REVIEW MSK Imaging

# MR imaging of artificial musculoskeletal tissues: bridging the gap between basic science and clinical reality

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## Abstract

The fields of regenerative medicine and tissue engineering are growing rapidly in an attempt to combine scaffolds, cells and growth factors that will develop into artificial implants for the regeneration of damaged human tissues. In vitro development of artificial musculoskeletal tissues can offer solutions to complex clinical problems such as the repair of large cartilage defects, the compensation for bone loss during revision joint arthroplasty and the repair of full-thickness tendon tears. However, clinical applications are currently limited due to safety and efficiency considerations but also due to the lack of co-operation between basic scientists, engineers and clinicians. Imaging of engineered tissue constructs can aid the design of patient-specific tissues, offering sensitive and specific monitoring of the in vitro development process, evaluation of treatment efficacy in preclinical models, non-invasive evaluation of the healing process and early detection of post-treatment complications. Despite its wide use in regenerative medicine, magnetic resonance imaging (MRI) has mainly been utilised for the assessment of artificial constructs in the *in vitro* and preclinical stage. This review presents the use of MRI for the evaluation of artificial tissues at all stages, from *in vitro* development to clinical implantation, highlighting the need for interdisciplinary collaboration.

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## *1. Introduction*

The need to develop artificial tissue constructs for the regeneration of damaged tissues bypassing the requirement for human grafts has led over the past twenty years to the rapid development of the fields of tissue engineering and regenerative medicine [1]. These interdisciplinary fields combine the principles of biomedical engineering, materials science, biology and medicine for the development of artificial tissues that can be used for the regeneration of heart, lung, brain, liver, skin and a wide variety of musculoskeletal tissues mainly bone, cartilage and tendon [2]. The development of such tissues presents tremendous opportunities for the treatment of cases involving critical-sized bone defects (e.g. bone loss following revision joint arthroplasty), extensive cartilage lesions (e.g. large osteochondral defects following sports injuries or trauma) and full-thickness tendon tears (e.g. degenerative and/or traumatic chronic rotator cuff tears). Autograft harvesting for the treatment of such conditions is linked with donor site morbidity, increased risk of infection and increased operation times, while offering limited material which may not suffice for the treatment of large lesions [3, 4].

Traditionally, engineered tissue grafts consisted of a biomaterial scaffold seeded with stem cells that can be differentiated towards specialised mature tissue cells under the influence of chemical and biological cues [5]. However, numerous variations of this traditional model have been attempted over the years by varying or even omitting the cells, the biomaterial or the differentiation signals. The tissue constructs can be either implanted to the lesion directly or following a period of in vitro cultivation in 2D or 3D culture systems. A wide variety of bioactive materials has been developed with properties similar to the healthy tissues, which can promote cell growth and/or differentiation. Apart from stem cells (embryonic stem cells, induced pluripotent stem cells & mesenchymal stem cells), mature primary cells including chondrocytes and tenocytes have been used in such tissues with variable efficiency [3, 6].

Nonetheless, despite the fact that research efforts on the discovery of optimal biomaterial-cell-bioactive cue combinations are constantly increasing, only a limited number of artificial tissues has successfully passed the clinical trial stage to make its way to the clinic [1]. In order to fulfil the safety and potency requirements of regulatory bodies, the process of graft development should be personalised and tightly monitored from the *in vitro* to the *in vivo*. Such monitoring will ensure the successful healing of damaged tissues, will confirm the absence of complications and the gradual replacement of the artificial graft by normal human tissue [7, 8].

Imaging techniques have been widely used in the preclinical and clinical graft development stage for the evaluation of artificial tissue development. Microscopy (widefield/confocal) is routinely used for the evaluation of tissues grown in the lab but cannot be used for in vivo imaging. Computed tomography (CT) scanning has been used for the structural characterisation of materials intended for tissue engineering and for the assessment of tissue healing *in vivo* in preclinical models. CT scanning has also been combined in the preclinical setting with positron emission tomography (PET) in order to obtain information about the metabolic activity of implanted tissues [9].

Nonetheless, the lack of ionising radiation, the outstanding soft tissue contrast and the superb spatial resolution render magnetic resonance imaging (MRI) advantageous over other techniques, for the imaging of artificial musculoskeletal tissues [10]. The outstanding ability of MRI to depict soft tissues enables the evaluation of tissue regeneration and the development of post-treatment complications, while enabling the 3D reconstruction of the lesions for accurate preoperative planning and the design of patient- and lesion-specific artificial tissues. MRI has been separately employed by engineers and basic scientists for the preclinical evaluation of tissues in the lab and by clinicians for medical imaging purposes. Interestingly, preclinical MRI rarely utilises or relates to clinically relevant MRI protocols, while developments in preclinical imaging are also rarely translated to the clinical setting. The purpose of this review is to present

published literature regarding the use of MRI for regenerative medicine purposes and importantly to highlight how interdisciplinary collaboration can bring MRI to the forefront of regenerative medicine for the ultimate benefit of patients.

## *2. Evaluation of tissue regeneration with MRI*

### *a. Bone regeneration*

Bone has the inherent ability to regenerate itself following injury. Stem cells and a wide variety of other cell types including macrophages and neutrophils are recruited from the bone marrow and the surrounding soft tissues, contributing to a process leading to the formation of callus. Nonetheless, in cases of extensive bone loss including severely comminuted fractures, bone resection for tumour reconstruction or revision joint arthroplasty, critical-sized bone defects are created which require the use of bone grafts for efficient healing. Currently, autografts represent the gold standard for the treatment of bone defects due to their excellent biocompatibility, immunocompatibility, osteoinductive and osteoconductive capacity [3, 11]. However, the limited amount of autograft that can be obtained without harming another bone, as well as donor site morbidity and the increased operating time, necessitate the development of artificial alternatives offering the advantages of autografts while avoiding the associated shortcomings. Over the past two decades, the rapidly growing field of bone tissue engineering has provided a multitude of engineered bone grafts, most of which have unfortunately remained at the preclinical evaluation stage [12]. Various types of natural and synthetic materials have been utilised in bone tissue engineering including but not limited to ceramics (hydroxyapatite, $TiO_2$ etc.), metals (porous tantalum, NiTi etc.), synthetic and natural polymers (alginate, collagen, PLGA etc.) [13]. These materials have been tested *in vitro* and implanted *in vivo* establishing their appropriateness for bone regeneration either on their own or following the functionalisation with bioactive molecules including proteins participating in bone extracellular matrix. Differences in the composition of such materials including water and fat content are translated to variable MRI appearance post-implantation, which needs to be taken into account when evaluating them for clinical purposes. Additionally, various stem cell types have been combined with the previously mentioned scaffolds or injected in scaffold-free formulations directly into critical-sized defects in an attempt to enhance fracture healing [14].

