In-Situ Gelling Stimuli-Sensitive PEG-Based Amphiphilic Copolymer Hydrogels

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Abstract In-situ gelling stimuli-sensitive block copolymer hydrogels exhibit sol–gel phase-transitions in response to external stimuli, due to the formation of reversible polymer networks caused by physical interactions. In-situ gelling stimuli-sensitive block copolymer hydrogels show many advantages, such as simple drug formulation and administration procedures, no organic solvent, site-specificity, a sustained drug release behavior, less systemic toxicity, and ability to delivery both hydrophilic and hydrophobic drugs. Poly(ethylene glycol)s with relatively low molecular weight are hydrophilic, nontoxic, absent of antigenicity and immunogenicity, and can be directly excreted by the kidneys. PEG-based amphiphilic copolymers have attracted extensive interest for their unique self-assembly and biocompatibility. The PEG-based amphiphilic copolymers exhibit unique changes in micellar architecture and aggregation number in response to changes near physiological temperature; therefore, in-situ gelling systems made of the PEG-based amphiphilic copolymers have received worldwide investigation. This article stresses the recent development and biomedical evaluation of the in-situ gelling stimuli-sensitive PEG-based amphiphilic copolymers that are capable of responding to changes in temperature and/or pH.

Introduction

The hydrogels systems are three-dimension hydrophilic polymer networks that can absorb considerable water and exhibit considerable flexibility [1, 2]. Many polymer networks can undergo reversible volume phase transitions or sol–gel phase transitions in response to the external physical or chemical stimuli, such as temperature, pH, ionic strength, light, electromagnetic radiation, and biomolecules, are called stimuli-sensitive or intelligent hydrogels, which have received increasing attention for their great potential in industrial applications, such as drug delivery systems (DDS), tissue engineering, and separation. Under the influence of external stimuli, hydrogels based on covalently crosslinked networks can undergo drastic volume phase transitions, while some physical networks show reversible sol–gel phase transitions without marked volume changes. Reversible hydrogels formed by stimuli-induced sol–gel phase transitions are called in-situ forming hydrogels and exist as flowable aqueous solutions (or sol state) before administration but immediately turn into standing gels after administration. These are of considerable interest for their applications in drug delivery and tissue engineering [3, 4].

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Table 2. LCSTs of several typical thermosensitive polymers

Polymer	LCST (°C)	References
Poly(N-isopropylacrylamide) (PNIPAM)	32	[5]
Poly(N,N-diethylacrylamide) (DEAM)	25	[6]
Poly(N-ethylmethacrylamide) (PNEMAM)	58	[6]
Poly(methyl vinyl ether) (PMVE)	34	[7]
Poly(2-ethoxyethyl vinyl ether) (PEOVE)	20	[8]
Poly(N-vinylisobutyramide) (PNVIBAM)	39	[9]
Poly(N-vinylcaprolactam) (PNVCa)	30~50	[10]
Poly(organophosphazenes)	25.0~98.5	[11]
Poly(N-(2-hydroxypropyl) methacrylamide mono/di lactate) (PHPMAM-mono/di lactate)	13~65	[12]

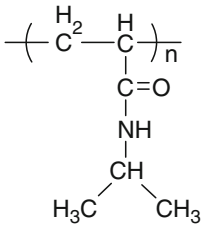
Thermo-sensitive hydrogels exhibit volume phase transitions or sol–gel phase-transitions at critical temperatures, i.e., lower critical solution temperatures (LCST) or upper critical solution temperatures (UCST). The LCST polymers exhibit swelling-to-shrinking (or sol-to-gels) transition with increasing temperature, whereas the UCST systems undergo the opposite transitions. Typical LCST polymers include poly(*N*-isopropylacrylamide) (PNIPAM) [5], poly(*N*,*N*-diethylacrylamide) (PDEAM) [6], poly(vinyl ether)s (PVE) [7, 8], poly(*N*-vinylalkylamide) [9], poly(*N*-vinylcaprolactam) (PVNCa) [10], polyphosphazene derivatives [11], and poly(*N*-(2-hydroxypropyl) methacrylamide mono/di lactate) (PHPMAM-mono/di lactate) [12, 13]. The LCSTs of several typical thermosensitive polymers are listed in Table 1.

