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The Effects of Stem Cells on the Recovery of Rotator Cuff Injuries

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Abstract

Rotator cuff injuries lead to impaired shoulder function, weakness, decreased range of motion, and pain. Rotator cuff injuries account for more than 75,000 surgical repairs annually. Rotator cuff injuries are reported to increase with age. Direct annual healthcare expenses related to shoulder disorders is approximately $7 billion in the United States. Stem cell transplant procedures show promising potential as treatment for rotator cuff injuries. Rotator cuff surgery may lead to extensive medical complications or potential side effects. Researchers have investigated the effectiveness of stem cells for rotator cuff injuries in animal and human subjects. Mesenchymal stem cells are the preferred source for therapy for orthopedic procedures as they differentiate into a variety of tissues including muscle, bone, fat, and cartilage. Stem cell transplantation has shown enhanced tissue quality, improved rate of healing, reduced pain, improved clinical outcomes, sustained functional gains, and reduced incidence of rotator cuff re-tear injuries. Researchers believe the use of stem cells may offer alternative options for patients with orthopedic injuries, including rotator cuff tears, to improve function and decrease recovery time.

Keywords

Rotator cuff injury; Stem cells; Treatment

Introduction

Rotator cuff injuries account for more than 4.5 million annual physician visits and more than 75,000 surgical repairs performed annually [1-4]. Each year, 18 million Americans report shoulder pain including a large percentage of rotator cuff disease [5]. A 10% lifetime incidence of shoulder disorder pain is reported United States, including 15 new cases per 1,000 reported in the at-risk population. Rotator cuff tears are reported to be a common problem and increase with age from 4% incidence in individuals age 40 to 60 years to more than 54% in individuals over 60 years [4,6]. At primary care visits, shoulder pain is reported to be the third most common musculoskeletal complaint. Direct annual healthcare expenses attributed to shoulder disorders is approximately $7 billion in the US. Rotator cuff disorders are the most common underlying cause, with estimates varying between 65% and 85% depending upon the setting and age of the study population [7]. Approximately 25% of individuals in their 60s and 50% of individuals in their 80s have full thickness rotator cuff tears. The first surgical intervention for rotator cuff injury was reported in 1911 [4].

The shoulder is a complex joint involved with movement of the upper extremities with the axial skeleton or trunk. It plays an important role in the dexterity and function of the arms and hands, which separates functionality of human beings apart from other mammals. Strong demands of strength, endurance, and flexibility are placed on the shoulder through daily activities, and therefore may result in musculoskeletal complaints. The shoulder is composed of a several soft tissues that overlay the skeleton. Bone anatomy involves the scapula, a flat triangular bone forming the posterior aspect of the shoulder girdle. The scapula has 17 muscular attachments including an anterior projection named the glenoid which forms half of the primary shoulder joint. The shoulder complex is composed of 4 smaller joints: glenohumeral (GH) joint, acromioclavicular (AC), sternoclavicular (SC), and scapulothoracic (ST) joints [8].

The superior shoulder suspensory complex is a bony and soft tissue ring involving these joints and is responsible for the coordination between the axial skeleton and the upper extremity. This complex works together with many ligamentous and muscular attachments. The GH articulation is surrounded in the shoulder joint capsule. Superiorly, this joint is covered by the acrimony, a bony anterior-superior scapular projection. The acrimony articulates with the clavicle and serves as the anterior connection to the axial skeleton. Rotator cuff is comprised of a collection of four muscles: supraspinatus, teres minor, infraspinatus and subscapularis, and their associated tendons. All of these muscles play a key role in shoulder movement as they provide strength and stability to the joint during movement [1,8] (Figure 1).

Common rotator cuff injuries may involve the tendon; which can become inflamed or tear due to mechanical overuse. Rotator cuff injuries lead to impaired shoulder function, weakness, decreased range of motion, and pain. The supraspinatus tendon is the most frequently injured tendon due to the anatomical location inferior to the acromion bone and this affects the tendon during overhead motion [1,7]. The glenoid has an increased upward tilt that leads to increased risk of tears of the supraspinatus tendon [10].

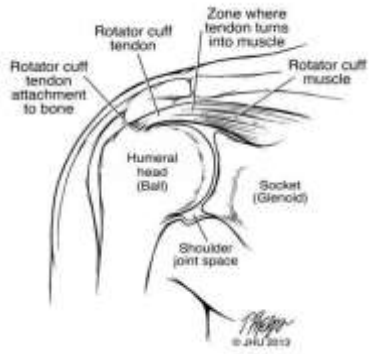

Figure 1: Shows anatomy of a normal shoulder [9].

An increased prevalence of rotator cuff tears was found in shoulders with a flatter slope of the acromion, increased anterior projection, and decreased lateral acromial angle on lateral radiographs. These changes reduced the size of the supraspinatus outlet and lead to increased pressure or friction on the rotator cuff tendons and progressive tearing [10] (Figure 2).

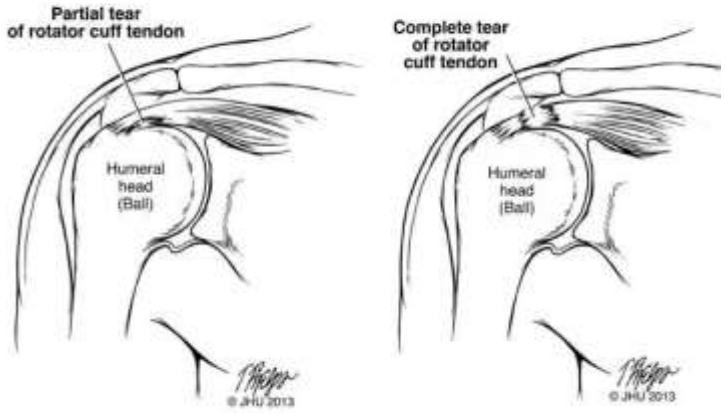

Figure 2: Shows partial and complete rotator cuff tear [9].