To date, MRI has showed limited preclinical use for the evaluation of bone healing with the use of engineered bone grafts. MRI has been used to assess the engraftment of mesenchymal stem cells (MSCs) when injected in the fracture site. Most of these studies utilise paramagnetic particles such as superparamagnetic iron oxide nanoparticles (SPIO) to label MSCs in an attempt to track their fate post implantation for bone healing purposes. Lalande et al. [15] used maghemite cores bonded to dextran and functionalised with poly(ethylene glycol) SPIO to label MSCs derived from adipose tissue. The authors utilised T2* gradient echo (GRE) sequences to assess nanoparticle uptake *in vitro* followed by the application of T2* TrueFISP sequences to assess the fate of labelled MSCs *in vivo*, showing that stem cells could uptake the nanoparticles and could be detected as areas with low signal intensity evident for up to 28 days post-implantation. Despite the long-term presence of the cells, MRI demonstrated a decrease in low intensity signal over time, indicative of the declining number of the original MSCs still surviving in the scaffolds [15]. Similar SPIOs have been also incorporated in gelatin sponges and assessed with T2-w sequences demonstrating an inverse relationship between T2 values and particle concentration utilising a 7 T magnet for in vivo experiments. The authors showed that the scaffolds containing nanoparticles were homogeneously hypointense and had different T2 appearance post-implantation than empty scaffolds, which had a heterogeneous intensity. Four weeks post-implantation, signal increased in the nanoparticle and decreased in the control group [16].

Healing of bone defects with biomaterials incorporating stem cells and bone factors has been evaluated with MRI in a limited number of studies. The majority of them utilise magnetic fields (7-12 T) with limited application in clinical practice [17, 18], applying a non-standardised variety of pulse sequences (which are not parts of routine clinical protocols for the evaluation of bone), including but not limited to Rapid Acquisition with Relaxation Enhancement (RARE) [19], SPoilt Gradient Recalled (SPGR) [20], Steady State Free Precesion (bSSFP) [18] and Fast Imaging with Steady state free Precession (TrueFISP) [15]. The variability in methods does not allow the adoption of a generalisable protocol for use in the evaluation of artificial grafts. This is further complicated

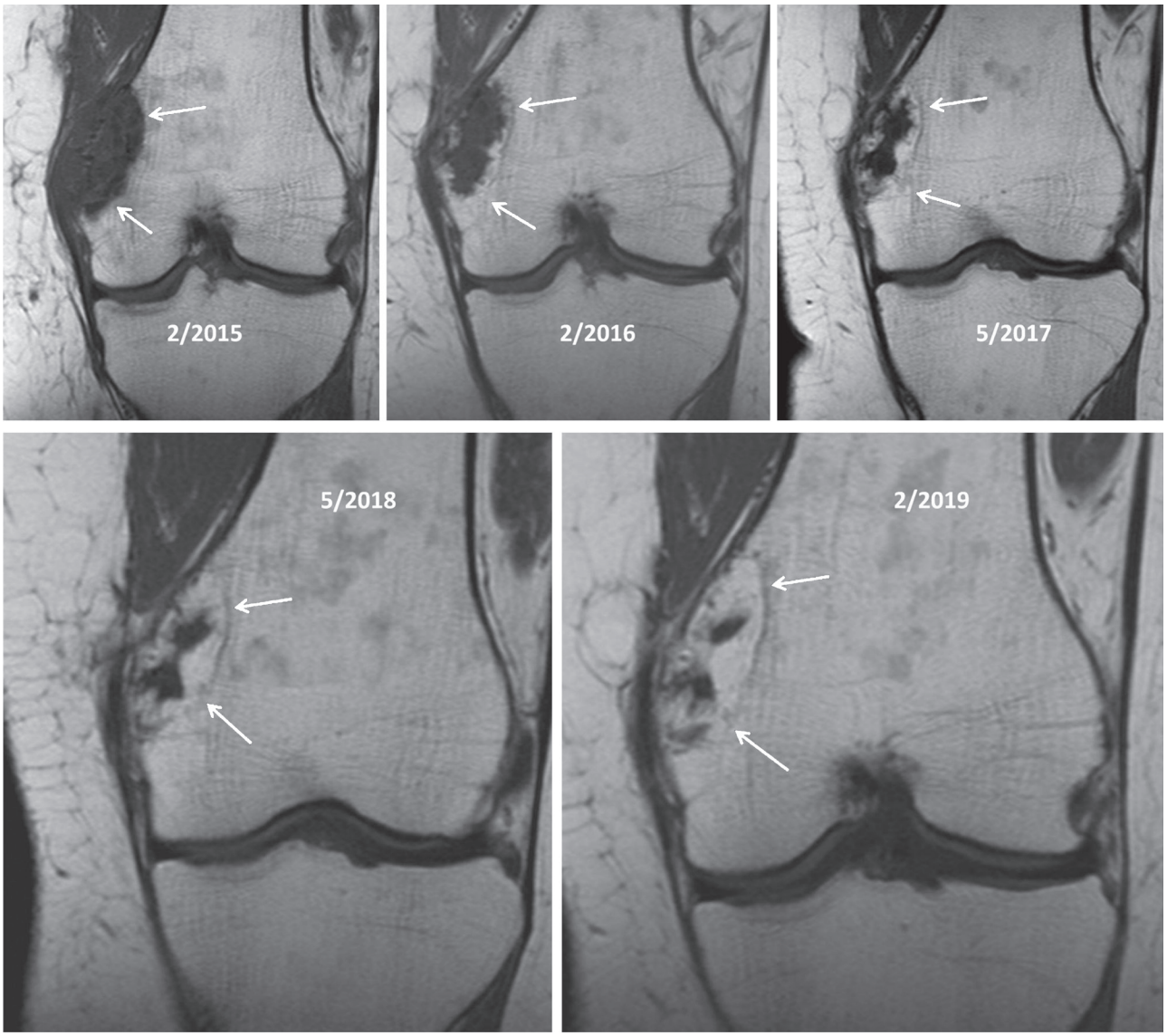


***Fig. 1.*** *A 56-year-old female underwent surgical excision of a low-grade chondrosarcoma of the distal medial femoral metaphysis. The defect was filled with hydroxyapatite. Sequential coronal T1-w MR images show the gradual filling of the defect with normal bone marrow (arrows).*

