

Review

Cell colonization in degradable 3D porous matrices

Benjamin J. Lawrence and Sundararajan V. Madihally*

School of Chemical Engineering; Oklahoma State University; Stillwater, Oklahoma USA

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Cell colonization is an important in a wide variety of biological processes and applications including vascularization, wound healing, tissue engineering, stem cell differentiation and biosensors. During colonization porous 3D structures are used to support and guide the ingrowth of cells into the matrix. In this review, we summarize our understanding of various factors affecting cell colonization in three-dimensional environment. The structural, biological and degradation properties of the matrix all play key roles during colonization. Further, specific scaffold properties such as porosity, pore size, fiber thickness, topography and scaffold stiffness as well as important cell material interactions such as cell adhesion and mechanotransduction also influence colonization.

Introduction

Advancements in tissue culture technique have allowed better understanding of cellular events and regulations in two-dimensional architecture. Interactions between the cells and the underlying matrix elements control cellular attachment, proliferation and activity. Dynamic cellmatrix interactions orchestrate the morphogenesis of cells. During the process, not only do cells undergo morphogenesis but they also remodel the matrix components. The general dogma is that a chemical¹ or mechanical² stimulus signaled through focal adhesion points³ changes the polymerization state of the cytoskeletal actin and subsequent cellular activity.⁴ A complex cascades of events follow including the activation of intracellular signaling pathways with tyrosine phosphorylation of focal adhesion kinases (FAKs) that change the differentiation, proliferation and migration of cells.⁵

Recent understanding of the components required to proliferate human embryonic stem cells in two-dimension, without lineage commitment, has opened a new window of opportunity to develop new cellular therapies and to generate functionally replaceable devices and/or tissue parts.⁶ Transforming these concepts into useful applications is currently limited due to the complexity of interactions that affect the differentiation and proliferation of stem cells. Despite much anecdotal evidence suggesting the plastic nature of mature cells, this possibility has not been explored. For example, chondrocytes (cells present in the cartilage) lose their

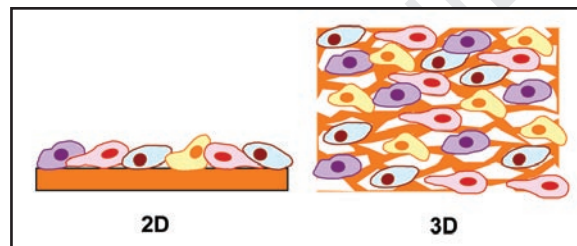


Figure 1. 2D and 3D cell culture.

round morphology in monolayer culture and assume a fibroblast-like phenotype. The phenotypic change subsequently leads to an altered biosynthesis of matrix proteins, increasing synthesis of collagen type I instead of collagen type II (a chondrogenic marker protein). After reseeding the differentiated chondrocytes on alginate microparticles (www.promocell.com), the cells redifferentiated into chondrocytes.

Much of the cell culture experiments performed in molecular biology are cultivated on two-dimensional (2D) tissue culture treated surfaces. Many in vitro experiments have shown that cells have different responses in colonization, proliferation and differentiation on 3D scaffolds than on traditional 2D-tissue culture. In 2D substrata, cultured cells are restricted to spread and attach to a flat rigid glass or tissue culture plastic surface (Fig. 1). Hence, effects of biophysical properties of the matrix that provide a spatio-temporal effect in the body are not part of the effect. However, biophysical properties significantly influence cell adhesion and functions in three-dimensional (3D) environment.⁷ 3D matrices provide physical cues to guide cell colonization as well as chemical cues of cell-binding sites to support cell attachment and spreading (Fig. 2).

To obtain useful functions and understand morphogenesis of tissues, one has to recreate the microenvironment that is conducive to drive the stem cells to required cell type. A number of investigators have focused on evaluating the role of various soluble factors on the regulation of lineage development in 2D culture systems. Although these results will help understand the interactions, evaluating 3D configurations are critical (1) to understand the spatio-temporal effects, (2) to evaluate the reorganization of various compartments and organ formation and (3) to develop devices that can be used in clinical applications. Three-dimensional porous matrices offer a spatio-temporal configuration similar to in vivo conditions. They have been generated and utilized in various tissue regeneration strategies⁸⁻¹⁰ or surrogate models for evaluating disease mechanisms. The

