EXPERT REVIEWS

Temperature-sensitive polymers for drug delivery

Expert Rev. Med. Devices 9(4), 339–351 (2012)

Scott D Fitzpatrick[1], Lindsay E Fitzpatrick[2], Ajit Thakur[1], Mohammad A Jafar Mazumder[3,4] and Heather Sheardown*[1,3]

[1]*School of Biomedical Engineering, McMaster University, 1280 Main Street West, Hamilton, ON, L8S 4L7, Canada*
[2]*The Institute of Biomaterials and Biomedical Engineering, University of Toronto, 164 College Street, Toronto, ON, M5S 3G9, Canada*
[3]*Department of Chemical Engineering, McMaster University, 1280 Main Street West, Hamilton, ON, L8S 4L7, Canada*
[4]*Department of Chemistry, King Fahd University of Petroleum & Minerals, University Blvd, Dhahran-31261, Saudi Arabia*
**Author for correspondence: Tel.: +1 905 525 9140 ext. 24794 sheardow@mcmaster.ca*

The ability to undergo rapid changes in response to subtle environmental cues make stimuli-responsive materials attractive candidates for minimally invasive, targeted and personalized drug delivery applications. This special report aims to highlight and provide a brief description of several of the significant natural and synthetic temperature-responsive materials that have clinical relevance for drug delivery applications. This report examines the advantages and disadvantages of natural versus synthetic materials and outlines various scaffold architectures that can be utilized with temperature-sensitive drug delivery materials. The authors provide a commentary on the current state of the field and provide their insight into future expectations for temperature-sensitive drug delivery, emphasizing the importance of the emergence of dual and multiresponsive systems capable of responding precisely to an expanding set of stimuli, thereby allowing the development of disease-specific drug delivery vehicles.

Keywords: drug delivery • intelligent polymers • lower critical solution temperature • *N*-isopropylacrylamide • smart polymers • sustained release • targeted • temperature-sensitive • thermoresponsive

Stimuli-responsive materials have gained considerable attention for their potential to create targeted, tunable and personalized therapeutics that respond directly to the physiologic environment. With recent advances in medical polymer technology, it is becoming increasingly possible to tailor drug-releasing scaffolds to produce on-demand delivery of therapeutic payloads locally in response to physiological requirements [1]. Stimuli-responsive polymers, also termed intelligent, smart, environmentally responsive and sensitive polymers, are a class of materials that undergo significant and rapid physicochemical changes in response to small changes in environmental conditions. There are a number of different classes of responsive polymers, which respond to a distinct set of stimuli, including light, pH, temperature, ultrasound, magnetism, electric field, enzymes, antibodies, or the presence of specific molecules, such as glucose [2]. In addition, the specific response varies depending on the system. Polymers may undergo changes in hydrophobic/hydrophilic balance, solubility, hydration, conformation, shape, degradation or micellization in response to the presence, or absence, of an external stimulus [2]. It is therefore possible to use these materials to generate drug delivery scaffolds that respond with predictable, controllable, predefined response profiles to impart a large degree of control and tunability over drug dosing. The ability to produce targeted on-demand drug release has implications for a number of different clinical applications, including hormone replacement therapy, chemotherapy, rhythmic heart disorders, diabetes, birth control and posterior segment ocular drug delivery. Of the various stimuli-responsive materials, temperature-sensitive polymers are the most widely studied, and this special report, while not exhaustive, will focus on several of these materials that have particular importance in drug delivery applications. A list of natural and synthetic thermoresponsive homopolymers and copolymers that are relevant to drug delivery is provided in Table 1.

Thermoresponsive polymers & their applicability for controlled release of drugs

Thermoresponsive polymers utilize subtle changes in temperature to trigger macroscopic changes in material properties. Polymers that possess a lower critical solution temperature (LCST) typically undergo a sol–gel phase transition when heated above their LCST, whereas polymers that become soluble upon heating are said to possess an upper critical solution temperature (UCST) [3]. Both systems can be exploited for drug delivery purposes. LCST copolymers can simply be mixed with drug as a liquid suspension at

 10.1586/ERD.12.24 ISSN 1743-4440

Table 1. A list of natural and synthetic thermoresponsive homopolymers and copolymers with their corresponding thermal phase transition temperatures.

Study (year)	Name	Abbreviation	LCST/ UCST (°C)	Ref.
Synthetic homopolymers				
Laukkanen *et al.* (2004)	Poly(*N*-vinylcaprolactam)	PVCL	31	[91]
Schild (1990)	Poly(*N*-isopropylacrylamide)	PNIPAAm	32	[92]
Cao *et al.* (2007)	Poly(*N*-n-propylacrylamide)	PNPAm	25	[93]
	Poly(*N*,*N*-ethylmethylacrylamide)	PEMA	70	
Liu and Zhu (1999)	Poly(*N*-ethylacrylamide)	PEA	82	[94]
Geever *et al.* (2011)	Poly(*N*,*N*-diethylacrylamide)	PDEAAm	~28–32	[19]
Uguzdogan *et al.* (2005)	Poly(ethoxypropylacrylamide)	PEPA	~32	[95]
Yamazaki *et al.* (1998)	Poly(*N*,*N*-bis(2-methoxyethyl) acrylamide)	PBMEAm	49	[96]
	Poly(*N*-(3-methoxypropyl)acrylamide)	PMPAm	>60	
Persson *et al.* (2000)	Poly(vinyl methyl ether)	PVME	34	[97]
Yuk *et al.* (1997)	Poly(2-dimethylamino)ethyl methacrylate)	PDMA	50	[98]
Liu and Armes (2001)	Poly(proprylene oxide)	PPO	10–20	[99]
Chiu *et al.* (1986)	Poly(2-ethyl-2-oxazoline)	PEOZ	~62	[100]
Uyama and Kobayashi (1992)	Poly(2-isopropyl-2-oxazoline)	PIPOZ	~36	[101]
Urban (2011)	Polyphosphazenes		~25–99	[81]
Synthetic multi-block copolymers				
Fusco (2006)	Poly(ethylene oxide)-poly(propylene oxide)-poly(ethylene oxide) (Pluronics®)	PEO–PPO–PEO	10–100	[102]
	L42†	PEO_4–PPO_{22}–PEO_4	37	
	L62†	PEO_6–PPO_{34}–PEO_6	32	
	L63†	PEO_9–PPO_{32}–PEO_9	34	
Polyester/PEG block copolymers				
Gao *et al.* (2010)	Poly(lactic acid-*co*-glycolic acid)-polyethylene glycol-poly(lactic acid-*co*-glycolic acid)	PLGA–PEG–PLGA	~37	[103]
Jeong *et al.* (1997)	Poly(ethylene glycol)–b-poly(D,L-lactic acid-*co*-glycolic acid)–b-poly(ethylene glycol)	PEG–PLA–PEG	~37	[104]
Lutz *et al.* (2006)	Poly(oligo(ethylene glycol) methacrylate)	POEGMA	26–90	[105]
Badi and Lutz (2009)	s-poly(ethylene glycol)-b-poly(2-(2-methoxy ethoxy) ethyl methacrylate-*co*-oligo(ethylene glycol) methacrylate)	sPEG-b-P(MEO2MA-co-$OEGMA_{475}$)	35–41	[106]
Lutz *et al.* (2006)	Poly(2-(2-methoxyethoxy)ethyl methacrylate-*co*-oligo(ethylene glycol) methacrylate)	P(MEO2MA-co-OEGMA) 5–8% OEGMA units per chain	32–37	[105]
Gong *et al.* (2009)	Poly(ε-caprolactone)-poly(ethylene glycol)-poly(ε-caprolactone)	PCL–PEG–PCL	~15–50	[107]
		PCL_{1000}–PEG_{1000}–PCL_{1000} (15–35 wt%)	~18–25	[107]
		PCL_{1000}–PEG_{1500}–PCL_{1000} (15–35 wt%)	~39–46	
		PCL_{1500}–PEG_{1500}–PCL_{1500} (15–35 wt%)	~37–45	
Dayananda *et al.* (2008)		PCL_{1950}–PEG_{1750}–PCL_{1950} (20 wt%)	~42	[108]
		PCL_{2110}–PEG_{2000}–PCL_{2110} (20 wt%)	~44	

†Pluronic nomenclature: The first letter in the copolymer name indicates that the physical state of the starting polymer is a liquid (L). The last number indicates the weight content of the PEO block (in terms of weight percent), while the remaining numbers give an indication of the molecular weight of the PPO block (taken from) [102]. Adapted from [111].

Table 1. A list of natural and synthetic thermoresponsive homopolymers and copolymers with their corresponding thermal phase transition temperatures (cont.).

