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STEM CELLS IN ORTHOPAEDIC SURGERY (J DRAGOO AND K JONES, SECTION EDITORS)

Cell Therapy—a Basic Science Primer for the Sports Medicine Clinician

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Abstract

Purpose of Review The emergence of cell-based therapies has brought much excitement to the field of orthopedic sports medicine. However, the significant inconsistency of reporting has led to the poor understanding, misinformation, and false expectations for patients and clinicians alike. In this paper, we aim to clarify the available cell-therapy treatments and summarize some of the latest research.

Recent Findings Although this technology is in early development, our understanding of cell biology has grown significantly over the last decade. Furthermore, it is becoming evident that tissue specificity may play a significant role in determining the effectiveness and overall clinical benefit attributed to cell therapy.

Summary Cell therapy is an emerging field with tremendous potential for clinically significant benefit. However, in its current state, clinical application of these treatments is limited by federal regulations, variability in formulation, and limited understanding of the biologic activity of various cell formulations.

Keywords Cell therapy · Bioengineering · Regenerative medicine · Orthobiologics · Sports medicine · Orthopedics

Introduction

Injuries to musculoskeletal connective tissues, including tendon, ligament, meniscus, articular cartilage, and muscle, are increasingly common in active individuals and are becoming more frequent due to the combination of an aging population and the desire of older individuals to remain highly active [1]. These tissues have intrinsically poor healing potential following injury. Furthermore, they undergo age-related degenerative changes that compromises their function and capacity for healing and regeneration [2, 3]. When coupled with the lack of currently available effective therapies, it is clear that there is tremendous unmet need in the management of many sports medicine conditions. All together, the growing number of people suffering from musculoskeletal disease and the challenges in treating these patients serves as a powerful driving force for clinicians to pursue new therapies and management strategies.

Over the last several decades, regenerative medicine has emerged as a promising field with the potential to provide clinically relevant solutions for debilitating musculoskeletal disease. Regenerative medicine aims to harness the capacity to restore native tissue function and structure, potentially facilitating superior outcomes and decreased patient morbidity. The field of "regenerative medicine" encompasses several approaches including cell therapy, biologically active small molecules and cytokines, gene therapy applications, scaffold materials, and blood-derived products. However, the cell sits at the center of any of these approaches, since no healing or tissue regeneration can occur without cells. Advancements in diverse fields including developmental biology, molecular genetics, physiology, and computational biology have greatly improved our understanding of the complex cellular systems and their potential application in clinical medicine.

In addition to the need for improved understanding of stem cell biology, there is a critical need to further define the underlying cellular and molecular mechanisms of diverse tissue pathology in the patients we are treating. For example, the

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tissue changes present in the setting of chronic, age-related degeneration will differ from that following acute, traumatic injury. The effects of age, gender, co-existing medical co-morbidities such as diabetes, and smoking need to be defined. The critically important role of genetic and epigenetic factors needs to be determined. Finally, recent studies are making clear that various immune cell subtypes play an important role in the regulation and biologic activity of stem/progenitor cells.

Regulatory Environment

The United States Food and Drug Administration (FDA) retains regulatory control over medical, pharmaceutical, and biological products. This encompasses all human cell- and tissue-based products (HCT/P). In Title 21, Part 1271 of the Code of Federal Regulations (CFR), the FDA outlines its coverage of stem cells as part of HCT/P [4••]. Further guidance of the clinical use for HCT/P has been outlined by the Public Health Service Act (PHSA) and the Food and Drug Cosmetic Act (FDCA) [4••, 5]. What was initially expected to be a simple area of regulation resulted in a complex field with great potential for new therapeutic agents and approaches. In 2001, the FDA promulgated a plan to promote the safe use of HCT/Ps in human patients. This was followed in 2005 by the development of a 3-tiered approach to regulate the risks associated with the use HCT/Ps. Under section 361 of the PHSA, category 1 products are considered to be lower-risk products and do not require guidance under HCT/P laws [4••, 6]. Category 2 products are also considered lower-risk products; however, they must meet the following criteria: [1] minimal manipulation, [2] homologous use, [3] no combination products, and [4] cannot act systemically and cannot be dependent on the metabolic activity of living cells [4••]. Category 3 products define HCT/Ps considered to be higher-risk and do not meet the criteria outlined under category 2. Category 3 products are regulated under section 351 of PHSA, requiring clinical evaluation and investigational new drug (IND) approval by the FDA; this allows for more extensive screening to ensure products approved for clinical use follow stringent FDA guidelines.

