# Supramolecular Hydrogels for Regenerative Medicine

**A.C.H. Pape and Patricia Y.W. Dankers**

## Contents



**Abstract** Regenerative medicine is the science of re-creating or repairing living functional tissue, often inside the body. Biomaterials for regenerative medicine are inspired by the extracellular matrix (ECM), which provides the natural scaffold for cells inside the body. The use of supramolecular hydrogels as man-made tunable replacements for the ECM is being investigated because hydrogels offer an aqueous environment. In addition, supramolecular systems offer modularity and dynamics, also found in the ECM. This chapter gives an overview of translational research on different supramolecular hydrogels, showing systems that have been used in vivo in the field of regenerative medicine. We discuss the chemical structures and biomedical applications of various natural compounds, biosynthetic compounds, biohybrid systems, and fully synthetic materials. Furthermore, we discuss tuning of the mechanical properties and functionalization of these hydrogels with bioactive compounds. Both characteristics are essential for their function in contact with cells and for the creation of a regenerative niche, thereby controlling cellular adherence, proliferation, homing, and differentiation.

A.C.H. Pape • P.Y.W. Dankers (✉)
Laboratory of Chemical Biology, Institute for Complex Molecular Systems, Eindhoven University of Technology, Eindhoven, The Netherlands

Laboratory of Chemical Biology, Department of Biomedical Engineering, Eindhoven University of Technology, Eindhoven, The Netherlands
e-mail: p.y.w.dankers@tue.nl



S. Seiffert (ed.), *Supramolecular Polymer Networks and Gels*, Advances in Polymer Science 268, DOI 10.1007/978-3-319-15404-6_7

**Keywords** Biomedical applications • Regenerative medicine • Supramolecular hydrogels • Translational research

## 1 Introduction

In the field of regenerative medicine, scientists aim to re-create or repair living, functional tissue up to the size of complete organs. Complete organs are one of the greatest promises of regenerative medicine, as these organs could potentially replace donor organs and provide a solution for the shortage of available organs for transplantation [1]. Regenerative medicine and other biomedical applications are naturally only concerned with aqueous systems and, therefore, hydrogels are widely investigated, for example, as scaffolds for tissue engineering and as drug delivery vehicles [2–7]. Although polymeric hydrogels have already proven their application in medicine (e.g., as contact lenses), supramolecular hydrogels are still emerging as biomaterials, mainly because of a lack of design rules [8, 9].

Supramolecular hydrogels consist of supramolecular polymers that are able to form freestanding three-dimensional (3D) networks when swollen in water. Supramolecular polymers consist of monomers or oligomers linked together by noncovalent, well-defined, directional interactions such as hydrogen bonding arrays, steric interactions, metal–ligand complexes, hydrophobic interactions, van der Waals forces, π–π stacking, and other electrostatic effects [10]. Via these supramolecular interactions, both long polymers and physically cross-linked networks can be formed [11]. Supramolecular polymers have found use in many other polymer applications such as adhesives, cosmetics, and coatings [12].

Supramolecular systems offer distinct advantages as biomaterials over chemically cross-linked gels. First, because of their noncovalent nature, supramolecular systems are inherently responsive to stimuli such as variations in temperature and pH. Second, the noncovalent bonding allows a unique mix-and-match principle to be used for tuning properties. Third, supramolecular materials biodegrade faster than chemically cross-linked gels as a result of the small molecular precursors that these materials are made of. Furthermore, these supramolecular systems are proposed to be able to display dynamic reciprocal behavior, as found in the natural environment of cells [13]. Supramolecular biomaterials are proposed to be able to spatiotemporally adapt to changes exerted by cells and their natural environment. To fulfill this promise, important features of supramolecular systems are their hierarchical structure/assembly, their dynamic and nonlinear behavior, and their biochemical properties.

The use of supramolecular hydrogels in regenerative medicine is inspired by the extracellular matrix (ECM), which provides the natural scaffolding for cells inside the body [14]. Mimicking the ECM is a major objective for tissue engineering, in order to create a regenerating niche for cells [15]. The natural ECM is inherently

dynamic, and the structural support is given by macromolecules such as proteoglycans and fibrous proteins; therefore, supramolecular hydrogels might be an ideal replacement [16]. Furthermore, the ECM provides cells with handles for attachment and signaling, and it regulates the transport and presentation of soluble components such as growth factors. These three roles of the ECM – mechanical support, cell signaling, and soluble factor transport/presentation – need to be fulfilled by an ideal ECM mimic.

The mechanical properties of the ECM in natural tissue show a highly variable stiffness, ranging from hundreds of pascals in the mammary gland to megapascals for articular cartilage [17] (Fig. 1). These tissues resist tensile forces using fibrillar proteins such as collagens, whereas hydrated proteins and glycosaminoglycans resist compressive forces [15]. Cells can sense this mechanical microenvironment and respond to the ECM by changing their own mechanical properties via the cytoskeletal network [18]. Therefore, for adherence, proliferation, and differentiation of cells, the mechanical components of designed cell matrices are essential [19–21]. However, hydrogels generally possess inferior mechanical properties, such as mechanical toughness and resistance to friction, compared with natural tissues [5]. Therefore, it is important to improve the mechanical properties of hydrogels and tailor them for their specific application.

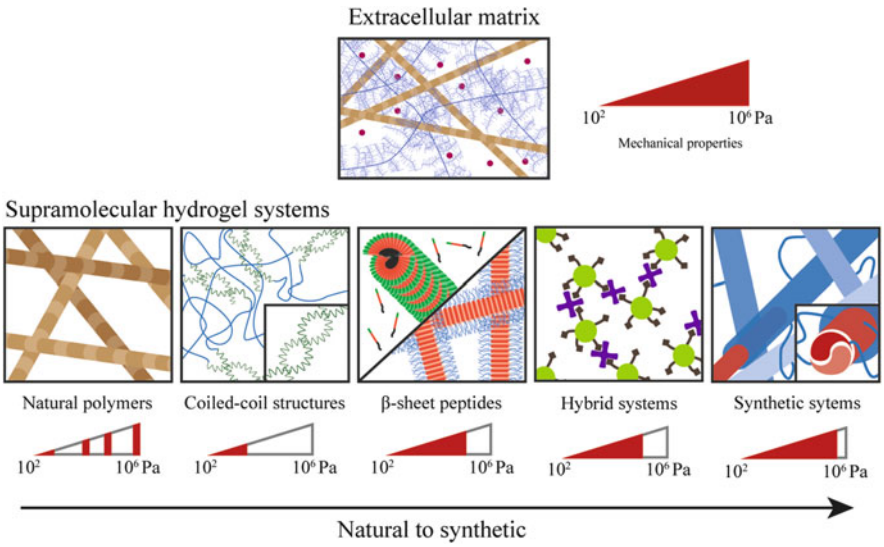


**Fig. 1** Supramolecular systems described in this chapter. *Top*: The extracellular matrix inspires most of the work and consists of fibers to resist tensile stresses such as collagens, fibers to resist compressive stresses such as the glycosaminoglycans, and soluble factors for cell signalling. *Bottom*: Natural systems such as collagen, coiled-coil structures, β-sheet peptides such as peptide amphiphiles and multidomain peptides, hybrid systems such as streptavadin–biotin cross-linked microparticles, and synthetic systems such as the UPy-based hydrogelators

Besides the physical factors, cell–matrix interactions are also regulated by biological factors. Thus, hydrogels should contain bioactive signals to facilitate and enhance these cell–matrix interactions. Furthermore, they also need to fulfill special functions such as the attraction of specific cells and controlled release of bioactives to regulate cell behavior. In the development of supramolecular hydrogels for biomedical applications, it is necessary to take into account the tuning and optimization of the mechanical properties of these hydrogels as well as their functionalization with bioactive components.

Numerous reviews describe supramolecular hydrogels in detail [22–28]; therefore, this article is focused on those supramolecular hydrogel systems designed for applications in regenerative medicine and tested in vivo, as translational research is essential to show the applicability of these materials. We discuss the chemical structures of the systems, their mechanical properties, and the introduction of bioactivity. Additionally, we show examples of various applications in regenerative medicine. Components derived from the ECM have been used extensively as hydrogelators, and we first discuss some of these natural components together with other natural supramolecular polymers that have been used in vivo (Sect. 2) (Fig. 1). Next, we discuss biosynthetic compounds, where man-made peptides are used to mimic the fibrous proteins found in the ECM (Sects. 3 and 4). Then, we discuss hybrid hydrogels, which combine natural structures with synthetic compounds (Sect. 5). In Sect. 6, fully synthetic compounds, inspired by the ECM but showing no resemblance in the chemical structure, are discussed.

## 2 Supramolecular Hydrogels Based on Natural Polymers

The use of natural polymers for the preparation of hydrogels is inspired by the ECM, which consists of many small and large natural polymers such as collagens and polysaccharides. The use of these hydrogelators and other natural polymers for tissue engineering has been reviewed extensively, for example, the use of elastins, glycosaminoglycans, hyaluronic acids, collagens, gelatins, and keratins [24, 29–33]. Often, chemical cross-linking is essential to obtain stable gels. Here, we describe natural polymers that form 3D networks via supramolecular interactions.

Collagen is the main structural component of the interstitial ECM and basement membrane. Different types of collagens exist, but all collagens share the repeated –Gly–Xaa–Yaa– sequence and have a high proline and hydroxyproline content [34]. Three peptides containing this repeating sequence form a triple helical chain that subsequently bundles together with other chains. These bundles align to form long collagen fibrils (Fig. 2a). Self-supporting gels can be formed via end cross-linking of these fibers. They show shear moduli of tens of pascals under physiological conditions, compressive moduli of tens of kilopascals, and tensile moduli of hundreds of kilopascals. This shows the importance of collagens in resisting tensile forces in the ECM [15, 35, 36]. The properties of collagen depend heavily on temperature and pH, and they can be altered by simply changing the pH during