Study (year)	Name	Abbreviation	LCST/ UCST (°C)	Ref.
Natural polymers and derivatives				
Chenite *et al.* (2000) and Molinaro *et al.* (2002)	Chitosan-glycerophosphate	Chitosan-GP	~37	[50,51]
Persson *et al.* (2000)	Methylcellulose	MC	50	[97]
Schild (1990)	Hydroxypropylcellulose	HPC	42	[92]
Persson *et al.* (2000)	Ethyl(hydroxyethyl)cellulose	EHEC	65	[97]
Miyazaki *et al.* (1998)†	Xyloglucan (with 44% removal of galactose)		22–27	[59]
Chilkoti *et al.* (2006)	Elastin-like polypeptides	ELP	0–100	[109]
Ge *et al.* (2010)		ELP[$V_5A_2G_3$-90]	49	[41]
Meyer *et al.* (2001)		ELP[$V_5A_2G_3$-150]	40	[48]
		ELP [$V_5A_2G_3$-160]	55	
Urry *et al.* (1991)		Poly(VPGVG)	27	[110]

†Pluronic nomenclature: The first letter in the copolymer name indicates that the physical state of the starting polymer is a liquid (L). The last number indicates the weight content of the PEO block (in terms of weight percent), while the remaining numbers give an indication of the molecular weight of the PPO block (taken from) [102]. Adapted from [111].

room temperature and delivered via minimally invasivel injection techniques directly to hard-to-access target tissues within the body. Heating to physiologic temperature drives a sol–gel phase transition, which entraps the infused drug within a solid depot and can provide sustained release of therapeutic concentrations of drug directly at the site of interest [4]. Drug-releasing polymer systems possessing a UCST may employ temperature-induced swelling or scaffold destabilization to rapidly release drug at a target site [5]. Localized heating (tumor tissues) or the application of an externally applied stimulus (ultrasound, infrared laser and so on) may be utilized to induce the local destabilization of a UCST drug-releasing copolymer scaffold to produce targeted release [6,7].

Thermoresponsive drug delivery scaffolds offer numerous advantages, such as eliminating the need for invasive surgical implantation and the ability to bypass physiological barriers, allowing delivery to hard-to-access locations within the body [8]. Furthermore, drug encapsulation within a scaffold may protect the therapeutic agent from enzymatic or environmental degradation. The release rate can be tailored to locally produce persistent levels of therapeutically relevant concentrations of drug, thus overcoming the ineffectiveness of simple injections, which are associated with an initial spike that may lead to toxic levels initially followed by a rapid decrease to levels that possess little to no therapeutic benefit (Figure 1) [8].

Synthetic thermoresponsive materials

Poly(*N*-isopropylacrylamide)

Polymer hydrogels that display reversible volume changes have gained considerable interest since Tanaka observed the tendency of polyacrylamide gels to undergo phase separation in response to temperature or fluid composition [9]. Poly(*N*-isopropylacrylamide) (PNIPAAm) is one of the most widely studied temperature-sensitive polymers; it exhibits a rapid sol–gel transition when heated above its LCST, approximately 32°C, allowing injection at room temperature and scaffold formation at body temperature [4]. One of the major limitations of PNIPAAm is that it is nondegradable. Degradable drug delivery materials afford elimination of the scaffold following exhaustion of the drug reservoir without the requirement for secondary surgical removal. Therefore, there has been an increased emphasis in recent years on introducing degradability into NIPAAm-based copolymers. Ultimately, it is desirable to preserve the thermal phase transition properties of NIPAAm, while promoting the eventual degradation and clearance from the body following exhaustion of the therapeutic effect. In 1999, Neradovic *et al.* synthesized NIPAAm-based polymers with 2-hydroxyethyl methacrylate–monolactate (HEMA–monolactate) [10]. As the hydrolytically labile lactate ester side groups were cleaved, the hydrophilicity of the copolymer increased, raising the LCST. If the LCST is raised above body temperature, the thermoreversible NIPAAm-based copolymers revert back into a hydrated liquid state, allowing uptake into systemic circulation and clearance from the body, as long as the molecular weight is below the renal filtration limit of 30–50 kDa [11]. Yoshida *et al.* designed NIPAAm-based copolymers that were crosslinked with degradable poly(amino acids), which showed a similar clearance mechanism [12]. Guan *et al.* synthesized a series of protein-reactive NIPAAm-based copolymers, possessing relatively high tensile strength that hydrolyze to produce soluble, nontoxic degradation products through copolymerization with HEMA–polylactide (HEMA–PLA), *N*-acryloxysuccinimide and acrylic acid [13]. Ma *et al.* further improved the mechanical properties through the incorporation of methacrylate–polylactide (MA–PLA) and HEMA [14]. Cui *et al.* developed a series of slow-degrading NIPAAm-based copolymers through copolymerization with dimethyl-γ-butyrolactone acrylate (DBA) [15]. These

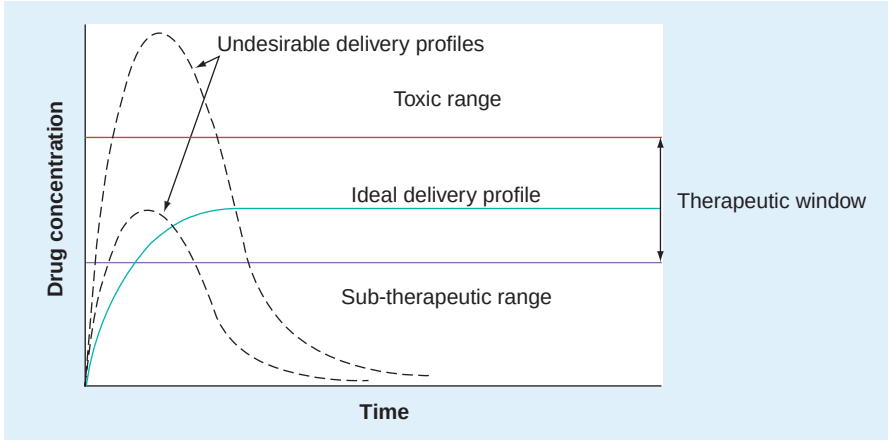

Figure 1. Representation of the ideal delivery profile in which drug concentration is maintained within the therapeutic window, below the toxic threshold, but high enough to exert a therapeutic effect.

copolymers undergo a hydrolysis-dependent opening of the DBA lactone ring structure, capable of increasing LCST above body temperature without producing any degradation products [15,16]. The Sheardown group prepared bioactive NIPAAm/DBA copolymers through copolymerization with *N*-acryloxysuccinimide [17] for posterior segment ophthalmic cell and drug delivery purposes.

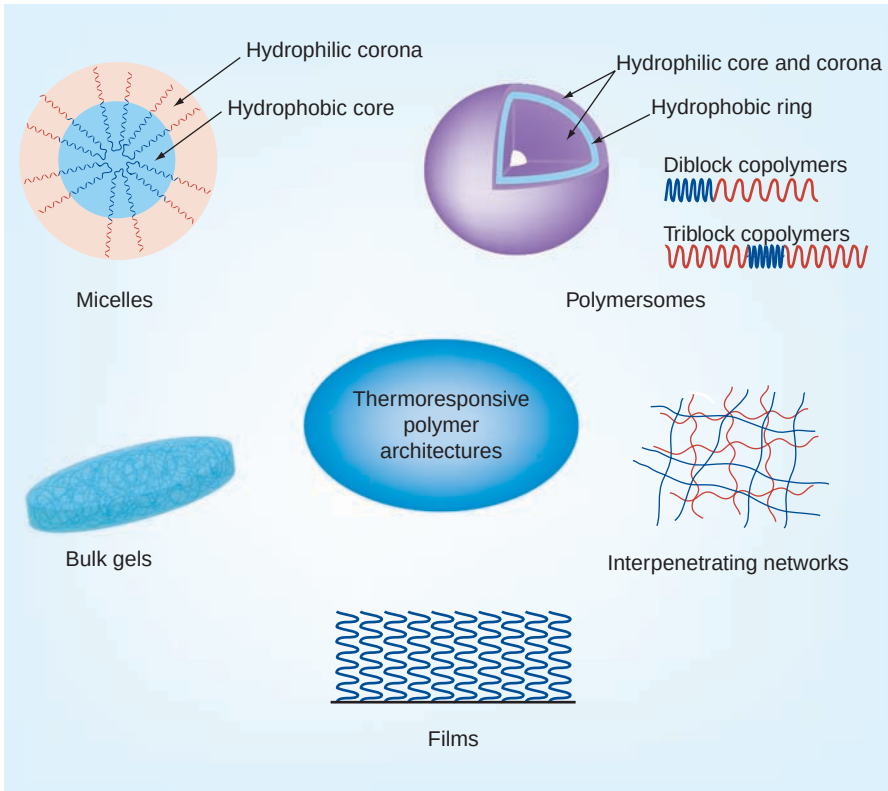

Figure 2. Illustration of some of the architectures that can be obtained using thermoresponsive copolymers for drug delivery applications. The architectures include bulk gels, micelles possessing a hydrophobic core and a hydrophilic corona, polymersomes that have a hydrophobic layer sandwiched between a hydrophilic core and corona, interpenetrating networks and polymer films.