Stem Cells

The term "stem cell" first appeared in literature in 1868 by the German scientist Ernest Haeckel when he theorized that all multicellular organisms originated from unicellular ancestors he called "stammzelle" [7]. Although his definition of a stem cell may have been broad, the principle idea that one cell can produce distinct varieties of cells and tissues sparked much interest. However, it was not until a century later that we began to understand what Haeckel meant by "stammzelle". In a series of papers, Owen and Friedenstein described how single precursor cells isolated from bone surfaces were capable of differentiation into various cell phenotypes, leading to what they called a stromal fibroblastic system [8–10]. These studies established that stromal tissue originated from a small population of stem and progenitor cells, whose ultimate fate could be determined by the microenvironment in which they reside. Although the definition of a "stem cell" is constantly evolving, at the most basic level a stem cell must [1] have the ability for self-renewal while maintaining an undifferentiated state and [2] possess the capacity for differentiation into mature, tissue-specific cells of various phenotypes [11].

Stem cells can be stratified to two major classes—embryonic and adult. Embryonic stem cells have the capacity to mature into any cell line in the body. These cells possess the ultimate "stem" capacity with no restrictions as to how they may differentiate. This unique quality also contributes to the challenges in controlling their growth and differentiation [12]. Because these cells are derived from human embryos, there are important ethical concerns that have limited the use embryonic stem cells for clinical application. Adult stem cells can differentiate into numerous cell lines; however, there are several key differences. Adult stem cells are multipotent, meaning they have a more limited capacity to differentiate into cells of similar origin. Adult stem cells can be harvested from readily available sources such as bone marrow, adipose tissue, synovium, or umbilical vein, making them an ideal candidate for cell therapy [3]. In the following paper, we will discuss basic concepts related to cell therapy, their potential for use by sports medicine clinicians, and the current limitations associated with these treatment strategies.

Mesenchymal Stem Cells

In his 1991 paper titled "Mesenchymal Stem Cell", Arnold Caplan described a population of plastic-adherent progeny cells, capable of bone and cartilage formation, repair, and turnover. He called these cells *mesenchymal stem cells*, referring to a single cell with the capability to differentiate and maintain numerous phenotypic lineages including bone, cartilage, tendon, and ligament [13]. However, his definition has since been challenged, even by himself [14]. In a recent paper, Caplan explored the numerous advancements in MSC research and elaborated how putative criteria and isolation methods for mesenchymal stem cells generates a heterogeneous population of multipotent cells [14]. Furthermore, he explains how the in vivo activity of MSCs to drive tissue healing and regeneration may be due to secretion of signaling molecules to the resident host cells, leading to the suggestion that the acronym "MSC" may represent *medicinal signaling cells*, as a more accurate description of their function [14, 15].

Most authors in this field use the criteria established by the International Society for Cellular Therapy (ISCT) when

discussing an "MSC" [11]. However, it should be noted that the original ISCT criteria defined mesenchymal *stromal* cells [11, 16]. These criteria defined stromal cells based on plastic adherence in tissue culture, a specific cell surface marker profile by flow cytometry, and ability to differentiate into several cell phenotypes (bone, cartilage, and adipose). These criteria were defined for *cultured* cells, which differ from the uncultured cell populations used in current "point of care" clinical applications. More importantly, the ISCT criteria are now felt to be outdated and not useful, as some of the cell markers label non-stem populations. Despite this, in this paper, we will use "MSC" in reference to cells defined by ISCT criteria for mesenchymal stromal cells given the ubiquity of this definition throughout the current literature.