*Correspondence to: Sundararajan V. Madihally; 423 Engineering North; School of Chemical Engineering; Oklahoma State University; Stillwater, Oklahoma 74078 USA; Tel.: 405.744.9115; Fax: 405.744.6338; Email: sundar.madihally@okstate.edu

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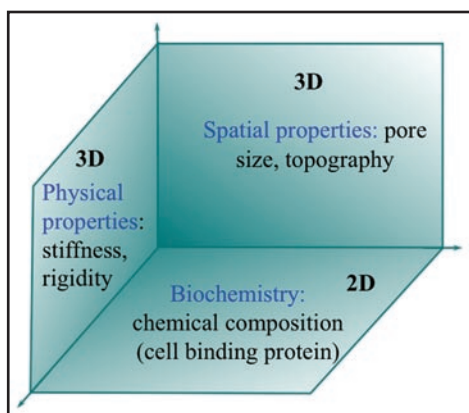


Figure 2. Factors influencing cell colonization in 3D.

purpose of this review is to summarize our understanding of factors affecting the cell colonization in 3-D porous structures.

Basics of Porous Structures

Tissue engineering has given promise for generating functionally replaceable 3D tissue parts, although currently the products obtainable are limited to avascular regions. Biodegradable scaffolds are used to support and guide the in-growth of cells i.e., they form the template for cell colonization. Scaffolding material eventually disappears leaving only the necessary healthy tissue in a topologically required form.^{8,9,11} The 3D matrix provides more space for cellular colonization and proliferation as well as provides a different set of physiological signals to the developing tissue. One option for creating tissue scaffolds is to use extracellular matrix (ECM) components derived from animal sources. For example porcine acellular dermis has been used for skin regeneration¹² and control of hypertrophic scarring.¹³ Small intestinal submucosa (SIS) is another material that has shown significant success in various tissue engineering applications.¹⁴ SIS is a dense connective tissue harvested from the small intestine. SIS promotes cell migration of numerous cell types and has been tested for regeneration of diverse tissues including large vascular grafts,¹⁵ venous valves and leaflets,¹⁶⁻¹⁸ skin,¹⁹ tendons²⁰ and wound dressing.²¹ For urinary tract reconstruction, SIS has been used for bladder augmentation,²²⁻²⁴ for ureter²⁵ and urethra^{22,26} replacement and to promote regeneration of transitional epithelium, smooth muscle and peripheral nerves with no evidence of immunological rejection.²⁷ Long-term studies show that SIS grafts can be remolded and replaced by the host and such regenerated tissues become histologically indistinguishable from native tissues.²³ Large-scale preparation of SIS is hindered by various physiochemical properties which affect the quality and reliability of the tissue regeneration in clinical settings. The physical and mechanical characteristics of the matrix, such as permeability, thickness, tensile properties, fatigue properties and ultrastructural properties, vary depending on the age of the animal, the sterilization technique and the location within the small intestine it is harvested from.²⁸

One way to overcome the heterogeneity in natural matrices uses polymers to fabricate degradable 3D porous matrices. Scaffolds generated from natural polymers such as alginates,²⁹⁻³¹ chitosan,³²⁻³⁸ collagen,³⁹ GAGs and elastin,⁴⁰⁻⁴³ gelatin⁴⁴⁻⁴⁶ and fibrin⁴⁷⁻⁴⁹ have also been used as scaffolding materials.⁵⁰⁻⁵² A commonly used system

is collagen/GAGs;^{53,54} collagen/GAG based skin equivalents are already in clinical use^{41,42} and under investigation for other applications such as heart valves, vascular grafts⁵⁵⁻⁶⁰ and vascular networks.⁶¹ However, weak mechanical strength, inadequate tailorability options in altering mechanical and degradation properties limit their usage. Synthetic polyesters such as poly (lactic acid) (PLA), poly (glycolic acid) (PGA), their copolymers (PLGA, PLLA, etc.),⁶²⁻⁷¹ and poly (caprolactone) (PCL)^{72,73} have generated immense interest as tissue engineering materials.⁷⁴ Further information on specific 3D matrices can be found in a variety of review articles.⁷⁵⁻⁷⁷ Various fabrication techniques such as free-form printing,⁷⁸ controlled rate freezing and lyophilization,³³ porogen-leaching,⁶² gas-foaming⁷⁹ and microfabrication⁸⁰ are available. Porous structures to be used as scaffolds should have the following basic properties: (1) biocompatible, bioresorbable and biodegradable during tissue regeneration process, (2) porous with an interconnected network to enable rapid tissue ingrowth through pores, and to allow unimpaired diffusion of nutrients, oxygen and wastes, (3) suitable surface properties (wettability, stiffness and compliance) to support cell attachment, proliferation and differentiation and (4) provide sufficient mechanical strength to withstand stresses at the site of implantation.

