Biological Treatment Approaches for Degenerative Disc Disease: Injectable Biomaterials and Bioartificial Disc Replacement

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Abstract

Degenerative disc disease (DDD) is a major cause of disability in the western world. Current treatment strategies only address the symptoms of DDD. To meet the clinical need of regenerative treatment strategies, biological treatment approaches have become of increasing interest in the past decade. Currently explored treatment strategies involve biomolecular treatments for early-stage degeneration, cell-based therapies involving differentiated cells as well as stem cells for advanced-stage DDD, as well as tissue engineering strategies for total disc replacement in terminal-stage disc degeneration.

The following chapter will provide a comprehensive overview about recent the recent progress in regenerative treatment strategies. This chapter will elucidate experimental in vivo studies as well as published and ongoing clinical trials.

Keywords

Annulus fibrosus repair · AF repair · Intervertebral disc regeneration · Tissue engineering · MSCs · Mesenchymal stem cells · Growth factors · Gene therapy · TE-IVD · Bioartificial disc · Biological IVD treatment

Pathology, Current Treatments, and Resulting Challenges

Low back pain (LBP) is one of the major causes of morbidity that leads to enormous costs for western healthcare systems (Schmidt et al. 2007; Hoy et al. 2010; McBeth and Jones 2007; CDC 2009; Katz 2006). An association between LBP and degenerative disc disease (DDD) has been established by recent studies, accounting DDD for up to 40% of all LBP cases (Pye et al. 2004; MacGregor et al. 2004; Freemont 2009). The intervertebral disc (IVD) contains the soft and gelatinous nucleus pulposus (NP), the surrounding fibrocartilaginous annulus fibrosus (AF), and the cartilaginous endplate (EP) which connects the IVD to the corpus vertebrae. DDD is characterized by extracellular matrix (ECM) degradation, release of proinflammatory cytokines, altered spine biomechanics, angiogenesis, and nerve ingrowth which is associated with increased pain sensation (Le Maitre et al. 2007; Rannou et al. 2003). Factors including mechanical stress, trauma, genetic predisposition, and inflammation can trigger and exacerbate DDD (Podichetty 2007) (Fig. 1).

Among the most commonly performed spinal procedures to treat disc herniation is lumbar discectomy, with an estimated 300,000 cases per year in the United States (Deyo and Weinstein 2001). However, while the neural tissue is decompressed by the discectomy, it leaves the annular defect untreated. Because of this, the risk of recurrent disc herniation through the open defect is elevated, which occurs in 6–23% of patients following discectomy. It is associated with compromised patient outcomes, the need for revision procedures, and increased healthcare costs (Carragee et al. 2003; Swartz and Trost 2003; Bruske-Hohlfeld et al. 1990; Ambrossi et al. 2009; Frymoyer et al. 1978). Aggressive surgical discectomy can reduce the rate of re-herniation, but is associated with more severe disc degeneration and back pain (Frei et al. 2001; Barth et al. 2008; O'Connell et al. 2011). Since the IVD does not possess a sufficient self-repair capacity, current treatment options for DDD range from conservative treatments to invasive therapies for severe and symptomatic courses of DDD, like spinal fusion or total disc replacement (TDR). However, long-term results do not show significant differences between invasive and conservative therapies, and complications are common (Peul et al. 2007; Lequin et al. 2013; Lurie et al. 2014).

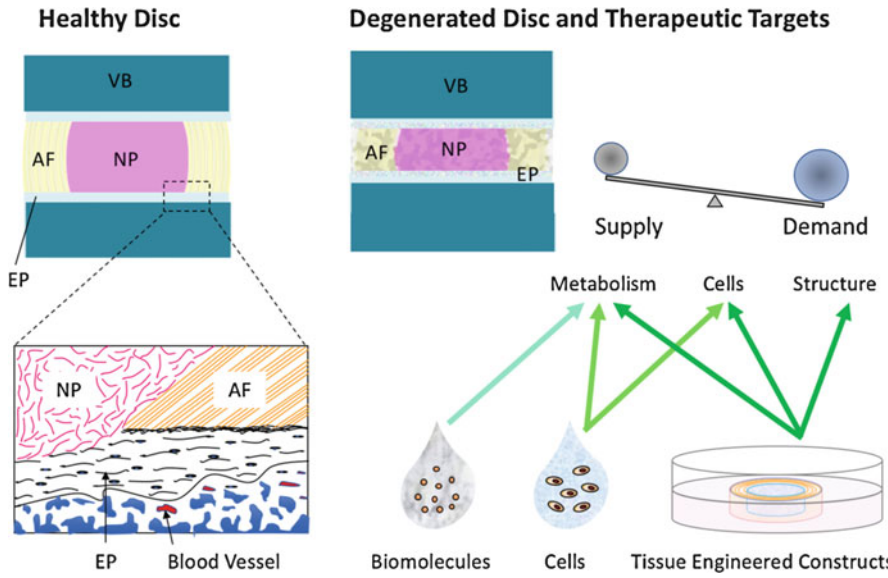

Fig. 1 Schematic pictures of the healthy disc show three components of the disc both macro- and microscopically. In degenerated discs, metabolism, cells, and structure encounter imbalance of supply and demand, one, some, or all of which each strategy will redress. *NP* nucleus pulposus, *AF* annulus fibrosus, *EP* endplate, *VB* vertebral body (Moriguchi et al. 2016)

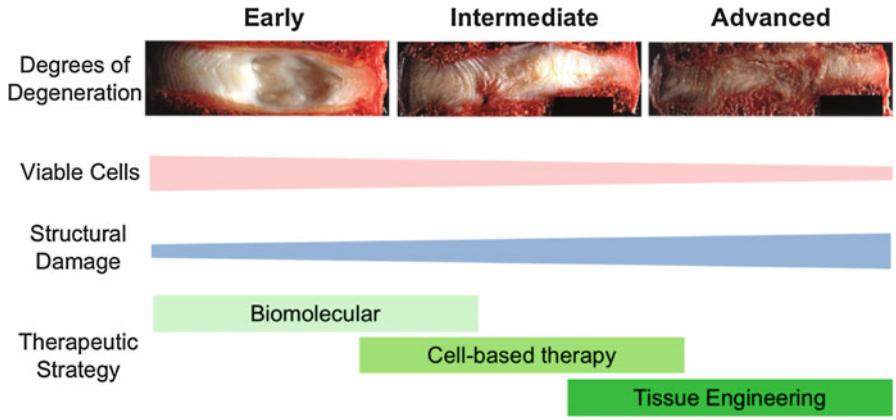

Fig. 2 Treatment strategies for different stages of IVD degeneration (Moriguchi et al. 2016)

To address the limitations of available treatments and enhancing patient outcome, biological approaches to IVD regeneration have become a growing area of interest.

Current strategies for regenerative biological disc treatment can be roughly categorized in three groups: biomolecular therapy, cell therapy, and tissue-engineered IVD construction (An et al. 2011; Zhang et al. 2011a; Maidhof et al. 2012) (Fig. 2).

In the early stage of IVD degeneration, which is defined by beginning structural changes and loss of hydration, a sufficient number of viable cells can still be found.

Thus, these treatment strategies involve recombinant genes, proteins, and stem cell therapies

(Fig. 2). These agents are meant to enhance selective protein expression by stimulating the remaining viable cells in order to promote an intrinsic self-healing within the IVD.