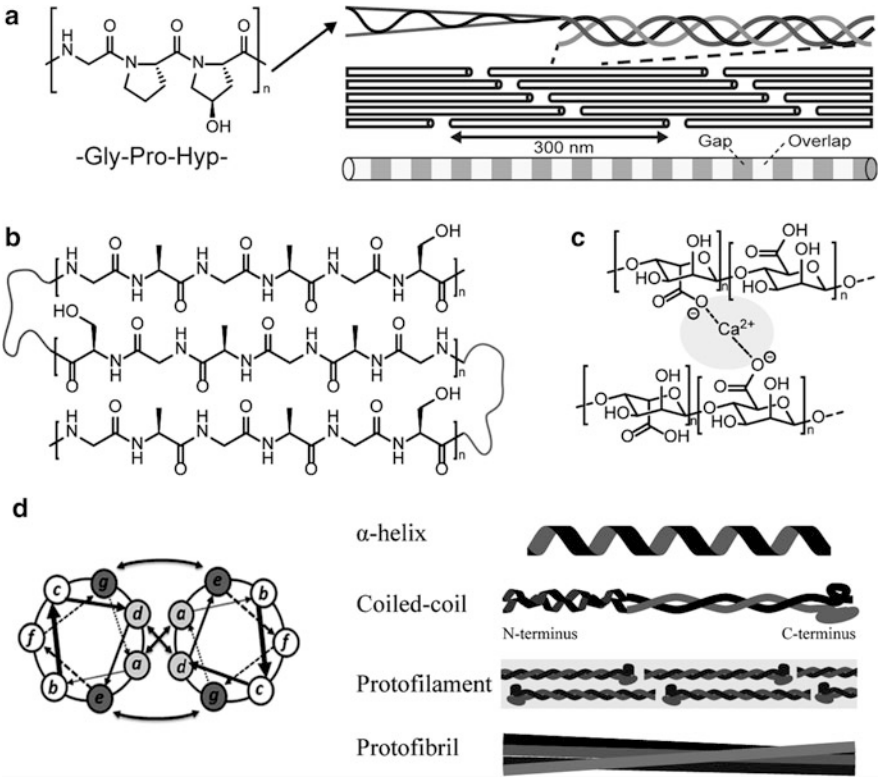


**Fig. 2** Supramolecular natural polymeric hydrogels discussed in this chapter. (**a**) Chemical structure of the most-repeated sequence in collagen, forming the α-chain that folds in a three-stranded superhelix [135]. These superhelices bundle to form the collagen fiber. (**b**) Representative chemical structure of fibroin and the antiparallel β-sheet formation connected by hydrophilic linkers. (**c**) Chemical structure of alginic acid, cross-linked by calcium ions (*highlighted*). (**d**) *Left*: Top view of two α-helixes of keratin forming a coiled coil by hydrophobic interactions. *Right*: Overview of subsequent formation of the fibril. The *left part* is adapted from [57] with permission of The Royal Society of Chemistry

polymerization [37, 38]. By increasing the pH, fibrils of collagen become longer and thinner, leading to increased moduli and failure stresses. However, under physiological conditions, no orders of magnitude difference in moduli can be obtained. Collagens contain cellular binding epitopes, for instance the CNYYSNSYSFWLASLNPER sequence in collagen type IV [39]. Cell binding can also be mediated by mixing glycoproteins with the collagen substrate [40] or by mixing in drugs. Collagenous scaffolds have been used extensively for biomedical applications because of their inherent biocompatibility. They have often been applied as skin grafts or together with other cells and biopolymers as disks, meshes, or foams in tissue such as cartilage and bone [41, 42]. As early as 1990, aliquots of collagenous hydrogels were used as a drug delivery system for sustained delivery of an anticancer drug in the mouse brain [43].

Natural polymers not present inside the body have also been tested for regenerative medicine. Silks are structural proteins, and the fibers from the *Bombyx mori* silkworm are the most-used silks for biomedical applications [44]. Silk mainly consists of fibroin, a protein rich in glycine and alanine that forms β-sheets and becomes insoluble in water, forming cross-links for the gel (Fig. 2b) [45]. Several techniques have been used to prepare silk scaffolds, such as freeze-drying, salt leaching, and gas foaming, which influence the mechanical properties of the silk fibroin gels [46]. Sonication and vortexing have also been used to aid gelation. The properties of silk fibroin hydrogels depend on the processing conditions. Gel formation is irreversible, and compressive moduli are largely determined by the pore sizes in the gel, which can be influenced by silk fibroin concentration, gelation temperature, and calcium ion concentration [47, 48]. In general, compressive moduli of 30–3,000 kPa are obtained. Increasing the concentration or temperature leads to accelerated formation of the physical cross-links. Without the presence of ions, this leads to increasing compressive strength and moduli. Bioactivity can be obtained by mixing in growth factors or other types of bioactive natural polymers, such as collagen, gelatin, and glycosaminoglycans [31]. Several examples of the use of these silk fibroin hydrogels in regenerative medicine exist. Silk fibroin hydrogels aided bone remodeling and maturation in a critical size defect in rabbits, but degraded within 12 weeks [49]. By encapsulation of vascular endothelial growth factor (VEGF) and bone morphogenetic protein 7 (BMP-7) in a sonication-induced silk fibroin hydrogel, bone regeneration was promoted in rabbits [50].

Alginate is a polysaccharide derived from algae that has many carboxylic acid groups and therefore can be cross-linked by calcium ions (Fig. 2c). The mechanical properties of hydrogels formed by alginates are determined by molecular weight when there is a low cross-linking density and by the guluronic acid content; hence, at high cross-linking density the properties depend on the amount of cross-links on the alginate [51]. By varying the polymer and salt concentration, compressive moduli of 5–200 kPa can be obtained. Often, alginate hydrogels are combined with other natural polymers such as collagens to influence cell attachment, or with bioactive signaling proteins such as growth factors to influence cell signaling. In one example, alginate gels were loaded with angiogenic growth factors and placed subcutaneously in the dorsal area of rats [52]. Sequential delivery of three growth factors [VEGF, platelet-derived growth factor, and transforming growth factor beta (TGF-β)] led to enhanced formation of stable and mature blood vessels after 3 months.

The use of human-hair-based keratin has also been explored, but only in a limited number of regenerative-medicine applications [53]. Keratin consists of a family of structural proteins, of which we have already discussed one member, fibroin. In the examples below, we discuss keratin from the human hair, α-keratin, which consists of two α-helices that form a coiled coil as a result of hydrophobic interactions (Fig. 2d). The process of converting hair into a hydrogel consists of oxidizing the keratin and subsequent breaking of the disulfide linkages. After cooling to room temperature, the remaining disulfide and hydrogen bonds form

the gel [30]. Temperature, concentration, and vortexing can be used to tune the shear moduli of the gel between 1 Pa and 1 MPa [54]. About 78 % of human hair keratins contain the cell-binding sequence RGD and, therefore, cells easily attach to keratin [55]. Keratin hydrogels have been used in a mouse tibia nerve injury model, where the hydrogel caused improved axon regeneration and functional recovery [55]. In another study, keratin gels were used to stop bleeding in a lethal liver injury in rabbits [56]. Enhanced 24-h survival rates were shown for rabbits treated with keratin hydrogels, and the keratin gels performed better than commercial hemostatic agents used to stop bleeding.

Natural materials offer many advantages, such as inherent biocompatibility and the natural presence of bioactive sequences. However, problems can arise with batch-to-batch variability, contamination by pathogens (when derived from animal origin, for example, collagen I), and contamination by other non-compatible components. For example, silk contains, in addition to the structural protein fibroin, a glue between the fibers, sericin, which needs to be removed because it can lead to problems with biocompatibility and hypersensitivity [44]. Furthermore, tuning the mechanical properties of natural hydrogels is only possible in a limited range. To be able to adapt materials for different applications with the full range of mechanical properties found in natural tissue, the mechanical tailoring possibilities of synthetic materials can be beneficial.

## 3 Biosynthetic Supramolecular Hydrogels Based on Coiled Coils

Biosynthetic hydrogels are synthetic systems with binding motifs copied from nature and allow exact control over the chemical structure. Therefore, designed supramolecular hydrogelators can be chemically modified to alter the mechanical properties and to include bioactive sequences.

A large body of literature concentrates on the use of coiled coils as cross-linkers for the development of biomaterials [57, 58]. Coiled coils are formed by two interacting α-helices, where, once folded, the subsequent orientation of hydrophobic amino acids leads to the formation of dimers in a superhelical fashion. These coiled coils are some of the most abundant oligomer folding motifs in nature. Tirrell and coworkers developed a large artificial protein consisting of a water-soluble $[(AG)_3PEG]_{10}$ random coil block flanked by two leonine-zipper blocks, all prepared by bacterial expression [59]. These end blocks can be selectively folded into an α-helix, forming coiled coils and subsequent gel networks with shear moduli of 200 Pa (Fig. 3a).

These leucine-zippers require the use of a trigger such as pH or temperature, for example, to encapsulate cells. Therefore, a two-component system was developed using recombinant protein engineering [60]. The first component consists of a small peptide spacer extended with WW domains (with conserved tryptophan residues,

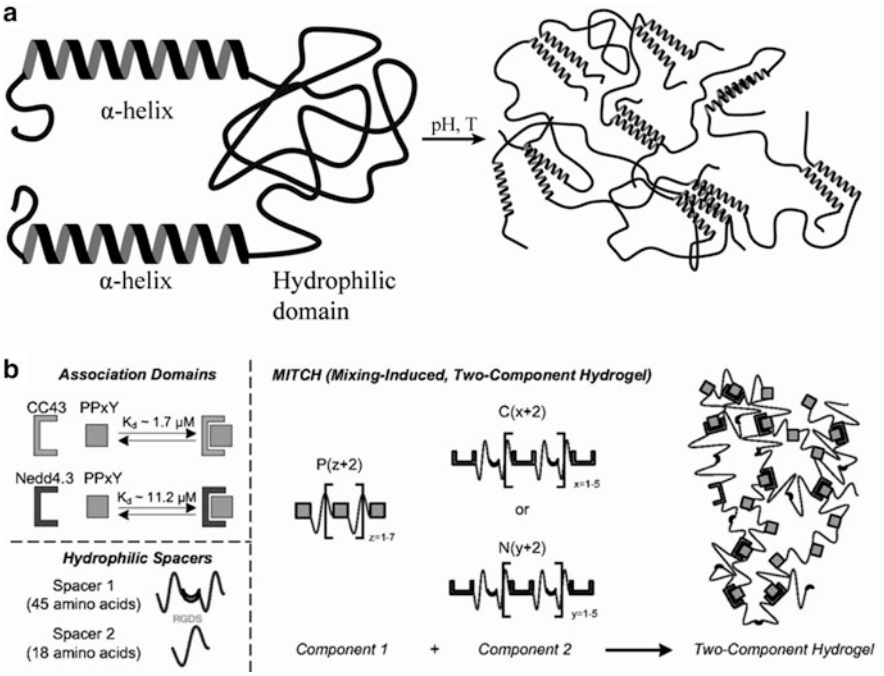


**Fig. 3** Coiled-coil hydrogelators. (**a**) Secondary structure of a coiled coil with soluble linkers (*left*), ultimately leading to a 3D network (*right*). (**b**) Engineered two-component hydrogel, where bioactivity is introduced in the hydrophilic spacer via the RGDS sequence. Adapted with permission from [60]

abbreviated by the single-letter amino acid symbol W), which binds to proline-rich peptides incorporated in the second component (Fig. 3b). Protein engineering gives full control over the sequence and, hence, over binding affinity and subsequent bulk properties. This two-component material forms a weak gel with shear moduli of 10–50 Pa, depending on the type of proline-rich sequence used. The binding affinity of the complex can be increased by an order of magnitude by changing the WW component or by changing the amount of repeating units of both binding domains and, thereby, changing the gel point and modulating the plateau modulus slightly [60]. These two-component gels show shear-thinning and self-healing behavior and are therefore suitable as injectable material. Because the material only forms a gel after mixing, simultaneous addition of cells allows mild incorporation of these cells into the material.

Because these peptides are made via protein engineering, cell-adhesion RGD peptide sequences can be encoded in the spacer between the binding motifs, thereby adding bioactivity to the hydrogels. By incorporation of the RGD-motif, PC-12 cells, human umbilical vein endothelial cells (HUVECs), and neural stem cells can be conveniently cultured in 3D. Implanted subcutaneously in mice, this material

shows improved retention of adipose-derived stem cells and is therefore promising as a cell carrier in stem cell injection therapies [61].

Poly(ethylene glycol) (PEG) has also been end-functionalized with these α-helical coiled-coil structures to form hydrogels with storage moduli of 1 kPa [62]. Additionally, it was shown that the self-assembly can only induce an immunogenic response in mice as a result of formation of oligomeric aggregates. The individual components do not induce a response, showing that the effect of the self-assembly can be different from the effect of its individual components and should be taken into account when studying supramolecular biomaterials [63].

