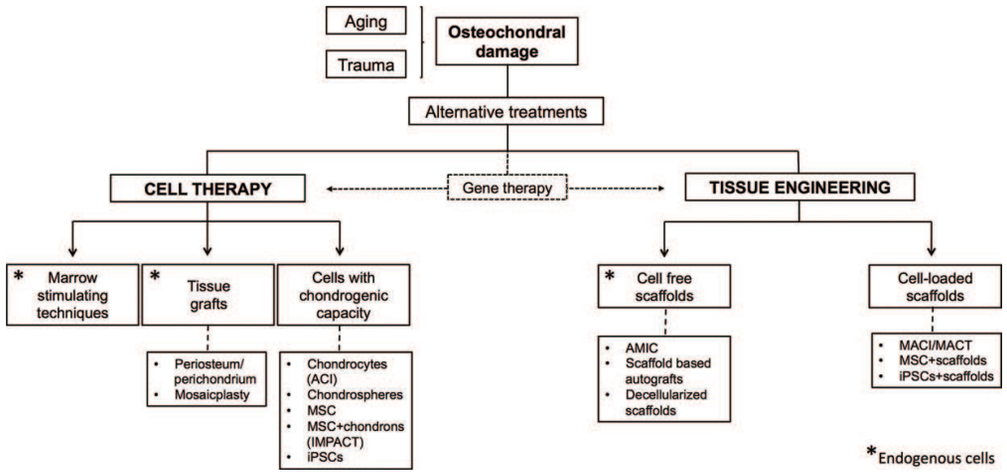

Figure 2. Diagram showing an overview of the alternative treatments for osteochondral damage.

2. Cell therapy

Cell therapy is a relatively new approach based on the regeneration or repair of a damaged tissue using autologous or allogenic cells.

2.1. Marrow stimulating techniques

Bone marrow stimulating techniques (MSTs) are based on the use of endogenous mesenchymal stromal cells (MSCs). This type of technique is used in the treatment of chondral lesions with less of 15 mm of diameter [19].

Penetration of subchondral bone is among the oldest and still the most commonly used method to stimulate regeneration of neocartilage [16, 20]. Arthroscopic techniques like drilling, abrasion arthroplasty or microfracture are different tools to perforate the subchondral

bone [12], allowing MSCs and growth factors from the bone marrow to infiltrate the lesion [15]. A blood clot is formed in the defect, acting as a scaffold and mediating the inflammatory response (through cytokines) [19].

However, it was described that endogen bone marrow angiogenic factors favor osteogenesis, instead of chondrogenesis, of bone marrow MSCs [11]. Generated repair tissue frequently ends up degenerating [21] and usually presents type I collagen (fibrocartilage phenotype) and lacks hyaline cartilage viscoelastic properties [22].

2.2. Tissue grafts

Tissue grafts have potential benefits in cartilage repair since they contain cell populations with chondrogenic capacity.

2.2.1. Implants of periosteum and perichondrium

In the 90s, autologous strips of perichondrium were used to treat chondral defects [23, 24]. Periosteum and perichondrium contain MSCs that are capable of chondrogenesis and act as a biological membrane [16]. However, the ability of periosteum MSCs to proliferate and differentiate into chondrocytes decreases with age [25].

The clinical outcomes of perichondrium implants are similar to those of subchondral perforation [26]. Calcification of the periosteum grafts had been mentioned as a problem in the long term [16].

2.2.2. Mosaicplasty

Autologous mosaicplasty is widely used for treating chondral and osteochondral defects. The most used technique is the osteochondral autologous transplantation (OAT), which consists in the translocation of osteochondral cylinders from not loading areas to the affected areas of the joint [15].

Even though good to excellent short-term subjective results were obtained, clinical and radiological midterms to long-term outcomes of mosaicplasty were moderate. Further limitations are donor-site morbidity, technical difficulty, special equipment, lesion size, and fibrocartilaginous repair [16, 27]. OAT might be more appropriate for lesions smaller than 2–3 cm^2 [28].

Another problem is the lack of congruence between the osteochondral cylinders implanted and the lesion area, and the differences in cartilage height of the defect and surrounding native cartilage, altering the distribution of stress and compression forces [16, 27].

Allogenic mosaicplasty has shown successful outcomes and its main advantage over autograft transplantation is the lack of donor-site morbidity. Nevertheless, the amount of transplanted bone has to be minimum because the allograft failure is mostly due to collapse of the subchondral bone [22].

Nowadays, synthetic cylindrical plugs for implant similar to OAT exist but studies have shown universal failure to incorporate these plugs into the subchondral bone, with formation of cysts [22].

In addition to fresh osteochondral grafts, particulated cartilage grafts, which are formed by combining fragments of cartilage with fibrin glue, may also be used. Superficial chondrocytes, released from the extracellular matrix as a consequence of the fragmentation of the cartilage, produce additional extracellular matrix that integrates the particulate graft with native cartilage and fills the defect [29].

2.3. Implantation of cells with chondrogenic capacity

Chondrogenic potential of different cell types (**Figure 3**) was tested for hyaline cartilage repair.

2.3.1. Autologous chondrocyte implantation

The autologous chondrocyte implantation (ACI) was firstly described by Peterson et al. [30]. This technique consists of harvesting a cartilage piece from a low-weight-bearing area of the joint and culture-expanding the chondrocytes to implant into the lesion. The lesion is sealed with autologous periosteum to avoid cell loss.

ACI is only applicable to small size (3–4 cm^2) focal lesions surrounded by healthy cartilage [15, 28]. Other limitations are dedifferentiation of chondrocytes during culture expansion, the low amount of chondrocytes obtained and multiple surgical procedures involved [31, 32]. Further, donor-site morbidity of cartilage and bone for chondrocyte and periosteum obtaining was observed [15, 33, 34].

ACI is considered superior to MSTs regarding the quality of the repaired tissue, although there are conflicting results [28].