by the fact that different scaffold compositions have different appearance prior to and at the end of the healing process [18]. MRI-based monitoring has been applied for different scaffolds, including mineralised polymers such as nano-hydroxyapatite poly (caprolactone) [21], polysaccharide-based materials such as pullulan-dextran in combination with fucoidan or hydroxyapatite [18], commercial DegraPol® [19], silk [22], gelatin [17, 23] and BioOss [24]. Despite the variability in appearance, some imaging common patterns have appeared, denoting progression of fracture healing, such as the gradual change of signal intensity over the course of differentiation where the scaffold typically acquires a signal intensity similar to normal surrounding bone [17, 24, 25] usually around 5 weeks [18]. Using clinical MR scanners of 1.5 T and hydroxyapatite-based poly (caprolactone) scaffolds, signal intensity of the graft at T1-w images gradually increased post-implantation and subsequently declined towards the end of the healing process [21]. However, it has been shown that with the use of proton density (PD-w), T1-w and T2-w sequences at 7 T, differentiation between mature and woven bone was not possible. None-

theless, PD-w images provided excellent signal-to-noise ratio allowing for quantification of bone regeneration [24]. Finally, it has been demonstrated that the appearance of grafts in contrast enhanced MRI is strongly influenced by the cells present in the graft [19].

Being the "holy grail" of bone tissue engineering, vascularisation of artificial bone tissue has been evaluated with a multitude of methods including MRI **(Fig. 1)**. Dynamic contrast enhanced MRI (DCE-MRI) has been used to assess vasculogenesis in VEGF loaded soft-tissue grafts used for critical size bone defect healing. The grafts did uptake contrast as early as 1 week post implantation with rapid maximum enhancement in the periphery followed by rapid washout, whereas over time uptake became gradual with minimal or no washout, demonstrating the development of vasculature [26]. MR angiography has also been used to evaluate vascular growth in a sheep arteriovenous model of bone regeneration where MSCs were combined with recombinant human bone morphogenetic protein-2 (rhBMP-2). Interestingly, the surface area of newly produced bone and the perfused area increased over time reaching a plateau approximately at 8 weeks post-treatment [27].

***b. Cartilage regeneration***

Development of strategies for the regeneration of damaged cartilage will benefit a series of patients with diseases ranging from osteoarthritis to traumatic cartilage defects and rheumatoid arthritis [28]. The lack of cartilage vascularisation is responsible for its inherent inability of self-repair post-injury. Existing surgical repair techniques (e.g. microfractures) aim at the repair of cartilage defects with the stimulation of fibrocartilage production or the transplantation of chondrocytes from healthy joint locations. However, these surgical techniques do not present viable and efficient solutions because of the limitations related to their application. Specifically, fibrocartilage consists mainly of collagen type I in comparison to normal hyaline cartilage which mainly contains collagen type II. Therefore, fibrocartilage does not possess the durability and the biomechanical properties of hyaline cartilage, limiting the effectiveness and life time of microfracture-induced repair. Additionally, chondrocyte transplantation is limited by the related donor site morbidity and the lack or absence of healthy tissue, hindering the repair of large cartilage defects [4]. For all the previously mentioned reasons, tissue engineering and regenerative medicine endeavour to utilise biomaterials, stem cells and chondrogenic signals in order to achieve the off-the-shelf production of high quality hyaline cartilage [4, 28].

MRI has been widely used in the *in vitro*, and *in vivo* preclinical setting for the evaluation of cartilage regeneration, due to its excellent tissue contrast and spatial resolution. However, it is important to note that the high spatial resolution in systems of 3T or more has been demonstrated to lead to false positive incidental findings on normal cartilage in patients [10]. This needs to be taken into account when evaluating *in vitro* and preclinical studies since a wide number of them utilise MR scanners, which have not yet been used clinically. In the case of cartilage examination, *in vitro* and *in vivo* preclinical studies are more aligned with clinical practice, applying sequences commonly used in clinical protocols such as T1-w, T2-w fast SE, PD-w and GRE [10, 29]. Nonetheless, the evaluation of cartilage damage and cartilage repair, with limited exceptions, is not performed by experienced musculoskeletal radiologists but by basic scientists without formal MRI training, who could potentially miss anatomical variations and MRI artefacts.

The integrity and MRI appearance of regenerating cartilage has been described in studies using scaffolds such as hyaluronic-based, photocrosslinkable carbamate dimer and PuraMatrix® peptide hydrogels, poly(lactic-co-glycolic acid) and silk scaffolds as well as decellularised cartilage extracellular matrix [22, 30-36]. These materials have been either used alone or combined with human bone marrow MSCs, allogeneic or autologous chondrocytes and the change in MRI appearance over time has been recorded **(Fig. 2)**. Trattnig et al. [30] implanted hyaluronic based hydrogels with autologous chondrocytes in patients and studied cartilage repair using a clinically relevant MRI protocol. Their results show that complete repair was achieved in 65% of patients whereas hypertrophy was noted in 9% of patients which eventually returned to normal over 3-4 months. The early (1-3 months) fluid-like appearance on FSE and GRE images gradually transitioned to the appearance of normal cartilage 6-12 months post-implantation. Importantly, bone marrow oedema is seen in normal healing which can persist for up to 24 months, even after the completion of cartilage healing. In another study [34], cartilage defects in goats were treated with Wharton's jelly MSCs, were seeded in decellularised cartilage ma-