Mid-stage degeneration is characterized by less active and rapidly disappearing viable cells and increasing structural damage. Here, cell transplantations and tissue-engineered biological scaffolds are utilized to recover the damaged IVD.

Finally, the most advanced stage of degeneration is described as severe structural damage to the whole disc and the lack of viable cell activity. For this stage of degeneration, the treatment approaches involve TDR with tissue-engineered constructs.

The following part of this chapter will provide an overview of the current biological treatment approaches for each of the previously described stages, including experimental in vivo studies as well as recent clinical trials.

Biomolecular Treatment (Moriguchi et al. 2016)

A defining compositional change in degenerated discs is the gradual decline of NP water content originating from the loss of proteoglycan. A decrease in swelling pressure within the NP is followed by the reduction of mechanical tension in the AF collagen fibers, resulting in abnormal loading of the spine. The consequence of these alterations often is the instability of segments with subsequent development of neck or back pain and narrowing of the spinal canal, which may induce neurological symptoms. In the early stages of degeneration, the disc undergoes an imbalance of anabolic and catabolic factors that leads to the degradation of extracellular matrix (ECM). Biomolecules such as recombinant proteins and genes can regenerate expression of target molecules through the increase in anabolic or decrease in catabolic factor production, hence facilitating ECM synthesis. The following section will review recent in vivo studies on biomolecules which are used to treat disc degeneration (Table 1).

Recombinant Protein and Growth Factor-Based Therapy

Protein solutions injected directly into discs may have the potential to stimulate cell growth or anabolic response that could reverse disc degeneration. Since the demonstration of the disc's responsiveness to exogenous growth factors in an ex vivo organ culture system (Thompson et al. 1991), various proteins capable of modulating cell growth, differentiation, and ECM synthesis have shown promising for treating DDD. Bone morphogenetic proteins, such as BMP2; BMP7, which is also known as osteogenic protein 1 (OP-1); and BMP14, or growth differentiation factor-5, as well as other transforming growth factor-beta (TGF-β) superfamily such as TGF-beta 1 or 3 have induced bone and cartilage formation. Their usage has been the part of extensive research in cases of spinal arthrodesis and disc regeneration (An et al. 2005, 2011; Imai et al. 2007; Walsh et al. 2004; Masuda et al. 2006; Chujo et al. 2006; Miyamoto et al. 2006; Huang et al. 2007; Chubinskaya et al. 2007; Leckie et al. 2012). In a single in vivo rabbit study by An H. et al., intradiscal OP-1 injection resulted in an increase in proteoglycan content of NP at 2 weeks and disc height at 8 weeks. This treatment has recently been moved on to a clinical trial. Though promising, protein injection is challenged by the short duration of its therapeutic effect. The solution for this may be the development of slow-release carriers or gene-based delivery systems.

Gene Therapy

Gene therapy induces the modification of intradiscal gene expression for a prolonged effect on degenerated discs. Genes that are potentially applicable therefore are delivered through either viral (mostly adenovirus) or non-viral vectors, which are then either directly injected into live tissue (in vivo gene therapy) or transfected into cells cultures in vitro prior to implantation into the IVD (Woods et al. 2011). In one of the pioneering in vivo studies in a rabbit model, NP cells were transfected with TGF-β1 expressing adenovirus vector.

Table 1 Recombinant proteins, growth factors, and gene therapy

Species	Model	Molecules	Dose	Outcome	Refs
Protein injection					
Rat	Compression	IGF-1∗	IGF-1 8 ng/8 ul/disc	GDF-5 and TGF-beta aid in expansion of inner annular fibrochondrocytes into the nucleus	Walsh et al. (2004)
		GDF-5	GDF-5 8 ng/8 ul/disc		
		TGF-beta	TGF-beta 1.6 ng/8 ul/disc		
		bFGF	bFGF 8 ng/8 ul/disc		
Rat	Compression	BMP7 (OP-1)	0.2 ug/uL/disc	OP-1 stimulates anabolic response characterized by the restoration of normal disc morphology	Chubinskaya et al. (2007)
Rabbit	Normal	BMP7 (OP-1)	2 ng/10 ul/disc	Increase in disc height	An et al. (2011)
Rabbit	Chemonucleolysis by C-ABC	OP-1	100 ul/10 ul/disc	Increase in disc height and PG content	Imai et al. (2007)
Rabbit	Needle puncture	BMP7 (OP-1)	100 ug/10 ul/disc	Improvement in disc height and MRI findings	Masuda et al. (2006)
Rabbit	Needle puncture	GDF-5	1100 ng,1, 100 ug/10 ul/disc	Increase in disc height	Chujo et al. (2006)
Rabbit	Needle puncture	OP-1	100 ug/10 ul/disc	Increase in disc height and PG content of the NP	Miyamoto et al. (2006)
Rabbit	Annular tear 5 × 7 mm	BMP2	100 ul/10 ul/disc	Exacerbated degeneration	Huang et al. (2007)
Rabbit	Nucleotomy	PRP	20 ul PRP + microsphere/disc	Less degeneration, more PG	Nagae et al. (2007)
Rabbit	Nucleotomy	PRP	20 ul PRP + microsphere/disc	Improvement in disc height and water content	Sawamura et al. (2009)
Rabbit	Annular puncture	PRP-releasate	20 ul/disc	Better X-ray and MRIs	Obata et al. (2012)
Sheep	Annular incision	BMP 13	300 ug/70 ul saline	BMP 13 prevents loss of hydration	Wei et al. (2009)
Gene therapy					
Rat	Degenerative model induced by unbalanced dynamic and static force	Lentiviral CHOP (C/EBP homologous protein) shRNA	1×10^6 PFU/2 ul/disc	Significant decrease of apoptotic incidence in cells treated with CHOP ShRNA at 7 weeks	Zhang et al. (2011b)
Rat	Normal	Plasmid DNA mixed with microbubbles	2 ug/2 ul/disc	Reported genes were expressed up to 24 weeks	Nishida et al. (2006)
Rabbit	Normal	Ad/CMV-hTGFβ1	6×10^6 PFU/15 ul/disc	Leads to double proteoglycan synthesis	Nishida et al. (1999)
Rabbit	Normal	Ad-LMP1	1×10^7 PFU/10 ul/disc	LMP1 overexpression increases PG, BMP2, and BMP7	Yoon et al. (2004)
Rabbit	Annular puncture	ADAMTS5 siRNA oligonucleotide	10 ug/10 ul/disc	Improvement in MRI and histological scores	Seki et al. (2009)

(continued)

Table 1 (continued)

Species	Model	Molecules	Dose	Outcome	Refs
Rabbit	Annulotomy	AAV2-BMP2 or-TIMP1	6×10^6 virus particles/15 ul/disc	AAV-BMP2 and -TIMP1 delayed degeneration	Leckie et al. (2012)
Rabbit	Post-annulotomy	Ad-Sox9	1×10^9 PFU/10 ul/disc	AdSox9 helped retain chondrocytic appearance, cellular morphology, and ECM at 5 weeks	Paul et al. (2003)

Proteoglycan synthesis showed to be increased by 100% in treated tissue (Nishida et al. 2006).