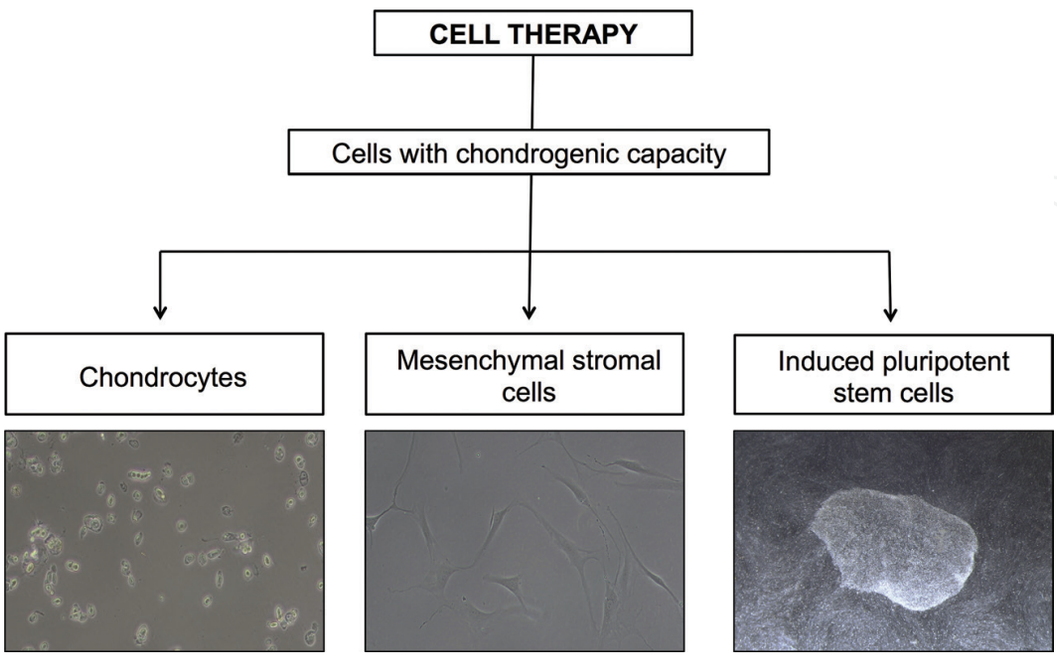

Figure 3. Diagram showing the different cell sources, most commonly used in cartilage treatment using cell therapy: chondrocytes (left), mesenchymal stromal cells (middle), and induced pluripotent stem cells (right).

2.3.2. Chondrospheres

The technique of chondrospheres consists of the generation and implantation of spheroids of autologous or allogenic articular chondrocytes [29]. Autologous chondrocytes are obtained from undamaged articular cartilage, expanded *in vitro*, and condensed in order to form spheroids, which then are coalesced. Chondrospheres have shown to be able to adhere, integrate into hyaline cartilage defect and produce cartilaginous extracellular matrix in mouse, mini pig, and horse cartilage defect models, as well as in artificial defects in human cartilage explants [35–37]. A phase III clinical trial is currently ongoing in Germany and Poland to investigate the efficacy of this technology compared to microfracture in the treatment of cartilage defects of knee joints [38].

2.3.3. Mesenchymal stromal cells

Human MSCs are nonhematopoietic multipotent progenitor cells with long-term self-renewal ability and the capacity to differentiate along multiple cell lineages, including cartilage, as well as immunomodulatory features [39–41]. MSCs are responsible for normal tissue renewal and for response to injury and may be an alternative to chondrocytes for the development of new therapeutic approaches for the treatment of cartilage defects.

In vitro and *in vivo* studies of clonally derived MSCs demonstrated that these cells consist of subsets that present different surface markers expression and different capacities for cellular differentiation [42]. These cells are considered a potential cell source for cell therapy since they can be easily collected from various tissues such as bone marrow [43], adipose tissue [44], synovial membrane [42], and amniotic membrane [45], among others. However, the equivalence of chondrogenic differentiation potential of MSCs derived from different tissues is a matter of considerable debate [46].

For cell therapy approaches, either autologous or allogenic MSCs can be used. MSCs do not express major histocompatibility complex class II (MHC II) and its co-stimulatory molecules, and barely express major histocompatibility complex class I (MHC I), so that they do not produce alloreactivity, avoiding rejection problems. This feature turns MSCs into a feasible cell source for allogenic transplantation [40, 47].

The therapeutic potential of autologous MSCs derived from different tissues to stimulate the regeneration of cartilage in OA has been reported in several preclinical studies [48, 49]. Bone marrow-derived MSCs suspended in hyaluronic acid and administrated by intra-articular injection have been used to promote cartilage repair in animal models such as guinea pig, mini pig, goat and donkey, leading to improvement in cartilage regeneration, less cartilage destruction and reduced osteophyte formation [50–53]. MSCs derived from other sources have also been used; for example, transplantation of synovial MSCs was used to repair osteochondral defects in rabbits [54], and intra-articular injection of adipose-derived MSCs was used to treat chronic osteoarthritis in dogs, showing significant improvement in MSCs-treated joints [55].

One of the MSCs transplantation techniques for cartilage focal lesions is a variation of ACI in which bone marrow MSCs are injected into defects and closed with periosteal membrane to be differentiated toward chondrocytes [56]. The first clinical study using MSCs to treat OA

was performed by Wakitani et al. [57]. In this study, bone marrow-derived MSCs were transplanted into the articular cartilage defect and covered with autologous periosteum. Although the arthroscopic and histological grading score was better in the cell-transplanted group than in the control one, the clinical improvement was not very clear. Since then, several clinical studies have been performed, mainly using intra-articular injection of autologous bone marrow-derived MSCs, showing some degree of improvement in terms of clinical outcomes and repaired cartilage tissue quality [58–60]. However, several studies described a lack of engraftment into cartilage defects [61] and it is important to highlight that most of the clinical trials are I and I/II phases, indicating the immaturity of MSC clinical applications in OA [49].

Limitations of this approach are that culture expansion is not avoided, cell yield is often low and MSCs differentiation capacity decreases with age of the donor [21]. This is a problem in regenerative therapies for degenerative diseases such as OA, where most of patients are aged [61]. Given that the age of patient and the size of the lesion affect the outcome, the cut-off points for the risk of failure have been suggested at age greater than 60 years and lesion size larger than 6.0 cm^2 [28].

