The Use of Bone Marrow Concentrate in the Treatment of Full-Thickness Chondral Defects

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Abstract This article is a critical analysis of a study, “Minimally Manipulated Bone Marrow Concentrate Compared with Microfracture Treatment of Full-Thickness Chondral Defects: A One-Year Study in an Equine Model,” by Chu et al. (*J Bone Joint Surg Am.* 100(2):138–146, 2018). The investigation compared two interventions in the management of full-thickness chondral defects in an equine model: autologous bone marrow concentrate without concomitant microfracture treatment versus microfracture treatment alone. This review analyzes the methodology and results of their investigation and examines how their findings may influence the continued development of therapeutic options for full-thickness cartilage injuries. The study utilized in vitro analysis, arthroscopic assessment, magnetic resonance imaging (MRI) evaluation, and histological analysis to compare the treatments and their influence on the quality of cartilage repair. Although Chu et al. reported similar results between groups, their findings offer insight into the role of arthroscopy, MRI, and histology in the evaluation of repair quality. We compare their findings to those of similar investigations, highlighting the limited therapeutic options and variable clinical outcomes related to the treatment of full-thickness articular cartilage defects.

Keywords microfracture · chondral defect · bone marrow concentrate

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Introduction

Chondral injuries contribute to the development of osteoarthritis through the loss of articular cartilage [4, 7, 8, 14, 23, 24, 26]. Despite the major health concern this poses, many studies have shown a limited ability for these chondral lesions to repair [4, 10, 14–16, 22]. These changes worsen progressively, leading to irreversible arthritic damage. Consequently, microfracture has been utilized to reduce the disease burden by accessing underlying bone to stimulate repair [4, 24]. The procedure seeks to enhance chondral resurfacing by perforating the subchondral bone plate, thereby releasing marrow elements such as mesenchymal stem cells (MSCs) and other growth factors [7, 18, 24]. Unfortunately, this technique has led to inconsistent clinical results [1, 4, 11, 12, 14, 18].

In addition, several studies have identified disadvantages of microfracture treatment, including hyperintense signal in the repair tissue indicating disorganized repair, variability in the quantity of repair tissue in the defect, failure of integration between the native and repaired cartilage, and osseous overgrowth [17–19]. While changes to the subchondral bone can be an expected consequence, their presence and severity can negatively affect clinical outcomes and subsequent treatments for cartilage repair [12, 13, 17–19].

Due to these concerns, interest has grown in the clinical use of bone marrow cell-based cartilage repair techniques to support the repair of full-thickness articular cartilage damage [4, 7–9, 13]. Bone marrow aspirate (BMA) contains MSCs that are pluripotent, with differentiation potential into muscle, fat, tendon, ligament, bone, and cartilage [2, 3, 20, 27]. BMA can be easily obtained from the iliac crest in humans and the sternum in horse models, allowing for study of its therapeutic potential in treating articular cartilage defects [4, 6, 9, 27]. Such cell-based therapy equips the clinician with a biological tool for regenerating the native hyaline-like cartilage through chondrocyte differentiation, rather than the spontaneous fibrous repair tissue elicited during the body's limited intrinsic repair [7, 27].

Based on the technical ease in accessing these cells coupled with their multi-lineage differentiation potential, this therapy is an intriguing option for the regeneration of cartilage in patients with articular injury [8, 9].

Microfracture surgery is a technically straightforward and cost-effective treatment of articular cartilage injuries [18]. However, the variability in clinical outcomes and the potential negative consequences of iatrogenic subchondral bone injury warrant consideration of other techniques. Management of articular cartilage injury with cell-based therapy alone would potentially provide the benefit of cell-based chondrogenic repair potential without the associated subchondral injury microfracture surgery causes. The article discussed in this review describes the outcomes of cell-based versus microfracture treatment in the management of articular injuries in an equine model.

Article

“Minimally Manipulated Bone Marrow Concentrate Compared with Microfracture Treatment of Full-Thickness Chondral Defects: A One-Year Study in an Equine Model.”

Chu CR, Fortier LA, Williams A, Payne KA, McCarrel TM, Bowers ME, Jaramillo D. *J Bone Joint Surg Am*. 2018;100(2):138–146.