For a list of thermoresponsive homopolymers and copolymers of PNIPAAm, see [18]. There are many other examples of *N*-substituted thermoresponsive polyacrylamides that may be suitable for drug delivery, such as poly(*N*,*N*-diethylacrylamide [19,20] and poly(*N*-vinyl caprolactam) [21]. For a comprehensive list, see [22]. Similar to PNIPAAm, linear polymers of poly(*N*,*N*-diethylacrylamide and poly (*N*-vinyl caprolactam) have LCST values of 32°C [19] and 31°C [18], respectively.

Poly(ethylene oxide)-based thermoresponsive copolymers

Triblock copolymers possessing an A–B–A configuration of poly(ethylene oxide)–poly(propylene oxide)–poly(ethylene oxide) (PEO–PPO–PEO), referred to as Pluronics or Poloxamers, are another family of thermogelling synthetic materials that have been extensively studied for their potential use as drug carriers [23]. Through manipulation of concentration, composition and molecular weight, these copolymers can be tuned to undergo reversible gelation at physiological temperature and pH [23]. The combination of hydrophilic ethylene oxide and hydrophobic propylene oxide units creates an amphiphilic copolymer that can self-associate into micelles under aqueous conditions when above a critical micelle concentration (CMC) (Figure 2). The CMC is highly temperature dependent, as below a critical micelle temperature, both ethylene oxide and propylene oxide blocks are relatively soluble in water [23]. As the temperature of the system increases, the PPO chains become less soluble, resulting in micelle formation. Pluronic micelles typically possess a diameter ranging from 10 to 100 nm and consist of a hydrophobic, PPO-rich core and a hydrated, hydrophilic PEO-rich shell [24]. The PPO core is capable of incorporating up to 30 wt % of water-insoluble drugs, while the PEO corona maintains the micelles in a dispersed state and improves drug stability by shielding the reserves from undesirable interactions with cells and proteins [24]. However, these hydrogels tend to possess poor mechanical strength, limited stability and high permeability, thereby limiting their effectiveness as sustained-release systems [24,25]. Cohn *et al.* have utilized a number of strategies to improve the mechanical integrity of PEO/PPO copolymers. Such strategies include the introduction of *in situ*

crosslinking end-groups, such as carbon-carbon double bonds [26], methacrylate groups [27] and triethoxysilane groups [27] and covalently linking PEO and PPO using carbonyl chloride and diacyl chloride coupling agents [28]. In recent years, there has been an extensive investigation into the synthesis of copolymers of polyethylene glycol (PEG) with degradable polyesters such as PLA, poly(lactide-co-glycolide) (PLGA) and poly(caprolactone) (PCL) to generate thermoresponsive copolymers with improved rigidity that degrade *in vivo*, allowing their ultimate clearance from the body [28–30]. PEG-containing copolymers that have attracted significant interest include PLGA–PEG–PLGA, PEG–PLA–PEG, PCL–PEG–PCL and poly(oligo[ethylene glycol] methacrylate), to name a few [1].

Polyphosphazenes

Polyphosphazenes are a thermoresponsive family of hybrid organic–inorganic polymers. These polymers contain an inorganic backbone consisting of alternating nitrogen and phosphorous atoms connected by alternating single and double bonds [31]. Attached to every phosphorous group are two organic groups that impart a high degree of versatility for modification and manipulation of properties and functionality, which can be utilized to impart thermoresponsive properties [4]. Numerous approaches have been explored to develop hydrolytically susceptible copolymers that have highly controllable degradation kinetics, capable of breaking down over periods ranging from hours to years [31–34]. Polyphosphazenes have demonstrated good compatibility with numerous cell lines in culture [35,36] and *in vivo* [37]. Furthermore, the degradation byproducts, namely ammonia, phosphate and alcohol, are well tolerated, and the copolymers can be designed to possess fast *in situ* gelation with tunable release kinetics, making polyphosphazenes attractive candidates for drug delivery [31,38]. For an in-depth review of polyphosphazenes, see [31].

Natural thermoresponsive materials

Elastin-like polypeptides

Elastin-like polypeptides (ELPs) are synthetic elastin-inspired polymers with a pentapeptide amino acid repeat structure, Val–Pro–Gly–Xaa–Gly, where the Xaa guest residue can be any natural amino acid except proline [39,40]. Below the phase transition temperature (T_t), ELPs exist as a clear homogeneous aqueous solution. When heated above their transition temperature, the solution becomes turbid through ELP coacervation into droplets [41]. The ELP droplet size and distribution can be manipulated through concentration and temperature [41], while the transition temperature can be adjusted by varying the concentration, molecular weight, salt content and ELP composition (i.e., through modifying the Xaa guest residue, using variable amino acid sequences or by functionalization with other proteins or polymers) [41]. ELPs are interesting candidates for drug delivery, as they possess tunable characteristics, are well tolerated *in vivo* and degrade into simple amino acid residues [42]. In addition, the molecular weight and composition of ELPs can be precisely controlled through genetic engineering approaches to form narrowly dispersed polymers, which allows an increased level of control over drug-release performance [43]. Furthermore, ELPs can be expressed in high quantities from *Escherichia coli* and can be easily purified as a result of their thermogelling behavior [44]. The Chilkoti group has extensively studied ELPs for their ability to target tumor tissues [45–48]. In one strategy, the group passively targeted tumor tissues by employing drug-conjugated ELPs with a T_t that was well above physiologic temperature. The small, soluble, ELPs took advantage of the enhanced permeability of tumor vasculature to accumulate within tumor tissues following systemic delivery [47]. In another approach, ELP–drug conjugates were designed to thermally target tumor tissues. The peptides were engineered to possess a T_t between 37 and 42°C and externally applied stimuli induced localized hyperthermia, causing the ELP–drug conjugates to aggregate and adhere to the vessel walls [48]. Mild hyperthermia was also used to drive the localized assembly of micelles that possessed a tumor-targeting ligand on the hydrophilic corona, leading to enhanced cellular uptake [46]. In another strategy, ELPs with a subphysiologic T_t were injected directly into tumorous tissues to produce a sustained-release drug depot directly within the tumor tissues for extended treatment [45].

Chitosan

Chitosan is a polysaccharide that is derived from chitin [4]. While on its own, chitosan is not thermoresponsive, it becomes thermoresponsive when it is mixed with glycerophosphate (GP) [4]. At elevated temperatures, GP forms strong hydrogen bonds with chitosan, which leads to gel formation [4]. However, chitosan/GP mixtures tend to possess slow gelation rates. Therefore, for applications requiring more rapid gelation, the derivative chitosan chloride can be used to expedite the gelling process [49]. Chitosan scaffolds also suffer from relatively rapid release of loaded protein and drugs possessing low molecular weight; complete release is often achieved within several hours [4,50]. There are also concerns about the suitability of chitosan/GP hydrogels for *in vivo* application as they have been shown to induce a relatively significant inflammatory response [51].

Cellulose derivatives

Several cellulose derivatives, such as methylcellulose (MC) and hydroxypropylmethylcellulose, display LCST behavior that can be exploited for drug delivery and tissue engineering applications [52]. MC and hydroxypropylmethylcellulose display LCST values between 40–50°C and 75–90°C, respectively [53]. However, these values can be substantially lowered using both physical and chemical methods, such as the addition of NaCl or a reduction in the hydroxypropyl content [53,54]. At low temperatures, the macromolecules exist in a fully hydrated state with little polymer–polymer interaction aside from physical entanglement [52]. Upon heating, intermolecular hydrophobic interactions between the methoxy groups result in gradual dehydration and gel formation [53]. Recently, a physical blend of hyaluronan and MC demonstrated rapid thermoreversible *in situ* gelation, degradability, good *in vivo* tolerance and potential for minimally invasive intrathecal drug delivery for spinal cord injuries [55,56]. Formulations

of hyaluronan and MC have also demonstrated favorable results as injectable cell scaffolds for retinal therapeutics [57].

Xyloglucan

In its native form, the xyloglucan polysaccharide does not form a gel [58]. However, Miyazaki *et al.* developed a thermally reversible xyloglucan hydrogel through partial degradation of xyloglucan from the seeds of *Tamarindus indica* [59]. When more than 35% of the galactose residues have been removed, xyloglucan exhibits temperature -responsive behavior under dilute aqueous conditions and possesses a relatively high storage modulus [23,25]. Xyloglucan gels have been examined as drug delivery scaffolds for oral [60], ocular [61], rectal [59], percutaneous [62] and intraperitoneal [63] applications.