2.3.4. Mesenchymal stromal cells combined with autologous chondrons

A novel cell therapy approach is based on combining autologous chondrocytes in their pericellular matrix (chondrons) and allogenic MSCs, which was called Instant MSC Product accompanying Autologous Chondron Transplantation (IMPACT) and performed by De Windt et al. [62]. In this phase I clinical trial, patients with focal cartilage defects were treated using a mix of 80–90% allogenic MSCs and 10–20% autologous chondrons combined with fibrin glue. In this approach, chondrons are "recycled" from debrided cartilage instead of being harvested from a low-weight-bearing area of the joint, as occurring in ACI. The combination of this recycled chondrons with allogenic human bone marrow MSCs stimulates cartilage regeneration and provides clinical improvement. Surprisingly, although the co-implantation of chondrons and MSCs provides better results in comparison with implantation of chondrons or MSCs alone [63], no allogenic cells were detected in the repaired cartilage after 1 year, suggesting that MSCs have trophic effects that stimulate chondrons to regenerate cartilage. The quality of the repaired tissue and the clinical outcome using the IMPACT technique was similar or even superior in comparison with ACI. Furthermore, IMPACT technique presents the advantage of allowing to perform both surgeries on the same day (the extraction of cartilage and the implantation of cells) [62].

2.3.5. Induced pluripotent stem cells

Pluripotent cells could provide an unlimited and renewable cell source that can be induced to differentiate into any cell type. In fact, pluripotent cells of embryonic origin [61, 64], embryonic human stem cells (hESCs), or induced to pluripotency [65], induced pluripotent stem cells (iPSCs), have shown to produce cartilage under specific conditions. iPSCs have been generated from adult cells (**Figure 4A**) using defined factors (**Figure 4B**) [66]. These cells present similar morphology (**Figure 4C**), proliferation capacity, genetic expression and epigenetic pattern, and pluripotency characteristics to hESCs [66, 67].

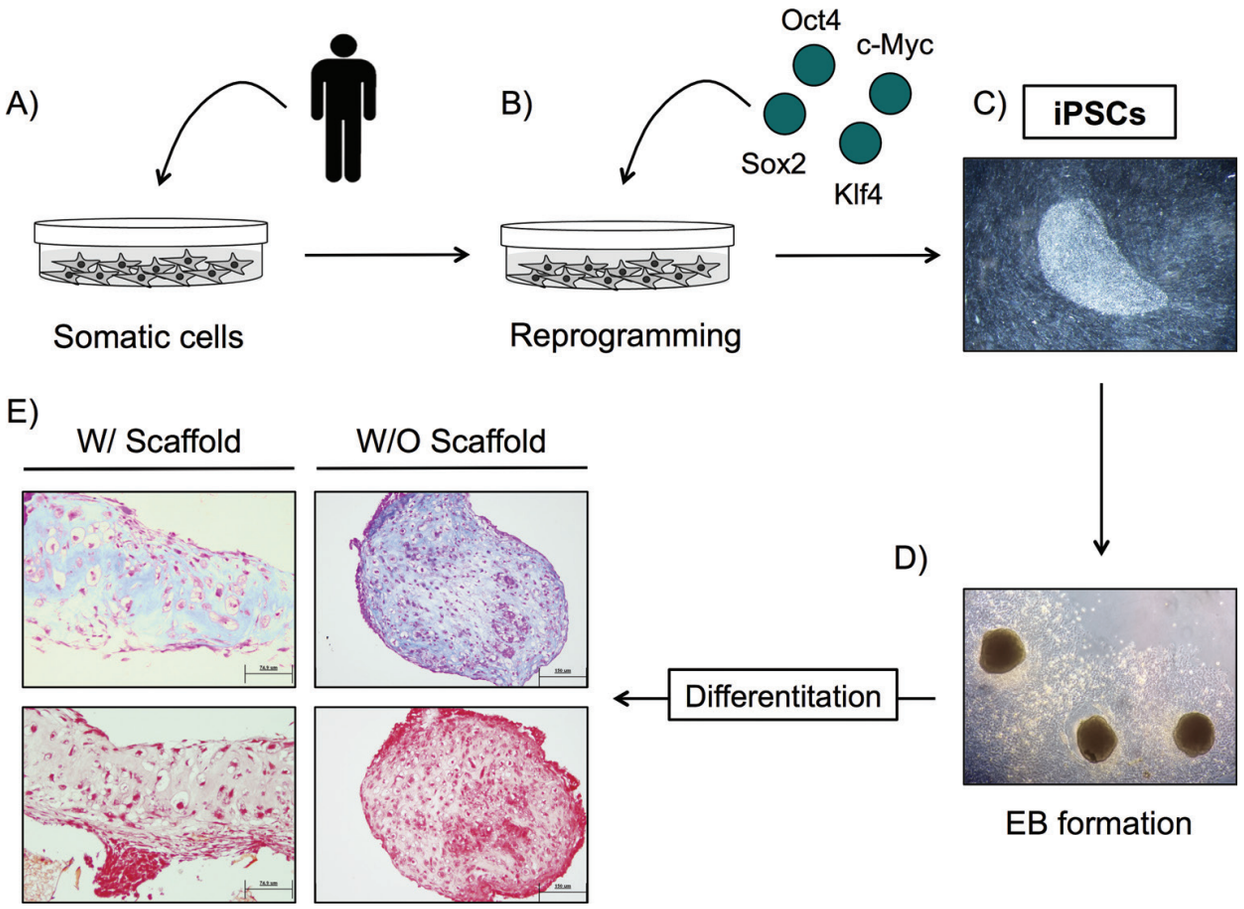

Figure 4. Scheme representing the role of iPSCs in tissue engineering. (A) Harvesting somatic cells from the patient. (B) Reprogramming the cells using the factors Oct4, Sox2, Klf4, and c-Myc. (C) iPSc colony obtained after reprogramming. (D) Embryoid bodies (EB) formation. (E) Differentiation of the iPSc toward chondrocytes with (W/) or without (W/O) scaffold.

iPSCs seem to be an alternative tool to chondrocytes for cartilage repair as they can be expanded before starting their differentiation (using or not embryoid bodies formation) toward chondrocytes (**Figure 4D**). Then, iPSC-derived chondrocytes can be cultured in three-dimensional culture with scaffold (**Figure 4E**, w/Scaffold), or cultured without a scaffold (**Figure 4E**, w/o Scaffold), to create cartilaginous tissues *in vitro* before transplantation to repair large defects [68].