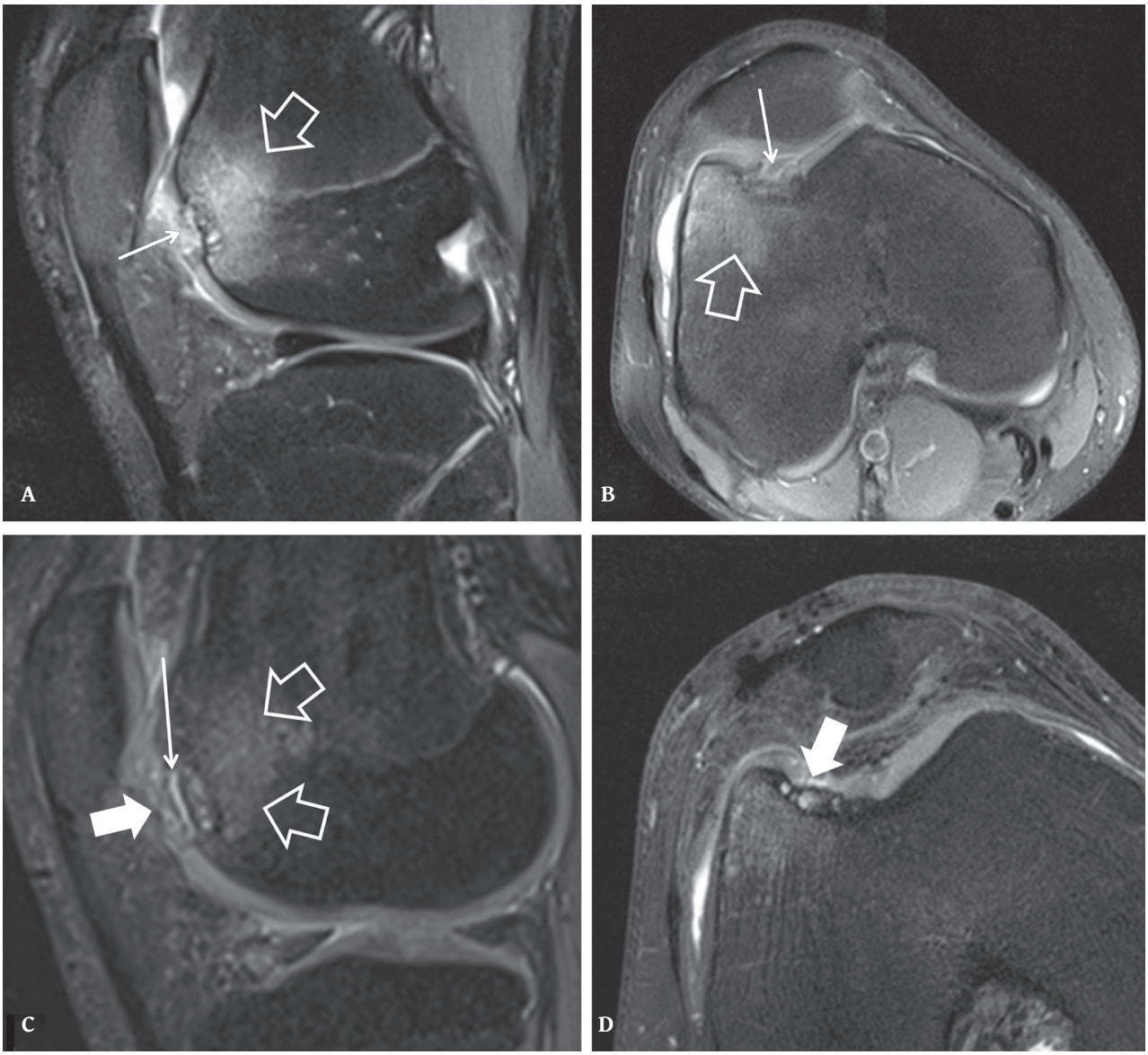


***Fig. 2.*** *A 16-year-old male athlete who underwent a matrix-assisted autologous chondrocyte implantation for a traumatic osteochondral knee injury. The patient showed no clinical improvement. Preoperative fat suppressed PD-w in the sagittal (**A**) and axial (**B**) planes show an osteochondral fracture (arrows) with bone marrow oedema (open arrows). The 3-year postoperative sagittal fat suppressed PD-w (**C**) and axial 3D T2-TrueFIsp (**D**) MR images show insufficient filling of the defect (thick arrow), delamination of the graft (long thin arrow) and persistent bone marrow oedema (open arrows) in keeping with failure of the procedure.*

trix and compared to defects treated with microfractures. At 6 months post-implantation authors describe the formation of "neo-cartilage" with high signal intensity on T2-w SE FS images, surrounded by subchondral bone marrow oedema which subsided by month 9 post-treatment, as the neo-cartilage acquired normal cartilage appearance. Importantly, in the microfracture group, the heterogeneous high signal intensity in the defect was still present after 9 months, indicating incomplete healing. High field sodium MRI has been proposed to evaluate the production of proteoglycans during stem cell chondrogenic differentiation *in vitro,* since positively charged sodium attached to negative charged proteoglycans [31], while gadolinium enhanced MRI has been used to assess glucosaminoglycan production in scaffolds seeded with MSCs and cultivated in rotating bioreactors