Importance of Spatial Architecture

The influence of spatial architecture of the porous matrix has been explored in various experiments. These studies^{8,9} have shown that besides chemical cues, 3D matrix physical properties such as stiffness,⁸¹ hydrophilicity, porosity,²⁹ pore size and void fraction^{82,83} can affect cell morphology, attachment and function (Fig. 2). Especially the spatial structures or the topography of scaffolds influence cell alignment, orientation,⁸⁴ multicellular organization,⁸² cell spreading⁸⁵ and cell attachment.⁸³ 2D surface features such as edges, grooves, steps, roughness and pores of substratum significantly influence cell behavior.^{84,86}

Important structural properties include both the mechanical properties inherent in the material, such as break stress, modulus of elasticity and stiffness, but also the properties of the scaffold's 3D architecture.⁸⁷ Both the microscale properties experienced by the cells and the bulk material properties that provide physical support for both the scaffold and the surrounding tissue become important during tissue regeneration. The major architecture features discussed below include porosity, pore size, fiber orientation, pore interconnectivity, topography and scaffold stiffness.

Porosity. A highly porous scaffold (>90% porosity) is desirable, since it can support the growth of tissue for the necessary nutrients transport.^{88,89} Porosity is a measure of the open pore volume within the matrix, often called the void fraction. Mathematically, porosity is defined as follows:

$$\text{Porosity} = \frac{\text{Open Pore Volume}}{\text{Total Volume}} \quad (1)$$

Several pore types are possible within a porous matrix (Fig. 3). Open pores have cellular access on both sides and allow for liquid flow and transport of nutrients through the porous matrix. Partially open pores are accessible on one side. They allow access for cell colonization, but mass transport of nutrients and waste products is limited to diffusion. Closed pores have no openings and are not accessible by cells. Other issues that complicate porosity are pore tortuosity and heterogeneous pore diameters. Consequently,

materials for tissue engineering concentrate on creating an open pore architecture.

Porosity also plays an important role in regulating cell adhesion and migration. High porosity provides a high surface area for cell-matrix interactions, sufficient space for ECM regeneration, uniform and efficient cell seeding.⁹⁰ Higher porosity could also lead to increased cell adhesion.⁹¹ Pore interconnectivity increases the overall surface area for cell attachment and facilitates cell ingrowth in the scaffolds. Increased interconnectivity and porosity also affect the deposition of ECM elements.⁹²

A functional approach to compare porous structures in a variety of materials is through their mass transport properties, such as permeability. The method of Raghavan et al., is one way to determine the permeability of the matrix,²⁸ quasi steady-state transport between two sealed chambers is assumed and the following equation is used:

$$\ln \left[\frac{C_1 - 2C_2}{C_1} \right] = - \left[\frac{A_m P}{V} \right] t \quad (2)$$

where C_1 is the initial concentration of the compound of interest, C_2 is the concentration of the compound of interest at time t . A_m is the area of the matrix normal to the mass flux, V is the volume of the test chamber and P is the permeability. Permeability is related to the diffusion coefficient by:

$$P = \frac{D_m \phi}{L} \quad (3)$$

where D_m is the diffusion coefficient, ϕ is the partition coefficient, L is the thickness of the membrane.

Pore size. There are a variety of ways to fabricate porous materials, but there are three key methods. Porous matrices may be derived from heterogeneous natural matrices, by forming void spaces within a polymer, or by creating a layered bed of polymer fibers (Fig. 4). One method for generating scaffolds uses a two phase polymer/porogen system where the porogen is subsequently removed, leaving a system of interconnected pores. The pores can be aligned by controlling porogen formation or alignment. Example of this technique are controlled rate freezing and lyophilization (CRFLT)³³ and salt leaching.⁹³ In CRFLT the pore alignment is controlled using the rate and direction of heat transfer.^{33,94} Creating a uniform pore size and distribution is a common problem when working with porous 3D matrices. Firstly, the porous structure is usually formed in an asymmetric fashion. Non-spherical pores form where one axis is much longer than the other. The spatial arrangement of pores within the material may also be an issue.