In addition, iPSCs seem to be an alternative tool to MSCs for cartilage repair. After *in vitro* chondrogenesis, iPSCs showed lower hypertrophic markers than MSCs [69]. The risk of iPSCs teratoma formation in cell therapy or tissue engineering can be avoided using pre-differentiated cells before implantation [70, 71]. Also, the use of iPSCs avoids the problem of *in vivo*-age-dependent and *in vitro*-passage-dependent MSC senescence [72].

Yamashita et al. [73] optimized a protocol of chondrogenic differentiation using human iPSCs to form homogenous cartilaginous particles. After the transplantation of these chondrogenic

particles into joint surface defects in immunodeficient rats and immunosuppressed mini pigs, they observed cartilaginous neotissue with potential for integration into native cartilage.

Nowadays, there are no clinical studies published about cartilage cell therapy using iPSCs. Although cell therapy or tissue engineering using iPSCs are promising tools, their clinical use is not legalized either by the scientific community or by existing international legislation yet, except in Japan.

3. Tissue engineering

The lack of efficient treatments for cartilage repair motivates the researchers to develop, by tissue engineering, biological tissue substitutes that can be implanted to replace the affected area of the joint [74]. Tissue engineering is not widespread yet in surgical procedures, although there are many combinations of different cells and supports being tested both *in vitro* and *in vivo*.

In this way, different strategies were developed for cartilage regeneration, based on the use of scaffolds and endogenous or exogenous cells. Whereas in *in vitro* studies scaffolds are usually combined with cells and bioactive factors, in most *in vivo* studies the scaffolds are used only combined with cells because those factors are present in the joint (e.g., AMIC described below).

In vitro administration of growth factors (transforming growth factor 1 or 3, bone morphogenetic proteins 2 or 7, and insulin growth factor 1, among others) have been used to induce chondrogenic differentiation of MSCs and iPSCs. However, the effect of application of these molecules is dose, timing of administration and cell type-dependent [75]. That is why, in recent years, scaffolds were functionalized with bioactive factors or other molecules for *in vivo* cartilage therapies, as a delivery system [76] or stimulation for MSCs. For example, the addition of proteoglycans to collagen biomaterials had improved bone marrow MSCs chondrogenic differentiation [43, 77].

A broad variety of biomaterials have been successfully developed to support proliferation, infiltration, or differentiation of allogeneic transplanted or endogenous MSCs to achieve functional tissue restoration [78]. Scaffolds/biomaterials should be a porous three-dimensional matrix that allow cell migration, adhesion and growth, and support the organization of the growing tissue [79].

However, despite the diffusion of new tissue-engineering techniques and the high number of scaffolds that have been developed and investigated for cartilage regeneration, the ideal matrix material has not been identified yet. Cartilage-engineering strategies have produced promising *in vitro* data, seeding chondrogenic cells on biomaterials with growth factors. However, thus far, no approach has led to the generation of long-term *in vivo* replacement tissue identical to native hyaline cartilage. There are different factors for the lack of stable functional tissue as inflammatory stress or biophysical stimuli [80].

3.1. Cell-free scaffolds and endogenous cells

Cell-free scaffolds are developed for one stage procedure techniques, since they can be implanted alone to attract the endogenous cells. In this case, the aim of using scaffolds is to obtain a suitable microenvironment to recruit and mobilize the host cells, from either the blood or a tissue specific (bone marrow, synovial fluid…) niche for self-repair. Several studies have detected the recruitment of endogenous synovial cells [81, 82] or exogenous-injected MSCs [50] in injured areas after the implantation of empty scaffolds.

Implantation of cell-free scaffolds avoids the issues around the *in vitro* cell culture process, as exogenous cell transplantation is not required. However, clinical results after implantation of cell-free scaffolds for OA treatment are few [3].

3.1.1. Autologous matrix-induced chondrogenesis

The autologous matrix induced chondrogenesis (AMIC) is a second generation MSTs. This is a one-step procedure combining subchondral microfracture with the attachment of a collagen scaffold to the lesion. The initially formed blood clot as produced by microfracturing is protected by the collagen scaffold [83]. The collagen scaffold is thought to stabilize the blood clot, helping to promote early mechanical stability and cartilage regeneration [29]. More complex scaffolds have also been tested in AMIC studies, for example, a biphasic scaffold consisting of calcium triphosphate in the osseous region and poly(lactic-co-glycolic acid) in the cartilaginous region [84].

Even though donor-site morbidity due to removal of periosteum from tibia is avoided, AMIC has similar clinical outcomes to ACI [85].

3.1.2. Scaffold-based autografts

Another approach is the use of scaffold-based autografts, in which harvested cartilage is mechanically minced and uniformly affixed to a biodegradable scaffold, using fibrin glue; then, the scaffold with the cartilage fragments is transferred to the lesion. When compared to microfracture, this scaffold-based autograft procedure resulted in an improvement of functional outcomes and cartilage development [86].

3.1.3. Decellularized extracellular matrix scaffolds

Decellularized extracellular matrix may be used as a scaffold with the potential to retain the bioactive factors needed to support specific tissue formation at the implantation site [87]. Cartilage matrix can be harvested from allogenic sources, then decellularized and used as a scaffold. This approach leads to the improvement of neocartilage formation in preclinical models, in comparison with the living-cartilage implantation [88]. One of the drawbacks of this technique is that the protocols required to decellularization of cartilage also imply some degree of destruction of extracellular matrix components [89]. Decellularized cartilage matrix has been used to treat osteochondral defects in a horse model, obtaining repair of both the

bone and cartilage phases [87]. Beside the tissue decellularization, extracellular matrix scaffolds can also be obtained from cultured cells [90].