The authors sought to compare the effects of autologous bone marrow concentrate (BMC) without concomitant microfracture versus microfracture treatment alone in the management of full-thickness chondral defects in an equine model. The study included eight horses with bilateral full-thickness chondral defects (15 mm in diameter) iatrogenically created by arthroscopic technique on the midlateral trochlear ridge. One limb of each animal was then randomly assigned to undergo microfracture alone, with the contralateral limb to undergo BMC therapy. For the microfracture-alone group, a standard microfracture awl was used to puncture the subchondral plate in the lesion bed. For the BMC-therapy group, a 60-mL aliquot of sternal bone marrow aspirate was collected from the horses and processed using SmartPrep 2 BMAC (Bone Marrow Aspirate Concentrate, Harvest Technologies). BMC was mixed 10:1 with bovine thrombin and calcium chloride to form a clot and then arthroscopically applied directly to the defect. The horses began free stall activity immediately post-surgery, an incremental walking program 2 weeks post-surgery, and free pasture exercise 3.5 weeks post-surgery until study completion.

One horse was lost to follow-up for reasons “unrelated to the study,” and the repair tissues of seven horses were analyzed by arthroscopy (by two blinded reviewers), MRI (by a blinded musculoskeletal radiologist), and histological assessment (by two consensus graders) at 1-year follow-up.

The investigation reported that culture-expanded cells from BMA, BMC, and Ficoll gradient isolated bone marrow cells displayed chondrogenic, osteogenic, and adipogenic potential. More specifically, pellet size increased in response to transforming growth factor (TGF)- β 1 in all culture-expanded groups and demonstrated positive Safranin-O/fast green staining. Pellet culture is a stable biomaterial free culture system routinely used as an in vitro technique for

chondrocyte induction of MSCs supplemented with TGF- β 1, the gold standard for chondrocyte and MSC differentiation studies [25]. The increase in pellet size indicates an increased cell growth with exposure to TGF- β 1, while positive Safranin-O/fast green staining indicates proteoglycan content in cartilage [21]. Arthroscopic evaluation was based on the International Cartilage Repair Society (ICRS) macroscopic scoring system; mean scores did not differ significantly between microfracture and BMC treatment groups ($n = 7$; mean 6.7 ± 3.6 versus 5.4 ± 2.0 , respectively; $p = 0.22$). Morphological MRI evaluating repair tissue fill, subchondral bone overgrowth, subchondral sclerosis, and synovial reaction revealed a trend toward less subchondral bone overgrowth in the BMC treatment group than in the microfracture group (mean grade, 1.6 ± 1.0 versus 2.3 ± 1.0 , respectively; $p = 0.09$). Overall, worse morphological MRI scores were found in the microfracture group than in the BMC group (8.4 ± 1.8 versus 6.3 ± 3.6 , respectively; $p = 0.027$). These morphological scores were based on four categories (repair fill percentage, bone overgrowth, sclerosis, and synovial reaction) scored from 0 to 12, with higher scores indicating inferior outcomes. Histological assessment revealed that both groups displayed fibrous to fibrocartilaginous tissues. There were no significant differences in ICRS histological scores for any individual criterion or total score ($p > 0.17$).

Commentary

The study by Chu et al. [4] is a valuable addition to the literature on cell-based therapy versus microfracture treatment in the management of articular cartilage injuries. The study's strengths include the randomized treatment design with blinded evaluations. In addition, the authors used multiple modalities to evaluate the integrity of repair through means of cell viability, arthroscopic evaluation, MRI analysis by a musculoskeletal radiologist, and histological analysis with consensus scoring by two graders.

This study also has several limitations. Most apparent is the low sample size of eight horses, including one lost to follow-up. As with any laboratory or animal-model study, there is concern for translational applicability to human patients.

The investigation found the arthroscopic application of BMC directly to large, artificially created, full-thickness chondral defects in horses resulted in repair tissue quality similar to that observed in the microfracture-therapy group at 1-year follow-up. Furthermore, histological evaluation of the repair tissue analyzed was similar between the groups without a statistically significant difference. However, MRI evaluation between groups revealed subchondral bone and synovium changes in the microfracture group, indicating a potentially detrimental iatrogenic effect of the surgical trauma associated with microfracture therapy.