Rotator cuff tears usually develop in the supraspinatus tendon and can lead to partial and then eventually full-thickness tearing. Once developed, the progression of a tear may be difficult to predict. Some tears continue to increase in size, and other tears may remain dormant and do not show signs of progression [10].

Rotator cuff tear may occur in young people associated with trauma (e.g., acute shoulder dislocation), but typically occur in middle-aged or elderly people and not attributed to trauma. Rotator cuff tears are a result of several biological and mechanical factors: degenerative tendon disorders including vascular, cellular, and tendon matrix changes, increased age, high body mass index, hypertension, hypercholesterolemia, smoking, and genetic factors [11,6].

Diagnosis of rotator cuff injuries include full medical and functional evaluations by orthopedic surgeons and physical therapists. Imaging modalities such as magnetic resonance imaging (MRI) and ultrasound have revealed high specificity and sensitivity in the diagnosis of full-thickness rotator cuff tears. Both tests have shown accuracy for detecting full-thickness tears in people considering surgery for shoulder pain. Both ultrasound and MRI may have shown poor sensitivity for detecting partial-thickness tears, and sensitivity of ultrasound is lower than MRI [7].

An important component of the shoulder physical exam includes range of motion. Normal range of motion for the shoulder includes: forward flexion from 150 to 180 degrees, extension from 40 to 60 degrees, abduction from 150 to 180 degrees, external rotation from 60 to 90 degrees, and internal rotation to the mid-thoracic level, or 50 to 70 degrees. Examination should include both active and passive range of motion [8].

Clinical researchers utilized a variety of standardized functional outcome measurement tools to evaluate overall pain, range of motion, and strength: Constant Murley Scale, Shoulder Pain and Disability Index (SPADI), University of California at Los Angeles (UCLA) Shoulder Scale, Disabilities of the Arm, Shoulder, and Hand (DASH), and other shoulder function scales [7] (Table 1).

Disabilities of the Arm, Shoulder, and Hand (DASH)
Rate your ability to complete the following tasks: • No Difficulty • Mild Difficulty • Moderate Difficulty • Severe Difficulty • Unable
○ Open a tight or new jar ○ Write ○ Turn a key ○ Prepare a meal ○ Push open a heavy door ○ Place an object on a shelf above your head ○ Do heavy household chores (e.g., wash walls, wash floors) ○ Garden or do yard work ○ Make a bed ○ Carry a shopping bag or briefcase ○ Carry a heavy object (over 10 lbs) ○ Wash or blow dry your hair ○ Wash your back ○ Put on a pullover sweater ○ Use a knife to cut food ○ Recreational activities which require little effort (e.g., card playing, knitting, etc.)

○ Recreational activities in which you take some force or impact through your arm, shoulder, or hand (e.g., golf, hammering, tennis, etc.) ○ Manage transportation needs (getting from one place to another) ○ Sexual activities ○ Social activities with family, friends, neighbors, or groups ○ Work or other daily activities ○ Sleeping because of pain in your arm, shoulder, or hand
Rate the severity of the following symptoms: • None • Mild • Moderate • Severe • Extreme
○ Arm, shoulder, or hand pain ○ Arm, shoulder, or hand pain when you performed any specific activity ○ Tingling (pins & needles) in your arm, shoulder, or hand ○ Weakness in your arm, shoulder, or hand ○ Stiffness in your arm, shoulder, or hand

Table 1: Lists items on the Disabilities of Arm, Shoulder and Hand (DASH) Standard Evaluation Tool [12].

Non-operative treatment options include physical therapy for muscle strengthening, scapular stabilization, stretching and flexibility exercises, glucocorticoid injection, non-steroidal anti-inflammatory drugs, acupuncture, iontophoresis, phonophoresis, transcutaneous electrical nerve stimulation, pulsed electromagnetic field, topical glyceryl trinitrate, and ultrasound [7].

Patients with symptomatic tears typically do not show positive results from non-operative options. Patients who respond to nonsurgical management will typically do so within the first 6 to 12 weeks [5,7].

Corticosteroid injections (CSI) may be used for short term pain control, but do not show improved healing results. Patients with rotator cuff injury who receive corticosteroid injections may show short term pain reduction and functional movement for 3 to 6 weeks, but not long-term results lasting longer than 24 weeks. Several recent clinical trials have demonstrated that CSIs may lead to increased risk of revision surgery after rotator cuff repair. Corticosteroid injection treatment should not be utilized if a rotator cuff repair is to be performed in the upcoming 6 months [13,14].

Platelet rich plasma (PRP) is an autologous concentration of human platelets in a small volume of plasma that is made from taking a sample of the patient's own blood and running it through a centrifuge which leaves a high concentration of platelets. These platelets secrete a number of proteins, cytokines, and other bioactive and growth factors that support healing and angiogenesis [4,15,16].

Since the 1990s, platelet rich plasma has been utilized in plastic and maxillofacial surgery. Platelet-rich plasma has gained increased attention in orthopedic sports medicine over the past several years. Several investigators have supported the use of platelet-rich plasma in the management of bone, muscle, tendon, and cartilage injury [17,18]. Growth factors released by platelets perform a wide range of functions that are regenerative including modulation of local inflammatory responses, proliferation and recruitment of stem cells, and stimulation of new blood vessel formation [18,19]. Concentrated platelets in PRP are suspended in a small volume of plasma and contains the 3 proteins in blood that act as cell adhesion molecules for osteo-conduction as well as a matrix for bone, connective tissue, and epithelial migration [16].