Coiled coils are interesting as designable cross-linkers because of the well-understood relation between the peptide sequence, the folded and dimerized structure, and the binding affinity. However, as described above, hydrogels based on coiled coils are generally weak, with shear moduli only up to 1 kPa. Furthermore, the use of recombinant protein expression or solid-phase peptide synthesis limits the scale on which these materials can be prepared. However, this approach allows tuning both the cross-links as well as the backbone to regulate the mechanical properties and incorporate bioactivity.

## 4 Biosynthetic Supramolecular Hydrogels Based on β-Sheets

Peptide-based hydrogelators are a special class of small molecular building blocks in which short peptide sequences are not only used to induce bioactivity, but also to induce self-assembly into a hydrogel. Several groups have investigated this class of hydrogels for application in tissue engineering, inspired by the self-assembly found in nature and because of the inherent biocompatibility and biodegradability of peptides. Several β-sheet-forming peptides have been developed for use in regenerative medicine and have shown promising potential in vitro; nevertheless, to the best of our knowledge, they have not yet been applied in vivo. For example, Schneider, Pochan, and coworkers developed the MAX8 sequence, an amphiphilic β-hairpin peptide of 20 amino acids of alternating hydrophobic valine and hydrophilic lysine or glutamic acid, folded by prolines in the center, yielding gels with shear moduli of a few hundred pascals [64]. The laboratory of Collier has developed a β-sheet-forming peptide, Ac-QQKFQFQFEQQ-Am (Q11), which slowly assembles into gels via long single fibrils in aqueous environments, resulting in gels with moduli of around 10 Pa [65–67].

Peptide amphiphiles (PAs) are well known to form functional supramolecular materials [68]. PAs consists of (i) a short hydrophobic domain, often an alkyl chain, linked to (ii) an oligopeptide that induces and guides self-assembly by the formation of β-sheets, and (iii) an oligopeptide containing a bioactive domain [69]. In water, these molecules assemble into high-aspect-ratio nanofibers, which entangle and

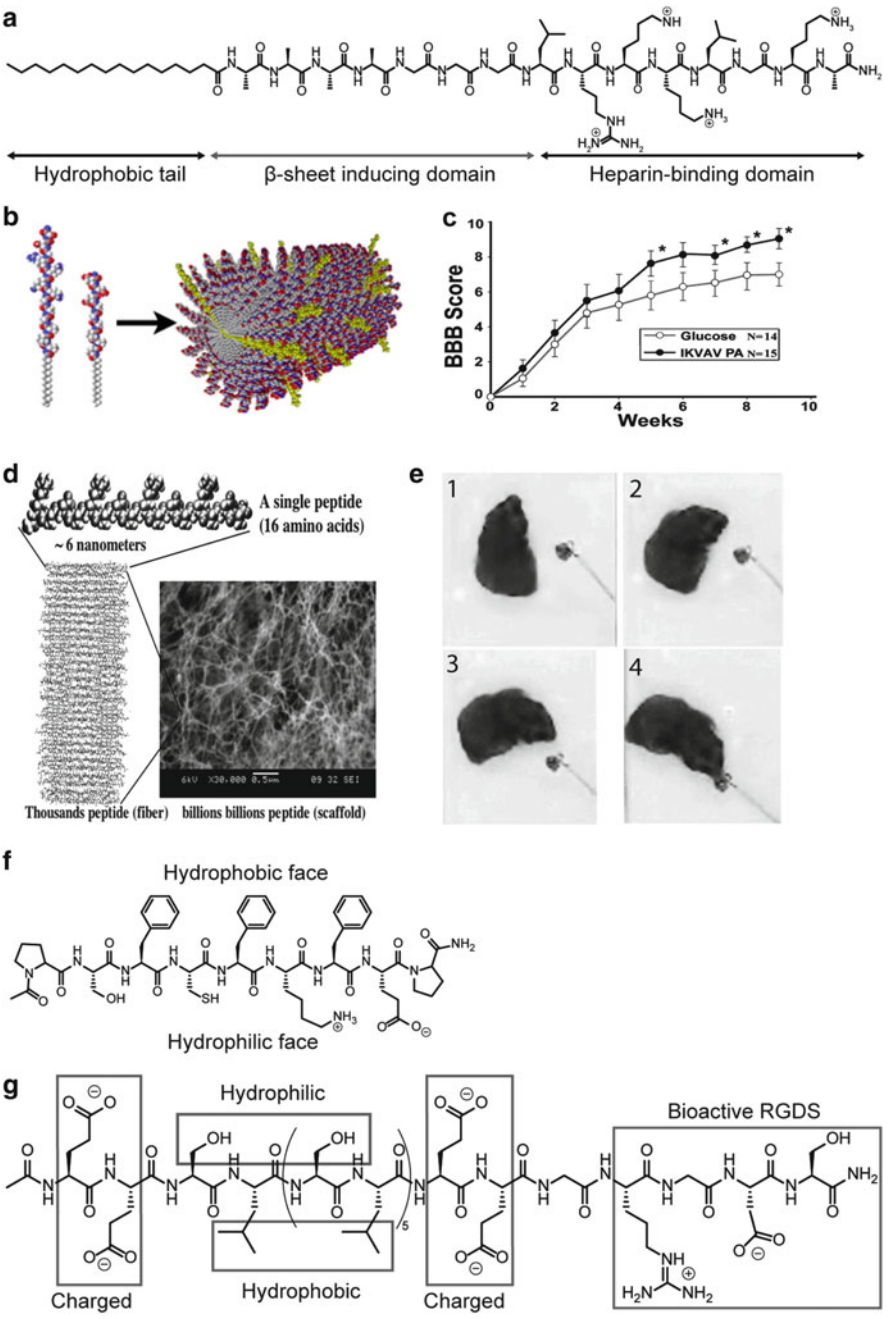


**Fig. 4** Self-assembling hydrogelators based on β-sheets. (**a**) Representative chemical structure of a peptide amphiphile, here without charged residues and with a heparin binding domain. (**b**) Peptide amphiphile with bioactive epitopes (*left*) and its assembly leading to formation of 1D fibers (*right*). Reprinted from [136], Copyright 2010, with permission from Elsevier. (**c**) Graph showing the enhanced functional recovery for animals treated with the peptide amphiphile, as assessed via the BBB score. Adapted with permission from [78]. Copyright 2008 Society for

form self-supporting hydrogels. This fibrillar structure mimics the ECM components, including the signaling peptides at the periphery.

The chemical structure of the PAs guides self-assembly into nanofibers and ultimately determines the mechanical properties of the hydrogel (Fig. 4a). The self-assembly of these amphiphiles is triggered by screening of the charges on the epitope by counter-ions or changes in pH [70]. These counter-ions also induce a change in the molecular geometry, which, together with the van der Waals forces, hydrogen bonding, and hydrophobic forces, induces self-assembly into cylindrical micelles. Within these nanofibers, parts of the peptide form β-sheets, leading to high-aspect-ratio objects (Fig. 4b). Mechanical properties originate from the strength and stiffness of these nanofibers and the density and strength of cross-links. Counter-ions have an effect on the fiber stiffness, through which the moduli can be modulated, leading to gels with shear moduli of 100 Pa to 10 kPa. The incorporation of carboxylic acids at the periphery of the PA allows cross-linking of fibers with divalent cations such as calcium, giving rise to different gelation kinetics and networks that can withstand more strain, although the moduli are not influenced [71].

Gels made from PAs have been used in several regenerative-medicine applications such as the induction of angiogenesis [72], bone regeneration [73], cartilage regeneration [74], enamel regeneration [75], and artery engineering [76]. We discuss three examples here, showing the different ways of introducing bioactivity into these PA systems.

Neural progenitor cells (NPCs) can be encapsulated inside PA gels [77]. By incorporating the laminin-mimicking IKVAV sequence, which promotes neurite sprouting and growth, cells were rapidly differentiated into neurons. Bioactivity was obtained by displaying a peptide-mimicking sequence on the periphery of the nanofibers. Furthermore, PAs in solution gel upon contact with tissue. In vivo experiments using a mouse spinal cord injury model showed promising results in the regeneration of the central nervous system [78]. Mice treated with IKVAV-PA showed enhanced regeneration of both descending motor axons and ascending sensory axons and showed functional recovery (Fig. 4c). This IKVAV-PA gel was used to deliver embryonic stem cells into the auditory nerves of rats [79].

Incorporation of a TGF-binding domain on the epitope of PA leads to capture and display of TGF-β1 in the hydrogels [74]. These materials were used to treat a full thickness chondral defect in a rabbit model. They promote the regeneration of articular cartilage by capturing growth factors from the host animal.

**Fig. 4** (continued) Neuroscience. (**d**) RADA16 peptide, the formation of a β-sheet, and SEM image of fibers in a gel. Adapted from [84]. (**e**) (*1–4*) Functional recovery of the vision of animals treated with a RADA16 hydrogel, as shown by the response to a light trigger. Adapted from [89]. Copyright (2006) National Academy of Sciences, USA. (**f**) Chemical structure of an amphiphilic PSFCFLFEP peptide. (**g**) Chemical structure of an ABA multidomain peptide, here with an additional bioactive sequence attached

As a third example, inclusion of a heparin-binding domain on the periphery of PAs leads to self-assembled nanofibers, where heparin can be displayed on the nanofiber surface (Fig. 4a) [80]. Subsequently, this bound heparin can be used to capture angiogenic growth factors such as VEGF and fibroblast growth factor 2 (FGF-2) [81]. Implanted in the rat cornea, these materials induced significant blood vessel growth compared with the growth factors alone, showing the added value of a structured delivery system.

Another class of peptide hydrogel formed by β-sheets are hydrogels that consist of ionic alternating hydrophobic and hydrophilic residues, such as the materials based on a β-sheet-forming segment found in a yeast protein developed by Zhang and coworkers [82]. This EAK16 (AEAEAKAKAEAEAKAK) peptide forms β-sheets distinct polar and hydrophobic surfaces, enabling further self-assembly into 20-nm fibers, leading to stable hydrogels in water. In these types of systems, the length of the peptide can be used to tune the mechanical properties, as demonstrated by increasing KFE8 to KFE12 with an accompanying change in the shear moduli from 400 Pa to 2 kPa. This effect can be used together with changing the concentration to obtain gels with shear moduli of 400 Pa to 15 kPa [83].

The RAD self-assembling peptide (Fig. 4d) has been used to locally deliver growth factors such as EGF to accelerate wound-healing in an in vitro model [84]. Both the EAK and RAD systems have been further developed to increase cell adhesion [85], and functional motifs such as laminin and collagen-mimicking sequences have been attached and optimized to culture cells in a 3D environment [86, 87]. These peptides have been used for the delivery of genes to cells [88], and they have proven their feasibility in regenerative medicine by enabling reconnection of brain tissue in hamsters [89]. The peptide scaffold was used to knit together tissue after an acute injury and resulted in increased recovery of the central nervous system and recovery of functional behavioral (Fig. 4e).