MSCs are the most studied cell type for tissue regeneration in MSK injuries. These cells demonstrate tremendous potential to regenerate tissue by directly replacing damaged cells, mitigating inflammation, providing important "signals" to resident host cells in a paracrine fashion, and promoting vascular ingrowth [14, 17]. Additionally, MSCs are considered to be evasive from the immune system; as they have relatively low expression of HLA class 1 surface markers permitting potential use for allogeneic therapy [18]. These cells are defined as multipotent stromal stem cells with the capacity to differentiate into cells of the mesoderm—osteoblast, adipocytes, and chondrocyte. The widely accepted criteria for defining mesenchymal stromal cells include [1] adherence to plastic in culture, [2] expression of cell surface markers CD105, CD73, and CD90, and lack of expression of CD45, CD34, CD14 or CD11b, CD79alpha or CD19, and HLA-DR surface molecules [11]. Along with the multi-lineage differentiation capacity, MSCs have been isolated from various tissues, providing significant advantages for cell source and accessibility. The following paragraphs will cover MSC source and preparations.

- Bone marrow-derived MSC (bmMSC) are harvested from bone marrow aspirate. This cell source is the most studied MSC for therapeutic use. As with other stromal cell populations, the total number isolated from a single bone marrow harvest is very small and considered to be below the therapeutic threshold. For this reason, the development of potency assays has been a critical step in creating effective therapies [18]. Although current regulatory guidelines prohibit the in vitro expansion of harvested cells for clinical applications, researchers have developed novel techniques to increase cell quantity while maintaining the high level of potency. One method involves a modified cryopreservation technique to preserve cell viability and overall ability to secrete growth factors, providing clinicians the opportunity to perform multiple harvests to reach a therapeutic cell concentration [19, 20].
- Adipose-derived MSCs (aMSC) have emerged as a viable source for cell isolation. One significant advantage to this cell source is the ease of accessibility. Adipose-derived MSCs can be isolated from culture of the stromal vascular fraction preparation derived from subcutaneous fat through liposuction or lipoaspiration [21, 22].
- Synovial derived MSC (snMSC) have gained recent attention; these cells are isolated from samples of synovium and related nearby tissues, making this cell population a challenging source for autologous transplant. Prior studies demonstrate an increased number of snMSCs in the joint after injury, suggesting an intrinsic healing capacity, further supporting to their therapeutic potential [23].
- Birth-tissue-derived MSC (btMSC) are harvested from the amnion, placenta, and umbilical cord blood. In vitro studies have revealed a greater capacity for expansion, particularly with placenta-derived MSCs. However, further research is necessary to understand their full differentiation potential [24, 25].

Endothelial Progenitor Cells-Vascular Stem Cell

The vascular niche has long been known to play a critical role in wound healing and tissue regeneration. Recent studies suggest that the vascular niche functions to maintain the tissue microenvironment and homeostasis. Building off this principle, endothelial progenitor cells (EPC) have emerged as a viable source to augment neovascularization and healing [26, 27]. EPCs can be characterized as +CD34 or +CD133, and endothelial markers, +CD31, +Flk-1/kinase insert domain receptor (KDR)/VEGF receptor2 (VEGFR2), +vascular endothelial- (VE-) cadherin, and +Tie2. However, there remains some controversy over what defines a bona fide EPC [28, 29]. One unique quality of EPCs is that they can routinely be found circulating in the blood, although basic studies suggest that the true source of these circulating cells is from bone marrow. Further studies have revealed that tissue-specific endothelial progenitor cells may also provide proliferative and regenerative signals to the tissue [30]. The use of EPCs for tissue regeneration has primarily been focused on tissue ischemia models, particularly chronic limb ischemia [31, 32]. Additionally, EPCs are being studied in bone fracture healing as well as ligament reconstruction and healing [26, 33, 34].

Tissue-Specific Endothelial Cells/HUVECs

It is established that the vascular niche harbors a population of cells which produce various cytokines that regulate and control the intrinsic progenitor cells in a tissue-specific manner. The substances produced by these cells are termed "angiocrine factors" and are tissue-specific [35••]. These cells can be isolated from endothelium and represent an important research

tool to study the role of the intrinsic stem cell niche in tissue repair. While these cells lack stem-like properties, they possess the unique ability to augment their microenvironment and ultimately promote healing and regeneration [36].

