■ REVIEW ARTICLE

Repair of defects in articular joints

PROSPECTS FOR MATERIAL-BASED SOLUTIONS IN TISSUE ENGINEERING

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Each year, more than one million procedures to treat defects of articular cartilage are performed in the knee alone.¹ While removal of loose bodies and the chondroplastic procedures of debridement and shaving account for approximately half of these, the remainder involve treatment of defects which are sufficiently severe to require the replacement or assisted regeneration of damaged hyaline cartilage.

Over the past decade, methods based on the principles of tissue engineering and regenerative medicine have emerged as the most promising approaches for the repair of defects in articular joints.^{2,3} These make use of combinations of cells, signalling molecules and scaffolds to augment the natural regenerative capacity of the body, with the aim of restoring healthy structure and function to damage or diseased tissue.⁴

Tissue engineering has produced success in a number of soft-tissue applications,^{5,6} but the regeneration of articular surfaces poses a unique challenge, in that articular cartilage has a limited capacity for self-repair and mechanical stimuli play a large role in the structure and function of cartilage and subchondral bone.

While these factors place special demands on all aspects of tissue engineering, scaffold design takes on a role of particular importance. This review addresses the design requirements for scaffolds used in the treatment of chondral and osteochondral defects. Particular emphasis is placed on recent trends in scaffold design and their potential for clinical application.

Current treatment concepts

Due to the limited capacity of articular cartilage for self-repair, surgeons have adopted a range of techniques in attempts to relieve pain, restore function and slow or halt the progression of disease. Selection of the appropriate method of treatment depends on the size, cause and depth of a defect. Large defects or advanced osteoarthritis are indications for total knee arthroplasty (TKA), while smaller

defects caused by trauma can be treated by limited arthroscopic methods.⁷ The management of full-thickness defects must take into consideration the limited and temporary spontaneous capacity for healing afforded by access to subchondral vessels. Partial-thickness lesions have no such capacity. Options for the treatment of intermediate-sized defects include techniques of marrow stimulation such as abrasion arthroplasty, subchondral drilling and microfracture. These may provide temporary relief of pain and can delay the need for TKA for several years. However, they result in fibrocartilaginous repair tissue whose deficiencies inevitably lead to breakdown under normal joint loading.^{8,9}

Over the past two decades, a number of new techniques based on transplantation and regeneration have been developed for the restoration of a hyaline or hyaline-like structure following injury to cartilage. However, only autologous chondrocyte transplantation with periosteal grafting and autologous osteochondral grafting have as yet been applied extensively clinically.¹⁰⁻¹³ Both these techniques have demonstrated the ability to promote both short- and long-term restoration of healthy hyaline or hyaline-like cartilage, but the clinical results show some drawbacks with each^{10,11,14-16} (Table I).

Regenerative repair of defects of articular cartilage

Tissue engineering can be defined as the application of the principles of engineering and life sciences toward the development of biological substitutes which restore, maintain or improve tissue function.⁴ It has been widely heralded as a possible means by which the ever-increasing demand for tissue replacements can be met.

Tissue engineering employs a multidisciplinary approach, drawing on the principles of cell biology, molecular developmental biology, materials science and biomechanics, to aid in the repair of tissues damaged beyond the natural healing capacity of the body. The under-

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Table I. Advantages and disadvantages of autologous chondrocyte implantation and autologous osteochondral grafting^{10,11,14-16}

	Clinical outcome	Drawbacks/concerns
Autologous chondrocyte transplantation	<ul style="list-style-type: none"> - hyaline-like repair tissue reported in 93% of patients at 1.5 to 10 years (n = 19+52)^{14,15} - candidate defect sizes 1 to 16 cm²¹⁰ - 76% of outcomes good-excellent* at 2 to 10 years post-op (n = 219)¹⁴ 	<ul style="list-style-type: none"> - donor site disturbance/morbidity - requirement for an overlying periosteal flap/membrane to prevent proteoglycan elution/difficulty of suturing periosteal flap to surrounding healthy cartilage - presence of an overlying fibrous tissue layer resulting from remnants of periosteal flap - potential inability of dedifferentiated cultured chondrocytes to regain chondrocytic phenotype - cost - donor site disturbances/morbidity
Autologous osteochondral grafting	<ul style="list-style-type: none"> - survival of transplanted hyaline cartilage in 85% of patients (n = 68)¹⁶ - candidate defect sizes 1 to 4 cm² - 91% of outcomes good-excellent† – at 3 to 6 years post-op (n = 126)¹¹ 	<ul style="list-style-type: none"> - limited size of defect repair/limited availability of donor tissue (3 to 4 cm² for patellofemoral peripheries)¹⁶ - potential for injurious compression of grafts due to unphysiological loading (low-weight-bearing donor site vs. high-weight-bearing graft site) - damage imparted to graft during removal from donor site and press-fitting into implantation site