Polymer architecture

A crucial parameter to consider when designing *in situ*-forming drug delivery scaffolds is the type of polymeric architecture that will be most suitable for the intended application. *In situ*-forming hydrogels can form numerous scaffold architectures, such as interpenetrating networks (IPNs), micelles, polymersomes, films and other variations (Figure 2). There are two main types of gels: physical gels and crosslinked gels. Physical gels are formed through the physical entanglement of polymer chains or micelle ordering, whereas crosslinked gels are covalently bound [8]. Covalently linked thermoresponsive networks undergo a change in their degree of swelling in response to temperature, while physically linked gels undergo a sol–gel phase transition [8]. Covalently linked networks can either be formed *in situ* or prior to implantation. *In situ* crosslinking minimizes invasiveness of instillation, but requires the use of crosslinking chemistry that is safe *in vivo*.

IPNs consist of two or more polymer networks that are bound through physical entanglement such that the networks can only be separated through bond breakage. IPNs offer a powerful tool for drug delivery as each polymer in the network can introduce specific properties, such as temperature sensitivity, and new properties can arise from the interaction of the various polymers within the network. Furthermore, it is relatively easy to manipulate properties by varying the polymer ratio within the IPN and modifying the polymers within the network. Liu *et al.* have designed transparent silicone/PNIPAAm IPN materials which show temperature transitions that are useful for drug loading and that show particular promise for the delivery of hydrophobic drugs [Liu L, Sheardown H, Manuscript Submitted]. Semi-interpenetrating copolymer networks (semi-IPNs) contain at least one crosslinked polymer network, either linear or branched [64]. Kim *et al.* prepared thermoresponsive semi-IPNs based on chitosan and poly(acryl amide) in which the hybrid synthetic and natural copolymers displayed high swelling ratios that were dependent on temperature, pH, ion concentration and electric field [64]. Nanoparticle IPNs consisting of poly(acrylic acid) (PAA) and poly(acryl amide) display UCST behavior and rapidly swell in response to heating above a critical temperature [65]. Chen *et al.* prepared semi-IPN nanogels based on hydroxypropylcellulse, which possesses an LCST around 41°C, and PAA, which, as mentioned, possesses UCST behavior [66]. By varying the chemical composition and the degree of crosslinking, the phase transition properties of these hydroxypropylcellulse–PAA nanogel semi-IPNs could be shifted from UCST to LCST. IPNs and semi-IPNs of thermoresponsive copolymers offer a high degree of flexibility and can be tailored to provide variable release profiles to suit a broad range of applications.

As discussed in the section entitled poly(ethylene oxide)-based thermoresponsive copolymers, amphiphilic block copolymers can spontaneously assemble into micelles with a hydrophilic corona and a hydrophobic core. Therefore, micelles may be particularly useful for cancer therapeutics, as many chemotherapeutics are small hydrophobic compounds with a poor therapeutic index [44]. Micelle drug carriers can increase drug accumulation in tumor tissues, while minimizing off-target effects through the enhanced permeability and retention effect, which allows extravasation of the small drug carriers through the leaky tumor vasculature, as mentioned in the section entitled 'Elastin-like polypeptides' [44]. Quan *et al.* designed an elegant thermoresponsive micelle carrier for tumor-triggered drug release [67]. Upon encountering the subtle physiological changes in tumor physiology (pH 6.8, T > 37°C), PEGylated arginine–glycine–aspartic acid (RGD) peptides were deprotected, allowing internalization by RGD receptor overexpressing tumor cells and destabilization of drug-loaded micelles for localized treatment. Wei *et al.* synthesized a series of NIPAAm-containing thermoresponsive shell crosslinked micelles [68]. They found that the crosslinked shell slowed drug release at temperatures below the LCST (25°C), but the rate accelerated dramatically above the LCST (37°C) as pNIPAAm gelation led to a deformation of the micelle structure.

Polymersomes, also known as polymer vesicles, are similar to micelles in that they are self-assembling amphiphilic block copolymers; however, they arrange to form a hydrophobic ring sandwiched between a hydrophilic core and corona [8]. The polymersome structure allows interior encapsulation of both hydrophilic and hydrophobic drugs, while the hydrophilic shell protects the entrapped drug from undesirable interactions and can help the drug delivery system to evade the immune system. The hydrophilic corona can act as a rate-controlling membrane to modulate the release of drug from the hydrophobic ring, which in turn can serve to impede release from the hydrophilic core [69]. Li *et al.* synthesized thermoresponsive, self-assembling polymersomes consisting of diblock copolymers of poly(*N*-[3-aminopropyl] methacrylamide hydrochloride) and PNIPAAm [70]. In aqueous conditions, the amphiphilic block copolymers existed as unimers at room temperature and transitioned to form vesicles when heated above their LCST, which could be adjusted between 30 and 40°C by varying the composition. The vesicle shells were then crosslinked by polyelectrolyte complexation. Qin *et al.* prepared thermoresponsive, doxorubicin-containing PEG–PNIPAAm-based polymersomes that self-associated upon heating above their LCST and could be destabilized, or ruptured, upon local cooling with either ice or penetrating cryoprobes [71]. These experiments demonstrate how temperature sensitivity can be utilized to create localized drug release following minimally invasive delivery. For an in-depth review of stimuli-responsive polymersomes in targeted drug delivery, see [72].

Thermoresponsive films can also be used as coatings on medical implants to create a stimuli-responsive material capable of modulating the microenvironment surrounding the implant. For example, a rate-controlling thermoresponsive film may increase its release rate in response to slightly elevated increases in temperature due to localized inflammation.

Using thermoresponsive materials, there are numerous design architectures that can be generated, and researchers must decide which type is suitable for their intended application. For an in-depth review on temperature-responsive polymer architecture, see [8] and [69].

Expert commentary

When designing drug delivery vehicles, an important design question to consider is whether to use natural or synthetic materials. Both choices possess inherent advantages and disadvantages. While natural materials offer great potential for inherent biocompatibility, synthetic materials offer greater flexibility for manipulation and tuning of system performance. With natural materials, we are limited in our ability to modulate the material's properties, unless we resort to the use of synthetic modification techniques. Furthermore, natural materials often possess indefinite composition, poor mechanical strength, variable and uncontrollable degradation kinetics, microbial contamination and compatibility issues [31]. Conversely, synthetic polymers allow a high degree of control over important design constraints, such as mechanical properties, degradation rates, pore size, morphology, scaffold shape and size, drug-release kinetics and biomimetic behavior [31]. It is the opinion of the authors that, moving forward, synthetic polymers inspired by, and potentially augmented by, natural materials will provide valuable tools for the design of novel drug-releasing scaffolds. Patenaude *et al.* synthesized novel hybrids of natural and synthetic materials based on PNIPAAm and various carbohydrate polymers [73]. These studies demonstrated a high degree of control over copolymer properties, such as swelling, degradation, phase transition, and mechanical properties, effectively combining the desired performance features of both natural (degradation and biological interactions) and synthetic (compositional diversity and thermal sensitivity) materials. Table 2 lists some of the advantages and disadvantages of natural and synthetic materials for medical application.

The current state of temperature-sensitive drug delivery copolymers offers minimally invasive implantation of sustained-release scaffolds to hard-to-access regions within the body through simple injection and *in situ* gelation. Furthermore, through utilization of the subtle temperature increase in tumor tissues, it is possible to tailor scaffolds to undergo a sol–gel phase transition upon encountering a tumor, thus targeting the subtle physiological differences and providing localized dosing. Several strategies, such as localized ultrasound application, can induce subtle temperature increases that lead to directed accumulation of drug carriers that possess gelling temperatures slightly above physiologic temperature, and penetrating cryoprobes can induce localized cooling to destabilize drug carriers. Thus, temperature-sensitive drug delivery scaffolds are particularly interesting for cancer therapeutics and applications where minimally invasive procedures are crucial, such as spinal cord [56] and ocular cell and drug delivery purposes [74]. However, temperature-responsive copolymers alone are limited in their ability to respond to the abundance of subtle differences that characterize specific diseased states. Therefore, when used in conjunction with additional stimuli-responsive materials, the degree of control vastly increases, as dual or multiresponsive materials can respond with controllable properties to a number of different physiological states.

There are two classifications of stimuli-responsive materials that can be used to further functionalize temperature-sensitive drug delivery scaffolds: materials that respond to internal stimuli present in the *in vivo* environment, and those that respond to externally applied stimuli. Light, magnetism, electrical impulses and ultrasound are examples of stimuli that can be externally applied to manipulate and regulate the performance of implanted scaffolds [75]. Ionic strength, pH, enzymes, antigen–antibody interactions, or the presence of specific chemicals are examples of internal stimuli that may drive a behavioral change in an

Table 2. A list of some of the advantages and disadvantages of using naturally derived and synthetic materials in medical applications.