Ruan et al. designed peptide-forming nanofibers via β-turn secondary folding [90]. The sequence of the peptide consists of the ionic amino acids lysine and glutamic acid on one end and two phenyl groups and a cysteine on the other end, creating an amphiphile (Fig. 4f). This amphiphilic PSFCFLFEP peptide also includes a proline to increase the formation of fibrils and to induce the β-turn. Hydrogels prepared with this peptide showed shear moduli of around 20 Pa, and addition of sodium chloride increased the moduli approximately tenfold [91]. The fibrils were able to gradually release hydrophobic model compounds such as pyrenes. The peptide hydrogel functioned as a hemostat, stopping bleeding of an injured surface on the rat liver faster than other hemostats.

Another example of a hydrogel based on β-sheet-forming peptides consists of a multidomain peptide (MDP) with an ABA structure [92]. The A-block consists of charged residues to keep the aggregates in solution, attached on both sides to a B-block consisting of alternating glutamine and leucine residues to create a facial amphiphile. Subsequent self-assembly of the B-block leads to poorly soluble aggregates in aqueous environments. This allows formation of a hydrogel. Both blocks of these MDPs can be changed to tune the mechanical properties of these gels, giving moduli ranging from 10 to 500 Pa [93]. Chemical cross-linking via

disulfides even leads to gels with moduli of 6 kPa. The gels undergo rapid shear thinning and recovery, and have therefore been used as injectable drug carriers [94]. Bioactive sequences such as the RGD sequence can be incorporated into the material during the solid-phase peptide synthesis of the MDPs (Fig. 4g). Gels were pre-loaded with the secretome from stem cells, which contains more than 36 secreted proteins, and subsequently used to study the effect on a lipopolysaccharide model of acute kidney injury in mice and an ischemia-reperfusion model of kidney injury in mice [95]. The preconditioned nanofibers protect the kidney against injury by delivery of the secretome of stem cells, which is proposed to be the cause of the improvement resulting from stem cell treatment of acute kidney injury.

Many excellent examples of the biomedical application of peptide-based hydrogelators demonstrate their promise in regenerative medicine. The relation between the design of the peptide and the secondary structure is well understood and helps in designing these types of materials. Furthermore, bioactive sequences are attached to these materials in the same way as the self-assembling domain is synthesized. However, the scale on which the solid-phase peptide synthesis used for the preparation of these systems can be performed is limited. The mechanical properties of these materials can be tuned to obtain moduli between 10 Pa and 10 kPa, giving a larger working range than natural hydrogels and hydrogels based on natural motifs. The maximally obtainable moduli are only one order of magnitude lower than those of tissues found in the body.

## 5 Hybrid Supramolecular Hydrogel Systems

Systems have been developed that combine both natural and naturally derived compounds with synthetic structures. Many groups have adapted peptide-based structures or have used natural cross-linkers to develop hydrogels. Inspired by the collagens in the ECM, collagen mimetic peptides (CMPs) have been developed [96]. The development of solid-phase peptide synthesis has enabled synthesis of more stable collagen-like peptides such as (proline–hydroxyproline–glycine)$_{10}$, ($POG_{10}$), which forms stable short helixes. To obtain hydrogels, the focus has been on the self-assembly of these structures into nanofibers and 3D structures. The $POG_{10}$ structure has been modified with three different metal-binding groups: histidine, nitrolotriacetic acid, and a bipyridyl moiety [97]. Subsequently, these structures were cross-linked using nickel ions. However, the mechanical properties of the resulting gels have not been determined. Bioactive sequences can be coupled covalently to promote cell adhesion [98]. By functionalization of growth factors with a His tag, these growth factors can also be supramolecularly complexed to the matrix using nickel ions.

Another interesting approach is the use of tubulin, which by itself forms long cylinders [99]. Forming a stable 3D gel using these cylindrical fibers requires cross-linking of the tubulin fibers with PEG polymers, yielding gels with shear moduli of

1.8 kPa. This cross-linking can only be performed when the tubulins are assembled into fibers to prevent blocking the supramolecular polymerization site of the tubulins. The gel still shows reversible gelation after cross-linking, demonstrating the supramolecular nature of this system. Furthermore, Wieduwild et al. prepared star-shaped PEGs end-functionalized with heparin-binding peptides, creating physical cross-links and subsequent gel formation upon addition of heparin; the obtained materials had shear moduli of around 2 kPa [100].

A different approach involves functionalization of chitosan with α-cyclodextrins and use of PEG as a thread through the α-cyclodextrins to form polypseudorotaxanes [101]. Cells can be encapsulated inside these gels, which showed fast degradation as a result of dethreading of the PEG. However, although these approaches show the promise of hybrid systems, the materials have not yet been tested in vivo.

Yang and coworkers developed a hydrogel based on two amino acid derivatives and bisphosphonate, which self-assemble into hydrogels with shear moduli of 20 Pa to 20 kPa via nanofibers [102]. The amino acid derivative contained the *N*-(fluorenyl-9-methoxycarbonyl) (Fmoc) group, which induces self-assembly via π–π stacking (Fig. 5a). A more systematic study of the use of aromatic–aromatic interactions in the formation of supramolecular peptide hydrogels has been performed, showing the universality of this approach [103]. Incorporation of a terminal carboxylic acid group allows tuning and cross-linking of fibers using different types of salts, and subsequent tuning of the gel moduli with salt and concentration to give moduli between 25 Pa and 75 kPa [104]. The method of preparation of the hydrogels also has an influence on their mechanical properties [21]. Slowly adjusting the pH to form homogenous gels elevates the storage moduli of the gels from 5 to 184 kPa. Simple functionalization of RGD peptides with an aromatic Fmoc group leads to direct cell adhesion to these 3D scaffolds [105]. Besides functioning as a 3D support for the gel, these small molecules can also reduce inflammation [102]: The biphosphonate component of the hydrogel developed by Yang and colleagues lowers into uranyl oxide toxicity. Topical application of the hydrogel to wound sites on skin of mice, previously administered with uranyl oxide, showed effective recovery of the skin to normal, showing the therapeutic effect of these gels.

Yang et al. also showed that glucosamine (a naturally occurring precursor for glycosaminoglycans, proteoglycans, and glycolipids) modified with naphthalene can form a hydrogel (Fig. 5b) [106]. After cooling a solution of the modified glucosamine to room temperature, ribbons are formed. These subsequently form large bundles and tangle to make a 3D hydrogel with a storage modulus of 1 kPa. This hydrogel assisted in wound healing and prevented scar formation, as shown in a mouse animal model.

Gels were also shown to form when biotinylated poly(lactic acid) (PLA)–PEG microparticles were cross-linked using avidin (Fig. 5c) [107]. Gels can be formed that can encapsulate cells, although in rheology the gels do not behave as viscoelastic solids and show a large frequency dependence of their shear moduli. The microparticles can be used as conventional carriers for drugs or signaling

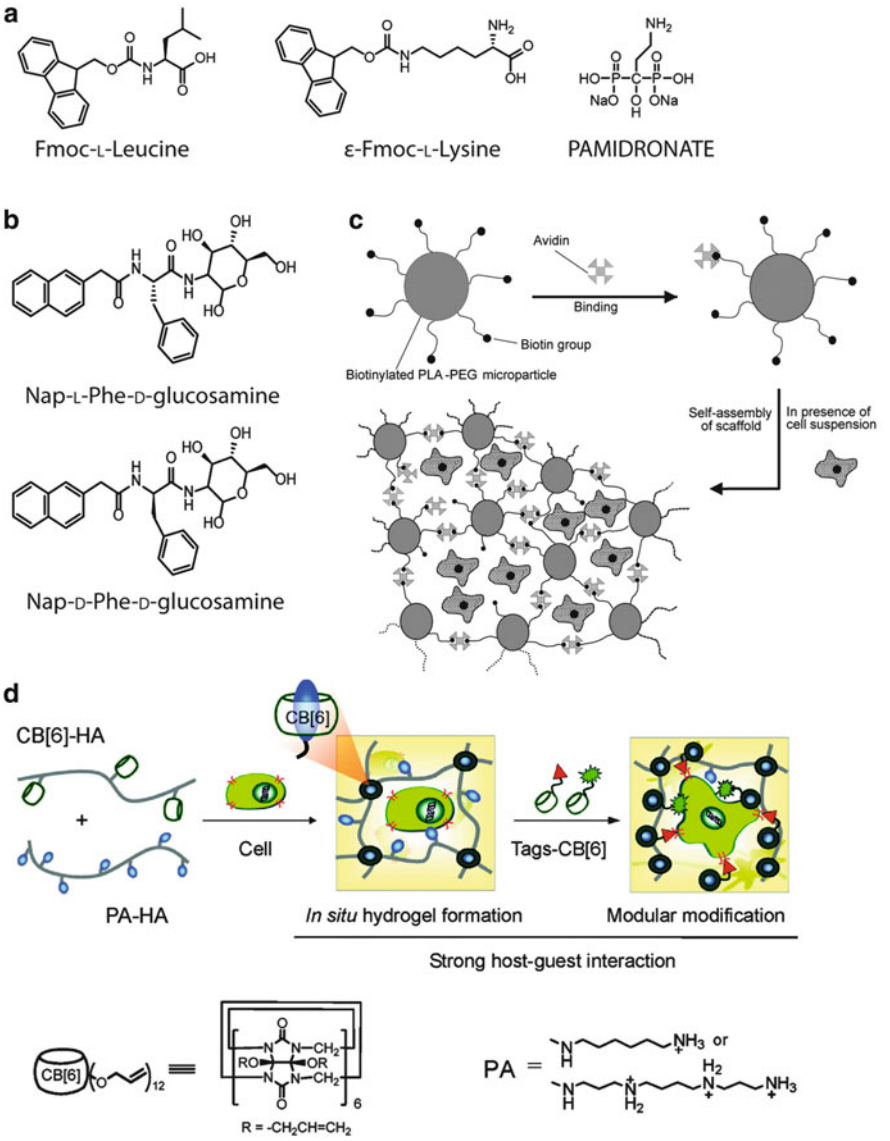


**Fig. 5** Hybrid supramolecular hydrogel systems. (**a**) Chemical structures of the three components of Fmoc-gels that assemble via π–π stacking. (**b**) Chemical structures of hydrophobically modified glucosamines. (**c**) Self-assembly of a hydrogel in the presence of cells [107]. Copyright 2003 Wiley-VCH Verlag GmbH&Co. KGaA, Weinheim. (**d**) Hyaluronic acid cross-linking by host–guest interaction of cucurbituril and amines, and the incorporation of cells and dyes. Reprinted with permission from [108]. Copyright 2012 American Chemical Society

molecules, providing a well-understood sustained release. Bone defect in femurs of chicken embryos were filled with these materials and the viability of the bone could be shown, proving applicability in regenerative medicine.

Hyaluronic acid has been modified with cucurbit[6]uril (CB[6]) and polyamines to form stable hydrogels with shear moduli of around 3 kPa when mixed [108]. The CB[6] can then also be used to include functional molecules in the hydrogel in a supramolecular fashion (as demonstrated with dyes to allow in vivo imaging) without influencing the mechanical properties of the hydrogel (Fig. 5d). Subsequently, RGD was incorporated by conjugation to CB[6] to promote cell adhesion of encapsulated cells in these gels, showing that this gel is a promising candidate for in vivo cell delivery. The gel can be formed under the skin of mice and functionalized in a supramolecular fashion inside the body.

The hybrid materials discussed here are still in their infancy and only initial experiments in regenerative medicine have been performed using them. Furthermore, not much knowledge has been gained yet on the mechanical tuning of these materials, except for their small peptide fragments. So far, the use of aromatic–aromatic interactions with small peptide fragments has shown promising properties for regenerative medicine, but more work is needed to demonstrate the applicability of these materials.