Since, a variety of proteins were discovered as promising targets for gene therapy: upstream proteins such as LMP-1 which regulates BMP2 and BMP7, ECM-degrading enzymes, disintegrin and metalloproteinase with thrombospondin motifs-5, their inhibitors (tissue inhibitor of metalloproteinase-1, TIMP-1), chondrocyte-specific transcription factors (SRY-box 9, Sox9), and apoptosis inducers (C/EBP homologous protein) (Leckie et al. 2012; Nishida et al. 1999, 2006; Yoon et al. 2004; Seki et al. 2009; Zhang et al. 2011b; Paul et al. 2003).Though gene therapy can be advantageous in its sustained effect, inherent risk of viral gene delivery systems becoming infectious or immunogenic has moved the focus toward non-viral gene delivery systems. The development of microbubble-enhanced ultrasound gene therapy and injection of small interfering RNA (siRNA) have proven to achieve long-standing transgene expression in IVD cells in vivo (Nishida et al. 2006; Zhang et al. 2011b). However, non-viral gene delivery systems are limited by low transfection efficiency, which must be overcome to enhance their clinical applicability. The feasibility of ex vivo gene therapy, which reduces the risks of infection and immunogenicity and plays an important role in the future of tissue engineering technology, has been explored in several studies (Xin et al. 2012; Leo et al. 2004).

Platelet-Rich Plasma

Platelet-rich plasma (PRP), an autologous blood product manufactured by the centrifugation of whole blood, offers a variety of proteins for the treatment of degenerative discs due to its high concentration of platelets. Upon activation, these platelets release a variety of multifunctional growth factors such as PDGF (platelet-derived growth factor), IGF-1 (insulin-like growth factor), TGF-β1, (transforming growth factor-beta 1), VEGF (vascular endothelial growth factor), and bFGF (basic fibroblast growth factor). When used in the early stage of disc degeneration, PRP may enhance disc hydration (Gullung et al. 2011). Various PRP technologies have emerged to retard the degenerative cascade, which include a gelatinous hydrogel scaffold, impregnated with PRP (Nagae et al. 2007; Sawamura et al. 2009; Obata et al. 2012) and soluble releasate derived from activated PRP (Obata et al. 2012). The in vivo efficacy of PRP in improving or maintaining disc height and hydration has facilitated its transition to ongoing clinical trials.

Cell-Based Therapy (Moriguchi et al. 2016)

The efficacy of biomolecules is limited when the degeneration of an IVD is more advanced, since there is a correlation between the progress of the degeneration and the decline of the number of cells responsive to injected genes and proteins (Gruber et al. 2002). Mid-stage degeneration is characterized by a decrease in the number of cells within the IVD tissue. Therefore, cell transplantation is a feasible treatment strategy at this stage. A number of in vivo studies report the efficacy of using a vast array of cell sources (Table 2).

Table 2 Cell therapy

Species	Model	Cell type	Dose	Outcome	Refs
Mouse	Post-annular injury	Allogenic bone marrow MSCs	BMSCs 1.0×10^3	ECM augmented in NP via autonomous differentiation and stimulation of endogenous cells at 12 weeks	Yang et al. (2009)
Mouse	Annular puncture	Multipotent stem cells derived from human umbilical cord blood	1.0×10^3 cells intradiscally, 1.0×10^6 cells intravenously	Unlike intradiscal injection, intravenous injection did not preserve the IVD architecture nor disc height at 14 weeks	Tam et al. (2014)
Sand rat	Discectomy	Autologous disc cells	1.0×10^4 cells/ 5 ul/2-mm^3 Gelfoam	Implanted disc engrafted with the host disc for up to 8 months	Gruber et al. (2002)
Rat	Normal	Bone marrow MSCs	5.0×10^5/50 ul hyaluronan gels	MSCs maintained viability and proliferated over 28 days	Crevensten et al. (2004)
Rat	Post-annular puncture	Human bone marrow MSCs	1.0×10^6/15 ul	Human MSCs survived for 2 weeks post transplantation, increasing disc height and MRI intensity	Jeong et al. (2009)
Rat	Post-annular puncture	Adipose-derived MSCs (ADSCs)	1.0×10^6/50 ul	Discs maintained disc height and restored MRI signal intensity	Jeong et al. (2010)
Rat	Nucleotomy	Co-culture of NP cells and MSCs	2.5×10^5 cells (25% NPCs and 75% MSCs)	Bilaminar co-culture pellet of NP cells and MSCs outperformed solely NP cells or MSCs at 5 weeks	Allon et al. (2010, 2012)
Rabbit	Nucleotomy	Allogenic NP cells	5.0×10^4 cells/ 20 ul	Histology indicated delayed degeneration at 16 weeks	Okuma et al. (2000)
Rabbit	Nucleotomy	Autologous articular chondrocytes	2.0×10^6/ 150 ul	Chondrocytes survived and produced hyaline-like cartilage at 6 months	Gorensek et al. (2004)
Rabbit	Normal	Allogenic bone marrow MSCs	1.0×10^5 cells	MSCs survived and enhanced PG synthesis	Zhang et al. (2005)
Rabbit	Post-nucleotomy	Autologous MSCs	4.0×10^4/40 ul atelocollagen	Improved disc height, MRIs, and histology at 48 weeks	Sakai et al. (2003,2005, 2006)
Rabbit	Post-annular Injury	Autologous bone marrow MSCs	1.0×10^5/25 ul	Injection of MSCs significantly increased PG synthesis in severely degenerated discs at 16 weeks	Ho et al. (2008)
Rabbit	Normal	Allogenic MSCs	1.0 × 105/15 ul	Injected cells engrafted into inner annulus fibrous at 24 weeks	Sobajima et al. (2008)
Rabbit	Post-puncture	Xenogeneic derivatives of embryonic stem cells	1.0 × 106 cells/ 20 ul	New notochordal cells observed; no immune response elicited	Sheikh et al. (2009)
Rabbit	Nucleotomy	Allogenic synovial MSCs	1.0 × 107 cells/ 100 ul PBS	Implanted cells labeled with DiI or GFP detected at 24 weeks. Disc height and MRI signal intensity were maintained	Miyamoto et al. (2010)
Rabbit	Compression	Allogenic bone marrow MSCs	0.08 ml of 1.0 × 106 cells/ ml	Combination of MSC injection and distraction led to better disc height and histology at 8 weeks	Hee et al. (2010)
Rabbit	Post-nucleotomy	Autologous NP cells and allogenic MSCs	1.0 × 106/20 ul	Both NP cells and MSCSs better maintained disc height and GAG content at 16 weeks	Feng et al. (2011)

(continued)

Table 2 (continued)

Species	Model	Cell type	Dose	Outcome	Refs
Canine	Post-nucleotomy	Disc cells	6.0 × 106 cells/1 ml/disc	Disc remained viable, produced ECM, better maintained disc height	Ganey et al. (2003)
Canine	Post-nucleotomy	Autologous MSCs	1.0 × 106/ml stem cells	MSCs led to better disc height, MRI, and histology grading at 12 weeks	Hiyama et al. (2008)
Canine	Post-nucleotomy	Bone marrow MSCs	105, 106, 107 cells	The disc treated with 106 MSCs had more viable cells than 105 and less apoptotic cells than 105 cells at 12 weeks	Serigano et al. (2010)
Porcine	Post-nucleotomy	Human MSCs	0.5 × 106/hydrogel carrier	Implanted cells survived and differentiated into disc-like cells at 6 mos	Henriksson et al. (2009)
Porcine	Nucleotomy	Allogenic juvenile chondrocytes and MSCs	7–10 × 106/0.5–75 ml fibrin carrier	JC outperformed MSCs in proteoglycan synthesis at 12 months	Acosta et al. (2011)

Differentiated Cells

Implanted differentiated disc chondrocytes are meant to produce demanded ECM components such as proteoglycan and collagen types II and I under hypoxia and nutrient stress and can meet the increased cellular and metabolic demands of the disc (Rajpurohit et al. 2002).