Human umbilical vein endothelial cells (HUVECs) were first isolated and characterized by Jaffe et al. in the 1970s [37]. However, their therapeutic potential in soft tissue regeneration was not realized until more recently. A study by Yuan et al. demonstrated a dose-dependent migratory effect on bovine meniscal fibrochondrocytes (bMFC) cultured in the presence of HUVECs [38]. Similarly, the use of HUVECs has gained much interest in in vitro tissue regeneration as a means to enhance capillary networks allowing for the growth and development of larger tissue constructs [39].

Intrinsic Progenitor Cells

An intrinsic progenitor cell describes a unique population of cells specific to the tissue in which they reside. These cells retain the capability to terminally differentiate into tissue-specific cells while maintaining a state of self-renewal. First described in the tendon by Bi et al., it is believed that these cells possess the innate ability to proliferate, repair, and regenerate tendon tissue [40] (Bi 2007). Since then, the identification of tissue-specific progenitor cells has expanded to numerous tissues including ligament, meniscus, and cartilage [41•, 42, 43, 44, 45, 46]. Although these cells possess great potential for therapeutic application, autologous harvesting of cells yields sub-therapeutic concentrations. Furthermore, in their natural state, these cells remain quiescent therefore requiring biological, mechanical, and/or spatiotemporal cues to initiate differentiation and repair of the tissue.

Induced Pluripotent Stem Cells/Embryonic Stem Cell

Embryonic stem cells are derived from the inner cell mass of the blastocyst in the developing embryo and these cells can differentiate into essentially any cell type. Due to pervasive ethical considerations, embryonic-derived stem cells are not readily available or practical for research or therapeutic applications. Furthermore, this class of cells possesses a teratogenic potential, with studies highlighting the dangers of injecting pluripotent cells in the undifferentiated state [47, 48]. In contrast, induced pluripotent stem cells (iPSC) are generated from somatic cells and thus do not have associated ethical concerns. iPSCs are produced by the transient over-expression of four specific transcription factors in differentiated "adult" cells, such as cells derived from skin or blood. The challenge with iPSCs lies in establishing GMP-grade manufacturing processes to produce cells at clinical scale (i.e., adequate quantities for clinical use). A further risk related to iPSCs is the potential for oncogenic mutations to occur during culture expansion of iPSCs [49]. Rigorous FDA regulations have restricted the use of embryonic stem cells and iPSCs to FDA-approved trials. Currently, there are no approved therapeutic indications for the use of induced pluripotent stem cells in sports medicine.

Biologics–PRP/BMAC/APS/ACS

Platelet-Rich Plasma—PRP

The classic definition of platelet-rich plasma (PRP) describes a preparation of plasma with a concentration of platelets 5 times greater than baseline, or greater than 1 million platelets per milliliter [50, 51••]. The applications for PRP have expanded significantly over the last decade. Particularly, the use of PRP in sports medicine applications has grown to encompass many common pathologies [52]. Although the precise mechanism of action is incompletely defined, the biologic activity is attributed to the abundance of cytokines and growth factors possessed within platelet α-granules and dense granules. PRP supports cell migration and proliferation, but also promotes an immunomodulatory function, attenuating pain and inflammation. Some early data support a positive effect of PRP in treating symptoms of knee osteoarthritis and overuse tendinopathy [53•]. However, clinical studies evaluating PRP demonstrate tremendous variability in outcomes, leading clinicians to question the true benefits of this treatment. A fundamental challenge in the evaluation of PRP is the wide variability in PRP formulations due to lack of standardization in sample preparation protocols. Important factors to consider when deciding to use PRP include platelet concentration, leukocyte content, timing of platelet activation, fibrin content, and numerous other plasma proteins beyond those derived from platelet granules. Furthermore, patient demographics (age, sex) and medical comorbidities likely have important effects on the biologic activity of autologous blood products such as PRP. Furthermore, most third-party insurance carriers do not pay for the use of PRP due to the lack of robust clinical evidence, placing a large financial burden on patients receiving this treatment. While PRP is still widely used, further high-quality studies will be necessary to validate its efficacy.

