Mooney 2003). Depending on the applications, however, not all of the above criteria must be simultaneously fulfilled within a particular hydrogel.

Transport properties

As the survival and function of cells rely largely on the availability of essential nutrients and timely removal of metabolic wastes, the transport property of cell-laden hydrogels is of the utmost importance. Although most cell-laden hydrogels contain large quantity of water, slight variation of polymer crosslinking density affects largely the diffusivity of large molecules (proteins, antibodies, polysaccharides, etc.) (Lin and Anseth 2009). Diffusion is the most common means of molecular transport within a cell-laden hydrogel. Thus, the transport property of a cell-laden hydrogel can be evaluated by the diffusivity of the molecule of interest. In a highly swollen hydrogel with no apparent intermolecular interactions between the molecule and the polymeric network, diffusivity of any molecule is positively correlated to the mesh size of the hydrogel and negatively correlated to the hydrodynamic radius of the molecule (Lin and Metters 2006). To increase molecular diffusivity within a cell-laden hydrogel, one can decrease the crosslinking density of the hydrogel. Such approach, however, leads to weakening of the cell-laden matrix. Hydrogels with independently controllable molecular transport and matrix stiffness should be highly valuable in both fundamental and applied research. These advanced hydrogel materials are often made of heterogeneous components, including composites, double-networks, or materials with hierarchical structures. Under some circumstances, one may wish to retard the transport of certain molecules, such as growth factors, cytokines, or chemokines. Adjusting up the network crosslinking is highly effective in reducing molecular transport. However, hydrogel crosslinking density is often positively proportional to the stiffness of the hydrogel (Anseth et al. 2002). Care must be taken if altering stiffness would lead to unexpected complications in data interpretation. Increasing efforts have been dedicated to developing 'affinity hydrogel' where the rate-limiting step of protein diffusion is governed by the engineered intermolecular interactions, rather than simple diffusion (Pratt et al. 2004; Lin and Anseth 2009; Impellitteri et al. 2012; Belair et al. 2014; Lin et al. 2015).

Depot for growth factors

Native ECM is rich in glycosaminoglycans (GAGs) that interact with biomacromolecules through non-specific electrostatic interactions. As such, ECM serves as a depot for storing and supplying growth factors to the cells. When hydrogels are used for 3D cell culture, the administration of solution growth factor to the encapsulated cells can be achieved simply by media supplement. However, this simple approach may not recapitulate the dynamic and complex growth factor availability *in vivo*. Designing biomimetic hydrogels with highly tunable delivery of soluble cues is not a trivial task, given that most hydrogels are highly swollen with high permeability. Various strategies have been integrated into the crosslinking of cell-laden hydrogels in an attempt to provide sustainable delivery of soluble cues. Early work by Hubbell and Sakiyama