Accumulated evidence in an array of animal models demonstrate the viability of autologous or allogenic cells in vivo as well as the integration into the host tissue. Thus, a reduction of ECM degradation, recovery of disc height, and MRI signal intensity can be achieved (Table 2). In fact, the pioneering preclinical study in an injured canine model showed that NP disc chondrocyte implantation contributed to ECM regeneration, retarding further disc degeneration (Ganey et al. 2003).

However favorable, disc cell transplantation showed several challenges: (1) donor site morbidity, (2) difficulty in expanding cells in vitro while maintaining cell phenotype, and (3) paucity of allograft donor tissue. Similar to differentiated disc cells, cultured articular chondrocytes (AC) are a well-established non-disc cell source in regenerative medicine (Brittberg et al. 1994). Their effortless extraction from non-weight-bearing parts of the knee and capacity to produce NP-like ECM when transplanted in vivo makes autologous (Gorensek et al. 2004) or allogenic (Acosta Jr et al. 2011) AC a safe and feasible cell source in IVD regeneration. Furthermore, potential immune evasion by juvenile articular chondrocytes supports their applicability in allogenic cell transplantation.

Stem Cells

Multipotent mesenchymal stem cells (MSCs), which are present in adult bone marrow or adipose tissue, can replicate as undifferentiated cells and then differentiate into lineages of mesenchymal tissue: bone, cartilage, fat, tendon, muscle, and marrow stroma (Pittenger et al. 1999). These somatic stem cells are a potentially ideal option for disc repair due to their accessibility and ability to differentiate along a chondrogenic lineage and produce the required proteoglycan and collagen for the disc ECM. The feasibility of MSCs to facilitate disc repair has been substantiated.

Yet it remains controversial whether differentiated cells or stem cells are superior in terms of regenerative capacity of disc morphology.

A porcine study comparing the utility of different cell sources found that committed articular chondrocytes are more suited for the use in

disc repair than MSCs due to their aptness for survival in the ischemic disc microenvironment (Acosta Jr et al. 2011). Interestingly, a comparative rabbit study found that MSC transplantation can serve as an ideal substitute for differentiated chondrocytes of disc NP owing to better accessibility with equivalent regenerative potential (Feng et al. 2011). Studies assessing the combination of both cells demonstrated that rather in vitro co-culture (Okuma et al. 2000) or co-implantation (Allon et al. 2010) yields better in vivo performance of the implanted cells. Nonetheless, pluripotent embryonic (Evans and Kaufman 1981; Martin 1981) and induced pluripotent stem cells (iPSCs) (Takahashi and Yamanaka 2006), unlike the lower potent MSCs, have unlimited proliferative and differentiate capacities, which can be strategically exploited in cell-based disc repair.

Sheikh H et al. extracted murine embryonic stem cells (ESCs) and differentiated them into chondro-progenitor cells. Upon implantation into rabbit injured discs, these cells induced notochordal cell formation at site of injury without xenograft-associated immune responses (Sheikh et al. 2009). Unstable in vitro differentiation into desired cell lineages and the potential risks of tumor formation in vivo are still major obstacles in the use of ESCs and iPSCs. However, if these issues are overcome, the use of stem cells may offer abundant potential for intervertebral disc repair.

Tissue Engineering Strategies

The implementation of tissue engineering (TE) pioneered by Langer and Vacanti in 1993 (Langer and Vacanti 1993) has fueled the efforts toward constructing functional biological substitutes for TDR as a novel treatment strategy for DDD. Recently, major efforts have been directed toward developing a replacement for either NP or AF using TE technology.

Tissue engineering originally consists of three, and more recently four components (Langer and Vacanti 1993): scaffolds, cells, growth factors, and physical conditioning using electrical or mechanical stimuli (Fig. 3). Since extensive loss of matrix and structural damages are exhibited in

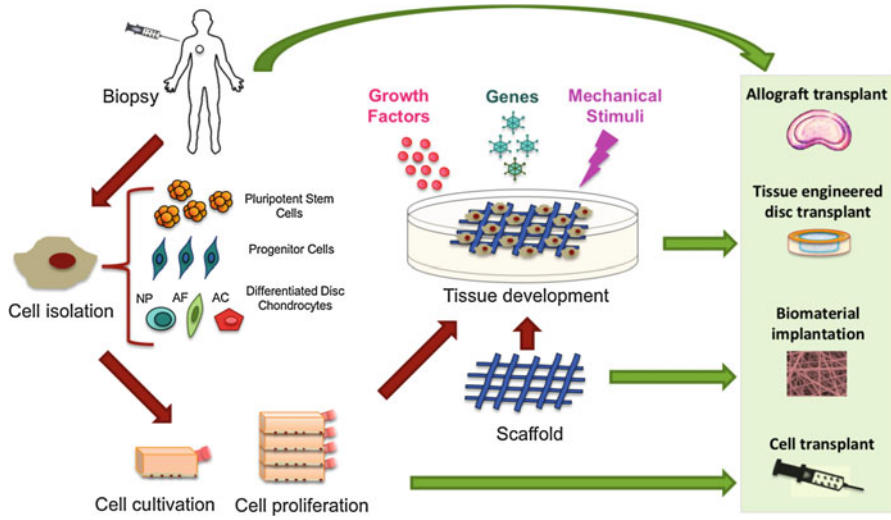

Fig. 3 Cells harvested from different sources can be expanded in vitro and transplanted in vivo in cell transplant for disc regeneration. Scaffolds can be combined with cells, and, if they have bio-mimicking properties, these treatments can be regarded as a part of tissue engineering strategy, which traditionally composes of cells, scaffolds, growth, and factors, but recently including gene treatment and mechanical conditioning. *NP nucleus pulposus cells*, *AF* annulus fibrosus cells, *AC* articular chondrocytes (Moriguchi et al. 2016)