* modified Noyes Cincinnati rating

† modified Hospital for Special Surgery rating

lying principles involve the induction of cellular remodeling of a scaffold to produce tissue with a predefined form and function. Three key ingredients are necessary, namely cells; an extracellular matrix (ECM), which may be either natural or synthetic, and inductive signals.^{17,18}

The use of tissue engineering for repair of articular cartilage poses unique challenges. The tissue structure of articular cartilage is heterogeneous, with a distinct zonal arrangement.¹⁹ It includes, most significantly, a transition from mineralised to unmineralised tissue, the regeneration of which must be induced. Furthermore, the avascularity of articular cartilage results in a low capacity for self-repair.

Another distinctive feature is the unique relationship of structure to function, in which both biochemical and mechanical stimuli strongly influence the integral relationship between the extracellular matrix and the cells that maintain it (Fig. 1).²⁰

Developments in scaffold design

Although a wide range of materials has been used to fabricate scaffolds for the repair of chondral and osteochondral defects²¹⁻³⁴ (Table II), research has failed to produce agreement as to the most appropriate class of biomaterials for regeneration of articular cartilage. Recent efforts have

Fig. 1

Structure-function relationship for cartilage and subchondral bone.

Table II. Scaffold materials for the repair of chondral and osteochondral defects²¹⁻³⁴

Material	Reference
Calcium phosphates (CaP)	
Hydroxyapatite (HAp)	21
Biological polymers	
Collagen	22
Fibrin	23
Alginates	24
Hyaluronan (HyA)	25
Chitosan	26
Collagen/glycosaminoglycan (GA)	27
Synthetic polymers	
Polylactic acid (PLA)	28
Polyglycolic acid (PGA)	29
Polycaprolactone (PCL)	30
Other	
Demineralised bone matrix	31
Devitalised cartilage	32
Periosteum	33
Bioactive glass	34

aimed to explore combinations of existing materials, as opposed to developing new ones.

Traditional approaches to tissue engineering have sought to address both surface lesions and osteochondral defects using compositionally homogeneous scaffolds. The failure of these approaches to produce a healthy, regenerated hyaline structure has necessitated recent initiatives to recognise that the conventional set of requirements for chondral and osteochondral regeneration scaffolds, namely that they be bioresorbable, possess low immunogenicity, be non-toxic, facilitate attachment to the defect site, promote cell attachment and possess adequate mechanical strength,^{35,36} are sufficient to meet the requirements for restoration of healthy joint structure and function.

Most prominent among these have been attempts to develop multiphase, compartmental scaffolds (Table III),

which incorporate separate osseous and cartilaginous components.^{23,37-44} These seek to address the separate, and sometimes contradictory, requirements for regeneration of bone and cartilage, while recognising that the restoration of a healthy articular surface depends on the success of both processes simultaneously.

Emerging issues for scaffold design

Role of mechanical stimulus. The functional and structural properties of healthy articular cartilage are conditioned to withstand the stresses to which they are most regularly subjected, with mechanical stimuli affecting cellular distribution,⁴⁵ metabolic activity^{46,47} and the mechanical properties of the tissue itself.⁴⁸ This integral relationship is governed by a negative feedback loop,²⁰ in which deformation of the extracellular matrix (ECM) induced by physiological loading or otherwise stimulates changes in the metabolic activity of the cell and matrix expression.⁴⁹ This in turn alters the structure of the ECM and the manner in which it deforms under load, resulting in a configuration best suited to the loading environment in which it exists (Fig. 1).

Preliminary results *in vivo* indicate that the effect of mechanical stimulus is equally influential on the remodelling of tissue-engineered cartilage constructs. While subcutaneous implantation of growth factor- and cell-loaded scaffolds has demonstrated the feasibility of chondrogenesis in the absence of mechanical stimulation,^{50,51} the regenerated tissues generally lack the hyaline structure and mechanical properties of articular cartilage.^{38,52} Superior histological scores have been observed in defects in femoral condyles, which are subjected to high loading, compared with those on the patellar groove, where loads are much lower, when treated with identical implant materials.⁴¹

Given the importance of this relationship of structure to function, a major objective of scaffold design is to provide an optimal loading environment under whose influence the properties of regenerated tissue can develop.⁵³ While exact