## 6 Synthetic Supramolecular Hydrogels

Besides functionalization of synthetic polymers with peptides or functionalization of peptides with synthetic cross-linkers, fully synthetic supramolecular systems have also been developed for use in regenerative medicine, although they have hardly been tested in vivo. Nature provides good inspiration for new synthetic biomaterials. We envision that many benefits can be gained by using completely synthetic systems, because such systems are infection-free, cheap, fully tunable, and scalable [5]. Before discussing the ureido-pyrimidinone (Upy) hydrogelators developed in our group, we discuss two classes of synthetic systems that are presently being developed for application in regenerative medicine but have not yet been tested in vivo. The first class consists of low molecular weight hydrogelators; the second class consists of hydrogels made from macromolecular monomers that are cross-linked via supramolecular interactions.

Low molecular weight hydrogelators form one-dimensional (1D) stacks and, subsequently, gel networks in water. The behavior of these molecules in organic solvents is well known, and the ability to tune these materials lies in the power of the chemistry to be scalable. Therefore, we consider these materials promising for application in regenerative medicine and give a few examples.

van Esch and coworkers developed hydrogelators based on 1,3,5-triamide cyclohexane with amino acid derivatives and water-soluble chains in the periphery [109]. These molecules interact via hydrophobic interactions and hydrogen bonds to form long 1D stacks, which at low concentration form hydrogels. A similar

C3-symmetric molecule, benzene-tricarboxamide, has been developed that forms a hydrogel via a similar mechanism [110]. Lee and coworkers synthesized coordination polymers by functionalization of pyridine ligands with dendritic tri(ethylene glycol) chains, which lead to helical chains after the addition of silver ions, forming a 3D network [111].

The second class consists of polymers that are cross-linked by different moieties. Cyclodextrin (CD) has been widely studied as a supramolecular cross-linker for hydrogels [112]. The cavity of β-CD can include hydrophobic molecules as guests. For example, adamantane can be grafted onto poly(acrylic acid), which can form a complex with poly(acrylic acid) functionalized with β-CD to yield a supramolecular hydrogel [113]. This versatile approach allows the use of different polymers and different guests. Some polymers that have been cross-linked to form hydrogels using this complex are chitosan [114], poly(acrylamide)s [115], and polyacrylates [116]. Other guests that have been used include cholesterol [117], alkyl chains [118], azobenzenes [119], and ferrocenes [120], to name a few. However, not many systems containing CD have been used in vivo because of toxicity and reduced performance [112].

Another popular supramolecular host used to form supramolecular hydrogels is cucurbituril (CB), as described above. The big CB[8] molecule allows encapsulation of two hydrophobic guests. For instance, naphthol and methyl-viologen attached to PEG can be complexed inside CB[8] to form cross-links via a ternary complex, leading to gels with plateau modulus of around 10–1,000 Pa [121]. Also a short butane linker has been used to connect napthol and methyl-viologen to prepare supramolecular hydrogels [122]. Although the CB[8] and the guest show limited water solubility, encapsulation of the guests leads to enhanced water solubility and, subsequently, to formation of a hydrogel.

The synthetic systems described above have not been tested in vivo and their mechanical properties have not been determined. In our group, bifunctional telechelic hydrogelators were developed based on PEG cross-linked with UPy groups at both ends of the polymer (Fig. 6a) [123]. In dilute solution, stacking of UPy dimers results in the formation of nanofibrous structures, aided by additional urea hydrogen bonds in a hydrophobic pocket formed by long alkyl spacers. Above a critical concentration, bundling causes formation of a transient network (Fig. 6b).

Analysis of the telechelic hydrogel with rheology and small angle X-ray scattering (SAXS) has shown that gel formation coincides with the formation of hydrophobic domains, where the fibers formed by the UPy stacks probably bundle to form cross-links [124]. The storage modulus and the relaxation time depend on concentration, temperature, and pH. By increasing the concentration, higher moduli and slower relaxation times are obtained, whereas increasing the temperature induces a switch from a hydrogel to a viscous liquid. The change from a viscous liquid to a hydrogel by increasing the pH allows the use of this system as an injectable delivery system with shear moduli of 100 Pa to 10 kPa. Interestingly, the addition of small monofunctional oligo(ethylene glycol) chains (modified only on one side with a UPy group) to the bifunctional UPy-hydrogelator gives a stronger gel than that formed from the individual components (Fig. 6c). In this

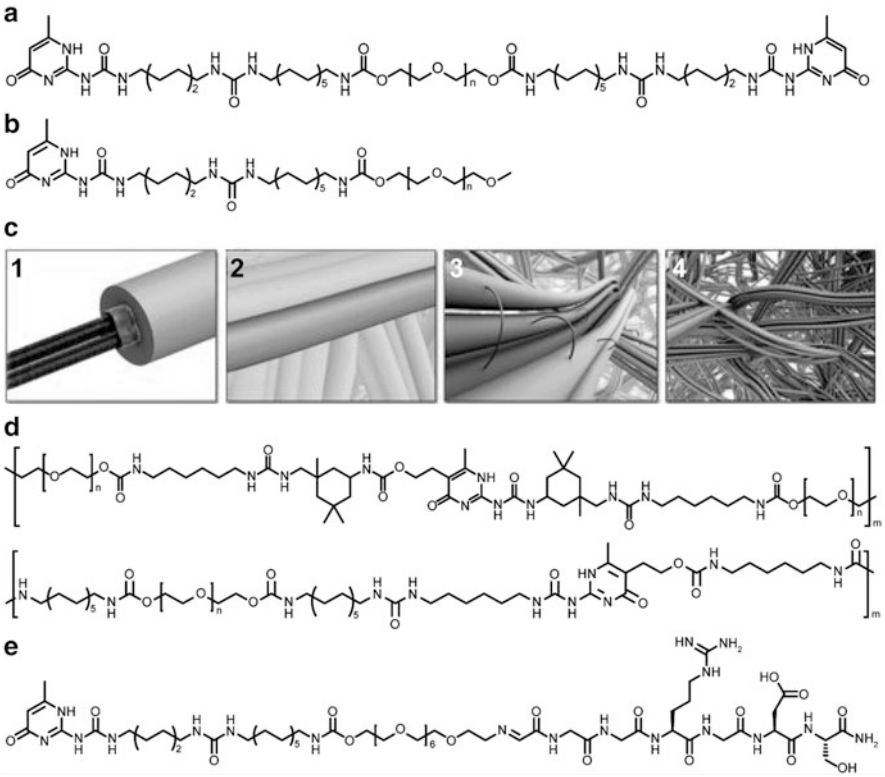


**Fig. 6** Ureido-pyrimidinone (UPy)-based hydrogelators. (**a**) Chemical structure of telechelic UPy-functionalized PEG. (**b**) Chemical structure of monofunctional UPy-functional PEG monomethylether. (**c**) (*1–4*) Formation of fibers and subsequent bundling, leading to a 3D hydrogel [131]. Copyright (c) Wiley-VCH Verlag GmbH & Co. KGaA, Weinheim. (**d**) Chemical structures of chain-extended UPy- modified PEGs [127]. (**e**) Chemical structure of RGD-peptide functionalized with the UPy moiety [128]

mixture, the transient network is vitrified [125]. The synergetic network formed shows a storage modulus of 20 kPa, corresponding to a tenfold increase over that of the bifunctional hydrogelators at the same concentration. After mixing, the gel loses its responsiveness to temperature but is still responsive to pH. By incorporating UPy groups in the main chain of a PEG polymer, the hydrogels can be extended further while retaining the storage moduli (Fig. 6d) [126, 127]. These hydrogels show tensile moduli of 480–920 kPa, reaching almost the modulus of articular cartilage. This opens the opportunity for carefully designed gels by mixing-and-matching of the components for specific applications.

Although not yet applied in hydrogel materials, several UPy-modified peptides have been synthesized, such as UPy–RGD and a UPy–collagen I binding peptide, which in principle can be used for bioactivation of these hydrogels (Fig. 6e) [128]. These UPy–peptide conjugates have already proven their value in activating

solid supramolecular polycaprolactone–UPy-based materials for regenerative medicine [129, 130].

Bifunctional UPy–PEG hydrogels have been applied to deliver the growth factor BMP7 under the kidney capsule in rats, and are able to promote fibrous tissue formation by diminishing the amount of infiltrating inflammatory cells and myofibroblasts. The bifunctional hydrogels have also been used for minimally invasive delivery of growth-factor proteins IGF and HGF in the infarcted myocardium in a porcine model using a long thin catheter. This was possible as a result of the pH responsiveness of the hydrogelators. At basic conditions with a pH of approximately 8.5–9, the UPy-hydrogelator formed a solution, whereas at the site of injection, the hydrogelator solution was neutralized to form a hydrogel drug delivery depot in situ [131]. Delivery of the growth-factor proteins stimulated endogeneous cardiac regeneration, that is, diminished collagen deposition, improved cardiac function, reduced hypertrophy, improved cell proliferation, and promoted formation of more capillaries [131, 132].

Although synthetic supramolecular hydrogelators offer several advantages over biomimetic systems and could be an interesting addition to the field, not many fully synthetic systems have been tested in vivo. This might be partially due to the easy availability of non-supramolecular conventional synthetic hydrogels, which have a proven track-record in medicine. However, as new systems are being developed, cheaper and scalable hydrogelators that more closely mimic the ECM can become available. We have shown that, with supramolecular hydrogels, it is possible to obtain good control over the mechanical properties (both strength and dynamics). Furthermore, bioactivity can be conveniently introduced into supramolecular hydrogels by using a mix-and-match approach.

## 7 Perspective

This chapter demonstrates that supramolecular hydrogels are promising materials for use in regenerative medicine. The examples show how a modular approach allows convenient tuning of mechanical properties and controlled addition of bioactive compounds. The use of the resulting soft dynamic and bioactive materials can provide a more natural-like environment for cells interacting with the biomaterial. Mimicking the native environment, and more specifically the ECM, has inspired a large body of the work performed in this field. However, the ECM is much more complex than the currently used biomaterials. There is not a single supramolecular biomaterial yet that is able to perform in a similar way as the complex set of components in the ECM. For example, the ECM consists of different components that are involved in resisting tensile, compressive, and shear forces, whereas in general only one of these is measured and considered for the hydrogels discussed in this chapter. Furthermore, many biological tissues show strain stiffening, which is essential for their physiological function [133], but nonlinear behavior is observed and studied only in a limited number of synthetic systems, such as

polyisocyanopeptides hydrogels [134]. In addition, although short peptide sequences such as cell-adhesive RGD peptides offer an easy way to incorporate cell-attachment sites in biomaterials, these peptides are not very specific for inducing cell signaling. Fortunately, the use of supramolecular systems allows mixing of different (bioactive) components, both naturally derived and synthetically produced. This can lead to more complex systems offering all the structural and bioactive components required for specific functions. Although many systems are in the developmental phase, more translational research is required to show the true applicability of these systems in vivo.

**Acknowledgments** Our work is funded by the Ministry of Education, Culture and Science (Gravity program 024.001.035), the Netherlands Organisation for Scientific Research (NWO), the European Research Council (FP7/2007–2013) ERC Grant Agreement 308045, and conducted within the LSH TKI framework. The authors thank E.W. Meijer for many useful discussions.