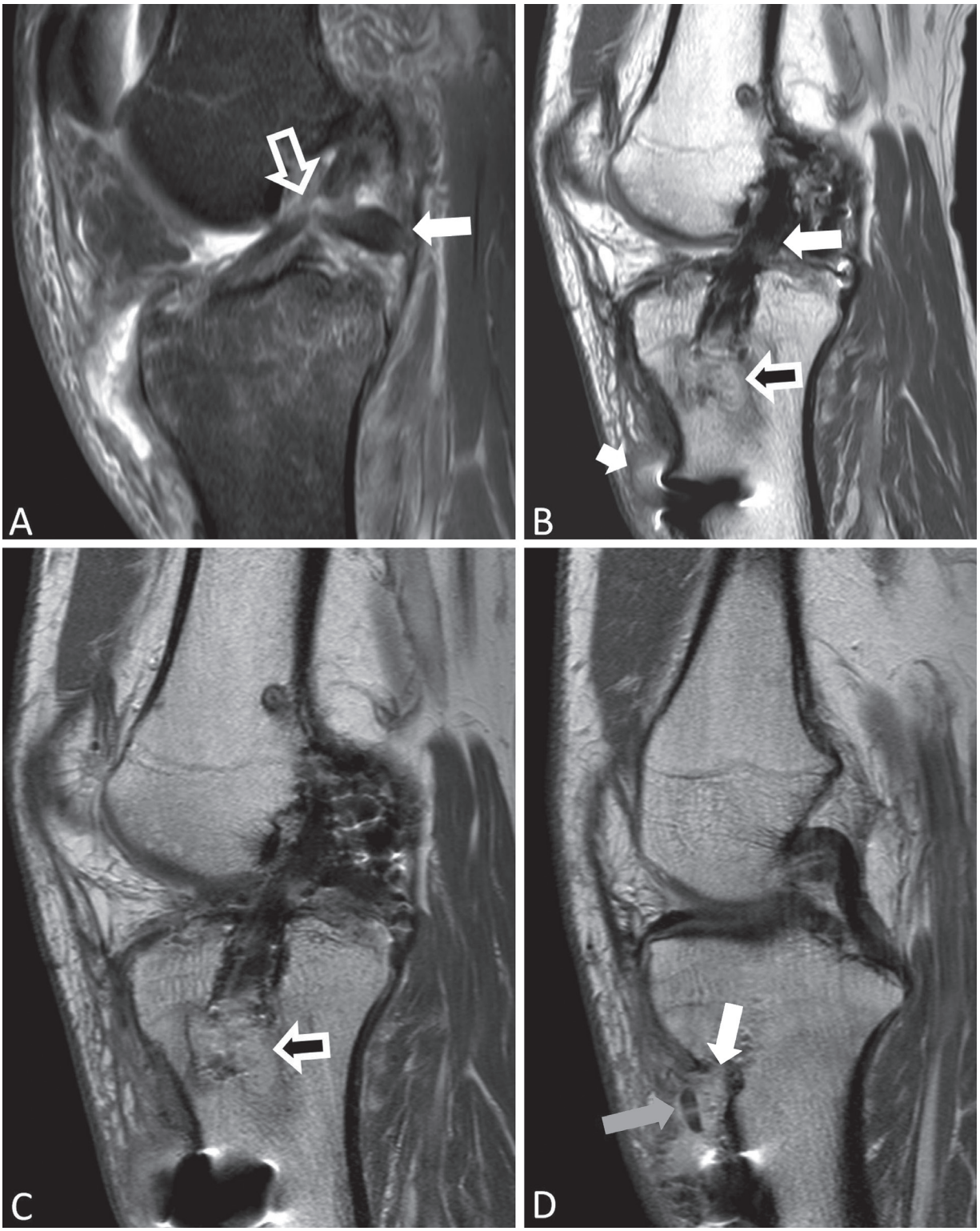


***Fig. 3.*** *A 58-year-old man with anterior cruciate ligament (ACL) rupture who underwent repair with a synthetic graft. The patient reports anterior swelling and mild pain.* ***A.*** *The preoperative sagittal fat suppressed PD-w MR image shows the ruptured ACL (open arrow) and a meniscal fragment (thick arrow). The sagittal PD-w MR image* ***(B)*** *30 months postoperatively shows the intact graft (thick arrow), an abnormal signal in the distal tibial tunnel (black arrow) and soft tissue changes anterior to the tibia (short arrow). The corresponding contrast enhanced sagittal T1-w MR images* ***(C, D)*** *show enhancement in the tibial bone marrow (black arrow), cortical disruption (white arrow) and soft tissue abscess formation anterior to the tibia (grey arrow).*

[22]. Similarly, Man et al. demonstrated that the production of fibrocartilage in rabbit cartilage defects did not result to the acquisition of signal intensity similar to adjacent normal cartilage, in comparison to defects treated with allogeneic chondrocytes in chitosan-demineralised bone matrix hydrogels, where regenerated cartilage acquired normal appearance within 24 weeks [32]. *In vivo* cartilage regeneration has been also monitored by Huang et al. with the use of T2* mapping, demonstrating that the T2* values tended to increase from deep to superficial, acquiring values similar to native cartilage later during the healing process [35]. T2 relaxation times throughout the cartilage depth have been also examined in defects treated with PLGA, where the T2 signal depended on the depth and the orientation of the specimen in the magnetic field. The PLGA treated defects showed a signal similar to normal cartilage by week 12. By week 24 a laminar outline of the newly formed cartilage could be distinguished, which was not present in the control group [33].

As previously described for bone tissue engineering, labelling of stem cells with paramagnetic nanoparticles has been also used for the evaluation of cartilage healing with MRI. Iron oxide nanoparticles have been used to label MSCs in Puramatrix ® and chitosan-glycerophosphate hydrogels or directly label acellular nanocrystal/silk fibroin scaffolds to enable visualisation of the cells and/or the scaffold matrix in GRE, PD-w, T2 and T2* sequences *in vivo* [37-39] and *in vitro* [20, 40]. These studies have shown that stem cell labelling with paramagnetic nanoparticles is a viable technique for the monitoring of the post-implantation fate of scaffold/MSC constructs used for the treatment of cartilage defects.

***c. Tendon regeneration***

Limited number of reports have evaluated tendon regeneration using artificial grafts. The complexity of tendon architecture hinders the development of artificial tendon grafts with mechanical properties mimicking native tissue. To date, the majority of studies applying MRI for the evaluation of tendon healing focus on the effects of MSC injection in tendon tears. Clinical studies have been performed, evaluating the potential augmentation of tendon repair with single injections of stem cells. Specifically, Hernigou et al. used MRI (FSE, T1-w fs, T2-w fs sequences) to demonstrate that bone marrow MSC injection in rotator cuff tears leads to 100% success within six months from injection, whereas only 67% was healed without MSC treatment [41]. Similarly, Kim et al. used conventional clinical MRI to assess healing and the presence of tears/re-tears in patients who received a single injection of adipose tissue MSCs in fibrin glue post rotator cuff arthroscopy, showing that despite the lack of clinical differences to the control group at 28-months, the structural repair was superior in the MSC group [42]. Similar results were reported by Rotini et al. who used human dermal matrix scaffolds without added stem cells for large rotator cuff repair in humans, and utilised MRI (PD-w and FSE T2-w fs sequences) at 12 months to assess healing [43]. In the preclinical setting, Achilles tendon tears in rats have been treated with bovine crossliked or porcine collagen scaffolds where 7 T MRI (T2-w Turbo-RARE sequence) revealed the location of the scaffold at the end of the follow up period, uncovering failure of approximately half cases solely due to suture and not scaffold failure [44]. In rabbit anterior cruciate ligament (ACL) tears, Setiawati et al. used 5T 3D SPGR fs sequences to show that injection of bone marrow MSCs with VEGF produced higher signal intensities than the controls which decreased over time, with a concomitant decline in ACL canal diameter [45].