In contrast to the permanent networks formed by chemical crosslinking, the in-situ forming hydrogels are transient (or reversible) physical networks and can be reversibly transformed into sol state by varying the environmental conditions [14]. Typical in-situ forming thermosensitive hydrogels based on natural polymers include methylcellulose [15] and chitosan/glycerophosphate blends [16]. Examples of in-situ forming hydrogels based on synthetic polymers are poly(*N*-isopropylacrylamide-*co*-acrylic acid) (P(NIPAM-*co*-AA)) [17], poly(*N*-isopropylacrylamide)-*block*-poly(ethylene glycol) (PNIPAM-*b*-PEG) [18], and poly(ethylene oxide)–*b*-poly(propylene oxide)–*b*-poly(ethylene oxide) (PEO–PPO–PEO) [19]. Poly(ethylene glycol) (PEG) with lower molecular weight (MW < 10,000) is known as a kind of polymer that is hydrophilic, nontoxic, absent of antigenicity and immunogenicity, and can be excreted by kidney [20, 21]. PEG-based amphiphilic copolymers exhibit unique changes in micellar architecture and aggregation number in response to a temperature change at physiological temperatures [22, 23]. The in-situ gelling systems based on the PEG-based amphiphilic copolymers have also attracted increasing attention.

This article is focused on the development and biomedical evaluation of the in-situ gelling stimuli-sensitive PEG-based amphiphilic copolymers and Pluronic-based amphiphilic copolymers.

Thermogelling PEG–PNIPAM Block Copolymers

Poly(*N*-isopropylacrylamide) (PNIPAM), shown in Scheme 1, is one of the most popular thermosensitive polymers. The linear PNIPAM chain undergoes a rapid coil-to-globule transition in aqueous solution at ~32°C, its LCST, while the crosslinked PNIPAM



PNIPAM

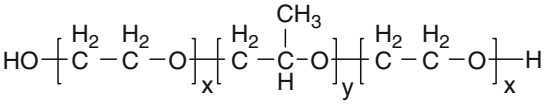
Scheme 1. Chemical structure of PNIPAM.

network shows an abrupt swelling–deswelling transition at LCST [5]. This unique hydration-to-dehydration transition is attributed to the isopropyl side groups, as the associated water separation in the gels (syneresis) due to the entropic increase when the temperature is increased above LCST [24]. The LCST of PNIPAM can be elevated or reduced by incorporating *N*-isopropylacrylamide (NIPAM) with a more hydrophilic monomer or a more hydrophobic monomer [25]. Homo- and co-polymers of PNIPAM have been synthesized by different methods, including conventional radical polymerization [5] and controlled radical polymerization, such as reversible addition fragmentation chain transfer (RAFT) [26], atom transfer radical polymerization (ATRP) [27], and Cerium (IV) redox-initiation polymerization [28]. The NIPAM monomer is cytotoxic, but PNIPAM is nontoxic and PNIPAM with limited molecular weight is excreted by glomerular filtration without long-term accumulation in vivo [29]. PNIPAM and its copolymers have been extensively developed because of their potential industrial applications such as in separation [30], conductivity control [31], controlled drug delivery [32], and tissue engineering [33].

Compared with the volume phase transitions of the chemically crosslinked PNIPAM homopolymer systems, the in-situ forming PNIPAM-based hydrogels exhibit simple sol–gel phase transitions. These hydrogels contain a certain hydrophilic component that can retain the water above the LCST. For example, monomethoxy PEG–PNIPAM diblock copolymers (MPEG–PNIPAM), prepared by quasi-living polymerization using cerium (IV) redox [28], exhibit sol-to-gel-to-syneresis transitions with increasing temperature and the gels window is markedly influenced by the composition of the copolymer [18, 34]. The gelation of the AB diblock copolymer is driven by micellar ordered-packing and entanglement. A series of AB, BAB, $A(B)_4$, and $A(B)_8$ linear and star-shaped block copolymers with poly(ethylene glycol) (PEG) as the A block and PNIPAM as the B block was synthesized by cerium (IV) redox initiated free radical polymerization [18]. A 20 wt% copolymer aqueous solutions exist as a sol state at a lower temperature (5°C), whereas gelation occurred rapidly when the temperature was increased to 37°C. In contrast to the micellar packing and entanglement mechanism for the diblock copolymers, the BAB, $A(B)_4$, and $A(B)_8$ type copolymers form a strong associative network due to the hydrophobic aggregation of the PNIPAM segments at temperatures above LCST. The PEG/PNIPAM block copolymer hydrogels show no significant syneresis (water separation in the gels) after 2 months at 37°C, as compared with a high degree of syneresis for the hydrogels based on the PNIPAM homopolymer or the systems composed of PNIPAM and hydrophobic blocks, such as the poly(methyl methacrylate)–PNIPAM diblock (PMMA–PNIPAM) [35] and polystyrene–PNIPAM–polystyrene triblock copolymers (PS–PNIPAM–PS) [36]. Similar block copolymers composed of PEG and PNIPAM are used for immobilization and culturing rabbit chondrocytes [37]. Higher PNIPAM contents and polymer concentration lead to higher cell viability after a 7-day culture, probably due to a more appropriate mechanical strength and higher porosity.