## References

1. NIH (2010) NIH fact sheet – regenerative medicine. National Institutes of Health, Bethesda, http://report.nih.gov/nihfactsheets/Pdfs/RegenerativeMedicine(NIBIB).pdf. Accessed 9 Oct 2014
2. Peppas NA, Hilt JZ, Khademhosseini A, Langer R (2006) Hydrogels in biology and medicine: from molecular principles to bionanotechnology. Adv Mater 18:1345–1360. doi:10.1002/adma.200501612
3. Thiele J, Ma Y, Bruekers SMC, Ma S, Huck WTS (2013) 25th Anniversary article: designer hydrogels for cell cultures: a materials selection guide. Adv Mater 26:125–148. doi:10.1002/adma.201302958
4. Seliktar D (2012) Designing cell-compatible hydrogels for biomedical applications. Science 336:1124–1128. doi:10.1126/science.1214804
5. Gong J, Osada Y (2010) Soft and wet materials: from hydrogels to biotissues. High Solid Dispers 236:203–246
6. Skardal A (2014) Extracellular matrix-like hydrogels for applications in regenerative medicine. In: Connon CJ, Hamley IW (eds) Hydrogels in cell-based therapies. Royal Society of Chemistry, Cambridge, doi: 10.1039/9781782622055-00191
7. Ulijn RV (2006) Enzyme-responsive materials: a new class of smart biomaterials. J Mater Chem 16:2217. doi:10.1039/b601776m
8. Abul-Haija YM, Ulijn RV (2014) Enzyme-responsive hydrogels for biomedical applications. In: Connon CJ, Hamley IW (eds) Hydrogels in cell-based therapies. Royal Society of Chemistry, Cambridge, doi: 10.1039/9781782622055-00112
9. Hirst AR, Escuder B, Miravet JF, Smith DK (2008) High-tech applications of self-assembling supramolecular nanostructured gel-phase materials: from regenerative medicine to electronic devices. Angew Chem Int Ed Engl 47:8002–8018. doi:10.1002/anie.200800022
10. Jones RG, Ober CK (2012) Terminology for aggregation and self-assembly in polymer science (IUPAC Recommendations 2013). Pure Appl Chem 85:463. doi:10.1351/PAC-REC-12-03-12
11. Yui N (ed) (2002) Supramolecular design for biological applications. CRC, Boca Raton
12. Bosman AW, Sijbesma RP, Meijer EW (2004) Supramolecular polymers at work. Mater Today 7:34–39. doi:10.1016/S1369-7021(04)00187-7

13. Xu R, Boudreau A, Bissell M (2009) Tissue architecture and function: dynamic reciprocity via extra- and intra-cellular matrices. Cancer Metastasis Rev 28:167–176. doi:10.1007/s10555-008-9178-z
14. Frantz C, Stewart KM, Weaver VM (2010) The extracellular matrix at a glance. J Cell Sci 123:4195–4200
15. Collier JH, Rudra JS, Gasiorowski JZ, Jung JP (2010) Multi-component extracellular matrices based on peptide self-assembly. Chem Soc Rev 39:3413–3424. doi:10.1039/B914337H
16. Rehmann MS, Kloxin AM (2013) Tunable and dynamic soft materials for three-dimensional cell culture. Soft Matter 9:6737–6746. doi:10.1039/C3SM50217A
17. Levental I, Georges PC, Janmey PA (2007) Soft biological materials and their impact on cell function. Soft Matter 3:299–306. doi:10.1039/B610522J
18. Guilak F, Cohen DM, Estes BT, Gimble JM, Liedtke W, Chen CS (2009) Control of stem cell fate by physical interactions with the extracellular matrix. Cell Stem Cell 5:17–26. doi:10.1016/j.stem.2009.06.016
19. Trappmann B, Gautrot JE, Connelly JT, Strange DGT, Li Y, Oyen ML, Cohen Stuart MA, Boehm H, Li B, Vogel V, Spatz JP, Watt FM, Huck WTS (2012) Extracellular-matrix tethering regulates stem-cell fate. Nat Mater 11:642–649. doi:10.1038/nmat3339
20. Choquet D, Felsenfeld DP, Sheetz MP (1997) Extracellular matrix rigidity causes strengthening of integrin-cytoskeleton linkages. Cell 88:39–48. doi:10.1016/S0092-8674(00)81856-5
21. Wang H, Yang Z, Adams DJ (2012) Controlling peptide-based hydrogelation. Mater Today 15:500–507. doi:10.1016/S1369-7021(12)70219-5
22. Tsitsilianis C (2010) Responsive reversible hydrogels from associative "smart" macromolecules. Soft Matter 6:2372. doi:10.1039/b923947b
23. Kopecek J (2007) Hydrogel biomaterials: a smart future? Biomaterials 28:5185–5192. doi:10.1016/j.biomaterials.2007.07.044
24. Jonker AM, Löwik DWPM, van Hest JCM (2012) Peptide- and protein-based hydrogels. Chem Mater 24:759–773. doi:10.1021/cm202640w
25. Kopeček J, Yang J (2012) Smart self-assembled hybrid hydrogel biomaterials. Angew Chem Int Ed Engl 51:7396–7417. doi:10.1002/anie.201201040
26. Seiffert S, Sprakel J (2012) Physical chemistry of supramolecular polymer networks. Chem Soc Rev 41:909–930. doi:10.1039/C1CS15191F
27. Krieg E, Rybtchinski B (2011) Noncovalent water-based materials: robust yet adaptive. Chem A Eur J 17:9016–9026. doi:10.1002/chem.201100809
28. Weiss R, Terech P (eds) (2006) Molecular gels: materials with self-assembled fibrillar networks. Springer, Dordrecht
29. Whittaker J, Balu R, Choudhury NR, Dutta NK (2014) Biomimetic protein-based elastomeric hydrogels for biomedical applications. Polym Int 63:1545–1557. doi:10.1002/pi.4670
30. Silva R, Fabry B, Boccaccini AR (2014) Fibrous protein-based hydrogels for cell encapsulation. Biomaterials 35:6727–6738. doi:10.1016/j.biomaterials.2014.04.078
31. Van Vlierberghe S, Dubruel P, Schacht E (2011) Biopolymer-based hydrogels as scaffolds for tissue engineering applications: a review. Biomacromolecules 12:1387–1408. doi:10.1021/bm200083n
32. Patterson J, Martino MM, Hubbell JA (2010) Biomimetic materials in tissue engineering. Mater Today 13:14–22. doi:10.1016/S1369-7021(10)70013-4
33. Kim TG, Shin H, Lim DW (2012) Biomimetic scaffolds for tissue engineering. Adv Funct Mater 22:2446–2468. doi:10.1002/adfm.201103083
34. Engel J, Bächinger H (2005) Structure, stability and folding of the collagen triple helix. In: Brinckmann J, Notbohm H, Müller PK (eds) Collagen SE-2. Springer, Berlin, pp 7–33
35. Yang Y, Leone LM, Kaufman LJ (2014) Elastic moduli of collagen gels can be predicted from two-dimensional confocal microscopy. Biophys J 97:2051–2060. doi:10.1016/j.bpj.2009.07.035

36. Achilli M, Mantovani D (2010) Tailoring mechanical properties of collagen-based scaffolds for vascular tissue engineering: the effects of pH, temperature and ionic strength on gelation. Polymers (Basel) 2:664–680. doi:10.3390/polym2040664
37. Yamamura N, Sudo R, Ikeda M, Tanishita K (2007) Effects of the mechanical properties of collagen gel on the in vitro formation of microvessel networks by endothelial cells. Tissue Eng 13:1443–1453. doi:10.1089/ten.2006.0333
38. Roeder BA, Kokini K, Sturgis JE, Robinson JP, Voytik-Harbin SL (2002) Tensile mechanical properties of three-dimensional type I collagen extracellular matrices with varied microstructure. J Biomech Eng 124:214. doi:10.1115/1.1449904
39. Han J, Ohno N, Pasco S, Monboisse J-C, Borel JP, Kefalides NA (1997) A cell binding domain from the α3 chain of type IV collagen inhibits proliferation of melanoma cells. J Biol Chem 272:20395–20401. doi:10.1074/jbc.272.33.20395
40. Kleinman HK, Murray JC, McGoodwin EB, Martin GR (1978) Connective tissue structure: cell binding to collagen. J Invest Dermatol 71:9–11
41. Lee CH, Singla A, Lee Y (2001) Biomedical applications of collagen. Int J Pharm 221:1–22. doi:10.1016/S0378-5173(01)00691-3
42. Place ES, Evans ND, Stevens MM (2009) Complexity in biomaterials for tissue engineering. Nat Mater 8:457–470
43. Sutton R, Yu N, Luck E, Brown D, Conley F (1990) Reduction of vinblastine neurotoxicity in mice utilizing a collagen matrix carrier. Sel Cancer Ther 6:35–49. doi:10.1089/sct.1990.6.35
44. Altman GH, Diaz F, Jakuba C, Calabro T, Horan RL, Chen J, Lu H, Richmond J, Kaplan DL (2003) Silk-based biomaterials. Biomaterials 24:401–416. doi:10.1016/S0142-9612(02)00353-8
45. Jin H-J, Kaplan DL (2003) Mechanism of silk processing in insects and spiders. Nature 424:1057–1061
46. Nazarov R, Jin H-J, Kaplan DL (2004) Porous 3-D scaffolds from regenerated silk fibroin. Biomacromolecules 5:718–726. doi:10.1021/bm034327e
47. Kim U-J, Park J, Kim HJ, Wada M, Kaplan DL (2005) Three-dimensional aqueous-derived biomaterial scaffolds from silk fibroin. Biomaterials 26:2775–2785. doi:10.1016/j.biomaterials.2004.07.044
48. Kim U-J, Park J, Li C, Jin H-J, Valluzzi R, Kaplan DL (2004) Structure and properties of silk hydrogels. Biomacromolecules 5:786–792. doi:10.1021/bm0345460
49. Fini M, Motta A, Torricelli P, Giavaresi G, Nicoli Aldini N, Tschon M, Giardino R, Migliaresi C (2005) The healing of confined critical size cancellous defects in the presence of silk fibroin hydrogel. Biomaterials 26:3527–3536. doi:10.1016/j.biomaterials.2004.09.040
50. Zhang W, Wang X, Wang S, Zhao J, Xu L, Zhu C, Zeng D, Chen J, Zhang Z, Kaplan DL, Jiang X (2011) The use of injectable sonication-induced silk hydrogel for VEGF(165) and BMP-2 delivery for elevation of the maxillary sinus floor. Biomaterials 32:9415–9424. doi:10.1016/j.biomaterials.2011.08.047
51. Kuo CK, Ma PX (2001) Ionically crosslinked alginate hydrogels as scaffolds for tissue engineering: Part 1. Structure, gelation rate and mechanical properties. Biomaterials 22:511–521. doi:10.1016/S0142-9612(00)00201-5
52. Freeman I, Cohen S (2009) The influence of the sequential delivery of angiogenic factors from affinity-binding alginate scaffolds on vascularization. Biomaterials 30:2122–2131. doi:10.1016/j.biomaterials.2008.12.057
53. Rouse JG, Van Dyke ME (2010) A review of keratin-based biomaterials for biomedical applications. Materials (Basel) 3:999–1014. doi:10.3390/ma3020999
54. Yucel T, Cebe P, Kaplan DL (2009) Vortex-induced injectable silk fibroin hydrogels. Biophys J 97:2044–2050
55. Sierpinski P, Garrett J, Ma J, Apel P, Klorig D, Smith T, Koman LA, Atala A, Van Dyke M (2008) The use of keratin biomaterials derived from human hair for the promotion of rapid regeneration of peripheral nerves. Biomaterials 29:118–128. doi:10.1016/j.biomaterials.2007.08.023