Finally, a series of studies have performed MSC tracking with paramagnetic nanoparticle labelling. MSCs isolated from bone marrow or adipose tissue have been labelled, revealing that variable numbers of MSCs are retained [46, 47] in the tendon sheath and the synovial tissues without infiltrating the tendon matrix [48], appearing as high signal intensity regions on T2*-w images [47, 49]. These studies have created a doubt with regards to the effect of MSCs in tendon healing, since the absence of labelled MSCs within the regenerating matrix may indicate pure paracrine action or complete absence of healing effect [48].

ACL tendinous graft incorporation and complications, such as impingement with or without retear and infection **(Fig. 3)**, are common clinical indications for MRI.

## *3. Revolutionising regenerative medicine with the use of MRI: dream or reality?*

Developments in MRI over the past decades have revolutionised clinical medicine, enabling the diagnosis and treatment of conditions invisible to other imaging techniques. The revolution of tissue engineering has happened in parallel to the development of tissue engineer-

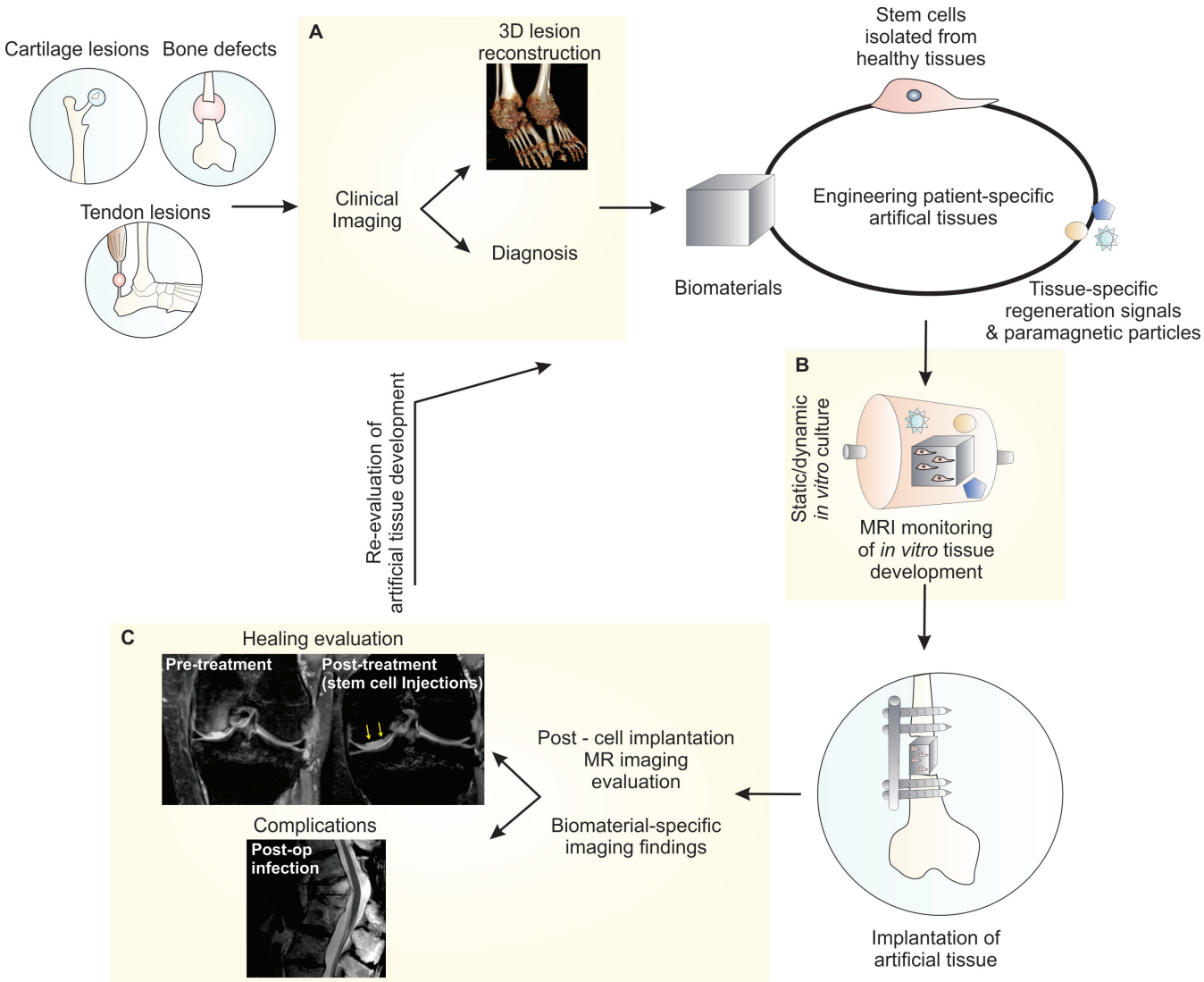


***Fig. 4.*** *The use of MRI in tissue engineering and regenerative medicine. MRI can be utilised at all stages of artificial tissue development, from lesion diagnosis and implant design (**A**) to in vitro (**B**) and in vivo (**C**) evaluation of artificial tissue development.*