3.2. Cell-loaded scaffolds

3.2.1. Matrix-associated chondrocyte transplantation

The matrix-associated chondrocyte implantation/transplantation (MACI or MACT) is a second generation ACI, which includes the employment of a bilayer collagen membrane [91]. Essentially, the concept is based on the use of biodegradable polymers as temporary scaffolds for *in vitro* growth of cells and their subsequent transplantation into the defect site. In this case, autologous chondrocytes are previously seeded in the scaffold before implantation into the lesion [12, 83]. Other types of scaffolds (hydrogels, fibrous scaffolds, decellularized ECM, or composites) were later used [85].

MACI presents lower rates of graft hypertrophy than first-generation ACI [92].

3.2.2. Mesenchymal stromal cells on scaffolds

Wakitani et al. [93] observed that MSCs embedded in a collagen gel could differentiate in *in vivo* animal models. Since these first studies, thousands of works were carried out using different types of scaffolds (hydrogels, sponges…), cells, and approaches for chondrogenic scaffolding.

Several *in vivo* studies tried to replicate the distinct osteochondral zones using tri- or bi-layered scaffolds of different composition and/or bioactive factors combined with MSCs. MSCs combined with scaffolds appear to engraft and contribute to cartilage repair, while MSCs injected as a free suspension into the joint do not engraft into the cartilage [61]. This happens because scaffolds can transport cells into the lesion and provide the proper environment for cell differentiation [75, 94].

It was described that cartilage tissue engineering from differentiation-induced *in vitro* MSCs has an inferior quality to that engineered from chondrocytes [95]. However, human amniotic MSCs with human amniotic membrane (as scaffold) showed better reparation in an *in vitro* repair model when compared with bone marrow MSCs and chondrocytes, and demonstrated good adhering capacity to the native cartilage [45]. Also, our group obtained good results using bone marrow MSCs and collagen/heparan sulfate scaffolds in an *in vitro* repair model (**Figure 5**) [96].

3.2.3. Induced pluripotent stem cells on scaffolds

Although tissue-engineering studies using iPSCs are scarce, several studies have shown their potential in chondral repair [21]. Liu et al. [48] have tested the chondrogenesis of murine cells derived from single embryoid bodies. After seeding these cells on polycaprolactone/gelatin scaffolds, they showed a good chondrogenic capacity.

Nowadays, 3D bioprinting into cartilage using iPSCs and bioinks (that act as scaffolds) is being developed [97].

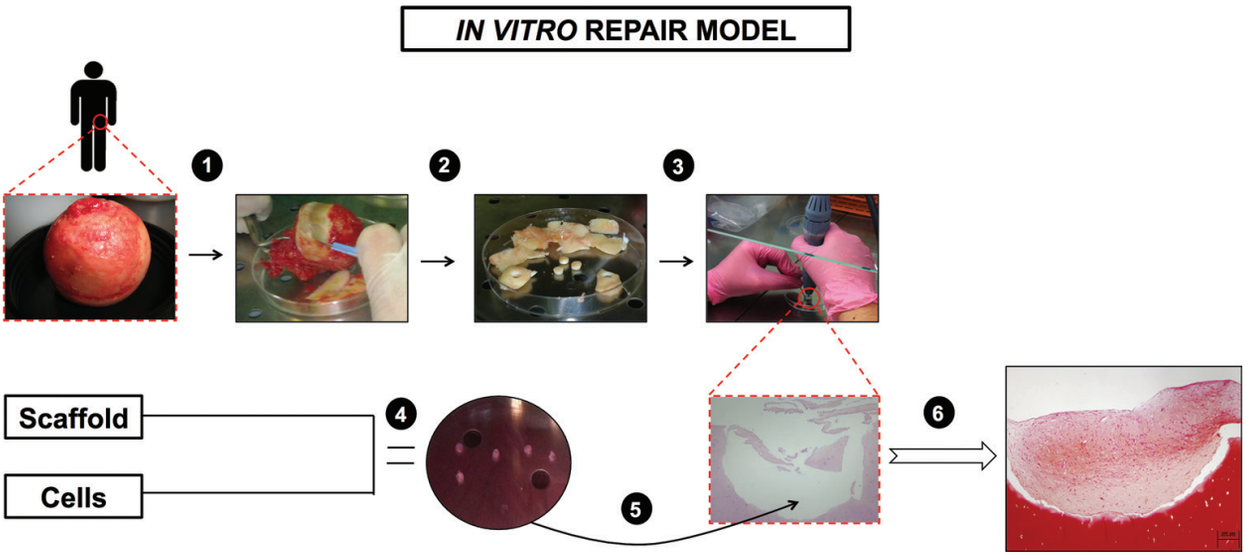

Figure 5. Scheme representing different steps during the development of an *in vitro* cartilage repair model. These steps are on one hand (1) to harvest cartilage explants from the joint (hip), (2) make cartilage punches, and (3) generate the lesion with a driller. On the other hand, (4) to seed the cells on the scaffold and (5) introduce the construct inside the lesion. (6) Safranin O staining showing the final result of the repair model after culture in chondrogenic medium during 2 months.

4. Gene therapy

Gene therapy involves the over-expression of the appropriate gene (anabolic factors, chondroinductor, or anti-inflammatory molecules) and cell type (chondrocytes or chondrogenic cells) for their use in cell therapy and tissue engineering.

Nowadays, no gene products have been approved for OA treatment and few clinical trials have been conducted. At present, only TGF-β gene therapy has been clinically investigated in USA and Korea [3].

5. Conclusions

Although many studies of cell therapy and tissue engineering have shown clinical and functional improvement in joints, these treatments generate a fibrocartilaginous tissue that is different from hyaline articular cartilage. The ability to regenerate articular cartilage that resists the degeneration process still remains elusive.

