efficiently deliver drugs, cells, or therapeutic agents as well as guide tissue repair or regeneration *in vivo*.

This chapter presents strategies used to molecularly engineer hydrogel materials and enhance their complexity and functionality. We first present opportunities to create hydrogels through either thiol-ene radical coupling or peptide self-assembly and then describe ways to tune them to exhibit bioactivity with both spatial and temporal control.

Thiol-ene Hydrogels

In addition to the requirements for good cytocompatibility, defined and reproducible composition, controlled cell adhesion and mechanical properties and degradability, 3D hydrogels should be easy to engineer and design (e.g., allowing the independant control of mechanical properties and cell adhesion) (Tibbitt et al. 2013; Anseth et al. 2002), be compatible with a range of patterning, printing and cell encapsulation technologies to allow their micro-structuring (Lowe et al. 2014) and should rely on very specific coupling chemistry for the precise control of molecular structure (Porel and Alabi 2014). Several "click" chemistry approaches have been proposed to achieve such goals (Beria et al. 2014; Jiang et al. 2014; Campos et al. 2008), but thiol-ene radical coupling displays important features that are particularly well suited for the design of biomaterials for tissue engineering and regenerative medicine: it does not require metal catalysts, it is fast and relatively insensitive to oxygen, it can be controlled by light (to avoid overheating the cell microenvironment during curing and for patterning and microfabrication) and it is based on relatively simple abundant functionalities (alkene and thiol moieties) yet is very chemoselective.

Thiol-ene radical coupling and structural design

Thiol-ene radical reactions occur between a thiol moiety and an alkene (or alkyne (Fairbanks et al. 2010)) in two stages (Fig. 1a) (Hoyle and Bowman 2010; Hoyle et al. 2004). Unlike Michael additions of thiols requiring the activation of alkenes, thiol-ene radical reactions occur at unactivated alkenes (Cramer and Bowman 2001). This confers to the reaction an improved chemoselectivity as other nucleophiles (i.e., primary amines) cannot react. This is attractive for the coupling of peptides or proteins to hydrogels and other biomaterials as the low abundance of cysteines in most proteins ensures the control of the reaction site without protection of other amino acids. The typical absence of alkene residues in proteins and many biomacromolecules also ensures very little cross-reactivity with other components of the system studied (during the formation of hydrogels in the presence of cells for example).

The parameters affecting the coupling efficiency and its kinetics have been studied, in particular for hydrophobic monomers. The chemical structure of the alkene was found to determine the rates of propagation (thiyl radical reacting with an alkene) and chain transfer (hydrogen abstraction to another thiol molecule) (Cramer et al. 2003b). Infrared spectroscopy allowed the quantification of the rates of reaction of different alkenes (Cramer et al. 2003a; Cramer and Bowman 2001). It was found that terminal