Regarding the in vitro analysis of the cells obtained, neither the BMA nor the BMC cells prepared using the standard point-of-care system underwent chondrogenesis in culture and therefore did not meet the International Society

for Cellular Therapy (ISCT) criteria of MSCs [5]. These criteria dictate that “first, MSC must be plastic-adherent when maintained in standard culture conditions,” with a minimum of two additional criteria describing surface molecules and the ability to differentiate into osteoblasts, adipocytes, and chondroblasts [4, 5]. The authors point out that this is an important finding that highlights the lack of MSCs in minimally manipulated preparations without culture expansion. After culture expansion, however, cells from both BMA and BMC displayed the appropriate equine cell surface markers with trilineage potential—therefore meeting ISCT criteria of MSCs. Additionally, after cell culture expansion, BMC samples yielded higher concentrations of MSCs than unprocessed BMA did.

In addition to the in vitro analysis, this study provides information on the utility of arthroscopy, MRI, and histological analysis in the evaluation of chondral lesions and their repair capabilities. Arthroscopy was shown to be effective in the surface appearance evaluation of chondral repairs but did not reveal subchondral lesions seen by MRI or histological analysis. Conversely, repair tissue surface fissuring was not apparent on MRI but was visualized arthroscopically. These findings point out the complex nature of the healing potential of articular cartilage and the difficulty in monitoring treatment effectiveness clinically, as no one modality sufficiently evaluates repaired tissue.

This study's protocols and outcomes differ from those of a previous equine study, by Fortier et al. in 2010, in which BMC augmentation of chondral defects treated with microfracture improved tissue quality compared to microfracture alone [6]. This investigation was performed by authors at the same institution as the paper presented in this review. The investigation reported on 12 horse models, also with 15-mm diameter full-thickness cartilage defects in the lateral trochlear ridge of the femur. The investigation reported better repair quality in the BMC-augmentation group over the microfracture group regarding macroscopic scoring and histological evaluation. Furthermore, quantitative MRI revealed increased fill of the defects and improved integration of the repair tissue in the BMC-augmentation group. The authors therefore concluded that delivery of BMC as an augmentation to microfracture could result in superior healing of acute full-thickness cartilage injury over microfracture treatment alone. Chu et al. [4] noted this study in their discussion and hypothesized that the contradictory findings may indicate other regenerative mechanisms at play, such as paracrine, anti-inflammatory, or immunomodulatory effects.

This study offers important insight into in vitro analysis, arthroscopic evaluation, advanced imaging techniques utilizing MRI, and histological analysis in the treatment of cell-based versus microfracture therapy in the management of articular cartilage injuries. These and other findings discussed in this review highlight the limited therapeutic options available to replace microfracture as a clinical standard. The investigation prompts further assessment of strategies to promote in vivo chondroprogenitor cell differentiation for the treatment of full-thickness chondral defects.

Compliance with Ethical Standards

Conflict of Interest: John M. Apostolakos, MD, MPH, and Lionel Lazaro, MD, declare that they have no conflicts of interest. Riley J. Williams, MD, reports receiving intellectual property royalties from and being a paid consultant to Arthrex, Inc.; owning stock or stock options in Cymedica, Inc., Pristine Surgical, R2T2 Laboratories, Inc., and RecoverX; receiving research support from Histogenics, Inc.; being a paid consultant to JRF Ortho and an unpaid consultant to R2T2 Laboratories, Inc.; and receiving publishing royalties from Springer.

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References

1. Bae DK, Yoon KH, Song SJ. Cartilage healing after microfracture in osteoarthritic knees. *Arthroscopy*. 2006;22(4):367–374.
2. Caplan AI. Mesenchymal stem cells. *J Orthop Res*. 1991;9(5):641–650.
3. Caplan AI. Review: mesenchymal stem cells: cell-based reconstructive therapy in orthopedics. *Tissue Eng*. 2005;11(7–8):1198–1211.
4. Chu CR, Fortier LA, Williams A, et al. Minimally manipulated bone marrow concentrate compared with microfracture treatment of full-thickness chondral defects: a one-year study in an equine model. *J Bone Joint Surg Am*. 2018;100(2):138–146.
5. Dominici M, Le Blanc K, Mueller I, et al. Minimal criteria for defining multipotent mesenchymal stromal cells. The International Society for Cellular Therapy position statement. *Cytotherapy*. 2006;8(4):315–317.
6. Fortier LA, Potter HG, Rickey EJ, et al. Concentrated bone marrow aspirate improves full-thickness cartilage repair compared with microfracture in the equine model. *J Bone Joint Surg Am*. 2010;92(10):1927–1937.
7. Gobbi A, Whyte GP. One-stage cartilage repair using a hyaluronic acid-based scaffold with activated bone marrow-derived mesenchymal stem cells compared with microfracture: five-year follow-up. *Am J Sports Med*. 2016;44(11):2846–2854.
8. Gobbi A, Karnatzikos G, Scotti C, Mahajan V, Mazzucco L, Grigolo B. One-step cartilage repair with bone marrow aspirate concentrated cells and collagen matrix in full-thickness knee cartilage lesions: results at 2-year follow-up. *Cartilage*. 2011;2(3):286–299.
9. Gobbi A, Karnatzikos G, Sankineani SR. One-step surgery with multipotent stem cells for the treatment of large full-thickness chondral defects of the knee. *Am J Sports Med*. 2014;42(3):648–657.
10. Kim HK, Moran ME, Salter RB. The potential for regeneration of articular cartilage in defects created by chondral shaving and subchondral abrasion. An experimental investigation in rabbits. *J Bone Joint Surg Am*. 1991;73(9):1301–1315.
11. Kreuz PC, Erggelet C, Steinwachs MR, et al. Is microfracture of chondral defects in the knee associated with different results in patients aged 40 years or younger? *Arthroscopy*. 2006;22(11):1180–1186.
12. Kreuz PC, Steinwachs MR, Erggelet C, et al. Results after microfracture of full-thickness chondral defects in different compartments in the knee. *Osteoarthritis Cartilage*. 2006;14(11):1119–1125.