Table 3 Tissue-engineered constructs

Species	Model	Construct	Outcome	Refs
Rat	Subcutaneous implantation	TE-IVD composed of a NP cell-laden alginate surrounded by an AF cell-laden PGL/PLA	Biochemical markers of matrix synthesis, increasing over time, were similar to native tissue at 12 weeks	Mizuno et al. (2004a)
Rat	Subcutaneous implantation	Porous type II collagen/hyaluronate-chondroitin-6-sulfate (CII/HyA-CS)	CII/HyA-CS scaffolds had satisfactory cytocompatibility and histocompatibility, as well as low immunogenicity	Li et al. (2010)
Rat	Subcutaneous implantation	Composite IVD consisting of demineralized bone matrix gelatin and collagen II/hyaluronate/chondroitin-6-sulfate scaffolds seeded AF and NP cells	Implant, similar to native disc in morphology and histology, increased proteoglycan synthesis over 12 weeks	Zhuang et al. (2011)
Rat	Total discectomy	TE-IVD composed of a NP cell-laden alginate surrounded by an AF cell-laden collagen layer	TE-IVD maintained disc space height, produced de novo ECM, and integrated into the spine – yielding intact motion segment with dynamic mechanical properties similar to that of native IVD	Bowles et al. (2011a)
Rat	Subcutaneous implantation	5.0×10^6 cells/ml in pentosan polysulfate-containing polyethylene glycol/hyaluronic acid	MPC/hydrogel composites formed cartilage-like tissue, well tolerated by the host	Frith et al. (2013)
Rabbit	Laser discectomy	2.0×10^6 cells/atelocollagen honeycomb shaped scaffold	AF cells survived and produced hyaline-like cartilage in the disc at 12 weeks	Sato et al. (2003)
Rabbit	Microdiscectomy	Cell-free implant composed of a polyglycolic acid (PGA) felt, hyaluronic acid (HA), and allogenic serum	Implantation of a cell-free PGA-HA implant immersed in serum after discectomy improved disc hydration and preserved disc height 6 months after surgery	Abbushi et al. (2008)
Rabbit	Post-nucleotomy	2.0×10^6 bone marrow MSCs/0.04 ml fibrin glue containing 10-ug/L TGF-β1 (MSC-PFG-TGF-β1)	MSC-PFG-TGF-β1 group had less degeneration and a slower decrease in disc height compared with both degenerative and acellular PFG-TGF-β1 group	Yang et al. (2010)
Rabbit	Nucleotomy	Allogenic NP cell-seeded collagen II/hyaluronan/chondroitin-6-sulfate (CII/HyA/CS) tri-copolymer construct	Viability of allografted NP cells, extracellular matrix deposition, and disc height maintenance; restoration of T2 MRI signal intensity observed at 24 weeks	Huang et al. (2011)
Rabbit	Post-puncture	5.0×10^3 allogenic bone marrow MSCs/10 ul hydrogel	MSCs suppressed collagen I in NP, reduced collagen aggregation, and maintained proper fibrillary properties and function	Leung et al. (2014)
Rabbit	Post-nucleotomy	1.0×10^6 human NP cell line infected with recombinant SV40 adenovirus vector (HNPSV-5) in atelocollagen	Deceleration of disc degeneration was evident after HNPSV-5 transplantation as shown by disc height and histologic examination at 24 weeks	Iwashina et al. (2006)
Canine	Total discectomy	Cell-allograft IVD composites made of allograft and NP cells, with in vitro transduced with recombinant adeno-associated virus (rAAV)-hTERT	The hTERT-loaded NP cells intervention could effectively resist the degeneration of the allogenic transplanted IVD at 12 weeks	Xin et al. (2012)

(continued)

Table 3 (continued)

Species	Model	Construct	Outcome	Refs
Canine	Post-nucleotomy	Autologous adipose tissue-derived stem and regenerative cells in hyaluronic acid carrier (ADRC/HA)	Disc that received ADRC/HA produced matrix and resembled native disc in morphology at 12 months	Ganey et al. (2009)
Canine	Nucleotomy	Cell-scaffold composite made of three-dimensional porous PLGA scaffolds and NP cells	Disc height, segmental stability, and T2-weighted MRI signal intensity were well preserved at 12 weeks	Ruan et al. (2010)
Porcine	Nucleotomy	Cell-scaffold composite made of NP cells and injectable hyaluronan-derived polymeric substitute material HYADDR (1.0 × 105 cells/ml)	Injected discs had a central NP-like region similar to the normal disc biconvex structure and viable chondrocytes forming matrix like that of normal disc at 6 weeks	Revell et al. (2007)
Porcine	Post-annular injury	1.25 × 105 autologous MSCs/ml in either hydrogel PhotoFix or hyaluronic acid	Stem cells in hydrogel treatment had significantly higher T2 MRI intensities and lower degeneration grade at 24 weeks than hydrogel alone treatment	Bendtsen et al. (2011)
Porcine	Partial nucleotomy	5.0 × 105 autologous bone marrow MSCs transduced with retrovirus encoding luciferase in 1 mL hyaluronan-enhanced albumin hydrogel	In vivo 3-day analysis showed persistent metabolically active implanted cells in the disc	Omlor et al. (2014)
Goat	Post-disc injury	2.5 × 105 allogenic bone marrow stromal cells/10 ul PBS + 30 ul chondroitin sulfate-based hydrogel	Significant increase in NP proteoglycan accumulation at 6 months	Zhang et al. (2011c)
Sheep	Total discectomy	Noncrystalline polylactide copolymer interbody cages filled with1.0 × 106 allogenic mesenchymal progenitor cell (MPC)-laden Gelfoam sponge formulated with the chondrogenic agent pentosan polysulfate (PPS)	Biodegradable cage-contained MPCs in combination with PPS produced cartilaginous tissue at 3 months	Goldschlager et al. (2010)
Sheep	Post-chondroitinase-ABC injection	4.0 × 106 or 0.5 × 106 human mesenchymal precursor cells (MPCs) suspended in hyaluronic acid	High-dose injection improved histopathology scores at 3 months, while low dose at 6 months	Ghosh et al. (2012)
Sheep	Nucleotomy	Allogenic or autologous disc cells (0.4–2.0 × 106 cells/0.5–1 ml hydrogel) in hydrogel containing hyaluronic acid and maleolyl-albumin	Biological repair of traumatic damage occurs in sheep discs at 6 months; hydrogel-supported disc cells may be beneficial	Benz et al. (2012)
Canine	Total discectomy	TE-IVD composed of a NP cell-laden alginate surrounded by an AF cell-laden collagen layer	Early displacement in some cases, if stably implanted TE-IVD maintained disc height, produced new ECM, and integrated into host tissue, intact motion segment with dynamic mechanical properties similar to that of native IVD	Moriguchi et al. (2017)

advanced stages of disc degeneration, development of biocompatible and biomimetic scaffolding materials based on engineering innovation can facilitate the recovery of native biological and biomechanical functionality. Numerous studies have assessed tissue-engineered components as well as whole disc constructs of the disc in vivo (Table 3).

Scaffold Development

Numerous scaffold materials, including altinate, silk-fibrin/HA composites, atelocollagen, synthetic polymers, and a collagen 2/hyaluronan/chondroitin-6-sulfate (C2/Hy/CS) composite, which mimic the mechanical and biochemical properties of the native NP, have been part of a study. Extensive research on hyaluronic acid, a native NP extracellular matrix component, has been performed in vivo (Revell et al. 2007; Abbushi et al. 2008; Ganey et al. 2009; Li et al. 2010; Huang et al. 2011). Resorbable cell-free implants consisting of a polyglycolic acid (PGA) felt, hyaluronic acid, and serum were used in a rabbit study. This resulted in improved disc hydration and height 6 months after microdiscectomy (Abbushi et al. 2008). The reason for the frequent use of cells together with bio-mimicking materials is to encourage de novo ECM production. The findings of Ganey T. et al. were that adipose-derived stem cells contribute significantly to the recovery of T2 intensity and disc height in a canine disc injury model. Synthetic polymers such as PGA or poly-L-lactic-co-glycolic acid (PLGA) have also been used either solely or in combination with hydrogels to construct cell-laden TE composites (Abbushi et al. 2008; Ruan et al. 2010).