Autologous Conditioned Serum and Autologous Protein Solution—ACS and APS

Further modulation of autologous blood products has resulted in the development of autologous protein solution (APS) and autologous conditioned serum (ACS). APS, like PRP, is processed by centrifugation and filtration of fresh whole blood samples. However, the production of APS introduces a second centrifugation step using modified devices containing polyacrylamide beads [54], further concentrating the protein components in whole blood while also retaining mononuclear cells

[55]. ACS production involves the incubation of whole blood samples in treated syringes for up to 24 h at 37 °C [56]. This process stimulates mononuclear cells to produce interleukin 1-receptor antagonist (IL-1RA), as well as other anti-inflammatory molecules [57]. Furthermore, the levels of pro-inflammatory cytokines, IL-1 and TNF-α, remain unchanged. The theoretical basis for the use of IL-1RA-rich products was established by studies performed in the early 1990s demonstrating how macrophages and monocytes serve as a robust source of IL-1RA, an inhibitor of the potent inflammatory mediator IL-1 [58, 59]. Although the use of autologous blood products sounds appealing in theory, there is need for more RCT and level 1 evidence to support the use of these formulations as effective therapeutics.

Alpha-2-Macroglobulin

Alpha-2-macroglobulin (A2M) has emerged as a potential targeted therapy for mitigating the degenerative environment common in many musculoskeletal diseases. Using large-scale screening with Western blotting and mass spectrometry, A2M was identified as major inhibitor of endogenous serum matrix metalloproteases—known drivers of inflammation and degeneration of connective tissues [60].

Bone Marrow Aspirate—BMA

Similar to PRP, the use of bone marrow aspirate (BMA) has grown significantly over the past decade. As the name suggests, BMA is harvested directly from bone marrow of the patient and centrifuged to create separate cell layers, effectively concentrating the cellular components of the aspirate. This process allows for concentration of hematopoietic stem cells (HSC), mesenchymal stem cells (MSC), platelets, and other mononucleated cells (WBC). Therefore, BMA allows for delivery of progenitor cells while adhering to the criteria of minimal manipulation set by the FDA. Although it is common practice for industry to label BMA as "stem cell" therapy, the prevalence of stem cells by formal criteria is approximately 0.005–0.01% of total cells [61, 62••, 63], propagating the poor understanding, misinformation, and false expectations that is pervasive in this field. BMA yields have shown to vary significantly based on source of harvest and inter-individual variability. It is known that aspirate from the iliac crest yields a higher concentration of progenitor cells when compared to other bones with hematopoetic marrow, such as femur and distal tibia. Furthermore, a study by Davies et al. demonstrated that this variability was not affected by patient age or other common demographic factors [64–66]. The use of BMA as a source of tissue augmentation has shown some promise in preclinical and clinical trials, despite lack of detailed understanding of the underlying biological processes. It is essential for clinicians to become accurately informed about the potential benefits and current limitations of cell therapy, and to understand that much of this remains unproven.

Cell Therapy for Soft Tissue Injury

ACL/MCL

Ligament repair and reconstruction are among the most common sports medicine procedures. Biologic approaches have been explored for augmentation of intrinsic ligament healing (i.e., knee medial collateral ligament) and for improved graft healing in ACL reconstruction. There are currently no approved stem cell therapies for ACL or MCL reconstruction in the USA. Bone marrow aspirate and PRP have been studied in preclinical and clinical trials, with some studies showing an added benefit to using these therapies. However, there is currently no consensus on whether or not PRP or BMA improve integration of a tendon graft in a bone tunnel, improve failure rates, and the overall clinical significance. To date, there is a paucity of rigorous data to support a role for cell therapy to improve mid-substance ligament healing, to accelerate ACL graft healing in a bone tunnel, or to improve maturation and remodeling of an ACL graft [67–69]. While the potential for cell therapy is significant, there remains a significant need for high-quality studies supporting the use of these therapies.