Table III. Multiphase scaffolds for osteochondral repair^{23,37-39,41-44}

Cartilaginous compartment*	Osseous compartment*	Joining method	Test site	Seeded cell type	Growth factors/drugs	Reference
PLGA	Bioglass	Solvent fusion	Femoromedial condyle/patellar groove	Autologous chondrocytes (uniform)	None	41
PLGA	Calcium sulphate	Solvent fusion	Femoromedial condyle/patellar groove	Autologous chondrocytes (uniform)	None	41
Hyaluronan	ICP	Infiltration	Femoromedial condyle	Marrow-derived progenitors	None	37
PGA	Collagraft (Collagen/HAp/TCP)	Suturing	Femoropatellar groove	Allogeneous chondrocytes/autologous bone marrow	None	42
PGA	PLGA/PEG	Suturing	Chondrocyte/periosteal cell culture	Chondrocytes/periosteal cells	None	43
PLA/PLGA	PLA/PLGA/TCP	Solvent fusion	Chondrocyte culture	Chondrocytes	None	44
Hyaluronan	HAp/TCP	Tissue glue	Subcutaneous	Mesenchymal stem cells	TGF- β /osteogenic supplements	38
Agarose	Decellularised bone	Infiltration	Autologous chondrocyte	Autologous chondrocytes (cartilage only)	None	39
Gelatin/fibrin	Gelatin/fibrin	None	Trochlear groove/femoral condyle	None	TGF- β /IGF-1/suramin	23

*PLGA, polylactic co-glycolic acid; PGA, polyglycolic acid; PLA, polylactic acid; ICP, injectable calcium phosphate; HAp, hydroxyapatite; TCP, tricalcium phosphate; PEG, polyethylene glycol

Fig. 2

Deformation of and stresses within a) natural articular joint, b) homogeneous scaffold and c) multiphase scaffold (E, elastic modulus; ϵ , transverse strain; τ , shear stress; c, cartilaginous component; b, osseous component).

replication of the highly complex mechanical behaviour of articular cartilage⁵⁴ is beyond the limits of current technology, the incorporation of a stiff, subchondral osseous component has sought to mimic natural loading (Fig. 2).

The bone cartilage interface

Natural joints are characterised by continuity of collagen fibrils between their respective zones, including the transition from mineralised to unmineralised tissue.¹⁹ The resultant system of smooth transitions, soft interfaces, imparts an intrinsic mechanical stability which allows joints to withstand physiological loading without mechanical failure.⁵⁵

In contrast, the majority of combination scaffolds which have been developed contain hard interfaces, forming a distinct boundary between two dissimilar materials. Suturing,⁴³ fibrin adhesive bonding³⁸ and other techniques^{37,39} have been used to strengthen this interface. The persistent incidence of dislocation of implants⁴² raises questions regarding the reproducibility of these methods.

Efforts to produce scaffolds with soft interfaces constitute an important focus of current research but have so far been limited to the use of synthetic polymers. Using computer-aided design (CAD)/computer-aided machining (CAM)

technology, layers with successively decreasing proportions of calcium phosphate can be deposited, with each bonded to the next via solvent bonding.⁴⁴

A number of calcium phosphate/collagen biomaterials, whose nanostructure closely resembles that of bone, have been developed in recent years.^{56,57} However, a major obstacle to their development into combination scaffolds is the lack of systematic means to control the ratio of calcium phosphate to collagen. The development of such a capacity may constitute a first step towards producing a biomimetic multiphase scaffold.

Phenotype-specific biomaterials

As the general understanding of the complex processes of chondrogenesis and osteogenesis has progressed, so too have efforts aimed at tailoring biomaterials to invoke specific cellular responses at the molecular level.⁵⁸

Certain biomaterials have come to be associated with specific tissue-engineering applications such as hyaluronan with cartilage regeneration⁵⁹ and calcium phosphates with the treatment of bone defects.⁶⁰ With some notable exceptions,⁶¹ these materials are generally not capable of inducing the formation of bone and cartilage types on their own.

Covalent immobilisation of specific proteins, peptides and other biomolecules on typical scaffolding materials can impart chondrogenic or osteogenic capacity by providing an environment that mimics that of the extracellular matrix, thus affording a favourable surface for cell adhesion.⁶² The most common of these molecules is the group of cytokines commonly referred to as growth factors.^{63,64} Function-specific peptides have also been used to tailor cellular response, while localised drug and antibiotic delivery is receiving increasing attention.^{65,66}

The clinical use of growth factors is currently limited, mainly due to lack of adequate methods of delivery.⁶⁷ Optimal delivery of growth factors and other inductive signalling molecules requires careful consideration of the mechanism through which the molecules interact with the scaffold. While we have a basic understanding of these mechanisms^{23-25,65,66,68,69} (Table IV), a system of delivery capable of sustained release has not yet been devised. Hunziker et al²³ have sought to use combinations of growth factors and antiangiogenic drugs to address the difficulties associated with preventing ossification of cartilaginous repair tissue. Through selective incorporation of the antiangiogenic drug suramin into the cartilaginous component, a biochemical or functional barrier to invasion by vasculature and marrow-derived cells was created. Invasion by vasculature, which is thought to favour osteogenesis at the expense of chondrogenesis,^{70,71} was successfully prevented when liposome-encapsulated suramin was incorporated into the cartilaginous component. Although the regenerated cartilage was not deemed to be 100% hyaline, and the mechanical properties of the interface have not been reported, the use of functional barriers constitutes a promising area of research.