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References

[1] Blanco-García FJ. Aspectos básicos. Chap. I. In: Batlle-Gualda E, Benito Ruiz P, et al., editors. Manual SER de la artrosis. 1st Ed. Madrid: IM&C; 2002

[2] Bomer N et al. Translating genomics into mechanisms of disease: Osteoarthritis. Best Practice & Research. Clinical Rheumatology. 2015;**29**(6):683-691

[3] Zhang W et al. Current research on pharmacologic and regenerative therapies for osteoarthritis. Bone Research. 2016;**4**:15040-15040

[4] Lories RJ, Luyten FP. The bone-cartilage unit in osteoarthritis. Nature Reviews. Rheumatology. 2011;**7**(1):43-49

[5] Blanco-García FJ, Tornero-Molina J. Artrosis. Chap. 112. In: Farreras P, Rozman C. Medicina Interna. 18th Ed. Barcelona: Elsevier, v.I; 2016.

[6] Kraus VB et al. Call for standardized definitions of osteoarthritis and risk stratification for clinical trials and clinical use. Osteoarthritis and Cartilage. 2015;**23**(8):1233-1241

[7] Allen KD, Golightly YM. Epidemiology of osteoarthritis: State of the evidence. Current Opinion in Rheumatology. 2015;**27**(3):276-283

[8] Ross MH, Pawlina W. Histología. Texto y atlas en color con biología celular y molecular. 5th ed. Madrid: Editorial médica panamericana; 2007

[9] Claus S et al. Cartilage-characteristic matrix reconstruction by sequential addition of soluble factors during expansion of human articular chondrocytes and their cultivation in collagen sponges. Tissue Engineering Part C-Methods. 2012;**18**(2):104-112

[10] Zhang L et al. An in vitro study of collagen hydrogel to induce the chondrogenic differentiation of mesenchymal stem cells. Journal of Biomedical Materials Research Part A. 2012;**100A**(10):2717-2725

[11] Mao JJ. Stem-cell-driven regeneration of synovial joints. Biology of the Cell. 2005; **97**(5):289-301

[12] Gomoll AH et al. The subchondral bone in articular cartilage repair: Current problems in the surgical management. Knee Surgery, Sports Traumatology, Arthroscopy. 2010;**18**(4):434-447

[13] Dhollander AAM et al. The use of scaffolds in the treatment of osteochondral lesions in the knee: Current concepts and future trends. The Journal of Knee Surgery. 2012; **25**(3):179-186

[14] Meretoja VV et al. The effect of hypoxia on the chondrogenic differentiation of co-cultured articular chondrocytes and mesenchymal stem cells in scaffolds. Biomaterials. 2013;**34**(17):4266-4273

[15] Fuentes-Boquete IM et al. Tratamiento de lesiones del cartílago articular con terapia celular. Reumatología Clínica. 2007;**3**:S63-S69

[16] Bhosale AM, Richardson JB. Articular cartilage: Structure, injuries and review of management. British Medical Bulletin. 2008;**87**(1):77-95

[17] Zscharnack M, et al. Repair of chronic osteochondral defects using predifferentiated mesenchymal stem cells in an ovine model. American Journal of Sports Medicine. 2010; **38**(9):1857-69

[18] Magnussen RA et al. Treatment of focal articular cartilage defects in the knee: A systematic review. Clinical Orthopaedics and Related Research. 2008;**466**:952-962

[19] Vilá Y, Rico J, et al. Treatment of osteochondral lesions of the talus with bone marrow stimulation and chitosan-glycerol phosphate/blood implants (BST-CarGel). Arthroscopy Techniques. 2015;**4**(6):e663-e667

[20] Schrock JB et al. Cost-effectiveness analysis of surgical treatment modalities for chondral lesions of the knee: Microfracture, osteochondral autograft transplantation, and autologous chondrocyte implantation. Orthopaedic Journal of Sports Medicine. 2017;**5**(5):2325967117704634

[21] Toh WS et al. Advances in mesenchymal stem cell-based strategies for cartilage repair and regeneration. Stem Cell Reviews and Reports. 2014;**10**(5):686-696

[22] Gomoll AH, Minas T. The quality of healing: Articular cartilage. Wound Repair and Regeneration. 2014;**22**:30-38

[23] Homminga GN et al. Perichondrial grafting for cartilage lesions of the knee. Journal of Bone and Joint Surgery. 1990;**72-B**:1003-1007

[24] Ritsila VA et al. Periosteal and perichondral grafting in reconstructive surgery. Clinical Orthopaedics and Related Research. 1994;**302**:259-265

[25] O'driscoll SW et al. The chondrogenic potential of periosteum decreases with age. Journal of Orthopaedic Research. 2001;**19**:95-103

[26] Díaz Prado SM, et al. Cell therapy and tissular engineering to regenerate articular cartilage. Chap. In: Komorowska MA, Olsztynska-Janus S, editors. Biomedical Engineering, Trends, Researches and Technologies. Ed. Intech, 2011

[27] Valderrábano V et al. Knee-to-ankle mosaicplasty for the treatment of osteochondral lesions of the ankle joint. The American Journal of Sports Medicine. 2009;**37**:105S-111S

[28] Rai V et al. Recent strategies in cartilage repair: A systemic review of the scaffold development and tissue engineering. Journal of Biomedical Materials Research Part A. 2017;**105**(8):2343-2354

[29] Makris EA et al. Repair and tissue engineering techniques for articular cartilage. Nature Reviews. Rheumatology. 2015;**11**(1):21-34

[30] Peterson L et al. Chondrocyte transplantation–An experimental model in rabbits. Transactions of the Annual Meeting of the Orthopaedic Research Society. 1984;**9**:218

[31] Demoor M et al. Cartilage tissue engineering: Molecular control of chondrocyte differentiation for proper cartilage matrix reconstruction. Biochimica et Biophysica Acta-General Subjects. 2014;**1840**(8):2414-2440

[32] Rackwitz L et al. Functional cartilage repair capacity of dedifferentiated, chondrocyte- and mesenchymal stem cell-laden hydrogels in vitro. Osteoarthritis and Cartilage. 2014;**22**(8):1148-1157

[33] Liao CJ et al. Injecting partially digested cartilage fragments into a biphasic scaffold to generate osteochondral composites in a nude mice model. Journal of Biomedical Materials Research Part A. 2007;**81A**(3):567-577

[34] Dewan AK et al. Evolution of autologous chondrocyte repair and comparison to other cartilage repair techniques. BioMed Research International. 2014;**2014**:272481

[35] Litzke LE et al. Repair of extensive articular cartilage defects in horses by autologous chondrocyte transplantation. Annals of Biomedical Engineering. 2004;**32**(1):57-69

[36] Schubert T et al. Long-term effects of chondrospheres on cartilage lesions in an autologous chondrocyte implantation model as investigated in the SCID mouse model. International Journal of Molecular Medicine. 2009;**23**(4):455-460

[37] Meyer U et al. Cartilage defect regeneration by ex vivo engineered autologous microtissue–preliminary results. In Vivo. 2012;**26**(2):251-257

[38] US National Library of Medicine. ClinicalTrials.gov [Online] 2014. http://clinicaltrials.gov/show/NCT01222559.