Biological Annulus Fibrosus Repair

In mid-stage DDD, a commonly occurring pathology is the lumbar disc herniation. Due to the progressive degeneration, the IVD shows reduced hydration. The inadequate hydration of the disc leads to fissure formation, eventually allowing the soft NP to herniate through the defect and thus compress neighboring neural structures (Freemont 2009).

Lumbar discectomy is one of the most commonly performed spinal procedures to treat disc herniation, with an estimated 300,000 cases performed annually in the United States (Deyo and Weinstein 2001). While efficient in relieving acute symptoms by removing the herniated part of the NP and decompressing neural structures, the AF defect typically remains untreated after discectomy. Persistent AF defects increase the risk of re-herniation, which may lead to additional operations including more invasive procedures such as TDR and instrumented fusion (Carragee et al. 2003; Swartz and Trost 2003; Bruske-Hohlfeld et al. 1990; Ambrossi et al. 2009; Frymoyer et al. 1978; Laus et al. 1993).

Previous studies of intervertebral disc repair, which aim to halt, delay, or reverse intervertebral disc degeneration, were primarily focused on NP regeneration (Masuda et al. 2004; Bae and Masuda 2011; Sakai and Grad 2015; Wang et al. 2014; Kepler et al. 2011; Blanquer et al. 2015; Mern et al. 2014). However, the majority of these strategies are delivered through a punctured AF, which can generate a degenerative cascade within the disc affecting IVD biomechanics, cellularity, and biosynthesis even upon modest injury (Elliott et al. 2008; Iatridis et al. 2009; Korecki et al. 2008; Hsieh et al. 2009). Annular defects can emerge not only from needle punctures through the AF to reach the NP but also from the early process of IVD degeneration. Given the sensitivity of the AF, lesions from NP treatment can provoke further degeneration, inducing leakage of the delivered material and eventual failure of the regenerative treatment. In fact, one prospective study with 10-year follow-up found that discography performed with a small needle puncture accelerated disc degeneration rate of same-side disc herniation and changes to the endplate (Carragee et al. 2009). A different study demonstrated that injecting MSCs through the AF into the NP led to cell leakage and augmented osteophyte formation (Vadalà et al. 2012). Combining an injectable NP regenerative strategy with a sealant that repairs annular defects is the optimal strategy that can circumvent leakage of implanted cells or material while enhancing therapeutic outcome. Previous approaches to annular repair have involved mechanical treatments such as suturing and annuloplasty devices, which failed to improve annular healing strength in long-term clinical trials (Ahlgren et al. 2000; Chiang et al. 2011; Bailey et al. 2013). Although several NP regenerative studies and a few in vitro AF studies (Nerurkar et al. 2009) provide critical insight on the

Table 4 Annular repair

Species	Model	Treatment	Outcome	Refs
Rat	Degradation tests with subcutaneous implantation	Fibrin-genipin adhesive hydrogel (fib-gen)	60% of fib-gen remained at 8 weeks and nearly all resorbed at 16 weeks; kinetics show better in vivo longevity compared to fibrin	Likhitpanichkul et al. (2014)
Rat	Needle puncture	Injection of cross-linked high-density collagen (HDC) gels	Cross-liked HDC capable of repairing annular defects most likely due to enhanced stiffness of HDC at 5 weeks	Grunert et al. (2014b)
Porcine	Needle puncture	Injection of Gelfoam, platinum coil, bone cement, and tissue glue	Injection of Gelfoam better improved integrity of punctured disc than the other three to potentially prevent recurrent disc herniation at 2 months	Wang et al. (2007)
Sheep	Box annulotomy	Patch and plug with small intestinal submucosa (SIS) and titanium bone screw	SIS-based treatment led to better maintenance of hydration and intradiscal pressure at 26 weeks after annulotomy	Ledet et al. (2009)
Sheep	Box annulotomy	Triphase AF implant composing two outer phases of absorbable polyglycolic acid (PGA) and a centric phase of a nonabsorbable polyvinylidene fluoride (PVDF) mesh	Implant-treated discs had more reparative tissue. But, contrast media leakage tests under provocative pressure did not show a difference between groups	Hegewald et al. (2015)
Sheep	Microdiscectomy	Allogenic mesenchymal progenitor cells (MPCs) + pentosan polysulfate (PPS) embedded in a gelatin/fibrin scaffold	Discs treated with MPC + PPS showed higher PG content than the untreated or ones treated with solely scaffold at 6 months	Oehme et al. (2014)
Sheep	Box annulotomy	Injection of cross-linked high-density collagen (HDC) gel into annulus defect	IVDs treated with HDC gel showed histologically less degeneration. Imaging difference was not significant	Pennicooke et al. (2017)

reparative process within the AF tissue (Wei et al. 2009; Sakai et al. 2006; Sato et al. 2003; Zhang et al. 2011c), there are a very limited number of in vivo studies focusing primarily on annular repair (Table 4). Current efforts in the biological treatment for in vivo AF repair include either development of injectable material in conjunction with biologics such as biomolecules/cells or construction of rigid implants derived from synthetic polymer or biological tissue.

In order to introduce alternative methods, injectable biomaterials have recently gained further popularity in the field. Injectable genipin cross-linked fibrin collagen gel was suggested to integrate with human AF tissue and presented promising biomechanical and cell-seeding properties in vitro (Schek et al. 2011). Our group successfully tested a high-density collagen gel in vitro and in vivo using a needle puncture rat tail model. Furthermore, we have recently translated this project to a large animal (ovine) model, which demonstrated positive histologic results at 16 weeks following injury (Pennicooke et al. 2017).

Collectively, these studies demonstrate an ability to formulate and deliver injectable biomaterials to the lumbar spine of sheep to seal AF defects, promote sufficient tissue healing, and prevent further disc degeneration.

In another large animal study conducted by Oehme et al., injected mesenchymal progenitor cells combined with chondrogenic agent pentosan

polysulfate maintained disc height, disc morphology, and NP proteoglycan content post microdiscectomy in a sheep model (Oehme et al. 2014). Despite the few studies dedicated to annular repair, more attention is now being paid to this field given its enhancement of even NP-targeted therapy.

Bioartificial Total Disc Replacement Therapies

In advanced stages of DDD with significant structural damage and the absence of viable cell activity, the injection of biomolecules or cell transplantation is no longer a feasible option.

A current surgical treatment strategy for advanced DDD is the total removal of the IVD followed by the fusion of the whole segment including the adjacent vertebrae. However, fusion may result in pseudoarthrosis or adjacent segment disease, which may lead to reoperation and long-distance fusion procedures (Maldonado et al. 2011; Sugawara et al. 2009; Bydon et al. 2013). To prevent these complications and to preserve mobility in the treated segment, TDR by synthetic prosthesis has become an alternative treatment strategy. Yet, current mechanical prosthetic TDR devices have not been able to reproduce the biomechanical properties of the natural IVD. Additionally, recent studies have demonstrated that current TDR devices are not without their disadvantages as they also entail the risk of adjacent segment disease (Maldonado et al. 2011; Kelly et al. 2011).

In this case, the total replacement using a tissue-engineered intervertebral disc with the ability to integrate into the host environment is a promising treatment strategy. The current standard in whole IVD implantation involves NP and AF composites that replace the structurally damaged tissues of a severely degenerated disc.