Several researchers reported positive outcomes related to clinical us of PRP for orthopedic patients including reduced pain in early postoperative period following surgery, increased vascularization response, lower failure to heal and re-tear rate in small to medium tears, accelerated functional recovery, and improved tendon healing [11,20-23].

Surgery is usually considered when other treatments are not successful. Rotator cuff surgery was first implemented in 1911. Surgery options may include open surgery, mini-open surgery, or arthroscopic surgery. Surgery includes removing part of the bone to broaden the tendon passage and repair of the torn tendons. Most rotator cuff surgery is now performed arthroscopically or through mini-open surgical approach. Outcomes have been reported as favorable for open, mini-open, and arthroscopic repairs [1,4,5,7] (Table 2).

Open rotator cuff surgery includes five fundamental principles: repair of deltoid origin, subacromial decompression with division of the coracoacromial ligament, release of cuff to obtain freely mobile muscle-tendon units, secure transosseous fixation of tendon to greater tuberosity, and rehabilitation with early passive motion. Open rotator cuff repair has shown good to excellent outcomes for pain relief (85%-100%) and functional improvement (75% - 95% of patients) [4].

American Academy of Orthopedic Surgeons Appropriate Use Criteria (AUC) Treatment of Full-Thickness Rotator Cuff Tears
(1) nonsurgical management is always appropriate if patients have a positive response to conservative care (2) repair maybe appropriate for a reparable tear even if patients respond to nonsurgical treatment

(3) repair is the appropriate treatment in healthy symptomatic patients who failed conservative management (4) debridement/partial repair and/or reconstructions may be appropriate in chronic massive tears (5) arthroplasty is a maybe appropriate option for healthy patients with painful pseudoparalysis and an irreparable tear

Table 2: Lists American Academy of Orthopedic Surgeons Appropriate Use Criteria (AUC) for treatment of full-thickness rotator cuff tears [5].

Mini-open surgical technique allows surgeons the ability to evaluate the glenohumeral joint and perform subacromial decompressions without taking down the deltoid. Mini-open surgical repair is started arthroscopically. The surgeon can then extend the lateral portal to enable mini-open access and complete the surgery. Studies report good to excellent results for pain relief and functional improvement (85% - 95% of patients). Open repair surgery compared with mini-open surgery found equally effective results for pain relief and functional outcomes. Shorter hospital stays and quicker return to daily activities was reported for the mini-open surgery group compared to the open surgery group [4].

Arthroscopic surgery involves small skin incisions, access to glenohumeral joint for inspection and treatment of intra-articular lesions, and less dissection of soft tissue. Intra-articular sores are identified and corrected arthroscopically by glenohumeral shoulder surgery. Advocates for arthroscopic surgery repairs support that arthroscopy gives surgeons the ability and flexibility to completely visualize and analyze a rotator cuff tear. Results of arthroscopic surgery repair are similar to open surgery and mini-open surgery repairs, with 85%-95% of patients reporting improved pain relief and functional outcomes. With the advantages of smaller skin incisions, no deltoid detachment, and less soft tissue dissection, arthroscopic techniques continue to show increased popularity with surgeons for the treatment of rotator cuff tears [4].

Single row surgical method of anchor fixation in which one row of suture anchors is placed in the greater tuberosity on the lateral aspect of the rotator cuff footprint. This method does not completely recreate the native footprint insertion of the supraspinatus tendon on the greater tuberosity but spot-welds it and can potentially lead to incomplete healing [4].

Double-row surgical technique involves a second row of suture anchors placed medially closer to the articular margin and one set of sutures shared between the two rows acting to compress the rotator cuff on the original area as well as providing additional surface area to promote healing. Improved functional outcomes were reported for patients with large to massive tears (>3 cm) that underwent double-row surgery. Double row procedures have greater difficulty, increased time required, and increased expense compared with single-row repairs. Researchers suggested double-row fixation should

be considered for patients that have large tears and/or have higher functional demands following surgery [4,5].

Transosseous-equivalent repair surgery involves a medial row of suture anchors placed with sutures tied in a mattress fashion. Suture limbs are preserved and bridged over the footprint insertion with distal-lateral interference screw fixation. This repair is different from double-row fixation because it does not require a second row of suture anchors and maximized tendon-to-bone compression. Studies have suggested transosseous repair provides more contact area and pressure over the footprint compared with double-row fixation and equivalent strength compared to open transosseous techniques. Short term clinical studies have shown positive functional and structural outcomes for transosseus surgery compared with double-row fixation surgery [4].

Several patient related factors affect healing potential for rotator cuff repair: patient age, tear size, and tear chronicity. Increased patient age has shown poor healing results after open, single-row arthroscopic, and double row arthroscopic repair. Patients over 65 years of age reported 43% healing as compared to patients under 65 with a reported healing rate of 86% for single tendon tears that underwent arthroscopic surgery. Researchers have also shown tear size correlates with tendon healing as larger tears showed worse healing rates after single-row and double-row rotator cuff repairs. Also, poor rotator cuff muscle quality has been correlated with decreased rotator cuff healing [6].

Patients typically wear a sling for 3-6 weeks after surgery and undergo rehabilitation with physical therapy for up to six months after surgery. Potential risks of surgery include complications related to the surgery or anesthesia such as pulmonary embolism, surgical site infection, postoperative adhesive capsulitis, peripheral nerve injury, chronic pain, and failed rotator cuff repair [4,7] (Table 3).