For example, in scaffolds frozen at constant temperature pores are formed in the direction of heat transfer. If a sample insulated on one side and placed in a freezer then areas near the surface of the scaffold (exposed to cold air) will freeze faster than areas deeper within the scaffold. The faster rate of freezing near the surface causes small ice crystals (and thus smaller pores) to be formed near the top edge of the scaffold material.

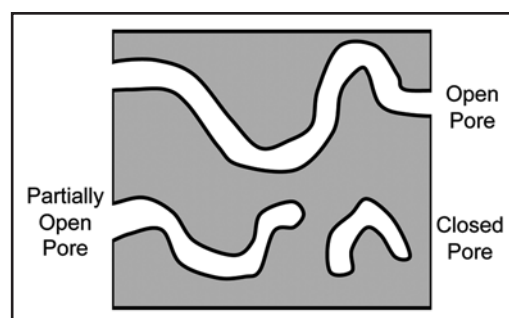


Figure 3. Types of pores within the scaffold matrix.

Pore size refers to the distance between solid sections of the porous matrix. Pore size is typically reported as the diameter of circular pores or the major axis for noncircular pores. Pore size affects cell binding, migration, depth of cellular in-growth, cell morphology and phenotypic expression.⁹⁵ Importantly, appropriate pore size provides structural advantages to allow cells to spread into the pores through “bridges” from adjacent cells. There is an “optimum size range” for supporting cell ingrowth. Outside this range, cells fail to spread and form networks. The optimal pore size range depends on the materials as well as cell types.⁹⁶ Many mature cell types including endothelial cells (ECs) are unable to completely colonize scaffolds with the pore sizes $>300 \mu\text{m}$ due to the difficulty in crossing large bridging distances.⁹⁷⁻¹⁰¹ An “optimum pore size range” for supporting cell ingrowth for majority of the mature cell types (except osteoblasts and osteocytes) is in the range of $100-150 \mu\text{m}$.⁹⁷ Recently, we showed that gelatin-chitosan scaffolds (3:1) with $50-80 \mu\text{m}$ pore size diminished the viability of fibroblasts and ECs, relative to 100 to $150 \mu\text{m}$ pore size chitosan scaffolds.⁴⁴ Pore sizes not only affect cell growth, but also affect scaffolds properties. For example, the elasticity of microporous scaffolds increases as the number of pores within the scaffold increases.¹⁰²

Fiber thickness. The matrix may also be characterized based on the microscale thickness of the individual material fibers. In some cases where the material is formed from a bed of stacked fibers fiber thickness is characterized as the diameter of the individual fibers. The fibers may be distributed randomly, as in electrospinning¹⁰³⁻¹⁰⁵ or form a highly organized system with regular repeating pore units, as in solid freeform fabrication.¹⁰⁶ Thus the fiber thickness, length, width and shape (circular rectangular, etc.) must be evaluated. However, defining fiber thickness may not be suitable when pores

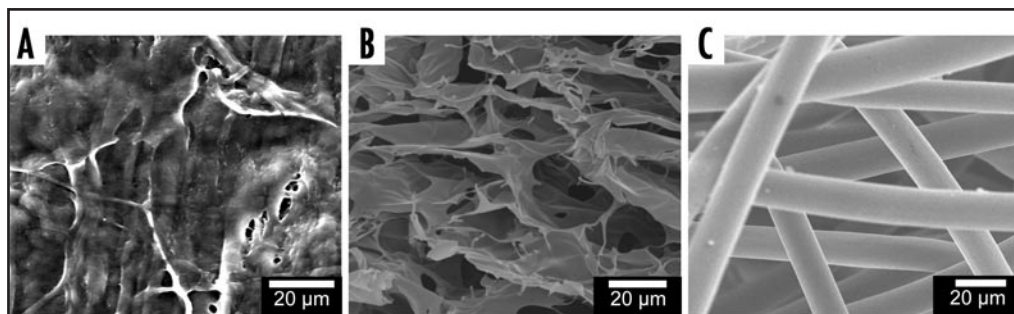


Figure 4. Types of porous polymer structures. (A) Heterogeneous structures, porcine SIS. (B) Pore based structures, freeze dried chitosan scaffolds. (C) Fiber based structures, poly (glycolic acid) mesh.

are formed by CRFLT and salt leaching technique. Utilizing freeze dried scaffolds the material forms an interconnected series of planes that connect the pores.