The first tissue-engineered whole IVD, implanted in vitro within the subcutaneous dorsum of athymic mice, comprised of NP cell-laden polyglycolic and polylactic acid (PGA/PLA) and AF cell-laden alginate (Mizuno et al. 2004a, 2006).

More than a decade ago, our group was the first to develop a tissue-engineered disc, composed of NP cells seeded into an alginate hydrogel, surrounded by a polyglycolic acid and polylactic acid scaffold seeded with AF cells (Mizuno et al. 2004b, 2006). This de novo construct was successfully implanted in the subcutaneous space of the dorsum of athymic mice and demonstrated the feasibility of creating a composite IVD including both AF and NP tissues Several other studies have reported the development of composite tissue-engineered IVD constructs, using combinations of materials such as demineralized bone matrix gelatin with type II collagen, hyaluronate and chondroitin-6-sulfate (C2/HyA-CS) (Zhuang et al. 2011), electrospun polycaprolactone and agarose (Martin et al. 2014), and self-assembled NP cells seeded onto calcium polyphosphate (Hamilton et al. 2006).

More recently, we developed a TE-IVD construct composed of an NP cell-laden alginate nucleus encircled by an AF cell-laden collagen annulus (Bowles et al. 2010, 2012). The efficacy of this construct, namely, maintaining disc height and physiological hydration as well as integrating into the host tissue, has been demonstrated through its implantation in a rat tail in vivo model (Bowles et al. 2011a; Gebhard et al. 2010, 2011; Grunert et al. 2014a; James et al. 2011). Although these results are promising, the rat tail has several dissimilarities with the human spine in terms of anatomy and biomechanical properties (O'Connell et al. 2007, 2011; Lotz 2004). Importantly, the rat tail has a significantly different biomechanical loading profile, as the IVDs of the human spine are exposed to higher axial loads. Furthermore, the rat tail lacks a spinal canal containing nervous tissue as well as posterior bone and joint elements. To move our approach closer toward clinical utilization and to mimic the biomechanical loads and anatomy of a human IVD more accurately, we transitioned to a larger animal model.

In a preliminary study, we performed TDR using TE-IVDs in the cervical spine of skeletally mature beagle dogs. Within this, we demonstrated

the ability of our TE-IVDs to integrate into the host tissue of a larger animal without any signs of inflammatory response (Moriguchi et al. 2017). Notably, these implants performed quite well when stably implanted in the intervertebral space. However, there was a persistent challenge in ensuring that implants remained firmly implanted in the intervertebral space.

Nonetheless, the addition of growth factors or bioactive molecules can encourage de novo ECM deposition. Goldschlager et al. demonstrated that adult allogenic mesenchymal progenitor cells (MPCs) formulated with a chondrogenic agent pentosan polysulfate (PPS) could synthesize a cartilaginous matrix when implanted into a biodegradable carrier and cage and over time might serve as a bioactive interbody spacer following anterior cervical discectomy (Goldschlager et al. 2010). Furthermore, the integration of tissue engineering and gene therapy has been attempted by a Chinese group that developed a tissue-engineered IVD using an allogenic disc transduced with hTERT gene within its NP cells. When implanted in a canine model, the hTERT-loaded NP cells manifested enhanced antidegenerative effect than unloaded NP cell (Xin et al. 2012). Such constructions of whole disc implants, the most ambitious therapeutic strategy yet, are met with extensive biological and functional challenges in vivo. Yet, the progressing field of TE continues to yield promising modifications to meet the higher demands of implanted discs.

Clinical Studies

Several of the above-described regenerative treatment approaches have already been utilized in a clinical setting. However, to date only a few clinical trials have been published on this topic (Table 5).

In the following section, several representative published clinical studies for the different treatment approaches will be presented.

In 2002, Meisel et al. started a multicenter prospective, randomized, controlled, non-blinded EuroDISC study comparing the safety and efficacy of autologous disc chondrocyte transplant (ADCT) implanted 12 weeks post discectomy. The 2-year interim analysis revealed a significant reduction of low back pain as well as retained disc height in the autologous disc cell transplantation (ADCT) group compared to the discectomy only control group (Meisel et al. 2006, 2007). The ADCT product is currently evaluated in a Phase II clinical trial under the product name NOVOCART® Disc (Meisel 2012; Tschugg et al. 2017).

While to date there is no clinical study using tissue-engineered material, efforts have been made to create functional substitutes for NP (Berlemann and Schwarzenbach 2009; Boyd and Carter 2006). Among many clinical studies focusing on NP replacement, a single-center, non-randomized, prospective feasibility study was undertaken to investigate the use of NuCore Injectable Nucleus hydrogel (Spine Wave, Inc., Shelton, CT, USA) post microdiscectomy prevented early disc collapse to potentially slow the degenerative cascade of the spinal segment over time (Berlemann and Schwarzenbach 2009).

The feasibility of a whole allogenic disc transplantation has first been proven by a group in China. Ruan et al. successfully performed transplantation of fresh frozen disc allografts including endplates in five patients. Implants successfully integrated into the host tissue, over the course of 5 years without any inflammatory reaction, although no immunosuppressive therapy was administered (Ruan et al. 2007). The absence of any immunologic response strongly supports the hypothesis that the intervertebral disc space is immunoprivileged tissue. Although promising, the allogenic transplantation of spinal motion segments has several limitations in terms of availability of healthy donor discs and potential disease transmission.

As mentioned in the section above, a frequently discussed treatment strategy is the intradiscal injection of platelet-rich plasma (PRP) for treating DDD. In 2016, Tuakli-Wosornu et al. published the results of a prospective, double-blind, randomized controlled study. Twenty-nine patients with low back pain, refractory to conservative treatment, received intradiscal PRP injections, while 18 patients who received a

Table 5 Published clinical trials

Trial treatment	No. of patients	Study design	Follow-up (m)	Outcome	Refs
Autologous hematopoietic stem cell injection	10	Case series	12	No patients reported any improvement in their discogenic back pain	Haufe and Mork (2006)
Total disc replacement with allogenic IVD	5	Case series	60	Allograft engrafted disc space without apparent immunoreaction; all minus one disc preserved range of motion	Ruan et al. (2007)
Autologous disc chondrocyte transplantation (EuroDisc)	28	Control study	24	ADCT with discectomy shows more pronounced decrease in OPDQ than discectomy alone	Meisel et al. (2006, 2007)
Injectable biomimetic nucleus hydrogel	14	Case series	24	Significant improvement in leg and back pain after micro-discectomy	Berlemann and Schwarzenbach (2009)
Autologous bone marrow mesenchymal cell injection	2	Case series	24	Both patients showed improvements in the vacuum phenomenon as well as signal intensity of T2-weighed MRIs	Yoshikawa et al. (2010)
Autologous bone marrow mesenchymal cell injection	10	Case series	12	Rapid improvement of pain and disability. Disc height was not recovered, but disc hydration was significantly elevated	Orozco et al. (2011)
Allogenic juvenile chondrocytes injection (NuQu)	15	Case series	12	ODI, NRS, SF-36 improved from baseline. 89% of the patients showed improvement on MRI	Coric et al. (2013)
Injection of autologous bone marrow-concentrated cells	26	Case series	12	Statistically significant improvement in pain scores and impairment was demonstrated. Most dramatic improvement seen in patients with higher CFU-F concentrations. Rehydration of the discs observed in 8 of 20 patients	Pettine et al. (2015)
Intradiscal injection of PRP	47	Prospective double-blinded randomized controlled study	12	Significant improvement in pain scales after 2 months, maintained at the 12-month follow-up	Tuakli-Wosornu et al. (2016)
Intradiscal injection of stromal vascular fraction with PRP	15	Case series	12	Significant improvement in VAS, no worsening, no radiographic changes	Comella et al. (2017)

placebo injection with a contrast agent served as a control group. At the 2-month follow-up, the PRP group showed significant improvement in pain scales. Patients maintained these improvements also in the 12-month follow-up (Tuakli-Wosornu et al. 2016).