13. Madry H, Orth P, Cucchiari M. Role of the subchondral bone in articular cartilage degeneration and repair. *J Am Acad Orthop Surg*. 2016;24(4):e45–46.
14. Magnussen RA, Dunn WR, Carey JL, Spindler KP. Treatment of focal articular cartilage defects in the knee: a systematic review. *Clin Orthop Relat Res*. 2008;466(4):952–962.
15. Mankin HJ. The reaction of articular cartilage to injury and osteoarthritis (first of two parts). *New Engl J Med*. 12 1974;291(24):1285–1292.
16. Mankin HJ. The response of articular cartilage to mechanical injury. *J Bone Joint Surg Am*. 1982;64(3):460–466.
17. Minas T, Gomoll AH, Rosenberger R, Royce RO, Bryant T. Increased failure rate of autologous chondrocyte implantation after previous treatment with marrow stimulation techniques. *Am J Sports Med*. 2009;37(5):902–908.
18. Mithoefer K, Williams RJ, 3rd, Warren RF, et al. The microfracture technique for the treatment of articular cartilage lesions in the knee. A prospective cohort study. *J Bone Joint Surg Am*. 2005;87(9):1911–1920.
19. Mithoefer K, Venugopal V, Manaqibwala M. Incidence, Degree, and clinical effect of subchondral bone overgrowth after microfracture in the knee. *Am J Sports Med*. 2016;44(8):2057–2063.
20. Pittenger MF, Mackay AM, Beck SC, et al. Multilineage potential of adult human mesenchymal stem cells. *Science*. 1999;284(5411):143–147.
21. Schmitz N, Lavery S, Kraus VB, Aigner T. Basic methods in histopathology of joint tissues. *Osteoarthritis Cartilage*. 2010;18 Suppl 3:S113–116.
22. Shapiro F, Koide S, Glimcher MJ. Cell origin and differentiation in the repair of full-thickness defects of articular cartilage. *J Bone Joint Surg Am*. 1993;75(4):532–553.
23. Squires GR, Okouneff S, Ionescu M, Poole AR. The pathobiology of focal lesion development in aging human articular cartilage and molecular matrix changes characteristic of osteoarthritis. *Arthritis Rheum*. 2003;48(5):1261–1270.
24. Steadman JR, Rodkey WG, Rodrigo JJ. Microfracture: surgical technique and rehabilitation to treat chondral defects. *Clin Orthop Relat Res*. 2001(391 Suppl):S362–369.
25. Watts AE, Ackerman-Yost JC, Nixon AJ. A comparison of three-dimensional culture systems to evaluate in vitro chondrogenesis of equine bone marrow-derived mesenchymal stem cells. *Tissue Eng Part A*. 2013;19(19–20):2275–2283.
26. Widuchowski W, Widuchowski J, Trzaska T. Articular cartilage defects: study of 25,124 knee arthroscopies. *Knee*. 2007;14(3):177–182.
27. Wilke MM, Nydam DV, Nixon AJ. Enhanced early chondrogenesis in articular defects following arthroscopic mesenchymal stem cell implantation in an equine model. *J Orthop Res*. 2007;25(7):913–925.