Pluronic-Based In-Situ Forming Hydrogels

Widely used in pharmaceutical systems, Pluronics (BASF) and Poloxamers (ICI) are PEO–PPO–PEO triblock copolymers (Scheme 2). The PEO block is predominantly hydrophilic from 0 to 100°C, whereas the PPO block undergoes a hydrophilic-to-hydrophobic transition as temperature is increased about 15°C [38]. With increasing temperature, some concentrated Pluronic aqueous solutions exhibit a sol-to-gels phase transition at the lower critical gelation temperature (LCGT) and a gels-to-sol transition at the upper phase transition temperature. As the temperature increases, a unimers-to-micelles transition first occurs in the Pluronic solution and the micelle number increases with further temperature increases (Fig. 1). When the micellar volume fraction is increased to a critical value (~0.53), the micelles are packed into a crystallization-like structure of hard-spheres and gelation occurs [38]. As the temperature increases further, the aggregation conformation of some Pluronic hydrogels changes from the spherical micelles closely packed in a cubic lattice into rod-like micelles packed in a hexagonal system; this results in a decrease in the intermicellar interactions and the upper gels-to-sol transition to occur at a higher temperature.



PEO-PPO-PEO

Scheme 2. Chemical structure of PEO–PPO–PEO.

The Pluronic hydrogels exhibit high viscosity, partial rigidity, and time persistence, due to the ordered micellar packing structure and micellar interactions. Some concentrated Pluronic aqueous solutions exist in the sol state at room temperature but form a gels at physiological temperatures. Consequently, Pluronics are used as injectable in-situ forming matrices for drug and cell deliveries. However, there are drawbacks to Pluronic hydrogels systems, such as weak mechanical strength, short residence time, high permeability, nonbiodegradability, and molecular weight limitations [3]. To overcome these drawbacks, several oligomers of Pluronic F127 were coupled using hexamethylene diisocyanate (HMDI) or phosgene as the coupling reagent [39]. These oligomers exhibited viscosities 15 times greater than F127 alone at 37°C.

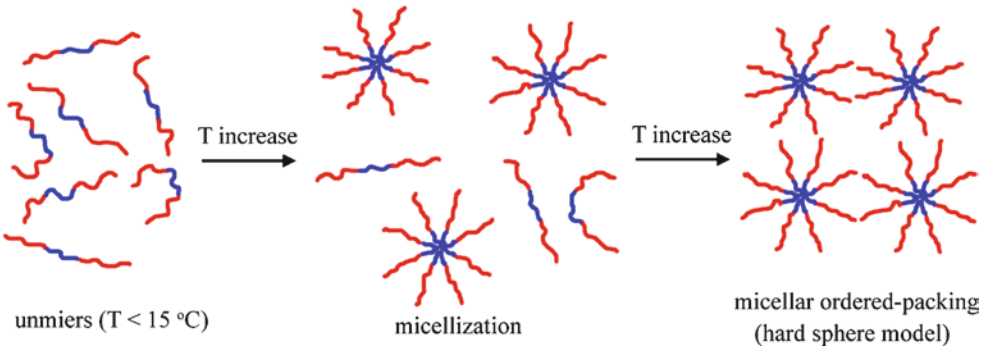


Fig. 1. Schematical illustration for the phase transition of PEO–PPO–PEO aqueous solution in response to temperature.

The in vitro release of an antirestenosis drug (RG-13577) from a 30 wt% p[F127]$_4$ hydrogels persisted over 40 days; in contrast, only 7 days from a 30 wt% F127 hydrogels. A series of biodegradable multiblock Pluronics with the molecular weights of 4,000–40,000 was prepared by coupling Pluronic P85 using terephthaloyl chloride [40]. The gels duration of the multiblock Pluronic is controllable from 8 h to 4 weeks by tailoring the molecular weight.