Meniscus

Meniscus injury is one of the most common injuries seen by clinicians. Given the important role of the meniscus in cartilage contact mechanics, preservation of the meniscus with repair when possible is favored over meniscectomy. However, meniscus healing is limited by several factors, including relatively poor vascularity, hypocellularity, fibrinolysates in the injured joint that prevent formation of an initial fibrin clot, and the complex mechanical loads on the healing meniscus.

Although cell therapy for meniscus repair has not been approved for use in the USA, there is much promise for the future. The application of bmMSC has been studied as a viable treatment strategy [70], and it has been established that the in vitro addition of MSC to electrospun scaffolds stimulates the production of glycosaminoglycan's (GAGs) and collagen-rich extra-cellular matrix consistent with the composition of native fibrocartilage tissue present in the central zone of the meniscus [71]. In a recent study, Piontek et al. describe a novel, all-inside meniscal repair technique using acellular collagen scaffold in conjunction with bone marrow aspirate to wrap meniscal lesions in patients who would otherwise

receive partial meniscectomy. At 2-year follow-up, 46 of 50 patients show statistically significant improvement on clinical assessment. Furthermore, MR imaging revealed evidence of healing in 38 of the 50 operated menisci, suggesting a novel and safe technique for meniscus repair [72•].

Synovial-derived cells appear to have significant potential to improve meniscus healing and regeneration. Several studies have reported improved meniscus healing with the use of snMSC [73•, 74]. In a series of studies, Sekiya et al. reported a significant increase in the presence of synovial-derived stem cells in the post-traumatic joint space, suggesting that these cells may play a significant role in meniscal and ligamentous healing [23]. Additionally, the use of HUVECs as paracrine signaling mediators stimulated enhanced migration of bovine meniscal fibrochondrocytes (bMFC) in vitro. Further evaluation revealed a paracrine effect, increasing expression of PDGF/R and VEGF/R through the function of endothelin-1 leading to improved integrative repair of explanted menisci [38]. In addition to intra-articular injection of stem cells, tissue-engineered scaffolds have emerged as a vehicle for delivery. Implanted scaffolds serve to provide structurally significant microarchitecture, as well as the necessary molecular cues to enhance and promote tissue regeneration. Furthermore, these scaffolds can be molded to a unique shape and size on an individual basis. Taking it a step further, the use of biologic and cell therapy in combination with scaffolds has revealed promising outcomes in pre-clinical studies. However, further investigation is necessary before clinical trials can be explored.

Tendinopathy

Tendinopathy is a common and debilitating disease prevalent among athletes and the general population. The underlying pathophysiology is due to cumulative microscopic matrix damage, with associated molecular inflammation and dysregulated activity of matrix metalloproteinases. The resultant clinical manifestations include activity-related pain, dysfunction, and progressive macroscopic tendon injury. This underlying pathophysiology is felt to be similar in diverse conditions such as rotator cuff tears, patellar tendonopathy ("jumper's knee"), lateral epicondylitis ("tennis elbow"), and Achilles tendon disorders. Currently available therapies including eccentric exercise therapy, NSAIDS, corticosteroid injections, and shockwave therapy are very limited and many patients continue to suffer from chronic pain and dysfunction. Given these limitations in our current treatment options, clinicians have looked to the use of cell therapy as a treatment option to modify the underlying biology and to restore tissue architecture, with the goal of mitigating the chronic degenerative response seen in recalcitrant cases.

Regeneration of the microstructure and composition of both normal tendon matrix and the native tendon-bone enthesis requires a complex interaction between cellular, biologic, and mechanical factors. While the exact mechanisms are not completely known, the need for mechanical loading in conjunction with appropriate cellular cues is essential for the development of tendon and the enthesis [75, 76]. There is very little data to support the use of cell therapy for treatment of overuse tendinopathy at this time. Current literature supports the possibility that cell therapy may be an effective adjuvant to improve tendon following surgical repair [77•, 78]. Cell therapy approaches may also be used as an adjunct to the use of currently available extracellular matrix patch materials. In this way, these scaffold materials may serve as an effective "carrier vehicle" for cell therapy. Extracellular matrix may provide important "signaling cues" for the cells [79–81].