	Natural materials	Synthetic materials
Advantages	Inherent biocompatibility Safe degradation byproducts Defined cellular and biological interactions Natural materials are well suited to 'nature-mimicking' strategies popular in tissue engineering Can provide a close approximation of native extracellular matrix	Synthetic flexibility and compositional diversity High degree of control over performance parameters, such as molecular weight, mechanical properties, elasticity, stimulus-response, release profile, degradation kinetics and so on Easily sterilized
Disadvantages	Limited number of natural polymers, therefore restricted range of attainable properties Batch-to-batch variability with indefinite composition Poor mechanical strength Can illicit an immune response Biological contamination Sterilization can be difficult	Carbon–carbon backbone is not inherently degradable, thus degradation strategies are often required, which can induce inflammation and cytotoxicity Often engineered in attempt to 'mimic' biological tissues, however, unable to recreate 'true' extracellular microenvironment Can illicit a foreign body reaction

implanted scaffold [76]. Internal stimuli have the ability to act as a negative-feedback loop and generate a direct response to the surrounding physiologic environment. By contrast, externally regulated stimuli-responsive materials require active manipulation from an outside source to generate a change in performance properties. While internal stimuli may provide better on-demand release profiles and tighter regulation of the pathological state, materials requiring external stimuli afford physicians a greater degree of control over the dosing parameters, which is particularly important should complications arise. For an in-depth review of recent advances and future perspectives of various stimuli-responsive materials, see [75–77].

Thermoresponsive & externally regulated stimuli responsive systems

There are many examples of dual thermo- and externally regulated copolymer systems. Temperature- and light-responsive materials were prepared from photochromic derivatives of ELP, in which one azobenzene moiety was incorporated for every 30 amino acid residues [78]. Irradiation at 350 nm induced a *trans–cis* isomerization, which increased the hydrophilicity of the material and shifted the phase transition temperature from 32–42°C. Irradiation with a longer wavelength was found to reform 50% of the hydrophobic *trans* isomer, thus driving phase separation. Such a system could be used to generate a pulsed-release profile for on-demand release. Zrinyi synthesized thermo- and magneto-responsive polymer beads by incorporating magnetic nanoparticles into crosslinked PNIPAAm and poly(vinyl alcohol) hydrogels [79]. In uniform magnetic fields, the gel beads arranged into linear, chainlike structures. However, in nonuniform fields, the gels formed random aggregates. This study demonstrates the ability to externally manipulate scaffold architecture with externally applied magnetism. This concept could be extended to manipulate gates in a channeled drug-release scaffold, thus creating on–off capabilities and a pulsatile release profile. Kim *et al.* prepared thermo- and electro-responsive IPNs from poly(vinyl alcohol) and PNIPAAm [80]. Electro-responsive materials tend to swell, shrink or bend in response to an applied electric field and are typically comprised of polyelectrolyte hydrogels [81]. As the charged ions are guided towards the cathode or anode side of the gel, deformation of the polyelectrolyte occurs [81]. Ultrasound is a noninvasive stimulus that has been shown to influence drug-release properties within the body by accelerating degradation in degradable polymers and enhancing the permeation of drug in both eroding and non-eroding scaffolds [81]. Ultrasound can be used to disrupt micelle architecture through acoustic destabilization, thus inducing release of the therapeutic payload [81,82]. It can also be used to induce localized heating and aggregation of thermoresponsive drug scaffolds.

Thermoresponsive & internally regulated stimuli-responsive systems

Thermoresponsive copolymers have also been combined with a number of materials that respond to internal stimuli. pH-responsive systems have received considerable attention owing to the significant variation of pH within the different locations of the body. In the GI tract, the stomach has an acidic pH between 1 and 3, whereas the pH in the duodenum ranges from 4.8 to 8.2 [3]. Cancer tissue has a slight acidic extracellur pH between 6.5–7.2, whereas normal tissues and blood possess a pH around 7.4 [3]. Furthermore, intracellular variations in pH can be exploited for targeted delivery; the early endosome, late endosome and lysosome have pH values of 6.0–6.5, 5.0–6.0 and 4.5–5.0, respectively [3]. pH-responsive polymers contain weak acids or weak bases, such as carboxylic acids or amines, and thus undergo changes in their ionization state in response to changes in pH [3]. Changes in the ionization state can lead to conformational changes, such as micelle formation or disruption, or changes in the swelling properties of crosslinked gels [2]. Ionizable polymers possessing a dissociation constant (pKa) that closely matches the pH of the target tissues can utilize the conformational pH-induced changes to release drug at a specific location. There are many examples of thermo- and pH-responsive polymers systems. Brazel and Peppas described the synthesis and characterization of temperature- and pH-responsive hydrogels of methacrylic acid and PNIPAAm [83] and Leung *et al.* synthesized microgels with a thermoresponsive core and pH-sensitive shell [84]. These studies demonstrate the potential to target tissues based on their pH and are particularly interesting for cancer therapeutics as tumor tissues possess an elevated temperature and a slightly acidic pH. In addition to pH, antigen-responsive materials are capable of undergoing significant property changes in response to highly specific stimuli recognition. Lu *et al.* reported the synthesis of thermo- and antigen-responsive hydrogels from the combination of a polymerizable antibody Fab′ fragment, which was prepared from an antifluorescein monoclonal antibody, with NIPAAm and *N,N′*-methylenebis(acrylamide) [85]. The resulting hydrogels underwent significant reversible volume changes in response to both temperature and the presence of antigens. Glucose-responsive copolymers are of considerable interest for their ability to detect glucose levels and deliver insulin as required [81]. Glucose-responsive polymers typically function either through enzymatic oxidation of glucose via glucose oxidase, through glucose binding with concanavalin A or through reversible bond formation with boronic acids [81]. Thermo- and glucose-responsive copolymers have been synthesized from comb-type graft copolymers of PNIPAAm-co-3-acrylamidophenylboronic acid) [86], and through covalently linking glucose oxidase to copolymers of NIPAAm, methacrylic acid and octadecylacrylate and subsequently immobilizing to the surface of liposomes [87]. Such scaffolds could introduce sustained-release scaffolds that detect blood sugar levels and modify their insulin-release profile accordingly, thus decreasing the frequency of insulin injections and allowing tighter regulation of blood sugar levels.

While temperature-sensitive materials have tremendous potential for targeted drug delivery, combination with dual and multi-responsive polymer systems has the potential to unleash and help realize the true capabilities of these drug release scaffolds to create personalized and on-demand release profiles.

Five-year view

It is becoming increasingly possible to synthesize drug delivery scaffolds consisting of multiple stimuli-responsive materials that can locally release a multitude of different pharmaceuticals on demand in response to internal physiological feedback and externally applied signals. Such control will allow drastically improved manipulation of the microenvironment of diseased tissues and improve the regulation of systemic conditions. As our understanding of the physiological signature of different diseases increases, so too will our ability to design drug-releasing scaffolds that produce a predictable and controllable response to disease-specific stimuli. As mentioned, temperature-sensitive systems alone are limited in their ability to respond to the surplus of stimuli that characterize a specific disease. Therefore, the true power of thermoresponsive drug-releasing scaffolds will be realized when they are combined with additional stimuli-responsive materials. Such dual and multiresponsive drug delivery scaffolds are beginning to emerge in the literature as temperature-sensitive polymers are combined with materials capable of external regulation through stimuli such as light [88], magnetism [89] and ultrasound [90]. However, a new generation of stimuli-responsive materials is emerging, wherein the identification of differentiating environmental factors characterizing various conditions is allowing the use of intricate internal stimuli to manipulate polymer properties to create a predefined response to disease-specific environmental cues, thus providing on-demand, personalized treatment. A thermoresponsive, glucose-sensitive copolymer that forms an insulin-loaded scaffold upon injection into the body with release that is dictated by blood sugar levels would mimic the body's natural regulation mechanism and help provide tighter regulation of blood sugar levels for diabetic patients. The generation of such nature-mimicking scaffolds will be driven by the improved understanding of biochemical pathways implicated in various diseases. Better characterization of the chemical signature of various diseases expands the engineer's toolbox for designing novel scaffolds capable of providing personalized treatment. Thus, future generations of drug delivery scaffolds will require a multidisciplinary approach to harness the true potential of stimuli-responsive materials.

Financial & competing interests disclosure

The authors would like to acknowledge NSERC and the NSERC 20/20 Ophthalmic Materials Network for funding. H Sheardown is the founder of the 20/20 NSERC Ophthalmic Materials Network, which had/has ties with and receives funding from the following industrial partners: Alimera Sciences, CIBA Vision Corporation, Custom Contact Lenses, GlaxoSmithKline, Fovea Pharmaceuticals, iCo Therapeutics, Siltech Corporation, Take Control Cosmedix, Vista Optics Limited and Walsh Medical Devices Incorporated. The authors have no other relevant affiliations or financial involvement with any organization or entity with a financial interest in or financial conflict with the subject matter or materials discussed in the manuscript apart from those disclosed.

No writing assistance was utilized in the production of this manuscript.