Orthopedic Surgeries for Rotator Cuff Tears	
open surgery	• repair of deltoid origin; subacromial decompression with division of the coracoacromial ligament • release of cuff to obtain freely mobile muscle-tendon units • secure transosseous fixation of tendon to greater tuberosity • rehabilitation with early passive motion • good to excellent outcomes for functional improvement (75% - 95% of patients) and pain relief (85%-100%) for patients
mini-open surgery	• ability to evaluate the glenohumeral joint and perform subacromial decompressions without taking down deltoid

	• can be initiated arthroscopically before extending lateral portal to facilitate mini-open access to complete repair • good to excellent results for pain relief and functional improvement (85% - 95% of patients) • equally effective results for pain relief and functional outcomes for open repair & mini-open repair • shorter hospital stays and quicker return to daily activities for mini-open surgery group compared to open surgery group
arthroscopic surgery	• small skin incisions • access to glenohumeral joint for inspection and treatment of intra-articular lesions • less dissection of soft tissue • gives surgeons ability and flexibility to completely visualize and analyze a rotator cuff tear • arthroscopic surgery repair results similar to open surgery and mini-open surgery repairs, functional outcomes & pain relief (85% - 95%) for patients
single row repair surgery	• anchor fixation, one row of suture anchors placed in greater tuberosity on lateral aspect of rotator cuff footprint • not completely recreate native footprint insertion of supraspinatus tendon on greater tuberosity • can potentially lead to incomplete healing
double row repair surgery	• involves second row of suture anchors placed medially closer to articular margin and one set of sutures shared between two rows • acting to compress rotator cuff on its native footprint and increasing surface area for healing • improved functional outcomes reported for patients with large to massive tears (>3 cm) • procedures have greater difficulty,

	increased time required, and increased expense compared with single-row repairs • should be considered for patients with large tears and/or higher functional demands following surgery
transosseous equivalent repair surgery	• involves medial row of suture anchors placed with sutures tied in mattress fashion • suture limbs preserved and bridged over footprint insertion with distal-lateral interference screw fixation • does not require a second row of suture anchors and maximized tendon-to-bone compression • provides more contact area and pressure over footprint compared with double-row fixation and equivalent strength • positive functional and structural outcomes compared with double-row fixation surgery

Table 3: Lists Orthopedic Surgeries for Rotator Cuff Tears [4,5].

Discussion

Stem cells are a promising area of research within the medical field for regenerative medicine due to their ability to self-renew and undergo differentiation along multiple lineages. Tissue specific stem cells can undergo multilineage differentiation, including mesenchymal stem/stromal cells (MSCs) or are largely restricted to a single lineage, such as muscle satellite cells. For musculoskeletal regeneration, multilineage stem cells derived from bone marrow stem cells or fat adipose-derived stem/stromal cells (ASCs) are the most commonly used stem cells [1,24,25].

Stem cells have the ability to differentiate into more than 200 different cell types in the body. They can create new cells in existing healthy tissues and also help repair tissues in damaged or injured structures when they differentiate into multi lineages and become multipotent under appropriate conditions. The cells create progenitor cells and have more specialized functions such as red blood cells, bone, brain cells, or cartilage [25-30].

Stem cells may be considered embryonic or adult stem cells. Additionally, these stem cells can be further classified as multipotent, totipotent, or pluripotent. Totipotent cells are only present in early embryo and capable of becoming an entire organism. Totipotent cells are not used widely for clinical use due to ethical concerns. Pluripotent cells can differentiate to every cell and can develop into cells of all

the three germ layers: endoderm, ectoderm or mesoderm. Multipotent cells are part of a specific germ layer and become organ-specific progenitors. Adult stem cells and cord blood cells are multipotent cells [1,25,31,32].

Mesenchymal stem cells (MSCs) are the most preferred source for cell therapy because they can differentiate into a variety of tissues including muscles, bones, fat, and cartilage. MSCs originate from the mesoderm and can be obtained from many sources including tendon, bone, skin, umbilical cord, adipose tissue, blood, and amnion. Bone marrow, adipose tissue, and muscle derived MSCs are most commonly used because they are abundantly available and easily obtained. MSCs have a good potential to develop into adipocytes, myoblasts, chondrocytes, and osteoblasts [25,31,33-36].

Skeletal muscle comprises different types of stem progenitor cells such as satellite cells and non-satellite stem cells including MSCs, interstitial stem cells, fibro/adipogenic progenitors/mesenchymal stem cells, muscle side population cells, and muscle resident pericytes. All of these stem cells have the ability to participate muscle regeneration process [37,38-40]. Satellite stem cells are a common progenitor cell that can differentiate into osteoblasts, adipocytes, chondrocytes, and myocytes [41]. When activated, satellite cells proliferate, migrate from the myofibers, express specific myogenic markers, and become muscle precursor cells. Recent studies on muscle satellite stem cells reviewed the possible use in repair of muscles and regeneration of tissues including bone and cartilage [41].

Stem cell trials in animals for orthopedics

A study evaluating 33 animal models revealed that the rat possessed a shoulder anatomy most similar to human with a prominent supraspinatus tendon passing beneath an enclosed bony arch and insertion into the proximal humerus at the greater tuberosity. These anatomical characteristics can lead to supraspinatus tendon impingement in both humans and rats when overhead forelimb activities are completed. This may lead to degeneration of the tendon over time [1].

The rotator cuff has a limited ability for intrinsic healing without surgical repair. Several investigators evaluated spontaneous rotator cuff healing in animal models. No evidence of rotator cuff healing was found at three weeks in a 12 mm tear in a rabbit supraspinatus tear model. Inadequate rotator cuff repair was found in a rat supraspinatus tear model where 78% of tendons had persistent defects at 12 weeks after a two mm defect was created. These results suggest very limited potential for spontaneous rotator cuff healing and repair without surgical intervention [6].