Rotator Cuff Repair

Rotator cuff tears are a common and debilitating injury, often managed by surgical repair. Due to its complex structure and poor intrinsic healing quality of the enthesis, the success rate of these procedures is often unsatisfactory, despite advancements in surgical technique. Current clinical guidelines do not support the use of cell therapy; however, there is one study that reports improvement in rotator cuff tendon healing following surgical repair with bone marrow aspirate applied at the repair site, with a significantly higher rate of intact tendon repair at mean 10-year follow-up compared to untreated patients [78]. Taking it a step further, Rothrauff et al. studied the effects of adipose-derived stem cells delivered with hydrogels for acute and chronic massive rotator cuff repair in a rat model. Their study revealed decreased bone loss at the proximal humeral epiphysis in chronic, but not acute, tears regardless of delivery method suggesting a temporal relationship [82].

Knee Osteoarthritis

Knee osteoarthritis (OA) remains one of the most prevalent and debilitating orthopedic conditions worldwide, although little is known about the disease mechanism and factors that determine progression. For this reason, cell therapy has emerged as a potentially promising solution for these patients. The majority of studies have examined treatment of knee osteoarthritis, with variable results reported. Several studies using culture-expanded bone marrow cells have reported positive effects on both symptoms as well as some evidence of a positive effect on cartilage structure. However, a single-blind, placebo-controlled study demonstrated no significant difference when compared to saline injections [83•]. In a small prospective study of patient with chondral lesions, one study demonstrated how the supplementation of hyaluronic acid-based scaffolds with bmMSC results in significant clinical improvement at 5-year follow-up [84•]. Additionally, the use of biologics such as PRP, hyaluronic acid, and APS/ACS has

been of great interest to mitigate pain and inflammation associated with knee OA. In a study by Gobbi et al., they demonstrated that repeat intra-articular injections of PRP for knee OA demonstrated significant improvement in pain scores at 12 and even 18 months; however, these results were not sustained at 24 months [85]. While short-term studies have shown promising results, the long-term efficacy of these treatments is yet to be determined.

Conclusion

Many sports injuries involve tissues with intrinsically poor healing potential, including tendon, ligament, meniscus, articular cartilage, and muscle. Both acute injury to these tissues and chronic degenerative changes due to overuse represent difficult management conditions for the treating physician. Cell therapy has tremendous potential to affect the underlying biological activity of these difficult-to-treat tissues. Autologous tissues such as bone marrow and adipose contain a small population of progenitor cells with the potential to induce tissue healing and regeneration. However, it is recognized that the number of true stem cells by any formal criteria is very low in any of these autologous tissues. Changes in the regulatory environment in the USA is necessary to allow cell sorting to isolate and then expand in culture the small population of true stem cells in these tissues. A further limitation is the significant variability in cell formulations derived from different patients, which makes interpretation of the literature very difficult. Advances in this field will require careful characterization and reporting of the composition and the biological activity of various cell formulations used in clinical trials so that we can begin to relate the outcomes for different tissues to the actual material delivered. Further understanding of the biologic activity of various cell formulations will also allow the ability to identify the optimal cell type and formulation for specific pathologies and tissues, permitting the clinician to better match the treatment to the underlying condition being treated, rather than the "one size fits all" approach that has been used with orthobiologics. Progress in these areas will ultimately allow us to begin to realize the tremendous promise of cell therapy in sports medicine.

Compliance with Ethical Standards

Conflict of Interest Dr. Scott Rodeo and Mr. Bijan Dehghani declare that they have no conflict of interest.

Dr. Scott Rodeo reports personal fees from Ortho RTI and personal fees from Flexion Therapeutics, Inc. outside the submitted work.

Human and Animal Rights and Informed Consent This article does not contain any studies with human or animal subjects performed by any of the authors.

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- • Of importance
- •• Of major importance

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