Key issues

- Temperature-sensitive drug delivery scaffolds allow minimally invasive instillation of sustained-release scaffolds for localized or systemic treatment.
- Encapsulation within a scaffold protects pharmaceuticals from undesirable interactions and enzymatic or environmental degradation. Furthermore, the scaffolds can be designed to generate sustained drug release, maintaining concentrations within the therapeutic window and avoiding complications frequently associated with simple injections.
- There are numerous temperature-responsive polymers and architectures to choose from when designing drug delivery scaffolds. Several design choices include whether to use degradable versus nondegradable scaffolds, natural versus synthetic materials and which architecture (simple hydrogel aggregates, interpenetrating networks, micelles, polymersomes, films and so on) is best suited for the intended application.
- Synthetic materials offer a high degree of flexibility, allowing manipulation of mechanical properties, degradation rates, pore size, morphology, scaffold shape and size, and drug release kinetics, whereas natural materials often possess uncontrollable degradation kinetics, microbial contamination and compatibility issues. Ultimately, combinations of natural and synthetic materials may overcome the limitations while capitalizing on the strengths of both types of materials.
- Temperature-sensitive drug delivery scaffolds are particularly well suited for delivery of chemotherapeutics. Scaffolds designed with a gelling temperature slightly above physiologic value utilize the subtle temperature increase in tumor tissues to drive scaffold formation and accumulation in tumor vasculature and the surrounding tissues.
- *In situ* gelling temperature-sensitive drug delivery scaffolds have tremendous potential for hard-to-access complications requiring minimally invasive techniques, such as ocular and spinal cord therapeutics.
- Combining temperature-sensitive polymers with additional stimuli responsive materials imparts the ability to respond to numerous external and internal stimuli, such as light, magnetism, electrical impulses, ultrasound (external) and ionic strength, pH, enzymes, antigen–antibody interactions or specific chemicals (internal). Such dual and multiresponsive drug delivery scaffolds offer a significant level of control over dosing characteristics and treatment personalization.
- The continued identification of differentiating environmental factors characterizing specific diseases will allow the development of increasingly intricate scaffolds capable of responding to disease-specific cues to provide negative feedback that attempts to mimic the body's natural regulation mechanisms.

References

1 Bikram M, West JL. Thermo-responsive systems for controlled drug delivery. *Expert Opin. Drug Deliv.* 5(10), 1077–1091 (2008).

2 Bajpai AK, Bajpai J, Saini R, Gupta R. Responsive polymers in biology and technology. *Polymer Rev.* 51(1), 53–97 (2011).

3 Schmaljohann D. Thermo- and pH-responsive polymers in drug delivery. *Adv. Drug Deliv. Rev.* 58(15), 1655–1670 (2006).

4 Li Z, Guan J. Thermosensitive hydrogels for drug delivery. *Expert Opin. Drug Deliv.* 8(8), 991–1007 (2011).

5 Hoogenboom R, Lambermont-Thijs HML, Jochems MJHC *et al.* A schizophrenic gradient copolymer: switching and reversing poly(2-oxazoline) micelles based on UCST and subtle solvent changes. *Soft Matter* 5(19), 3590–3592 (2009).

6 Rapoport N. Physical stimuli-responsive polymeric micelles for anti-cancer drug delivery. *Progr. Polymer Sci.* 32(8–9), 962–990 (2007).

7 Hirsch LR, Stafford RJ, Bankson JA *et al.* Nanoshell-mediated near-infrared thermal therapy of tumors under magnetic resonance guidance. *Proc. Natl Acad. Sci. USA* 100(23), 13549–13554 (2003).

8 Ward MA, Georgiou TK. Thermoresponsive polymers for biomedical applications. *Polymers* 3(3), 1215–1242 (2011).

9 Tanaka T. Collapse of gels and the critical endpoint. *Phys. Rev. Lett.* 40(12), 820–823 (1978).

10 Neradovic D, Hinrichs WLJ, Kettenes-Van Den Bosch JJ, Hennink WE. Poly (*N*-isopropylacrylamide) with hydrolyzable lactic acid ester side groups: a new type of thermosensitive polymer. *Macromol. Rapid Commun.* 20(11), 577–581 (1999).

11 Ruggiero A, Villa CH, Bander E *et al.* Paradoxical glomerular filtration of carbon nanotubes. *Proc. Natl Acad. Sci. USA* 107(27), 12369–12374 (2010).

12 Yoshida T, Aoyagi T, Kokufuta E, Okano T. Newly designed hydrogel with both sensitive thermoresponse and biodegradability. *J. Polym. Sci. A Polym. Chem.* 41(6), 779–787 (2003).

13 Guan J, Hong Y, Ma Z, Wagner WR. Protein-reactive, thermoresponsive copolymers with high flexibility and biodegradability. *Biomacromolecules* 9(4), 1283–1292 (2008).

14 Ma Z, Nelson DM, Hong Y, Wagner WR. Thermally responsive injectable hydrogel incorporating methacrylate–polylactide for hydrolytic lability. *Biomacromolecules* 11(7), 1873–1881 (2010).

15 Cui Z, Lee BH, Vernon BL. New hydrolysis-dependent thermosensitive polymer for an injectable degradable system. *Biomacromolecules* 8(4), 1280–1286 (2007).

16 Cui Z, Lee BH, Pauken C, Vernon BL. Manipulating degradation time in a *N*-isopropylacrylamide-based co-polymer with hydrolysis-dependent LCST. *J. Biomater. Sci. Polym. Ed.* 21(6), 913–926 (2010).

17 Fitzpatrick SD, Jafar Mazumder MA, Muirhead B, Sheardown H. Development of injectable, resorbable drug-releasing copolymer scaffolds for minimally invasive sustained ophthalmic therapeutics. *Acta Biomater.* 8, 2517–2528 (2012).

18 Liu CB, Gong CY, Huang MJ *et al.* Thermoreversible gel–sol behavior of biodegradable PCL–PEG–PCL triblock copolymer in aqueous solutions. *J. Biomed. Mater. Res. Part B Appl. Biomater.* 84(1), 165–175 (2008).

19 Geever LM, Lyons JG, Higginbotham CL. Photopolymerisation and characterisation of negative temperature sensitive hydrogels based on *N,N*-diethylacrylamide. *J. Mater. Sci. Lett.* 46(2), 509–517 (2011).

20 Patra L, Vidyasagar A, Toomey R. The effect of the Hofmeister series on the deswelling isotherms of poly (*N*-isopropylacrylamide) and poly (*N,N*-diethylacrylamide). *Soft Matter* 7(13), 6061–6067 (2011).

21 Beija M, Marty JD, Destarac M. Thermoresponsive poly(*N*-vinyl caprolactam)-coated gold nanoparticles: sharp reversible response and easy tunability. *Chem. Commun.* 47(10), 2826–2828 (2011).

22 *Advances in Polymer Science*. Aseyev V, Muller AHE, Tenhu H, Borisov O, Winnik FM (Eds). Springer Berlin Heidelberg, Berlin, Germany (2010).

23 Klouda L, Mikos AG. Thermoresponsive hydrogels in biomedical applications. *Eur. J. Pharm. Biopharm.* 68(1), 34–45 (2008).

24 Batrakova EV, Kabanov AV. Pluronic block copolymers: evolution of drug delivery concept from inert nanocarriers to biological response modifiers. *J. Control. Release* 130(2), 98–106 (2008).

25 Wells LA, Lasowski F, Fitzpatrick SD, Sheardown H. Responding to change: thermo- and photo-responsive polymers as unique biomaterials. *Crit. Rev. Biomed. Eng.* 38(6), 487–509 (2010).

26 Sosnik A, Cohn D, San Román J, Abraham GA. Crosslinkable PEO–PPO–PEO-based reverse thermo-responsive gels as potentially injectable materials. *J. Biomater. Sci. Polym. Ed.* 14(3), 227–239 (2003).

27 Cohn D, Sosnik A, Garty S. Smart hydrogels for *in situ* generated implants. *Biomacromolecules* 6(3), 1168–1175 (2005).

28 Sosnik A, Cohn D. Reverse thermo-responsive poly(ethylene oxide) and poly(propylene oxide) multiblock copolymers. *Biomaterials* 26(4), 349–357 (2005).

29 Cohn D, Lando G, Sosnik A, Garty S, Levi A. PEO–PPO–PEO-based poly(ether ester urethane)s as degradable reverse thermo-responsive multiblock copolymers. *Biomaterials* 27(9), 1718–1727 (2006).

30 Wang Y, Tan Y, Huang X, Xu G. Gelation behavior of thermo-responsive poly(ethylene oxide) and poly(propylene oxide) multiblock polycarbonates. *J. Macromol. Sci. A* 46(4), 397–404 (2009).

31 Kumbar SG, Bhattacharyya S, Nukavarapu SP, Khan YM, Nair LS, Laurencin CT. *In vitro* and *in vivo* characterization of biodegradable poly(organophosphazenes) for biomedical applications. *J. Inorg. Organomet. Polymer. Mater.* 16(4), 365–385 (2006).

32 Schacht E, Vandorpe J, Dejardin S, Lemmouchi Y, Seymour L. Biomedical applications of degradable polyphosphazenes. *Biotechnol. Bioeng.* 52(1), 102–108 (1996).