Dragoo *et al.,* in 2003 evaluated tissue-engineered cartilage and bone using *in vitro* techniques and placed cells into hind legs of five immunodeficient mice [42]. Radiological and histological analysis indicated processed lipoaspirate cells induced into the chondrogenic phenotype had the appearance of hyaline cartilage after six weeks. Cells transfected with the BMP-2 gene media produced profuse amounts of bone and were beginning to establish a marrow cavity. This study implies osteochondral defects can be treated with cartilage and bone that is engineered from infrapatellar fat pad tissues [42].

Lim *et. al.,* in 2004 investigated the effect of using mesenchymal stem cells (MSCs) to coat tendon grafts and the quality and rate of graft osteointegration for reconstruction of anterior cruciate ligament (ACL) [43]. Forty-eight adult rabbits underwent bilateral ACL reconstructions using hamstring tendon autographs. A fibrin glue carrier was utilized to coat grafts with MSCs in one limb and fibrin glue only was used in the other. Mature scar tissue with some Sharpey's-like fibers that spanned the tendon-bone interface was shown at eight weeks. At two weeks, the MSC-enhanced reconstructions had sizable areas of cartilage cells at the tendon-bone junction. A mature zone of cartilage was seen blending from bone into the tendon grafts by eight weeks. The MSC-enhanced grafts demonstrated higher failure load and stiffness at eight weeks. MSC-enhanced ACL reconstructions showed improved performance on biomechanical testing compared to controls. This study suggested that MSCs can be used to biologically alter the normal healing process of hamstring tendon grafts to their surrounding bony tunnels and lead to improved biomechanical ACL reconstruction [43].

A study by Kanaya *et. al.,* in 2007 examined the effects of intra-articularly injected mesenchymal stomal cells on acceleration of healing of a partially torn anterior cruciate ligament (ACL) [44]. The study was conducted with ninety-eight 12-week-old male Sprague-Dawley rats. The right ACL underwent partial transection, and a sham operation was performed on the left knee in the rats. Mesenchymal stromal cells extracted from bone marrow of green fluorescent protein transgenic Sprague-Dawley rats or saline was injected into the injury site of the rats. In the MSC+ group, the transected area was covered with healing tissues at two and four weeks after surgery. Four weeks after surgery, the histological score of the MSC+ group was better than the MSC- group. The transected area of the MSC- group remained void of any tissues during all times after surgery. This study suggested that injected mesenchymal stromal cells could accelerate healing of partially torn ACLs and intra-articular injection of mesenchymal stromal cells may be a possible option for treating partially torn knee ACLs [44].

Ju *et. al.,* in 2007 designed a study to investigate the effect of the implantation of the synovial MSCs on tendon-bone healing in rats [45]. Achilles tendon grafts of nineteen 12-year-old mature Sprague-Dawley rats had half of the Achilles tendon grafts inserted into a bone tunnel from the tibial plateau to the tibial tuberosity. Fluorescent marker DiI was used to identify the MSCs that filled the bone tunnel. Additional bone tunnel was not filled with the MSCs and was identified as the control. Tendon-bone interface was filled with DiI-positive cells at one week. By two weeks, the proportion of oblique collagen fibers was higher in the MSC group than the control group. At four weeks for both groups, there was tissue disappearance, and implanted tendon was attached directly to bone. It appeared that implantation of synovial MSCs into bone tunnel was found to accelerate early remodeling of tendon-bone healing. The effect, however, was not observed at four weeks [45].

A study conducted by Oe *et. al.,* in 2010 compared anterior cruciate ligament regeneration between animal groups with intra-articular injection of whole bone marrow cells (BMC), cultured mesenchymal stem cells (MSC), or saline [46]. Forty 13-week-old Fischer 344 rats were prepared with partially transected ACLs. All rats received injections of BMCs, MSCs, or saline into the articular cavity at one week after transection. At four weeks, donor cells were detected in the transected ACLs of the BMC and MSC groups and their ACLs appeared almost normal histologically. An increased volume of spindle cells

was discovered in the BMC group than the saline group at four weeks. At four weeks after injection, the BMC group tensile strength reached near normal levels. This study supports bone marrow transplantation using fresh whole BMCs as an effective treatment option for partial rupture of ACL [46].

Stem cells trials in humans for orthopedics

Stem cell use has the potential to revolutionize orthopedic practice including healing of bone defects caused by trauma, infection to cartilage defects, tumors, nerve tendon, and ligament healing [31]. Physician interest in stem cells and tissue engineering has increased over the past two decades related to orthopedic surgery, with additional focus on the musculoskeletal system [25].

Orthopedic surgeons have found success implementing stem cells for numerous orthopedic procedures including injuries and defects with bones, joints, ligaments, tendons, and cartilage. Stem cells have also been combined with scaffolds and used in bone tissue engineering to promote more rapid and better healing of tissues. Mesenchymal stem cells (MSCs) are the most preferred source of stem cells for orthopedic procedures because they can differentiate into a variety of tissues including- muscles, bones, fat, and cartilage [25,30,31].

Fractures-Bone Defects

Bone fractures and trauma-related joint injuries account for a large number of surgical procedures and are the primary issue of orthopedic surgery. Bone defects may occur from extrusion of bony fragments following high-energy trauma or gunshot injuries. Several researchers utilized membranes that contain MSCs for femoral defects and found that vascular endothelial growth factor, BMP-2, and TGFβ were elevated significantly in membranes. Other researchers investigated the use of bone marrow derived stem cells for fracture healing and found that BMDSCs enhanced bone formation and increased density of bone minerals [25,47-49].