Fiber orientation within a scaffold affect cell colonization. Scaffolds made of oriented polycaprolactone nanofibers (700 nm in diameter) were found to promote phenotypic differentiation of chondrocytes compared with 2D nonporous membranes.¹⁰⁴ Cells seeded on oriented fibrous structures tended to maintain phenotypic shape and guided growth according to nanofiber orientation. Another study showed that significantly more collagen was synthesized by fibroblasts on aligned nanofibers than randomly orientated fibers despite similar proliferation.¹⁰⁵ A hypothesis is that spindle-shaped and oriented fibroblasts in the direction of aligned fibers mimic *in vivo* condition better and thus produce more ECM. Further studies are necessary to understand the mechanisms involved in these cell-matrix and cell-cell interactions.

Topography. The surface characteristics of scaffold materials can be described by its topography, micro to nano-scale material surface features. Topography of scaffold surface influences spreading characteristics and activity of cells.¹⁰⁷ The existence of grooves may inhibit cell movement to bend its cytoskeleton¹⁰⁸ or reshape its actin filaments to adjust to the new topography.¹⁰⁹ Curtis proposed a term “topographic reaction” to describe that cells react as a response to substratum in microscale through changes in cell orientation, motility and adhesion.⁸⁶ Surface roughness can significantly increase cell migration area.¹¹⁰ Nanometer scale roughness has been shown to improve the adhesion and growth of both smooth muscle cells⁶⁴ and chondrocytes¹¹¹ on polymer scaffolds. However, the mechanisms for enhanced cell behavior are not completely understood. Additionally, the porous structure may be modified post fabrication by inclusion of nanoparticles¹¹² or etching the surface of the matrix.⁶⁴

Altered surface texture and charge could also affect cell spreading.^{7,64} In a separate study, we blended antibacterial chitosan with PCL and showed that blending compromises the antibacterial property of the material.^{113,114} Further, the blend membranes showed better support for fibroblast spreading and proliferation. Surface roughness analysis of blend membranes showed significant increase in roughness relative to chitosan membranes, and observed antibacterial activity could be partially attributed to changed topography. Nevertheless, decreased antibacterial activity could also be due to altered surface charge distribution. Additionally, colonization and proliferation of mammalian cells may be affected by altering the charge distribution within the porous structure.¹¹⁵

Cellular Interaction

Changes in cell-adhesion. An important part of cell colonization is cell signaling. Cellular adhesion, proliferation and differentiation can be modified using specific signaling molecules, such as growth or differentiation factors. The presence of specific cellular binding sites greatly enhances cell adhesion. The proliferation and differentiation of various cell types may be controlled by incorporating signaling molecules into the tissue engineering matrix.

To incorporate bioregulation of matrix elements, grafting a small peptide Arginine-Glycine-Aspartic acid (RGD) onto polymers is an approach taken by many investigators.¹¹⁶ The use of RGD is based on the understanding that the majority of communication across the cell wall takes place via integrins, which communicate with many

matrix elements through the RGD binding domain. Additionally, materials such as collagens, glycosaminoglycans (GAGs) and their analogues (e.g., Dextran sulfate) can be incorporated into the scaffold structure in order to direct cellular growth and provide binding sites.^{115,117} Our group has previously shown that the presence of binding sites improves cellular adhesion. Endothelial cells were grown on films of chitosan, gelatin or a blend of the two and then subjected to shear stresses similar to those present in arteries or veins. Cells remained attached to the blends containing gelatin, but no cells were found adhered to the pure chitosan film.⁷

Bioregulation of the porous matrix can also be achieved by the incorporation of growth factors. For example growth factors such as vascular endothelial growth factor (VEGF)¹¹⁶ and basic fibroblast growth factor (bFGF)^{118,119} as well as ECM components such as fibronectin¹²⁰ are important in angiogenesis. Growth factors are typically proteins with short half-lives. Therefore, a controlled release system is needed in order to protect the growth factors and provide a sustained signal. Cells will also receive signals from other cells within their vicinity. Therefore, various co-culture techniques allow communication between multiple cell types.¹²¹

Many cell types such as fibroblasts, mesenchymal stem cells, epithelial cells and neural crest cells show different adhesions when grown on 3D matrices as opposed to 2D cell culture.¹²²⁻¹²⁴ A possible reason is that the 3D architecture could distribute binding sites in a variety of special locations rather than on only the single plane of rigid substrate as in traditional 2D architecture.^{124,125} Cells may have cytoskeletal adaptor proteins on 3D matrix in addition to proteins present in 2D focal adhesions.^{124,126} For example, focal adhesion kinase (FAK) in 3D matrix adhesion is poorly phosphorylated at its major tyrosine phosphorylation site for cell adhesion. Such differences in cell adhesion between 2D and 3D matrices lead to different signal transduction and subsequent alteration in cellular rearrangement.