Table IV. Growth factor interactions with scaffold materials^{23-25,65,66,68,69}

Material*	Growth factor binding mechanism
Calcium phosphates	
Hydroxyapatite (HAp)	Electrostatic interaction of anionic groups and HAp surface ⁶⁸
Tricalcium phosphate (TCP)	Surface adhesion ⁶⁹
Biological polymers	
Collagen	Interactions with functional groups on polypeptide chains ⁶⁵
Fibrin	Binding through heparin incorporation ²³
Alginates	Regulation of diffusion through control of hydrogel mesh size ²⁴
Hyaluronan (HyA)	Retention following partial esterification ²⁵
Synthetic polymers	
PLA, PGA, Polycaprolactone	Surface adhesion or bulk incorporation ⁶⁶

* PLA, polylactic acid; PGA, polyglycolic acid

Cell selection

While a suitable scaffold is a key part of any strategy for the repair of defects in articular joints, a cellular component is also important to augment the tissue response. This is particularly true for the chondral portion of scaffolds because cartilage has a limited capacity for self-repair. Cell seeding of the osseous portion of scaffolds is also likely to be advantageous in providing an osteogenic component to the implant.

A common dilemma for both osseous and cartilaginous repair is that of cell selection, as it is still not clear which type of cell, or from which stage of cell lineage, should be chosen. In animal studies, several alternatives have been investigated, but since various scaffolds and models have been used, direct comparison is not possible.

For the chondral portion of combination scaffolds, the cells most commonly used are chondrocytes derived from articular cartilage,^{39,41,42} but marrow stromal cells have also been used.^{37,38} Selection of cell type is particularly important in this portion of the scaffold, as cells developing a fibroblastic or osteogenic phenotype are not replaced. There is evidence that the end-point of bone-marrow stromal cell differentiation is the osteoblast.⁷²

There are potentially more options in terms of cell choice for the subchondral portion of the scaffold, but of these, bone-marrow osteoprogenitors have been most commonly used and have been shown successfully to augment the osteoconductive response in defects of both animal and human bone.^{73,74} The ability to expand these cells in culture whilst maintaining their progenitor status⁷⁵ is particularly valuable as the amount of marrow available is limited.

The cells used in the repair of osteochondral defects should be autologous in order to avoid adverse immunological responses. However, as with all techniques relying on the harvest of autologous materials, lack of supply poses a potential problem. Alternatives to bone marrow as sources of autologous osteogenic and chondrogenic progenitors include adipose tissue and muscle, both of which could be harvested prior to surgery.^{76,77}

Looking further ahead, the use of embryonic stem cells derived from human blastocysts may become possible.⁷⁸ Although there are ethical problems associated with the production of these cells, evidence of their ability to pro-

duce both cartilage and bone has indicated the potential of this approach.^{79,80}

Practical considerations

The practical considerations of time, space and limited apparatus dictate that operative procedures which are overly complex or require a high level of surgical skill may not be practicable. The assembly of separate bone and cartilaginous portions of a scaffold poses the problem of reproducibility, as use of adhesives such as fibrin glue³⁸ requires significant care to avoid variation in the thickness of the interface and pore impregnation, while suturing⁴² requires the respective layers to be sufficiently robust to avoid damage. Recent development of pre-assembled multiphase scaffolds has addressed these difficulties to a certain degree,^{41,44} but an ideal interface-free multiphase scaffold has yet to be realised.

The geometric problem of filling irregular defects provides another practical challenge. Unlike conditions in controlled animal models, the geometry of clinical defects cannot be predicted, and thus the clinical applicability of prefabricated anatomically-shaped constructs³⁹ is questionable. A feasible solution to this problem may lie in mosaicplasty where combinations of cylindrical plugs with varying diameter would allow filling of 80% to 90% of the defects.¹³

Conclusion

The requirements for the tissue engineering of articular joints pose a unique challenge for scaffold design. Development of new treatments to induce restoration of articular joint structure and function has prompted the emergence of new scaffold technologies, which include multiphase scaffolds and functional barriers, among others. In addition to meeting the traditional requirements for scaffold design, new approaches must seek to address the complex interactions of biological and mechanical factors which influence the structure of regenerated tissue in articular joints.

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