Nonunion

An average of 6.2 million fractures occurs annually and 5% - 10% of the fractures resulted in nonunion. Severe complications and higher rates of patient morbidity and mortality can result from nonunion. Bone marrow aspirates that contain mesenchymal stem cells have been utilized to enhance nonunion injury healing. Tissue engineering that involved the use of stem cells with scaffolds, demineralized bone matrix, and tri-calcium phosphate were evaluated and were useful for bridging bone defects. Bone marrow aspirates containing mesenchymal stem cells have been used in seven patients for nonunion of long bones and reported successful results [25,31,50-52].

Spinal injuries

Spinal cord injury is one of the most frequent reasons for severe neurological damage. Stem cells have been utilized for axonal regeneration. Researchers performed bone marrow derived stem cell transplantation for 20 patients with spinal cord injury and reported positive outcomes. An additional 20 patients with spinal cord injury underwent umbilical cord blood derived mononuclear cell transplantation with 75% of patients demonstrating improved motor function [25,53-55].

Osteoarthritis-cartilage defects

Osteoarthritis is a degenerative joint disorder affecting weight bearing joints. The joint cartilage degenerates and joint space becomes narrow in the early stages. Patient comfort and activity level are affected. Stem cell studies have shown encouraging results for osteoarthritis and provided joint cartilage regeneration to become a considerable alternative for nonsurgical options [25,56,57].

Cartilage defects

Regeneration capacity of damaged hyaline cartilage is limited, and thus for a fast and efficient repair, materials are needed. Mesenchymal cells have been recommended because they have demonstrated reproducible and reliable effects on cartilage repair. Several studies have shown human bone marrow stem cells (hMSCs) to have regenerative and chondrogenic potential. Researchers reported clinical application of MSCs for the treatment of osteoarthritic knees with improved arthroscopic and histological outcomes. Bone marrow MSCs were also combined with TGF-beta for treating full-thickness articular cartilage defects and reported positive results [25,58-62].

Ligament-tendon injuries

Approximately 50% of musculoskeletal injuries involve soft tissue injuries including tendons or ligaments. Complete ligament and tendon healing require an increased recovery period up to one to two-and-a-half years postinjury. Re-injury risks are increased due to decreased mechanical strength. Researchers report stem cell therapy improves ligament-tendon healing. Some researchers reported lower doses of MCSs were more efficient for improved healing than higher doses of MSCs. Continued research to suggested for this area to optimize appropriate stem cell protocols [25,63-66].

Anterior cruciate ligament lesions

Knee injuries are common and 17%-61% require surgical repair. Mesenchymal stem cells and ACL fibroblasts have been found to have regenerative effects on anterior cruciate ligament lesions. MSCs have promoted early bone tendon interface healing by increasing Sharpey's fibers proportion. MSCs used with bone morphogenic protein 2 have shown improved biomechanical properties of the bone tendon interface including maximal load and stiffness [25,31,67-69].

Femoral head osteonecrosis

Femoral Head Osteonecrosis is a progressive disease that is typically seen in young adults caused by femoral head nutrition disruption and joint degeneration. Researchers investigated local MSC for treatment of femoral head avascular necrosis and found that MSC therapy decreased the total knee arthroplasty requirement for patients [25,70].

Osteogenesis imperfecta

Osteogenesis is one of the genetic disorders of mesenchymal cells typically including generalized osteopenia, fractures, and bone deformity. Complete cure is not possible with the current treatment interventions. Researchers stated that there was an increase in bone mineral density and decrease in long-term fractures for patients with osteogenesis imperfecta treated with BM-MSCs [25,71].

Ulnar collateral ligament tear

Ulnar collateral ligament (UCL) tears of the elbow are injuries common in overhead athletes. Surgical reconstruction of the UCL has led to improved outcomes, but not all athletes are able to return to their previous competition level. The time required for return to sport typically averages one to two years. A single baseball player served as the subject for researchers to investigate the use of alternative methods to reconstruct UCL, including MSC, PRP, and dermal allograft. No complications were found and three months postoperatively, intact dermal allograft without any surrounding inflammatory response was revealed by MRI. An additional MRI 17 months postoperatively showed intact dermal allograft without deformity or retraction at the surgical site. The patient was currently one year and nine months postop from UCL reconstruction and doing well without any indication of instability or ulnar neuropathy. The patient's postoperative course was reported to be uncomplicated, and he had successful return to throwing 86 miles per hour. Additional research studies with higher levels of evidence are needed to compare augmentation technique with conventional UCL reconstruction to determine if overhead athletes can have significant improvements in return to play and rate of return to play [72].

Professional athletes use of stem cells

Many professional athletes, including hundreds of National Football League (NFL) players and others, have utilized stem cell treatments to promote injury healing and improved recovery times. Over the past five years, many NFL players have publicly promoted stem cell treatments to support continued careers after injuries. NFL players' endorsement of stem cell therapy may encourage others to view these procedures as safe and effective, even without evidence based scientific data to support the clinical use of stem cells. [73,74].

Success for stem cell therapy with athletes has been primarily anecdotal, since many athletes have the ability to recover more quickly than other people, due to youth, overall muscle tone, genes, and training protocols. Athletes may assume that stem cells are what led to their improved recovery; however, rest, ice, heat, cortisone shots, nonsteroidal anti-inflammatory medications, physical therapy, and massage can be contributing factors [73,74] (Table 4).