56. Aboushwareb T, Eberli D, Ward C, Broda C, Holcomb J, Atala A, Van Dyke M (2009) A keratin biomaterial gel hemostat derived from human hair: evaluation in a rabbit model of lethal liver injury. J Biomed Mater Res B Appl Biomater 90:45–54. doi:10.1002/jbm.b.31251
57. Apostolovic B, Danial M, Klok H-A (2010) Coiled coils: attractive protein folding motifs for the fabrication of self-assembled, responsive and bioactive materials. Chem Soc Rev 39:3541–3575. doi:10.1039/b914339b
58. Wang C, Stewart RJ, Kopecek J (1999) Hybrid hydrogels assembled from synthetic polymers and coiled-coil protein domains. Nature 397:417–420. doi:10.1038/17092
59. Petka W, Harden J, McGrath K, Wirtz D, Tirrell D (1998) Reversible hydrogels from self-assembling artificial proteins. Science 281:389–392. doi:10.1126/science.281.5375.389
60. Wong Po Foo CTS, Lee JS, Mulyasasmita W, Parisi-Amon A, Heilshorn SC (2009) Two-component protein-engineered physical hydrogels for cell encapsulation. Proc Natl Acad Sci USA 106:22067–22072. doi:10.1073/pnas.0904851106
61. Parisi-Amon A, Mulyasasmita W, Chung C, Heilshorn SC (2013) Protein-engineered injectable hydrogel to improve retention of transplanted adipose-derived stem cells. Adv Healthcare Mater 2:428–432. doi:10.1002/adhm.201200293
62. Jing P, Rudra JS, Herr AB, Collier JH (2008) Self-assembling peptide-polymer hydrogels designed from the coiled coil region of fibrin. Biomacromolecules 9:2438–2446. doi:10.1021/bm800459v
63. Rudra JS, Tripathi PK, Hildeman DA, Jung JP, Collier JH (2010) Immune responses to coiled coil supramolecular biomaterials. Biomaterials 31:8475–8483. doi:10.1016/j.biomaterials.2010.07.068
64. Haines-Butterick L, Rajagopal K, Branco M, Salick D, Rughani R, Pilarz M, Lamm MS, Pochan DJ, Schneider JP (2007) Controlling hydrogelation kinetics by peptide design for three-dimensional encapsulation and injectable delivery of cells. Proc Natl Acad Sci USA 104:7791–7796. doi:10.1073/pnas.0701980104
65. Collier JH, Messersmith PB (2003) Enzymatic modification of self-assembled peptide structures with tissue transglutaminase. Bioconjug Chem 14:748–755. doi:10.1021/bc034017t
66. Rudra JS, Tian YF, Jung JP, Collier JH (2010) A self-assembling peptide acting as an immune adjuvant. Proc Natl Acad Sci USA 107:622–627. doi:10.1073/pnas.0912124107
67. Jung JP, Nagaraj AK, Fox EK, Rudra JS, Devgun JM, Collier JH (2009) Co-assembling peptides as defined matrices for endothelial cells. Biomaterials 30:2400–2410. doi:10.1016/j.biomaterials.2009.01.033
68. Aida T, Meijer E, Stupp S (2012) Functional supramolecular polymers. Science 335:813–817. doi:10.1126/science.1205962
69. Webber MJ, Kessler JA, Stupp SI (2010) Emerging peptide nanomedicine to regenerate tissues and organs. J Intern Med 267:71–88. doi:10.1111/j.1365-2796.2009.02184.x
70. Stendahl JC, Rao MS, Guler MO, Stupp SI (2006) Intermolecular forces in the self-assembly of peptide amphiphile nanofibers. Adv Funct Mater 16:499–508. doi:10.1002/adfm.200500161
71. Greenfield MA, Hoffman JR, Olvera de la Cruz M, Stupp SI (2009) Tunable mechanics of peptide nanofiber gels. Langmuir 26:3641–3647. doi:10.1021/la9030969
72. Webber MJ, Tongers J, Newcomb CJ, Marquardt K-T, Bauersachs J, Losordo DW, Stupp SI (2011) Supramolecular nanostructures that mimic VEGF as a strategy for ischemic tissue repair. Proc Natl Acad Sci USA 108:13438–13443. doi:10.1073/pnas.1016546108
73. Mata A, Geng Y, Henrikson KJ, Aparicio C, Stock SR, Satcher RL, Stupp SI (2010) Bone regeneration mediated by biomimetic mineralization of a nanofiber matrix. Biomaterials 31:6004–6012. doi:10.1016/j.biomaterials.2010.04.013
74. Shah RN, Shah NA, Del Rosario Lim MM, Hsieh C, Nuber G, Stupp SI (2010) Supramolecular design of self-assembling nanofibers for cartilage regeneration. Proc Natl Acad Sci USA 107:3293–3298. doi:10.1073/pnas.0906501107

75. Huang Z, Newcomb CJ, Zhou Y, Lei YP, Bringas P Jr, Stupp SI, Snead ML (2013) The role of bioactive nanofibers in enamel regeneration mediated through integrin signals acting upon C/EBPα and c-Jun. Biomaterials 34:3303–3314. doi:10.1016/j.biomaterials.2013.01.054
76. McClendon MT, Stupp SI (2012) Tubular hydrogels of circumferentially aligned nanofibers to encapsulate and orient vascular cells. Biomaterials 33:5713–5722. doi:10.1016/j.biomaterials.2012.04.040
77. Silva GA, Czeisler C, Niece KL, Beniash E, Harrington DA, Kessler H, Stupp SI (2004) Selective differentiation of neural progenitor cells by high-epitope density nanofibers. Science 303:1352–1355. doi:10.1126/science.1093783
78. Tysseling-Mattiace VM, Sahni V, Niece KL, Birch D, Czeisler C, Fehlings MG, Stupp SI, Kessler JA (2008) Self-assembling nanofibers inhibit glial scar formation and promote axon elongation after spinal cord injury. J Neurosci 28:3814–3823. doi:10.1523/JNEUROSCI.0143-08.2008
79. Palmgren B, Jiao Y, Novozhilova E, Stupp SI, Olivius P (2012) Survival, migration and differentiation of mouse tau-GFP embryonic stem cells transplanted into the rat auditory nerve. Exp Neurol 235:599–609. doi:10.1016/j.expneurol.2012.03.014
80. Behanna HA, Rajangam K, Stupp SI (2007) Modulation of fluorescence through coassembly of molecules in organic nanostructures. J Am Chem Soc 129:321–327. doi:10.1021/ja062415b
81. Rajangam K, Behanna HA, Hui MJ, Han X, Hulvat JF, Lomasney JW, Stupp SI (2006) Heparin binding nanostructures to promote growth of blood vessels. Nano Lett 6:2086–2090. doi:10.1021/nl0613555
82. Zhang S, Holmes T, Lockshin C, Rich A (1993) Spontaneous assembly of a self-complementary oligopeptide to form a stable macroscopic membrane. Proc Natl Acad Sci USA 90:3334–3338. doi:10.1073/pnas.90.8.3334
83. Caplan MR, Schwartzfarb EM, Zhang S, Kamm RD, Lauffenburger DA (2002) Effects of systematic variation of amino acid sequence on the mechanical properties of a self-assembling, oligopeptide biomaterial. J Biomater Sci Polym Ed 13:225–236. doi:10.1163/156856202320176493
84. Schneider A, Garlick JA, Egles C (2008) Self-assembling peptide nanofiber scaffolds accelerate wound healing. PLoS One 3:e1410. doi:10.1371/journal.pone.0001410
85. Zhang S, Holmes TC, DiPersio CM, Hynes RO, Su X, Rich A (1995) Self-complementary oligopeptide matrices support mammalian cell attachment. Biomaterials 16:1385–1393. doi:10.1016/0142-9612(95)96874-Y
86. Gelain F, Bottai D, Vescovi A, Zhang S (2006) Designer self-assembling peptide nanofiber scaffolds for adult mouse neural stem cell 3-dimensional cultures. PLoS One 1:e119. doi:10.1371/journal.pone.0000119
87. Genové E, Shen C, Zhang S, Semino CE (2005) The effect of functionalized self-assembling peptide scaffolds on human aortic endothelial cell function. Biomaterials 26:3341–3351. doi:10.1016/j.biomaterials.2004.08.012
88. Zhao X, Pan F, Xu H, Yaseen M, Shan H, Hauser CAE, Zhang S, Lu JR (2010) Molecular self-assembly and applications of designer peptide amphiphiles. Chem Soc Rev 39:3480–3498. doi:10.1039/b915923c
89. Ellis-Behnke RG, Liang Y-X, You S-W, Tay DKC, Zhang S, So K-F, Schneider GE (2006) Nano neuro knitting: peptide nanofiber scaffold for brain repair and axon regeneration with functional return of vision. Proc Natl Acad Sci USA 103:5054–5059. doi:10.1073/pnas.0600559103
90. Ruan L, Zhang H, Luo H, Liu J, Tang F, Shi Y-K, Zhao X (2009) Designed amphiphilic peptide forms stable nanoweb, slowly releases encapsulated hydrophobic drug, and accelerates animal hemostasis. Proc Natl Acad Sci USA 106:5105–5110. doi:10.1073/pnas.0900026106
91. Ruan L-P, Luo H-L, Zhang H-Y, Zhao X (2009) Investigation on structure and properties of a novel designed peptide. Macromol Res 17:597–602. doi:10.1007/BF03218915