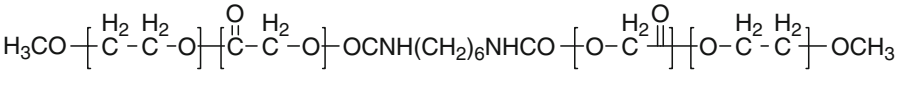
PEO/PPO alternating multiblock copolymers were synthesized by coupling PEO and PPO using carbonyl chloride or diacyl chloride [41]. The hydrogels based on the multiblock copolymers have markedly higher viscosities than Pluronic F127 hydrogels. In addition, the alternating ether-carbonate structure within the backbone renders the copolymers biodegradable. A family of pentablock copolymers composed of Pluronic F87 flanked by two short polyester blocks [poly(D,L-lactide) (PLA) or poly(ε-caprolactone) (PCL)] have been developed [42, 43]. The pentablock copolymers retained the thermoreversible sol–gel transition properties and have a lower critical micellization temperature (CMT) in comparison with F87. The in vitro release of hydrophilic procain hydrochloride (PrHy) and hydrophobic 9-(methylaminomethyl) anthracene (MAMA) was performed using PLA$_6$–F87–PLA$_6$ and PCL$_4$–F87–PCL$_4$ hydrogels, respectively. No initial burst was observed in either release curve, while the drug release from the hydrogels copolymer at 37°C was almost equal to or faster than that from the corresponding polymer solution at 25°C.

A Pluronic-based poly(ether-ester-urethane) multiblock copolymer was synthesized by coupling the oligo(ester)-Pluronic(F127)-oligo(ester) pentablock copolymers using HMDI [44]. The rheology and degradation behavior of the copolymer hydrogels are tunable by varying the polyester block length and the degree of the chain extension.

Thermogelling PEG/PLGA Amphiphilic Block Copolymers

Aliphatic polyesters, such as poly(lactic acid) (PLA), poly(ε-caprolactone) (PCL), and poly(glycolic acid) (PGA), are biodegradable and biocompatible polymers; therefore, they have potential biomedical applications as well as being environmental-friendly materials [45]. PEG-polyester copolymers have also been developed as unique polymers, such as amphiphilicity, self-assembly, permeability, biocompatibility, and biodegradability [46].

The first biodegradable thermosensitive hydrogels, based on the PEG-polyester block copolymer, were ABA-type PEG–poly(L-lactide)–PEG triblock copolymers (Scheme 3) [47]. These PEG–PLLA–PEG triblock copolymers are prepared in two steps: first, the MPEG–PLLA diblock copolymers are synthesized by ring-opening polymerization of L-lactide (LLA) using the monomethoxy PEG (MPEG, Mw = 5,000) as the macroinitiator; then the PEG–PLLA–PEG triblock copolymers are obtained by coupling MPEG–PLLA using HMDI. The molecular weight (Mw) of the PLLA block varied from 2,000 to 5,000. The concentrated PEG–PLLA–PEG solutions exhibited a gels-to-sol transition as temperatures increased.



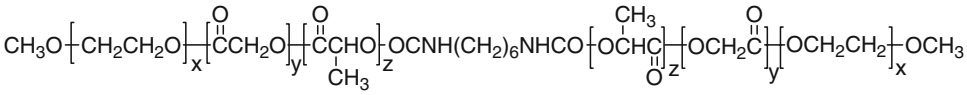
PEG-PLLA-PEG

Scheme 3. Chemical structure of PEG–PLLA–PEG.

The in vitro release of fluorescein isothiocyanate (FITC) labeled dextran from the PEG–PLLA–PEG (5,000–2,040–5,000) hydrogels was investigated. Polymer aqueous solutions containing a drug were first prepared at 45°C, and then drug-loaded hydrogels were formed by lowering the temperature to 37°C. The release of dextran from the 35 wt% gels is a constant rate up to 12 days without an initial burst, in contrast to an initial burst in the release curve of the 23 wt% gels (Fig. 3). The influence of the hydrophilic/hydrophobic balance, block length, hydrophobicity, and stereoregularity of the hydrophobic block on the gels–sol transition properties of some PEG/polyester diblock and triblock copolymers has been reported [48, 49].