33 Laurencin CT, Koh HJ, Neenan TX, Allcock HR, Langer R. Controlled release using a new bioerodible polyphosphazene matrix system. *J. Biomed. Mater. Res.* 21(10), 1231–1246 (1987).

34 Lee KY, Mooney DJ. Hydrogels for tissue engineering. *Chem. Rev.* 101(7), 1869–1879 (2001).

35 Conconi MT, Lora S, Baiguera S *et al.* *In vitro* culture of rat neuromicrovascular endothelial cells on polymeric scaffolds. *J. Biomed. Mater. Res. A* 71(4), 669–674 (2004).

36 Bhattacharyya S, Lakshmi S, Bender J *et al.* Preparation of poly bis(carboxylato phenoxy)phosphazene non-woven nanofiber mats by electrospinning. In: *Architecture and Application of Biomaterials*

and Biomolecular Materials. Wong JY, Plant AL, Schmidt CE *et al.* (Eds). Materials Research Society, PA, USA, 157–163 (2004).

37 Langone F, Lora S, Veronese FM *et al.* Peripheral nerve repair using a poly(organo)phosphazene tubular prosthesis. *Biomaterials* 16(5), 347–353 (1995).

38 Al-Abd AM, Hong KY, Song SC, Kuh HJ. Pharmacokinetics of doxorubicin after intratumoral injection using a thermosensitive hydrogel in tumor-bearing mice. *J. Control. Release* 142(1), 101–107 (2010).

39 Chilkoti A, Dreher MR, Meyer DE. Design of thermally responsive, recombinant polypeptide carriers for targeted drug delivery. *Adv. Drug Deliv. Rev.* 54(8), 1093–1111 (2002).

40 Ge X, Filipe CD. Simultaneous phase transition of ELP tagged molecules and free ELP: an efficient and reversible capture system. *Biomacromolecules* 7(9), 2475–2478 (2006).

41 Ge X, Hoare T, Filipe CD. Protein-based aqueous-multiphasic systems. *Langmuir* 26(6), 4087–4094 (2010).

42 Bessa PC, Machado R, Nürnberger S *et al.* Thermoresponsive self-assembled elastin-based nanoparticles for delivery of BMPs. *J. Control. Release* 142(3), 312–318 (2010).

43 Meyer DE, Chilkoti A. Genetically encoded synthesis of protein-based polymers with precisely specified molecular weight and sequence by recursive directional ligation: examples from the elastin-like polypeptide system. *Biomacromolecules* 3(2), 357–367 (2002).

44 McDaniel JR, Callahan DJ, Chilkoti A. Drug delivery to solid tumors by elastin-like polypeptides. *Adv. Drug Deliv. Rev.* 62(15), 1456–1467 (2010).

45 Liu W, MacKay JA, Dreher MR *et al.* Injectable intratumoral depot of thermally responsive polypeptide-radionuclide conjugates delays tumor progression in a mouse model. *J. Control. Release* 144(1), 2–9 (2010).

46 Dreher MR, Simnick AJ, Fischer K *et al.* Temperature triggered self-assembly of polypeptides into multivalent spherical micelles. *J. Am. Chem. Soc.* 130(2), 687–694 (2008).

47 MacKay JA, Chen M, McDaniel JR, Liu W, Simnick AJ, Chilkoti A. Self-assembling chimeric polypeptide-doxorubicin conjugate nanoparticles that abolish tumours after a single injection. *Nat. Mater.* 8(12), 993–999 (2009).

48 Meyer DE, Shin BC, Kong GA, Dewhirst MW, Chilkoti A. Drug targeting using thermally responsive polymers and local hyperthermia. *J. Control. Release* 74(1–3), 213–224 (2001).

49 Chang Y, Xiao L, Du Y. Preparation and properties of a novel thermosensitive *N*-trimethyl chitosan hydrogel. *Polymer Bull.* 63(4), 531–545 (2009).

50 Chenite A, Chaput C, Wang D *et al.* Novel injectable neutral solutions of chitosan form biodegradable gels *in situ*. *Biomaterials* 21(21), 2155–2161 (2000).

51 Molinaro G, Leroux JC, Damas J, Adam A. Biocompatibility of thermosensitive chitosan-based hydrogels: an *in vivo* experimental approach to injectable biomaterials. *Biomaterials* 23(13), 2717–2722 (2002).

52 Ruel-Gariépy E, Leroux JC. *In situ*-forming hydrogels – review of temperature-sensitive systems. *Eur. J. Pharm. Biopharm.* 58(2), 409–426 (2004).

53 Van Vlierberghe S, Dubruel P, Schacht E. Biopolymer-based hydrogels as scaffolds for tissue engineering applications: a review. *Biomacromolecules* 12(5), 1387–1408 (2011).

54 Sarkar N. Thermal gelation properties of methyl and hydroxypropyl methylcellulose. *J. Appl. Polymer. Sci.* 24(4), 1073–1087 (1979).

55 Gupta D, Tator CH, Shoichet MS. Fast-gelling injectable blend of hyaluronan and methylcellulose for intrathecal, localized delivery to the injured spinal cord. *Biomaterials* 27(11), 2370–2379 (2006).

56 Kang CE, Poon PC, Tator CH, Shoichet MS. A new paradigm for local and sustained release of therapeutic molecules to the injured spinal cord for neuroprotection and tissue repair. *Tissue Eng. Part A* 15(3), 595–604 (2009).

57 Ballios BG, Cooke MJ, van der Kooy D, Shoichet MS. A hydrogel-based stem cell delivery system to treat retinal degenerative diseases. *Biomaterials* 31(9), 2555–2564 (2010).

58 De Freitas RA, Busato AP, Mitchell DA, Silveira JLM. Degalatosylation of xyloglucan: effect on aggregation and conformation, as determined by time dependent static light scattering, HPSEC-MALLS and viscosimetry. *Carbohydr. Polymer.* 83(4), 1636–1642 (2011).

59 Miyazaki S, Suisha F, Kawasaki N, Shirakawa M, Yamatoya K, Attwood D. Thermally reversible xyloglucan gels as vehicles for rectal drug delivery. *J. Control. Release* 56(1–3), 75–83 (1998).

60 Itoh K, Tsuruya R, Shimoyama T *et al.* *In situ* gelling xyloglucan/alginate liquid formulation for oral sustained drug delivery to dysphagic patients. *Drug Dev. Ind. Pharm.* 36(4), 449–455 (2010).

61 Miyazaki S, Suzuki S, Kawasaki N, Endo K, Takahashi A, Attwood D. *In situ* gelling xyloglucan formulations for sustained release ocular delivery of pilocarpine hydrochloride. *Int. J. Pharm.* 229(1–2), 29–36 (2001).

62 Takahashi A, Suzuki S, Kawasaki N *et al.* Percutaneous absorption of non-steroidal anti-inflammatory drugs from *in situ* gelling xyloglucan formulations in rats. *Int. J. Pharm.* 246(1–2), 179–186 (2002).

63 Suisha F, Kawasaki N, Miyazaki S *et al.* Xyloglucan gels as sustained release vehicles for the intraperitoneal administration of mitomycin C. *Int. J. Pharm.* 172(1–2), 27–32 (1998).

64 Kim SJ, Shin SR, Kim NG, Kim SI. Swelling behavior of semi-interpenetrating polymer network hydrogels based on chitosan and poly(acryl amide). *J. Macromol. Sci. Pure. Appl. Chem.* A42(8), 1073–1083 (2005).

65 Owens DE, Iii, Jian Y, Fang JE, Slaughter BV, Chen Y-H, Peppas NA. Thermally responsive swelling properties of polyacrylamide/poly(acrylic acid) interpenetrating polymer network nanoparticles. *Macromol.* 40(20), 7306–7310 (2007).

66 Chen Y, Ding D, Mao Z *et al.* Synthesis of hydroxypropylcellulose-poly(acrylic acid) particles with semi-interpenetrating polymer network structure. *Biomacromolecules* 9(10), 2609–2614 (2008).

67 Quan CY, Chen JX, Wang HY *et al.* Core-shell nanosized assemblies mediated by the alpha-beta cyclodextrin dimer with a tumor-triggered targeting property. *ACS Nano* 4(7), 4211–4219 (2010).

68 Wei H, Cheng C, Chang C *et al.* Synthesis and applications of shell cross-linked thermoresponsive hybrid micelles based on poly(*N*-isopropylacrylamide-co-3-(trimethoxysilyl)propyl methacrylate)-b-poly(methyl methacrylate). *Langmuir* 24(9), 4564–4570 (2008).

69 Stuart MaC, Huck WTS, Genzer J *et al.* Emerging applications of stimuli-

responsive polymer materials. *Nat. Mater.* 9(2), 101–113 (2010).

70 Li Y, Lokitz BS, McCormick CL. Thermally responsive vesicles and their structural 'locking' through polyelectrolyte complex formation. *Angew. Chem. Int. Ed. Engl.* 45(35), 5792–5795 (2006).