92. Dong H, Paramonov SE, Aulisa L, Bakota EL, Hartgerink JD (2007) Self-assembly of multidomain peptides: balancing molecular frustration controls conformation and nanostructure. J Am Chem Soc 129:12468–12472. doi:10.1021/ja072536r
93. Aulisa L, Dong H, Hartgerink JD (2009) Self-assembly of multidomain peptides: sequence variation allows control over cross-linking and viscoelasticity. Biomacromolecules 10:2694–2698. doi:10.1021/bm900634x
94. Bakota EL, Wang Y, Danesh FR, Hartgerink JD (2011) Injectable multidomain peptide nanofiber hydrogel as a delivery agent for stem cell secretome. Biomacromolecules 12:1651–1657. doi:10.1021/bm200035r
95. Wang Y, Bakota E, Chang BHJ, Entman M, Hartgerink JD, Danesh FR (2011) Peptide nanofibers preconditioned with stem cell secretome are renoprotective. J Am Soc Nephrol 22:704–717. doi:10.1681/ASN.2010040403
96. He L, Theato P (2013) Collagen and collagen mimetic peptide conjugates in polymer science. Eur Polym J 49:2986–2997. doi:10.1016/j.eurpolymj.2013.05.033
97. Pires MM, Przybyla DE, Chmielewski J (2009) A metal-collagen peptide framework for three-dimensional cell culture. Angew Chem Int Ed Engl 48:7813–7817. doi:10.1002/anie.200902375
98. Hernandez-Gordillo V, Chmielewski J (2014) Mimicking the extracellular matrix with functionalized, metal-assembled collagen peptide scaffolds. Biomaterials 35:7363–7373. doi:10.1016/j.biomaterials.2014.05.019
99. Sano K-I, Kawamura R, Tominaga T, Nakagawa H, Oda N, Ijiro K, Osada Y (2011) Thermoresponsive microtubule hydrogel with high hierarchical structure. Biomacromolecules 12:1409–1413. doi:10.1021/bm101578x
100. Wieduwild R, Tsurkan M, Chwalek K, Murawala P, Nowak M, Freudenberg U, Neinhuis C, Werner C, Zhang Y (2013) Minimal peptide motif for non-covalent peptide–heparin hydrogels. J Am Chem Soc 135:2919–2922. doi:10.1021/ja312022u
101. Tran NQ, Joung YK, Lih E, Park KM, Park KD (2010) Supramolecular hydrogels exhibiting fast in situ gel forming and adjustable degradation properties. Biomacromolecules 11:617–625. doi:10.1021/bm100047y
102. Yang Z, Xu K, Wang L, Gu H, Wei H, Zhang M, Xu B (2005) Self-assembly of small molecules affords multifunctional supramolecular hydrogels for topically treating simulated uranium wounds. Chem Commun 2005(35):4414–4416. doi: 10.1039/b507314f
103. Ma M, Kuang Y, Gao Y, Zhang Y, Gao P, Xu B (2010) Aromatic–aromatic interactions induce the self-assembly of pentapeptidic derivatives in water to form nanofibers and supramolecular hydrogels. J Am Chem Soc 132:2719–2728. doi:10.1021/ja9088764
104. Chen L, Pont G, Morris K, Lotze G, Squires A, Serpell LC, Adams DJ (2011) Salt-induced hydrogelation of functionalised-dipeptides at high pH. Chem Commun 47:12071–12073. doi:10.1039/C1CC15474E
105. Zhou M, Smith AM, Das AK, Hodson NW, Collins RF, Ulijn RV, Gough JE (2009) Self-assembled peptide-based hydrogels as scaffolds for anchorage-dependent cells. Biomaterials 30:2523–2530. doi:10.1016/j.biomaterials.2009.01.010
106. Yang Z, Liang G, Ma M, Abbah AS, Lu WW, Xu B (2007) D-Glucosamine-based supramolecular hydrogels to improve wound healing. Chem Commun 2007(8):843–845. doi: 10.1039/b616563j
107. Salem AK, Rose FRAJ, Oreffo ROC, Yang X, Davies MC, Mitchell JR, Roberts CJ, Stolnik-Trenkic S, Tendler SJB, Williams PM, Shakesheff KM (2003) Porous polymer and cell composites that self-assemble in situ. Adv Mater 15:210–213. doi:10.1002/adma.200390047
108. Park KM, Yang J-A, Jung H, Yeom J, Park JS, Park K-H, Hoffman AS, Hahn SK, Kim K (2012) In situ supramolecular assembly and modular modification of hyaluronic acid hydrogels for 3D cellular engineering. ACS Nano 6:2960–2968. doi:10.1021/nn204123p
109. Van Bommel KJC, van der Pol C, Muizebelt I, Friggeri A, Heeres A, Meetsma A, Feringa BL, van Esch J (2004) Responsive cyclohexane-based low-molecular-weight hydrogelators with modular architecture. Angew Chem Int Ed 43:1663–1667. doi:10.1002/anie.200352396

110. Leenders CMA, Albertazzi L, Mes T, Koenigs MME, Palmans ARA, Meijer EW (2013) Supramolecular polymerization in water harnessing both hydrophobic effects and hydrogen bond formation. Chem Commun 49:1963–1965. doi:10.1039/C3CC38949A
111. Kim H-J, Lee J-H, Lee M (2005) Stimuli-responsive gels from reversible coordination polymers. Angew Chem Int Ed 44:5810–5814. doi:10.1002/anie.200501270
112. Van de Manakker F, Vermonden T, van Nostrum CF, Hennink WE (2009) Cyclodextrin-based polymeric materials: synthesis, properties, and pharmaceutical/biomedical applications. Biomacromolecules 10:3157–3175. doi:10.1021/bm901065f
113. Li L, Guo X, Wang J, Liu P, Prud'homme RK, May BL, Lincoln SF (2008) Polymer networks assembled by host–guest inclusion between adamantyl and β-cyclodextrin substituents on poly(acrylic acid) in aqueous solution. Macromolecules 41:8677–8681. doi:10.1021/ma8020147
114. Auzély-Velty R, Rinaudo M (2002) New supramolecular assemblies of a cyclodextrin-grafted chitosan through specific complexation. Macromolecules 35:7955–7962. doi:10.1021/ma020664o
115. Koopmans C, Ritter H (2008) Formation of physical hydrogels via host–guest interactions of β-cyclodextrin polymers and copolymers bearing adamantyl groups. Macromolecules 41:7418–7422. doi:10.1021/ma801202f
116. Gosselet NM, Beucler F, Renard E, Amiel C, Sebille B (1999) Association of hydrophobically modified poly (N,N-dimethylacrylamide hydroxyethylmethacrylate) with water soluble β-cyclodextrin polymers. Colloids Surf A Physicochem Eng Asp 155:177–188. doi:10.1016/S0927-7757(99)00026-6
117. Van de Manakker F, van der Pot M, Vermonden T, van Nostrum CF, Hennink WE (2008) Self-assembling hydrogels based on β-cyclodextrin/cholesterol inclusion complexes. Macromolecules 41:1766–1773. doi:10.1021/ma702607r
118. Hashidzume A, Tomatsu I, Harada A (2006) Interaction of cyclodextrins with side chains of water soluble polymers: a simple model for biological molecular recognition and its utilization for stimuli-responsive systems. Polymer 47:6011–6027. doi:10.1016/j.polymer.2006.06.021
119. Tomatsu I, Hashidzume A, Harada A (2006) Contrast viscosity changes upon photoirradiation for mixtures of poly(acrylic acid)-based α-cyclodextrin and azobenzene polymers. J Am Chem Soc 128:2226–2227. doi:10.1021/ja058345a
120. Nakahata M, Takashima Y, Yamaguchi H, Harada A (2011) Redox-responsive self-healing materials formed from host–guest polymers. Nat Commun 2:511
121. Appel EA, Biedermann F, Rauwald U, Jones ST, Zayed JM, Scherman OA (2010) Supramolecular cross-linked networks via host-guest complexation with cucurbit[8]uril. J Am Chem Soc 132:14251–14260. doi:10.1021/ja106362w
122. Liu Y, Yu Y, Gao J, Wang Z, Zhang X (2010) Water-soluble supramolecular polymerization driven by multiple host-stabilized charge-transfer interactions. Angew Chem Int Ed 49:6576–6579. doi:10.1002/anie.201002415
123. Dankers PYW, Hermans TM, Baughman TW, Kamikawa Y, Kieltyka RE, Bastings MMC, Janssen HM, Sommerdijk NAJM, Larsen A, Bosman AW, Popa R, Fytas G, Meijer EW, van Luyn MJA, Popa ER (2012) Hierarchical formation of supramolecular transient networks in water: a modular injectable delivery system. Adv Mater 24:2703–2709. doi:10.1002/adma.201104072
124. Pape ACH, Bastings MMC, Kieltyka RE, Wyss HM, Voets IK, Meijer EW, Dankers PYW (2014) Mesoscale characterization of supramolecular transient networks using SAXS and rheology. Int J Mol Sci 15:1096–1111. doi:10.3390/ijms15011096
125. Kieltyka RE, Pape ACH, Albertazzi L, Nakano Y, Bastings MMC, Voets IK, Dankers PYW, Meijer EW (2013) Mesoscale modulation of supramolecular ureidopyrimidinone-based poly (ethylene glycol) transient networks in water. J Am Chem Soc 135:11159–11164. doi:10.1021/ja403745w

126. Guo M, Pitet LM, Wyss HM, Vos M, Dankers PYW, Meijer EW (2014) Tough stimuli-responsive supramolecular hydrogels with hydrogen-bonding network junctions. J Am Chem Soc 136:6969–6977. doi:10.1021/ja500205v
127. Dankers PYW, van Luyn MJA, Huizinga-van der Vlag A, van Gemert GML, Petersen AH, Meijer EW, Janssen HM, Bosman AW, Popa ER (2012) Development and in-vivo characterization of supramolecular hydrogels for intrarenal drug delivery. Biomaterials 33:5144–5155. doi:10.1016/j.biomaterials.2012.03.052
128. Kieltyka RE, Bastings MMC, van Almen GC, Besenius P, Kemps EWL, Dankers PYW (2012) Modular synthesis of supramolecular ureidopyrimidinone-peptide conjugates using an oxime ligation strategy. Chem Commun 48:1452–1454. doi:10.1039/c1cc14728e
129. Mollet BB, Comellas-Aragones M, Spiering AJH, Sontjens SHM, Meijer EW, Dankers PYW (2014) A modular approach to easily processable supramolecular bilayered scaffolds with tailorable properties. J Mater Chem B 2:2483–2493. doi:10.1039/C3TB21516D
130. Dankers PYW, Harmsen MC, Brouwer LA, van Luyn MJA, Meijer EW (2005) A modular and supramolecular approach to bioactive scaffolds for tissue engineering. Nat Mater 4:568–574. doi:10.1038/nmat1418
131. Bastings MMC, Koudstaal S, Kieltyka RE, Nakano Y, Pape ACH, Feyen DAM, van Slochteren FJ, Doevendans PA, Sluijter JPG, Meijer EW, Chamuleau SAJ, Dankers PYW (2014) A fast pH-switchable and self-healing supramolecular hydrogel carrier for guided, local catheter injection in the infarcted myocardium. Adv Healthcare Mater 3:70–78. doi:10.1002/adhm.201300076
132. Koudstaal S, Bastings MC, Feyen DM, Waring C, van Slochteren F, Dankers PW, Torella D, Sluijter JG, Nadal-Ginard B, Doevendans P, Ellison G, Chamuleau SJ (2014) Sustained delivery of insulin-like growth factor-1/hepatocyte growth factor stimulates endogenous cardiac repair in the chronic infarcted pig heart. J Cardiovasc Transl Res 7:232–241. doi:10.1007/s12265-013-9518-4
133. Storm C, Pastore JJ, MacKintosh FC, Lubensky TC, Janmey PA (2005) Nonlinear elasticity in biological gels. Nature 435:191–194
134. Kouwer PHJ, Koepf M, Le Sage VAA, Jaspers M, van Buul AM, Eksteen-Akeroyd ZH, Woltinge T, Schwartz E, Kitto HJ, Hoogenboom R, Picken SJ, Nolte RJM, Mendes E, Rowan AE (2013) Responsive biomimetic networks from polyisocyanopeptide hydrogels. Nature. doi:10.1038/nature11839
135. Birk DE, Bruckner P (2005) Collagen suprastructures. In: Brinckmann J, Notbohm H, Müller PK (eds) Collagen SE-7. Springer, Berlin, pp 185–205
136. Webber MJ, Tongers J, Renault M-A, Roncalli JG, Losordo DW, Stupp SI (2010) Development of bioactive peptide amphiphiles for therapeutic cell delivery. Acta Biomater 6:3–11. doi:10.1016/j.actbio.2009.07.031