ing and attempts to combine the achievements in both fields have created hopes for a rapid translation of artificial tissues to the bedside with the assistance of state-of-the-art MRI techniques. Nonetheless, as demonstrated previously, only small steps forward have been noted to date, disproving the aforementioned prediction. The delayed progress noted in the field of MRI of artificial musculoskeletal tissues can be attributed to a number of factors. First of all, research in the field of clinically relevant MRI is performed by radiologists and medical physicists, without obvious collaboration with disciplines involved in tissue engineering and regenerative medicine, such as engineers, biologists and mathematicians. The lack of interdisciplinary collaboration contributes significantly to the delayed clinical translation of progress in both fields. In addition, the variable chemical composition of biomaterials used in musculoskeletal tissue regeneration results in variable MRI characteristics of tissue repair over time. Therefore, systematic assessment of MRI healing patterns with the use of a wide variety of biomaterial compositions is necessary in order to provide clear guidelines for the evaluation of healing progression with clinical MRI. Finally, there is an immense need for the homogenisation of protocols used in preclinical and clinical MRI so that results generated in preclinical scanners are applied and translated to practical knowledge. This limitation of current practice is also related to the lack of communication between disciplines involved

in tissue engineering and MRI, which is required more than ever in order to realise the dream of revolutionising regenerative medicine with the use of MRI. Overcoming these limitations will lead to the integration of MRI in all steps of tissue engineering practice, from lesion evaluation **(Fig. 4A)** to monitoring of *in vitro* graft development **(Fig. 4B)** and assessment of *in vivo* healing and complications **(Fig. 4C)**.

### *4. Conclusions-Future directions*

MRI has the potential to play a key role in the clinical translation of artificial musculoskeletal tissues. This can be achieved by means of interdisciplinary collaboration between radiologists, medical physicists, biologists and engineers working towards the development of artificial tissues. Such collaborations will enable the combination of state-of-the-art MRI techniques including diffusion tensor imaging, MRI-based radiomics and artificial intelligence to advanced tissue engineering strategies for the development of intelligent, personalised and high-quality therapies for musculoskeletal disease.

### *Conflict of interest*

*The authors declared no conflicts of interest.*

## References

1. Kim YS, Smoak MM, Melchiorri AJ, et al. An overview of the tissue engineering market in the United States from 2011 to 2018. *Tissue Eng Part A* 2018; 25: 1-8.
2. Vacanti JP, Langer R. Tissue engineering: the design and fabrication of living replacement devices for surgical reconstruction and transplantation. *Lancet* 1999; 354: S32-S34.
3. Amini AR, Laurencin CT, Nukavarapu SP. Bone tissue engineering: recent advances and challenges. *Crit Rev Biomed Eng* 2012; 40: 363-408.
4. Elisseeff J. Injectable cartilage tissue engineering. *Exp Opin Biol Ther* 2004; 4: 1849-1859.
5. Khademhosseini A, Langer R. A decade of progress in tissue engineering. *Nat Protoc* 2016; 11: 1775-1781.
6. Evans CH. Advances in regenerative orthopedics. *Mayo Clin Proc* 2013; 88: 1323-1329.
7. Wong VW, Sorkin M, Gurtner GC. Enabling stem cell therapies for tissue repair: Current and future challenges. *Biotechnol Adv* 2013; 31: 744-751.
8. Zhang YS, Yao J. Imaging biomaterial–tissue interactions. *Trends Biotechnol* 2018; 36: 403–414.
9. Appel AA, Anastasio MA, Larson JC, et al. Imaging challenges in biomaterials and tissue engineering. *Biomaterials* 2013; 34: 6615-6630.
10. Karantanas AH. What's new in the use of MRI in the orthopaedic trauma patient? *Injury* 2014; 45: 923-933.
11. Klontzas ME, Kenanidis EI, Heliotis M, et al. Bone and cartilage regeneration with the use of umbilical cord mesenchymal stem cells. *Expert Opin Biol Ther* 2015; 15: 1541-1552.
12. Shrivats AR, McDermott MC, Hollinger JO. Bone tissue engineering: state of the union. *Drug Discov Today* 2014; 19: 781-786.
13. De Witte TM, Fratila-Apachitei LE, Zadpoor AA, et al. Bone tissue engineering via growth factor delivery: from scaffolds to complex matrices. *Regen Biomater* 2018; 1-15.
14. Giannoudis PV, Dinopoulos H, Tsiridis E. Bone substitutes: an update. *Injury* 2005; 36: S20-S27.
15. Lalande C, Miraux S, Derkaoui SM, et al. Magnetic resonance imaging tracking of human adipose derived stromal cells within three-dimensional scaffolds for bone tissue engineering. *Eur Cells Mater* 2011; 21: 341-354.
16. Hu S, Zhou Y, Mark A, et al. Enhanced bone regeneration and visual monitoring via superparamagnetic iron oxide nanoparticle scaffold in rats. *J Tissue Eng Regen Med* 2018; 12: e2085-2098.
17. Peptan IA, Hong L, Xu H, et al. MR Assessment of osteogenic differentiation in tissue-engineered constructs. *Tissue Eng* 2006; 12: 843–851.
18. Ribot EJ, Tournier C, Aid-launais R, et al. 3D anatomical and perfusion MRI for longitudinal evaluation of biomaterials for bone regeneration of femoral bone defect in rats. *Sci Rep* 2017; 7: 6100.
19. Buschmann J, Welti M, Hemmi S, et al. Three-dimen-