Mechanotransduction. The mechanical forces a scaffold is subjected to during tissue regeneration should also be accounted for during cell colonization. Studies have shown that both hydrodynamic stresses^{44,127} and mechanical stresses¹²⁸ affect cell colonization. For example, endothelial cells⁴⁴ and chondrocytes¹²⁹ grown in a perfusion reactor align cells in the direction of flow. Beyond the structural modifications, shear stress initiates a number of signal transduction cascades leading to altered gene expression profiles and functional changes, particularly in endothelial cells.¹³⁰ *In vitro* studies have shown that shear stress activates mitogen-activated protein (MAP) kinases (including extracellular signal regulated kinase and c-Jun N-terminal kinase),¹³¹⁻¹³³ and kinases involved in focal adhesion such as FAKs, Src family kinases and phosphatidylinositol 3-kinase.¹³⁴ Mechanical forces affect cells in a variety of ways including opening or closing ion channels (changing mass transport properties across the cell membrane) and unfolding selected protein domains (providing access to a different set of binding sites).¹³⁵

Flow through the scaffold microarchitecture dictates the local shear stress rates experienced by the cells. Further the scaffold architecture controls the transport of nutrients within the samples. Channeling and other flow irregularities can result in local hypoxia or extracellular matrix washout.¹³⁵ The presence of flow within a reactor also affects the production of ECM elements, for example rat bone marrow cells produce greater mineralization in scaffolds under direct perfusion.¹³⁶

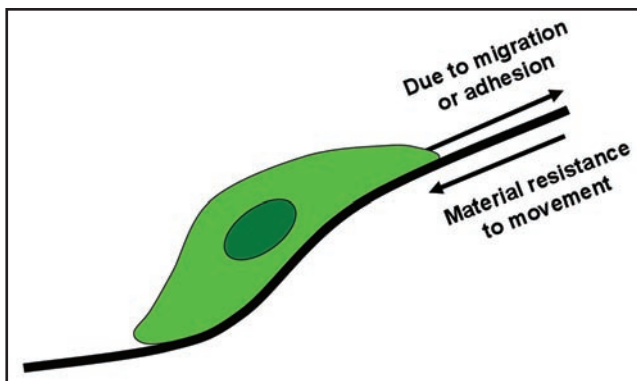


Figure 5. Micromechanical forces acting on a single cell within a porous structure.

Once the scaffold is placed into a flow system (either implanted *in vivo* or grown *in vitro* in a bioreactor) the effect of loading from both external forces and fluid flow can affect cell colonization. While the scaffold itself will be subjected to the bulk forces supplied by the tissue and fluid flow, the cells will experience the micromechanical properties of the individual fibers and local shear stresses within the porous structure (Fig. 5). The cell senses both the porous structure and other cells near where it is attached. An important step in characterizing the porous structure is to examine microenvironment surrounding the cells.

Cellular activity is influenced by the stiffness of the substrate.^{57,137,138} Stiffness is the resistance of the material to deformation, typically reported in force per distance. It is the slope of the load-extension curve. The dimensionality may be removed from the stiffness calculation by using the modulus of elasticity. The modulus of elasticity is the initial slope of the stress-strain curve. However, the thickness of the material must be measured to calculate the modulus of elasticity, and the thickness measurement becomes a major source of error when measuring very small thicknesses. The bulk stiffness controls the overall deformation of the matrix while each individual cell will encounter the stiffness of the individual fibers during colonization. Cells show reduced spreading and disassembly of actin even when soluble adhesive ligands are present in weak gels.^{139,140} This could be via the response of tractional forces between cells and materials; scaffold should be able to withstand cell contractile forces.¹³⁸ Maximum tractional force generated by a cell could be as much as 10–15% of substrate modulus.¹³⁹