[39] Dominici M et al. Minimal criteria for defining multipotent mesenchymal stromal cells. The International Society for Cellular Therapy position statement. Cytotherapy. 2006;**8**(4):315-317

[40] Gupta PK et al. Mesenchymal stem cells for cartilage repair in osteoarthritis. Stem Cell Research & Therapy. 2012;**3**(4):25

[41] Baghaban-Eslaminejad M, Malakooty-Poor E. Mesenchymal stem cells as a potent cell source for articular cartilage regeneration. World Journal of Stem Cells. 2014;**6**(3):344-354

[42] Hermida-Gómez T et al. Bone marrow cells immunomagnetically selected for CD271+ antigen promote in vitro the repair of articular cartilage defects. Tissue Engineering Part A. 2011;**17**(7-8):1169-1179

[43] Sanjurjo-Rodríguez C et al. Differentiation of human mesenchymal stromal cells cultured on collagen sponges for cartilage repair. Histology and Histopathology. 2016b;**31**(11):1221-1239

[44] Pak J et al. Potential use of mesenchymal stem cells in human meniscal repair: Current insights. Open Access Journal of Sports Medicine. 2017;**8**:33-38

[45] Muíños-López E et al. Human amniotic mesenchymal stromal cells as favorable source for cartilage repair. Tissue Engineering. Part A. 2017

[46] Kohli N et al. An in vitro comparison of the incorporation, growth, and chondrogenic potential of human bone marrow versus adipose tissue mesenchymal stem cells in clinically relevant cell scaffolds used for cartilage repair. Cartilage. 2015;**6**(4):252-263

[47] Chen FH, Tuan RS. Mesenchymal stem cells in arthritic diseases. Arthritis Research & Therapy. 2008;**10**(5)

[48] Liu Y et al. Therapeutic application of mesenchymal stem cells in bone and joint diseases. Clinical and Experimental Medicine. 2014;**14**(1):13-24

[49] Squillaro T et al. Clinical trials with mesenchymal stem cells: An update. Cell Transplantation. 2016;**25**(5):829-848

[50] Murphy JM et al. Stem cell therapy in a caprine model of osteoarthritis. Arthritis and Rheumatism. 2003;**48**(12):3464-3474

[51] Lee KB et al. Injectable mesenchymal stem cell therapy for large cartilage defects – A porcine model. Stem Cells. 2007;**25**(11):2964-2971

[52] Mokbel AN et al. Homing and reparative effect of intra-articular injection of autologus mesenchymal stem cells in osteoarthritic animal model. BMC Musculoskeletal Disorders. 2011;**12**:259

[53] Sato M et al. Direct transplantation of mesenchymal stem cells into the knee joints of Hartley strain guinea pigs with spontaneous osteoarthritis. Arthritis Research & Therapy. 2012;**14**(1):R31

[54] Koga H et al. Local adherent technique for transplanting mesenchymal stem cells as a potential treatment of cartilage defect. Arthritis Research & Therapy. 2008;**10**(4):R84

[55] Black LL et al. Effect of adipose-derived mesenchymal stem and regenerative cells on lameness in dogs with chronic osteoarthritis of the coxofemoral joints: A randomized, double-blinded, multicenter, controlled trial. Veterinary Therapeutics. 2007;**8**(4):272-284

[56] Im GI et al. Repair of cartilage defect in the rabbit with cultured mesenchymal stem cells from bone marrow. Journal of Bone and Joint Surgery. British Volume (London). 2001;**83**:289-294

[57] Wakitani S et al. Human autologous culture expanded bone marrow mesenchymal cell transplantation for repair of cartilage defects in osteoarthritic knees. Osteoarthritis and Cartilage. 2002;**10**:199-206

[58] Davatchi F et al. Mesenchymal stem cell therapy for knee osteoarthritis. Preliminary report of four patients. International Journal of Rheumatic Diseases. 2011;**14**(2):211-215

[59] Orozco L et al. Treatment of knee osteoarthritis with autologous mesenchymal stem cells: A pilot study. Transplantation. 2013;**95**(12):1535-1541

[60] Wong KL et al. Injectable cultured bone marrow-derived mesenchymal stem cells in varus knees with cartilage defects undergoing high tibial osteotomy: A prospective, randomized controlled clinical trial with 2 years' follow-up. Arthroscopy. 2013;**29**(12):2020-2028

[61] Whitworth DJ, Banks TA. Stem cell therapies for treating osteoarthritis: Prescient or premature? The Veterinary Journal. 2014;**202**(2014):416-424

[62] De Windt TS et al. Allogeneic mesenchymal stem cells stimulate cartilage regeneration and are safe for single-stage cartilage repair in humans upon mixture with recycled autologous Chondrons. Stem Cells. 2017;**35**(1):256-264

[63] Bekkers JE et al. Single-stage cell-based cartilage regeneration using a combination of chondrons and mesenchymal stromal cells: Comparison with microfracture. The American Journal of Sports Medicine. 2013;**41**(9):2158-2166

[64] Toh WS et al. Cartilage repair using hyaluronan hydrogelencapsulated human embryonic stem cell-derived chondrogenic cells. Biomaterials. 2010;**31**(27):6968-6980

[65] Koyama N et al. Human induced pluripotent stem cells differentiated into chondrogenic lineage via generation of mesenchymal progenitor cells. Stem Cells and Development. 2013;**22**(1):102-113