Recently the utilization of different stem cell lines has found their way to clinical use. In 2006 Haufe et al. was the first to publish clinical results, reporting about intradiscal autologous hematopoietic stem cell injections. However, in the 12-month follow-up, none of

the ten patients reported any improvement in back pain, and 80% of the patients required surgical spinal intervention within a year after injection. Mesenchymal stem cells (MSC) on the other hand showed more promising results in various clinical studies. Due to their relatively easy accessibility and expandability in vivo, the bone marrow has been used as a source for MSCs in several in vitro and in vivo studies. Pettine et al. were the first to utilize bone marrow-concentrated cells (BMCs) as a treatment for discogenic back pain. In 26 patients with chronic low back pain, BMCs harvested from the iliac crest were injected into the IVD. The 1-year follow-up revealed a reduction in pain as well as radiographic improvement in 40% of the patients (Pettine et al. 2015). Yoshikawa et al. reported a case series of two patients who received a collagen sponge soaked with 10^5 cells/mL suspension grafted into a degenerated disc. After 2 years, both patients demonstrated improvement in pain as well as increased hydration on MRI (Yoshikawa et al. 2010). Orozco et al. reported a rapid improvement of pain up to 85% after 3 months in ten patients who underwent intradiscal injection of bone marrow-derived MSCs. Despite the fact that the disc height remained unchanged, an improvement in disc hydration could be observed in the 12-month follow-up MRI (Orozco et al. 2011).

Apart from the bone marrow, the adipose tissue is an abundant source for mesenchymal stem cells (Ganey et al. 2009; Jeong et al. 2010). Due to easier accessibility and less invasive harvest, the utilization of adipose-derived stem cells became more recently of increasing interest. In a recent study, Comella et al. were the first to publish clinical results on the injection of stromal vascular fraction (SVF), containing adipose-derived stem cells as a treatment for low back pain. In this study, SVF was administered along with PRP into lumbar IVDs in15 patients with discogenic back pain. After a 12-month follow-up, patients showed significant improvement in pain scales. However, this study did not provide any radiographic outcome data (Comella et al. 2017).

Unpublished Clinical Trials

Within the last decade, a clear trend toward regenerative treatment approaches is recognizable. This trend is also represented by the increasing number of clinical studies currently emerging aiming to find new biological treatment approaches for DDD. The following will elucidate several promising ongoing clinical studies that are not published yet.

Due to the similar biological profile as disc chondrocytes and potential immunoprivileged property, allogenic juvenile articular chondrocytes are another promising cell source. In a prospective cohort study, Coric et al. demonstrated that NuQu, an injectable percutaneous fibrin-based delivery of juvenile chondrocytes attenuated otherwise medically refractory low back pain (Coric et al. 2013). A class II study has recently been completed. Despite these study's promising results, further investigation with a prospective, randomized, double-blinded, placebo-controlled study is necessary to make cell transplantation a valid therapeutic option for DDD.

Rathmell et al. are currently the first to evaluate the effects and safety of intradiscal injections with recombinant human growth and differentiation factor 5 (rhGDF5) in a clinical trial. GDF-5 belongs to the transforming growth factor-beta (TGF-β) family which is meant to influence the growth and differentiation of various tissues including the intervertebral disc (Xu et al. 2006). The intradiscal administration has shown to improve the reparative capacity of IVDs in a degenerative rabbit model (Chujo et al. 2006). Within a Phase I/II clinical trial, 32 patients receive a single intradiscal injection of rhGDF5 and will be observed over a 36-month follow-up (J R 2008).

Mesoblast Ltd. developed a commercially available lineage of in vitro differentiated allogenic mesenchymal precursor cells (MPCs). Currently, this product is being evaluated under the name Rexlemestrocel-L in a Phase III prospective, multicenter, randomized, double-blind, placebo-controlled study, comparing Rexlemestrocel-L only vs. Rexlemestrocel-L+ hyaluronic acid (Mesoblast Ltd. 2015).

The recently completed Phase II study included 100 patients with chronic low back

pain due to DDD. The outcomes of this study were promising; both treatment groups who received 6 million MPCs and 18 million MPCs, respectively, improved in VAS by 44.4% and 37.9%, whereas the two placebo groups who received saline or hyaluronic acid only improved by 11.8% and 15.8%, respectively. However, no significant improvement in radiographic outcomes could be observed (Mesoblast Ltd. 2019).

The data emerging from these ongoing clinical trials will reinforce findings from published studies and provide new insight for future biological disc repair.

Future Perspective

This present book chapter provides a comprehensive overview on the recent innovations and trends in biological disc repair (Takahashi and Yamanaka 2006). Biomolecular therapies have shown the potential of stimulating the intrinsic healing capacity of the intervertebral discs in early stages (Masuda et al. 2006; Chujo et al. 2006; Huang et al. 2011). In a more advanced setting, cellular therapies are increasingly demonstrating their potential as the understanding of underlying mechanisms of cell differentiation increases (Pittenger et al. 1999; Bernardo et al. 2007; Moroni and Fornasari 2013). A major challenge for cellular therapies remains the determination of the optimal cell type as well as the ideal carrier for application (Acosta Jr et al. 2005).

Another challenge is that all these treatments are inevitably associated with an annular damage caused by the needle puncture, which is necessary for the application of the therapeutic agent. Carragee et al. has shown in a prospective study of notable size that even a small needle puncture may disturb the integrity of the AF enough to accelerate the degeneration of the IVD (Carragee et al. 2009). Therefore, a sufficient annular repair strategy is mandatory in order to seal the defects caused by the necessary needle puncture.

Since the lack of viable cells in advanced DDD makes a stimulating agent, such as growth factors, impossible and the final stages of DDD do not possess enough extracellular matrix to offer an environment for viable cells (Roberts et al. 2006), a replacement will become inevitable. It is known that current mechanical prosthetic devices also involve the risk of adjacent segment disease and thus accelerate further degeneration of the whole spine (Maldonado et al. 2011; Kelly et al. 2011). Therefore, it is inarguable that a biological construct with the ability to integrate into the host tissue will be the better option. Considering the limitations of healthy allogenic transplants (Ruan et al. 2007), tissue engineering will be the best option for end-stage DDD. Although promising, the described in vivo studies for TDR using tissue-engineered constructs (Grunert et al. 2014a; Moriguchi et al. 2017; Bowles et al. 2011b) are still facing challenges that need to be solved before a transition to clinical use will be possible.

Despite all the above-described advances, we still have limited understanding of the physiological concept of a healthy IVD as well as the underlying pathomechanisms of disc degeneration. Also the pathophysiological correlation between back pain and degenerative disc disease is still not entirely explored. Therefore, extensive research about the physiological as well as the pathological processes in intervertebral discs is mandatory before the ideal treatment strategies can be developed.

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