sional co-cultures of osteoblasts and endothelial cells in DegraPol foam: histological and high-field magnetic resonance imaging analyses of pre-engineered capillary networks in bone grafts. *Tissue Eng Part A* 2011; 17: 291-299.
20. Saldanha KJ, Doan RP, Ainslie KM, et al. Micrometer-sized iron oxide particle labeling of mesenchymal stem cells for magnetic resonance imaging-based monitoring of cartilage tissue engineering. *Magn Reson Imaging* 2011; 29: 40-49.
21. Ganesh N, Ashokan A, Rajeshkannan R, et al. Magnetic resonance functional nano-hydroxyapatite incorporated poly(caprolactone) composite scaffolds for in situ monitoring of bone tissue regeneration by MRI. *Tissue Eng Part A* 2014; 20: 2783-2794.
22. Marolt D, Augst A, Freed LE, et al. Bone and cartilage tissue constructs grown using human bone marrow stromal cells, silk scaffolds and rotating bioreactors. *Biomaterials* 2006; 27: 6138-6149.
23. Ko IK, Song HT, Cho EJ, et al. In vivo MR imaging of tissue-engineered human mesenchymal stem cells transplanted to mouse: a preliminary study. *Ann Biomed Eng* 2007; 35: 101-108.
24. Elschner C, Korn P, Hauptstock M, et al. Assessing agreement between preclinical magnetic resonance imaging and histology: An evaluation of their image qualities and quantitative results. *PLoS One* 2017; 12: e0179249.
25. Wartella KA, Kelso ML, Kovar JL, et al. Multi-modal imaging for assessment of tissue-engineered bone in a critical-sized calvarial defect mouse model. *J Tissue Eng Regen Med* 2017; 11: 1732–1740.
26. Beaumont M, DuVal MG, Loai Y, et al. Monitoring angiogenesis in soft-tissue engineered constructs for calvarium bone regeneration: an in vivo longitudinal DCE-MRI study. *NMR Biomed* 2010; 23: 48-55.
27. Boos AM, Loew JS, Weigand A, et al. Engineering axially vascularized bone in the sheep arteriovenous-loop model. *J Tissue Eng Regen Med* 2013; 7: 654-664.
28. Mollon B, Kandel R, Chahal J, et al. The clinical status of cartilage tissue regeneration in humans. *Osteoarthr Cartil* 2013; 21: 1824-1833.
29. Crema MD, Roemer FW, Marra MD, et al. Articular cartilage in the knee: current MR imaging techniques and applications in clinical practice and research. *Radiographics* 2011; 31: 37-61.
30. Trattnig S, Pinker K, Krestan C, et al. Matrix-based autologous chondrocyte implantation for cartilage repair with Hyalograft® C : Two-year follow-up by magnetic resonance imaging. *Eur J Radiol* 2006; 57: 9–15.
31. Ajumdar SHM, Othirajan PAP, Orcemus DED, et al. High field sodium MRI assessment of stem cell chondrogenesis in a tissue-engineered matrix. *Ann Biomed Eng* 2016; 44: 1120-1127.
32. Man Z, Hu X, Liu Z, et al. Transplantation of allogenic chondrocytes with chitosan hydrogel-demineralized bone matrix hybrid scaffold to repair rabbit cartilage injury. *Biomaterials* 2016; 108: 157-167.
33. Zalewski T, Lubiatowski P, Jaroszewski J, et al. Scaffold-aided repair of articular cartilage studied by MRI. *Magn Reson Mater Phy* 2008; 21: 177–185.
34. Zhang Y, Liu S, Guo W, et al. Human umbilical cord Wharton's jelly mesenchymal stem cells combined with an acellular cartilage extracellular matrix scaffold improve cartilage repair compared with microfracture in a caprine model. *Osteoarthr Cart* 2018; 26: 954–965.
35. Huang H, Zhang X, Hu X, et al. A functional biphasic biomaterial homing mesenchymal stem cells for in vivo cartilage regeneration. *Biomaterials* 2014; 35: 9608–9619.
36. Degoricija L, Bansal PN, So SHM, et al. Hydrogels for osteochondral repair based on photocrosslinkable carbamate dendrimers. *Biomacromolecules* 2008; 9: 2863–2872.
37. Jing X, Yang L, Duan X, et al. In vivo MR imaging tracking of magnetic iron oxide nanoparticle labeled, engineered, autologous bone marrow mesenchymal stem cells following intra-articular injection. *Joint Bone Spine* 2008; 75: 432-438.
38. Andreas K, Georgieva R, Ladwig M, et al. Highly efficient magnetic stem cell labeling with citrate-coated superparamagnetic iron oxide nanoparticles for MRI tracking. *Biomaterials* 2012; 33: 4515–4525.
39. Chen Z, Yan C, Yan S, et al. Non-invasive monitoring of in vivo hydrogel degradation and cartilage regeneration by multiparametric MR imaging. *Theranostics* 2018; 8: 1146-1158.
40. Roeder E, Henrionnet C, Goebel JC, et al. Dose-response of superparamagnetic iron oxide labeling on mesenchymal stem cells chondrogenic differentia-

tion: a multi-scale in vitro study. *PLoS One* 2014; 9: e98451.
41. Hernigou P, Flouzat Lachaniette CH, Delambre J, et al. Biologic augmentation of rotator cuff repair with mesenchymal stem cells during arthroscopy improves healing and prevents further tears: a case-controlled study. *Int Orthop* 2014; 38: 1811-1818.
42. Kim YS, Sung CH, Chung SH, et al. Does an injection of adipose-derived mesenchymal stem cells loaded in fibrin glue influence rotator cuff repair outcomes? a clinical and Magnetic Resonance Imaging study. *Am J Sports Med* 2017; 45: 2010-2018.
43. Rotini R, Marinelli A, Guerra E, et al. Human dermal matrix scaffold augmentation for large and massive rotator cuff repairs: Preliminary clinical and MRI results at 1-year follow-up. *Musculoskelet Surg* 2011; 95: S13-23.
44. Gabler C, Saß J-O, Gierschner S, et al. In vivo evaluation of different collagen scaffolds in an Achilles tendon defect model. *Biomed Res Int 2018;* 2018: 6432742.
45. Setiawati R, Utomo DN, Rantam FA, et al. Early graft tunnel healing after anterior cruciate ligament reconstruction with intratunnel injection of bone marrow mesenchymal stem cells and vascular endothelial growth factor. *Orthop J Sports Med* 2017; 5: 1-8.
46. Scharf A, Holmes S, Thoresen M, et al. Superparamagnetic iron oxide nanoparticles as a means to track mesenchymal stem cells in a large animal model of tendon injury. *Contrast Media Mol Imaging* 2015; 10: 388-397.
47. Scharf A, Holmes SP, Thoresen M, et al. MRI-based assessment of intralesional delivery of bone marrow-derived mesenchymal stem cells in a model of equine tendonitis. *Stem Cells Int* 2016; 2016: 8610964.
48. Khan MR, Dudhia J, David FH, et al. Bone marrow mesenchymal stem cells do not enhance intra-synovial tendon healing despite engraftment and homing to niches within the synovium. *Stem Cell Res Ther* 2018; 9: 1-14.
49. Geburek F, Mundle K, Conrad S, et al. Tracking of autologous adipose tissue-derived mesenchymal stromal cells with in vivo magnetic resonance imaging and histology after intralesional treatment of artificial equine tendon lesions - A pilot study. *Stem Cell Res Ther* 2016; 7: 1–12.

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