In an effort to develop anti-scarring therapies in wound healing, understanding the process of increased collagen packing has been extensively investigated.^{141,142} A variety of cell- or matrix-based continuum modeling has also been attempted.^{143,144} It is very well established that fibroblasts hug the collagen fiber and induce tractional forces. The developed tractional forces lead to the generation of contractile forces which are essential for alignment of collagen fibers and tissue healing. Thus, one of the approaches to minimize scarring is to increase the tensile strength via wound dressing materials. 3D collagen sponges have been used alone^{145,146} or in conjunction with basic fibroblast growth factor,¹⁴⁷ fibronectin or hyaluronic acid.¹⁴⁸ Exogenous collagen increased wound tensile strength and increased degree of reepithelialization *i.e.*, early dermal and epidermal wound healing. Further, hyaluronic acid and

fibronectin may also be involved in faster wound healing via helping the migration of fibroblasts.¹⁴⁹ However, it is not clear whether these treatments reduced scarring in the long term. Nevertheless, the rigidity of the scaffold affect the formation of extracellular matrix which affect cellular activity.¹²³

Matrix Turnover

Cell colonization also involves the deposition of extracellular matrix elements. Cells can synthesize extracellular matrix components in response to different physical and chemical signals from surrounding 3D matrix. Unlike 2D architecture the degradation of a 3D matrix can create more space for cell expansion and migration. Scaffold degradation rate should be synchronized with the cell growth rate to ensure no space restriction due to slow degradation rate or the loss of structural support due to faster degradation. Key factors include the mode of degradation. For example, synthetic polyesters are hydrolytically degraded;⁷⁵ therefore, they will begin to break down if they are not protected from moisture. Conversely, natural polymers, such as chitosan, are enzymatically degraded and can be stored in hydrated condition.³³ Dynamic changes during the degradation process must also be accounted for. Some materials (hydrogels) can swell several times their dry weight.¹⁵⁰ It has also been shown that cellular constructs grown *in vitro* will shrink, possibly as a result of cellular attachment and contraction or as a result of hydrodynamic forces compressing the scaffold. The molecular weight of a polymer will also change over the course of degradation. Amorphous 50:50 PLGA shows an 80% drop in molecular weight over the course of eight weeks which reduces the polymer's tensile break stress by 75%.¹⁵¹ Another positive aspect of degradable polymer systems is that they can also be used for the controlled release of bioactive molecules. Degradable polymers have been used for drug delivery for years. Recently they have been used to deliver growth and differentiation factors within tissue scaffolds.¹⁵²

Matrix turnover significantly influences cellular phenotypic characters which in turn alters assembly of *de novo* synthesized matrix elements. Tissue remodeling in a variety of patho/physiological processes including embryogenesis,¹⁵³ normal tissue development, cancer¹⁵⁴ and wound healing^{155,156} has been evaluated. These studies implicate an array of molecules regulating the process which are regulated at transcriptional, translational and post-translation levels. Matrix metalloproteinases (MMP) form a degradative enzyme family with at least 20 members. MMPs mediate degradation of essentially all components of the ECM.¹⁵⁷ Loss of GAGs in arthritic patients has been attributed to the increased production of stromelysin (MMP-3).¹⁵⁸ Gelatin turnover is mediated either by MMP-2 (Gelatinase A), a constitutively produced homeostatic enzyme, or by MMP-9 (gelatinase B)¹⁵⁹ and upregulated in acute and chronic inflammations. MMP expressions are regulated by soluble mediators, presence of substrates, matrix elements¹⁶⁰ and adhesive interactions.¹⁶¹ In turn, MMPs influence rate of matrix synthesis. For example, cells exposed to hydrogels containing MMP-specific peptides show an increase in the transcriptional activity of collagen and proteoglycan synthesis.¹⁶² In addition, $\alpha_v\beta_3$ can bind to MMP-2,¹⁶¹ in an RGD-independent way, thereby localizing MMP-2 mediated matrix degradation to the endothelial cell surface.¹⁶³

Summary

In summary understanding cell colonization phenomena in 3D porous matrices important in a wide variety of biological applications, including vascularization, wound healing, tissue engineering, stem cell differentiation and biosensors. Colonization is controlled by both the 3D architecture features of the porous structure, include porosity, pore size, fiber orientation, pore interconnectivity, topography and scaffold stiffness and cell—material interactions, such as cellular adhesion, mechanotransduction and matrix turnover. Further understanding of the colonization process should lead to more accurate pathological and physiological tissue models, as well as improved clinical outcomes.

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