71 Qin S, Geng Y, Discher DE, Yang S. Temperature-controlled assembly and release from polymer vesicles of poly(ethylene oxide)-block-poly (*N*-isopropylacrylamide). *Adv. Mater.* 18(21), 2905–2909 (2006).

72 Meng F, Zhong Z, Feijen J. Stimuli-responsive polymersomes for programmed drug delivery. *Biomacromolecules* 10(2), 197–209 (2009).

73 Patenaude M, Hoare T. Injectable, mixed natural-synthetic polymer hydrogels with modular properties. *Biomacromolecules* 13(2), 369–378 (2012).

74 Fitzpatrick SD, Jafar Mazumder MA, Lasowski F, Fitzpatrick LE, Sheardown H. PNIPAAm-grafted-collagen as an injectable, *in situ* gelling, bioactive cell delivery scaffold. *Biomacromolecules* 11(9), 2261–2267 (2010).

75 Bawa P, Pillay V, Choonara YE, du Toit LC. Stimuli-responsive polymers and their applications in drug delivery. *Biomed. Mater.* 4(2), 022001 (2009).

76 Roy D, Cambre JN, Sumerlin BS. Future perspectives and recent advances in stimuli-responsive materials. *Progr. Polymer Sci.* 35, 278–301 (2010).

77 McCoy CP, Brady C, Cowley JF *et al.* Triggered drug delivery from biomaterials. *Expert Opin. Drug Deliv.* 7(5), 605–616 (2010).

78 Strzegowski LA, Martinez MB, Gowda DC, Urry DW, Tirrell DA. Photomodulation of the inverse temperature transition of a modified elastin poly(pentapeptide). *J. Am. Chem. Soc.* 116(2), 813–814 (1994).

79 Zrinyi M. Intelligent polymer gels controlled by magnetic fields. *Colloid. Polymer. Sci.* 278(2), 98–103 (2000).

80 Kim SJ, Park SJ, Lee SM, Lee YM, Kim HC, Kim SI. Electroactive characteristics of interpenetrating polymer network hydrogels composed of poly(vinyl alcohol) and poly(*N*-isopropylacrylamide). *J. Appl. Polymer Sci.* 89(4), 890–894 (2003).

81 *Handbook of Stimuli-Responsive Materials*. Urban MW (Ed.). Wiley-VCH Verlag GmbH & Co. KGaA, Weinheim, Germany (2011).

82 Munshi N, Rapoport N, Pitt WG. Ultrasonic activated drug delivery from Pluronic P-105 micelles. *Cancer Lett.* 118(1), 13–19 (1997).

83 Brazel CS, Peppas NA. Synthesis and characterization of thermo- and chemomechanically responsive poly(*N*-isopropylacrylamide-co-methacrylic acid) hydrogels. *Macromolecules* 28(24), 8016–8020 (1995).

84 Leung MF, Zhu JM, Harris FW, Li P. Novel synthesis and properties of smart core-shell microgels. *Macromol. Symp.* 226, 177–185 (2005).

85 Lu ZR, Kopeckova P, Kopecek J. Antigen responsive hydrogels based on polymerizable antibody Fab ' fragment. *Macromol. Biosci.* 3(6), 296–300 (2003).

86 Zhang S-B, Chu L-Y, Xu D, Zhang J, Ju X-J, Xie R. Poly(*N*-isopropylacrylamide)-based comb-type grafted hydrogel with rapid response to blood glucose concentration change at physiological temperature. *Polymer Adv. Tech.* 19(8), 937–943 (2008).

87 Jo SM, Lee HY, Kim JC. Glucose-sensitivity of liposomes incorporating conjugates of glucose oxidase and poly(*N*-isopropylacrylamide-co-methacrylic acid-co-octadecylacrylate). *Int. J. Biol. Macromolecules* 45(4), 421–426 (2009).

88 Ramanan VV, Hribar KC, Katz JS, Burdick JA. Nanofiber-nanorod composites exhibiting light-induced reversible lower critical solution temperature transitions. *Nanotechnology* 22(49), 494009 (2011).

89 Fan T, Li M, Wu X, Li M, Wu Y. Preparation of thermoresponsive and pH-sensitivity polymer magnetic hydrogel nanospheres as anticancer drug carriers. *Colloids Surf. B. Biointerfaces* 88(2), 593–600 (2011).

90 Nelson JL, Roeder BL, Carmen JC, Roloff F, Pitt WG. Ultrasonically activated chemotherapeutic drug delivery in a rat model. *Cancer Res.* 62(24), 7280–7283 (2002).

91 Laukkanen A, Valtola L, Winnik F. Formation of colloidally stable phase separated poly(*N*-vinylcaprolactam) in water: a study by dynamic light scattering, microcalorimetry, and pressure perturbation calorimetry. *Macromolecules* 37(6), 2268–2274 (2004).

92 Schild H. Microcalorimetric detection of lower critical solution temperatures in aqueous polymer solutions. *J. Phys. Chem. B* 94, 4352–4356 (1990).

93 Cao Y, Zhu X, Luo J. Effects of substitution groups on the RAFT polymerization of *N*-alkylacrylamides in the preparation of thermosensitive block copolymers. *Macromolecules* 40(18), 6481–6488 (2007).

94 Liu HY, Zhu XX. Lower critical solution temperatures of *N*-substituted acrylamide copolymers in aqueous solutions. *Polymer* 40(25), 6985–6990 (1999).

95 Uguzdogan E, Camh T, Kabasakal O *et al.* A new temperature-sensitive polymer: poly(ethoxypropylacrylamide). *Eur. Polymer J.* 41, 2142–2149 (2005).

96 Yamazaki A, Song J, Winnik F, Brash J. Synthesis and solution properties of fluorescently labeled amphiphilic (*N*-alkylacrylamide) oligomers. *Macromol.* 31, 109–115 (1998).

97 Persson J, Johansson HO, Galaev I, Mattiasson B, Tjerneld F. Aqueous polymer two-phase systems formed by new thermoseparating polymers. *Bioseparation* 9(2), 105–116 (2000).

98 Yuk S, Cho S, Lee S. pH/temperature-responsive polymer composed of poly((*N,N*-dimethylamino)ethyl methacrylate-co-ethylacrylamide). *Macromolecules* 30, 6856–6859 (1997).

99 Liu S, Armes SP. The facile one-pot synthesis of shell cross-linked micelles in aqueous solution at high solids. *J. Am. Chem. Soc.* 123(40), 9910–9911 (2001).

100 Chiu TT, Thill BP, Fairchok WJ. Poly(2-ethyl-2-oxazoline): a new water- and organic-soluble adhesive. In: *Advances in Chemistry*. American Chemical Society, Washington, DC, USA, 425–433 (1986).

101 Uyama H, Kobayashi S. A novel thermo-sensitive polymer. Poly(2-iso-propyl-2-oxazoline). *Chem. Lett.* 21(9), 1643–1646 (1992).

102 Fusco S. Perspectives on: PEO–-PPO–PEO triblock copolymers and their biomedical applications. *J. Bioact. Compat. Polym.* 21(2), 149–164 (2006).

103 Gao Y, Sun Y, Ren F, Gao S. PLGA–PEG–PLGA hydrogel for ocular drug delivery of dexamethasone acetate. *Drug Dev. Ind. Pharm.* 36(10), 1131–1138 (2010).

104 Jeong B, Bae YH, Lee DS, Kim SW. Biodegradable block copolymers as injectable drug-delivery systems. *Nature* 388(6645), 860–862 (1997).

105 Lutz JF, Akdemir O, Hoth A. Point by point comparison of two thermosensitive polymers exhibiting a similar LCST: is the age of poly(NIPAM) over? *J. Am. Chem. Soc.* 128(40), 13046–13047 (2006).

106 Badi N, Lutz JF. PEG-based thermogels: applicability in physiological media. *J. Control. Release* 140(3), 224–229 (2009).

107 Gong C, Shi S, Wu L *et al.* Biodegradable *in situ* gel-forming controlled drug delivery system based on thermosensitive PCL-PEG-PCL hydrogel. Part 2: sol–gel–sol transition and drug delivery behavior. *Acta Biomater.* 5(9), 3358–3370 (2009).

108 Dayananda K, He C, Lee DS. *In situ* gelling aqueous solutions of pH- and temperature-sensitive poly(ester amino urethane)s. *Polymer* 49(21), 4620–4625 (2008).

109 Chilkoti A, Christensen T, MacKay JA. Stimulus responsive elastin biopolymers: Applications in medicine and biotechnology. *Curr. Opin. Chem. Biol.* 10(6), 652–657 (2006).

110 Urry D, Luan C, Parker T. Temperature of polypeptide inverse temperature transition depends on mean residue hydrophobicity. *J. Am. Chem. Soc.* 113(11), 4346–4348 (1991).

111 Liu R, Fraylich M, Saunders BR. Thermoresponsive copolymers: from fundamental studies to applications. *Colloid. Polym. Sci.* 287(6), 627–643 (2009).