[66] Takahashi K et al. Induction of pluripotent stem cells from adult human fibroblasts by defined factors. Cell. 2007;**131**(5):861-872

[67] Zhao J et al. Induced pluripotent stem cells: Origins, applications, and future perspectives. Journal of Zhejiang University. Science. B. 2013;**14**(12):1059-1069

[68] Tsumaki N et al. iPS cell technologies and cartilage regeneration. Bone. 2015;**70**:48-54

[69] Ko JY et al. In vitro chondrogenesis and in vivo repair of osteochondral defect with human induced pluripotent stem cells. Biomaterials. 2014;**35**(11):3571-3581

[70] Zhang J et al. A human iPSC model of Hutchinson Gilford Progeria reveals vascular smooth muscle and Mesenchymal stem cell defects. Cell Stem Cell. 2011;**8**:31

[71] Liu Y et al. One-step derivation of mesenchymal stem cell (MSC)-like cells from human pluripotent stem cells on a fibrillar collagen coating. PloS One. 2012;**7**

[72] Mimeault M, Batra SK. Recent insights into the molecular mechanisms involved in aging and the malignant transformation of adult stem/progenitor cells and their therapeutic implications. Ageing Research Reviews. 2009;**8**(2):94-112

[73] Yamashita A et al. Generation of scaffoldless hyaline cartilaginous tissue from human iPSCs. Stem Cell Reports. 2015;**4**(3):404-418

[74] Liao J et al. Recent developments in scaffold-guided cartilage tissue regeneration. Journal of Biomedical Nanotechnology. 2014;**10**(10):3085-3104

[75] Kock L et al. Tissue engineering of functional articular cartilage: The current status. Cell and Tissue Research. 2012;**347**(3):613-627

[76] Feng Q et al. Sulfated hyaluronic acid hydrogels with retarded degradation and enhanced growth factor retention promote hMSC chondrogenesis and articular cartilage integrity with reduced hypertrophy. Acta Biomaterialia. 2017;**53**:329-342

[77] He X et al. Layer-by-layer assembly of type I collagen and chondroitin sulfate on aminolyzed PU for potential cartilage tissue engineering application. Applied Surface Science. 2012;**258**(24):9918-9925

[78] Taraballi F et al. Biomimetic collagenous scaffold to tune inflammation by targeting macrophages. Journal of Tissue Engineering. 2016;**7**:2041731415624667

[79] Baino F et al. Bioceramics and scaffolds: A winning combination for tissue engineering. Frontiers in Bioengineering and Biotechnology. 2015;**3**:202

[80] Vinatier C et al. Cartilage engineering: A crucial combination of cells, biomaterials and biofactors. Trends in Biotechnology. 2009;**27**(5):307-314

[81] Quintavalla J et al. Fluorescently labeled mesenchymal stem cells (MSCs) maintain multilineage potential and can be detected following implantation into articular cartilage defects. Biomaterials. 2002;**23**:109-119

[82] Sharma B et al. Human cartilage repair with a photoreactive adhesive-hydrogel composite. Science Translational Medicine. 2013;**5**:167ra6

[83] Benthien JP, Behrens P. The treatment of chondral and osteochondral defects of the knee with autologous matrix-induced chondrogenesis (AMIC): Method description and recent developments. Knee Surgery, Sports Traumatology, Arthroscopy. 2011;**19**(8):1316-1319

[84] Pearce CJ et al. Synthetic osteochondral grafting of ankle osteochondral lesions. Foot and Ankle Surgery. 2012;**18**(2):114-118

[85] Deng Z et al. Cartilage defect treatments: With or without cells? Mesenchymal stem cells or chondrocytes? Traditional or matrix-assisted? A systematic review and meta-analyses. Stem Cells International. 2016;**2016**

[86] Cole BJ et al. Outcomes after a single-stage procedure for cell-based cartilage repair: A prospective clinical safety trial with 2-year follow-up. The American Journal of Sports Medicine. 2011;**39**(6):1170-1179

[87] Benders KE et al. Extracellular matrix scaffolds for cartilage and bone regeneration. Trends in Biotechnology. 2013;**31**(3):169-176

[88] Peretti GM et al. Tissue engineered cartilage integration to live and devitalized cartilage: A study by reflectance mode confocal microscopy and standard histology. Connective Tissue Research. 2006;**47**(4):190-199

[89] Elder BD et al. Extraction techniques for the decellularization of tissue engineered articular cartilage constructs. Biomaterials. 2009;**30**(22):3749-3756

[90] Pei M et al. A review of decellularized stem cell matrix: A novel cell expansion system for cartilage tissue engineering. European Cells & Materials. 2011;**22**:333-343

[91] Barlett W et al. Autologous chondrocyte implantation versus matrix-induced autologous chondrocyte implantation for osteochondral defects of the knee. A prospective randomized study. Journal of Bone and Joint Surgery. British Volume (London). 2005;**87**:640-645

[92] Riboh JC et al. Comparative efficacy of cartilage repair procedures in the knee: A network meta-analysis. Knee Surgery, Sports Traumatology, Arthroscopy. 2016

[93] Wakitani S et al. Repair of rabbit articular surfaces with allograft chondrocytes embedded in collagen gel. Journal of Bone and Joint Surgery. British Volume (London). 1989;**71**:74-80

[94] Sharma S et al. Biomaterials in tooth tissue engineering: A review. Journal of Clinical and Diagnostic Research: JCDR. 2014;**8**(1):309-315

[95] Chiang H et al. Differences between chondrocytes and bone marrow-derived chondrogenic cells. Tissue Engineering Part A. 2011;**17**(23-24):2919-2929

[96] Sanjurjo-Rodríguez C et al. Tissue engineering in an in vitro model of human cartilage repair. Osteoarthritis and Cartilage. 2016a;**24**(Suppl. 1):S169-S170

[97] Nguyen D et al. Cartilage tissue engineering by the 3D bioprinting of iPS cells in a nanocellulose/alginate bioink. Scientific Reports. 2017;**7**(1):658

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