Mesenchymal Stem Cells in Orthopedic Surgery		
Type of MSC	**Pathology**	**Results**
cultured stem cells obtained from skin fibroblasts & modulated to grow collagen producing cells	patellar tendinopathy	improvement in pain & function
cultured cells differentiated into osteoblasts injected with fibrin	fracture healing rate & safety	acceleration of fracture healing
uncultured mononuclear cell installation obtained from bone marrow instilled following core decompression	avascular necrosis of head of femur	better clinical scores & radiological outcomes
uncultured cell installation obtained from bone marrow and	avascular necrosis of head of femur	improvement in pain & progression to collapse

implanted with core decompression		
cultured cells transplanted to area surrounding injury	complete & chronic cervical spinal cord injury	improvement in neurological function at 6 months in 10/20 patients
cultured bone marrow derived stem cells	articular cartilage repair	no significant improvement as compared to autologous chondrocyte implantation except for one stage procedure
uncultured bone marrow derived stem cells with hyaluronic acid membrane scaffold with platelet rich fibrin	osteochondral lesions of knee	no significant improvement as compared to autologous chondrocyte implantation except for one stage procedure
uncultured mononuclear cell concentrate obtained from peripheral blood and reinfused through arteriography	chronic spinal cord injuries	26/39 (66.7%) patients showed recovery of somatosensory evoked potentials
uncultured cells obtained from peripheral blood	knee cartilage repair	improvement in knee scores
cultured collagen producing stem cells obtained from skin	lateral epicondylitis	improvement in elbow scores & chondrocyte showing features of healing
uncultured cells implanted arthroscopically with collagen powder or hyaluronic acid scaffolds with platelet gel	osteochondral lesions of talus	improvement in ankle score & histologically tissue in various degrees of remodeling
uncultured mononuclear cells obtained from bone marrow aspiration with collagen sponge scaffold	filling of bone defects (trauma & tumor)	healing of all bone defects

Table 4: Lists types of MSC used for orthopedic surgery [31].

Stem Cells in Rotator Cuff Human Trials

Gomes et. al., in 2011 evaluated rotator cuff tear outcomes utilizing conventional surgical repair methods along with the addition of bone marrow mononuclear cells [75]. Fourteen patients with complete rotator cuff tears were repaired by transosseous stitches through a mini-open incision, followed by injection of bone marrow mononuclear cells into the tendon borders, which was obtained from the iliac crest just before surgery. MRI was utilized pre and post-surgery. After a 12-month minimum follow up period, there was an increase in the UCLA score from 12 to 31. After 12-month follow up period, MRI analysis showed tendon integrity in all 14/14 cases. Results of this study support implantation of bone marrow mononuclear cells in rotator cuff sutures as a safe and positive option to improve quality of tissue in affected tendons [75].

Mazzucca et. al., in 2011 evaluated if a one-time physiologic dose of insulin when compared with the growth factors insulin-like growth factor 1, growth differentiation factor 5, and B-fibroblastic growth factor were capable of differentiating bone marrow-derived mesenchymal stem cells into tendon [76]. Eleven patients scheduled for arthroscopic rotator cuff repair participated in this study and underwent aspiration of bone marrow. MSCs were divided into three groups: one group exposed to insulin, a second group treated with tendon-inducing growth factors, and the third group was a control group and not treated with any additional enhancements. MSCs that were treated with insulin demonstrated an increase in gene expression of tendon-specific markers, increased receptors on cell surface, and increased tendon-specific protein content when compared with control cells. Cells presenting with characteristics of tendon were the result of a single dose of insulin added to MSCs derived from bone marrow. The potential for MSCs to differentiate into tendon after a single dose of insulin may support additional research guided toward developing biologic options for rotator cuff repair augmentation [76].

A study by Hernigou et. al., evaluated the use of mesenchymal stem cells (MSCs) derived from the iliac crest of bone marrow to augment surgery for rotator cuff in 2014 [77]. Forty-five patients in the study group received concentrated bone marrow-derived MSCs in addition to single-row rotator cuff repair during the time of arthroscopy. Patients receiving MSCs during their surgical repair were compared to a 45-patient control group that did not receive MSCs. All patients underwent shoulder imaging studies by ultrasound every month from the first to the 24^{th} months. MRI was utilized to confirm rotator cuff healing or re-tear at three and six months postoperatively, one and two years, and at the most recent follow up appointment. MRI and ultrasound showed improved quality and healing rate for patients that received MSC injection during rotator cuff surgery. By six months, 45 of the 45 repairs (100%) with MSC augmentation were completely healed, compared to 30 of the 45 repairs (67%) that did not have MSC treatment. During the next ten years, bone marrow concentrate injection prevented additional ruptures. At the most recent follow-up appointments, 39 of the 45 patients (87%) had intact rotator cuffs in the MSC treated group, but just 20 of the 45 patients (44%) in the control group had intact rotator cuffs. These results promote bone marrow derived MSC augmentation use in rotator cuff repair as it showed reduced number of re-tears and enhanced rate of healing over extended period of time [77].

Havalas et. al., in 2015 researched the use of cultured human autologous mesenchymal stem cells (MSC) that were applied to the site of suture during arthroscopic repair of a rotator cuff tear [78]. The goal was to examine the safety of human cultured human MSCs use and study the therapeutic effect of their application. Ten patients were included in the study. There was an arthroscopic repair of the rotator cuff tear and an application of suspension of cultured MSCs to the suture site at the end of surgery. Patients showed improved clinical outcomes six weeks after surgery. Also, at three and six months all pre-operative scores improved. The MRI results six months post-surgery indicated well-integrated and fully healed tissue of rotator cuff tendon for all patients. No adverse effects of therapy were reported. This study supports the use of human cultured autologous MSCs for tissue repair [78].

A study designed by Centeno et. al., in 2015 examined the use of MSC enriched BMC injections for treatment of shoulder disability and pain due to rotator cuff tears and osteoarthritis [3]. A total of 115 shoulders in 102 patients were treated at the glenohumeral joint with autologous MSC enriched BMC injections for osteoarthritis and/or rotator cuff tears. The DASH Disabilities of the Arm, Shoulder, and Hand score, numeric pain scale, and subjective improvement rating scale were utilized to report clinical outcomes. Patients showed improved DASH scores from 36.1 to 17.1 and numeric pain scale from 4.3 to 2.4. Average subjective improvement was reported at 48.8%. This study showed encouraging results for patients treated with MSC enriched BMC injections for shoulder osteoarthritis and rotator cuff tears for improving function and reducing pain. Additional randomized clinical trials are suggested to further investigate the efficacy of MSC enriched BMC injections for treatment of rotator cuff tears and osteoarthritis [3].

Kim et. al., in 2017 evaluated clinical outcomes for patients that underwent rotator cuff surgery arthroscopically and additionally received an injection of adipose-derived MSCs [79]. One-hundred-eighty-two patients underwent arthroscopic surgery for a rotator cuff tear. There were 35 patients that were treated with arthroscopic rotator cuff repair alone (conventional group) and were matched with 35 patients that underwent arthroscopic rotator cuff repair with adipose derived MSC injections (injection group). Visual analog scale (VAS) for range of motion and pain, in addition to functional measures of the Constant Scale and University of California, Los Angeles (UCLA) shoulder rating scale were used to measure outcomes. Both groups, injection and control, showed improved scores after surgery without differences reported between groups. An MRI was used to assess repaired tendon structural integrity at 12 months after surgery and indicated the conventional group had a re-tear rate of 28.5% and the injection group had a re-tear rate of 14.3%. This research suggested injection of adipose-derived MSCs loaded in fibrin glue during rotator cuff repair could support decreased re-tear rate [79].

Centeno et. al., in 2019 compared autologous BMC and platelet product injections to exercise therapy for the treatment of partial and full-thickness supraspinatus tears [2]. Patients were randomized to either ultrasound-guided autologous BMC with MSC and PRP with platelet injection treatment or exercise therapy. Patients completed the Disability of the Arm, Shoulder, and Hand (DASH) scale, modified Single Assessment Numeric Evaluation (SANE), numeric pain scale (NPS), and a blinded MRI review. No serious adverse events were reported. At three and six months, patients reported improvements for the BMC treatment compared to exercise therapy for pain and function. At 24 months, patients reported 89% improvement, pain reduction, and sustained functional gains. MRI results reported a decrease of most tears post treatment of BMC. This research supports ultrasound-guided BMC and platelet product injections as a potentially useful and safe alternative for conservative exercise therapy of torn non-retracted supraspinatus tendons [2].

Future Research

A current research investigation, ClinicalTrials.gov Identifier NCT0375282, has been designed to evaluate the use of a single injection of adipose-derived regenerative cells into the partial-thickness rotator cuff tear compared to a single corticosteroid injection into the subacromial space. This is a prospective,

double-blinded, randomized, active-controlled, multi-site study. A maximum of 20 U.S. centers will be invited to enroll in the study and 246 subjects will be enrolled and assigned to two randomization groups. The estimated study completion date is August 2021 [80].

An additional single blinded randomized clinical trial, ClinicalTrials.gov Identifier NCT03688308, has been proposed using bone marrow aspirate concentrate in adults diagnosed with medium to large, single tears of the rotator cuff. At least 45 patients that meet eligibility will be enrolled into two groups. The control group will include patients who undergo arthroscopic surgery without BMAC. The experimental group will undergo arthroscopic surgery with the addition of BMAC. A double-row technique will be used when both groups undergo arthroscopic rotator cuff repair. Patients in the experimental group will also participate in bone marrow aspirate harvesting from the anterior inferior iliac crest that will be centrifuged and injected beneath tendon at the bone interface. MRIs and ultrasound will be completed post-operatively. Patient clinical outcomes will be evaluated at the pre-operative visits utilizing ASES, UCLA, and Constant scoring. The estimated study completion date is October 2021 [81].

An additional single-center, randomized, single-blind clinical trial, ClinicalTrials.gov Identifier NCT03279796, has been designed evaluating the use of adipose mesenchymal stem cells for tendon injuries. A maximum of 100 patients with a diagnosis of rotator cuff or lateral epicondylosis will be enrolled in this study. Adipose mesenchymal stem cells will be isolated from adipose tissue, cultured, and injected back into the tendon injury site. Patients in the experiment group will be injected with adipose mesenchymal stem cells and the control group will receive betamethasone injection. Clinical outcomes will be measured at follow up appointments utilizing Visual Analogue Scale (VAS), Constant-Murley Score, and Disability of Arm Shoulder and Hand (DASH). Ultrasound or MRI will be completed before the first injection and post procedure. The estimated study completion date is December 2021 [82].

Conclusions

Current treatment options for orthopedic procedures including rotator cuff injury are surgery as well as the use of MSC transplantation in both animal and human subjects. Current surgical intervention leads to increased recovery time as well as possible surgical complications. Researchers have found MSC transplantation demonstrated improved functional outcomes in animal models including improved treatment of osteochondral defects, improved healing process of hamstring tendon grafts to surrounding bony tunnels, accelerated healing of partially torn ACLs, and accelerated early remodeling of tendon-bone healing.

Human trials have shown- enhanced tissue quality of affected tendons, improved rate of healing, reduced pain, improved clinical outcomes, sustained functional gains, and reduced number of re-tears.

Several current research studies are currently in process for patients with rotator cuff injuries. One human study trial has been designed to compare the use of adipose-derived regenerative cell injection with corticosteroid injections. An additional study has been established to evaluate the results of arthroscopic surgical repair with and without BMAC. Additional research trials are needed to further

investigate the functional effects and efficacy for stem cell transplantation for human patients with rotator cuff injuries.

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