In general, a longer polyester block, shorter PEG block, higher hydrophobicity, or higher crystallizability of the polyester block lead to a lower critical gelation concentration (CGC) and a higher gels-to-sol transition temperature. The PEG-poly(D,L-lactide)-PEG (PEG–PLA–PEG) triblock copolymers with PEG block lengths of 2,000 and 5,000 were recently synthesized by coupling the corresponding MPEG–PLA diblock copolymers using adipoyl chloride [50]. A similar gels-to-sol transition was observed for the concentrated copolymer solutions. The H-bonding between PEG blocks was thought to be responsible for gelation at lower temperature. A series of star-shaped PLLA–PEG block copolymers was synthesized by coupling star PLLA with monocarboxy-MPEG using dicyclohexylcarbodiimide (DCC) [51, 52]. The concentrated block copolymer solutions showed a gels-to-sol transition with increasing temperature. The gelation and gels-to-sol transition were assumed to be attributed to micellar packing and the decrease in the micellar volume caused by the partial dehydration of the PEG block, respectively.

The PEG/PLA-based block copolymers, however, exhibited UCST behavior and did not show a sol-to-gels transition at lower temperatures; this is frequently observed in Pluronic systems. The use of UCST polymers may lead to an adverse effect to some drugs and proteins and bring inconvenience into practical applications. Additionally, the long PEG chains (Mw 5,000) in such block copolymers are likely to accumulate in the body after the degradation of the polyester block. More recently, new thermosensitive hydrogels made of the PEG–poly(D,L-lactide-*co*-glycolide)–PEG triblock copolymers (PEG–PLGA–PEG, Scheme 4) containing shorter PEG blocks (MW ≤ 750) were reported [53, 54]. The PEG–PLGA–PEG aqueous solutions showed a sol-to-gels transition as well as a gels-to-sol transition with rising temperature. Moreover, the gels window covered the physiological temperature (37°C), and the CGC and sol-to-gels transition temperature could be tuned by tailoring the block length and composition as well as by adding additives. In this case, the drugs could be mixed with the polymer solution at lower temperature and incorporated in the hydrogels after administration, such as injection. The sol-to-gels transition mechanism of PEG–PLGA–PEG was investigated by temperature-dependent ^{13}C nuclear magnetic resonance (NMR) spectra and dynamic light scattering (DLS), which indicates a mechanism of micellar growth and close packing. It is noteworthy that the CGC (~16 wt%) of PEG–PLGA–PEG system does not fit the theoretic calculation result from the "hard sphere crystallization model." A "soft sphere model" was assumed for the close packing of the PEG–PLGA–PEG micelles, indicating



PEG-PLGA-PEG

Scheme 4. Chemical structure of PEG–PLGA–PEG.

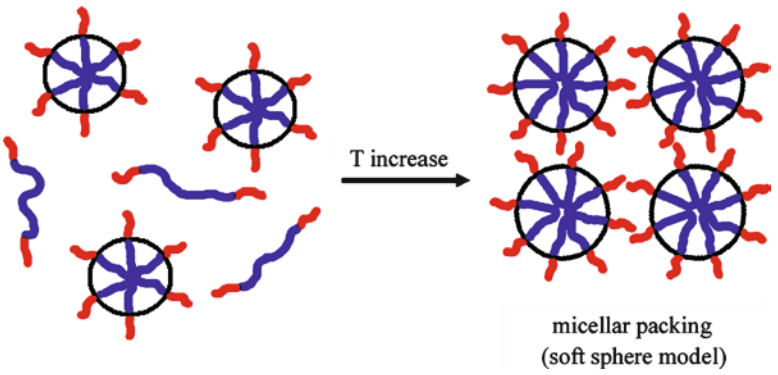
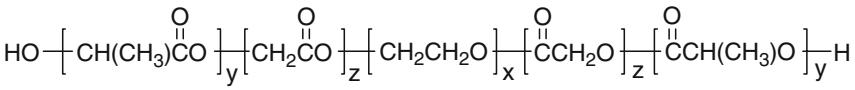


Fig. 2. Schematical illustration for the phase transition of PEG–PLGA–PEG aqueous solution in response to temperature.

marked micellar phase-mixing and overlapping for the later system (Fig. 2). Additionally, the much longer duration of the PEG–PLGA–PEG hydrogels in comparison with the Pluronic hydrogels may be attributed the fact that the dissolution of the PLGA hydrophobic cores is much more difficult than that of the PPO cores. The upper gels-to-sol transition was proved to be due to the breakage of micellar structure caused by partial dehydration of the PLGA and PEG block [54, 55]. In a separate study, the gelation behavior of the PEG–PLGA–PEG (750–3,500–750) triblock copolymer aqueous solutions was studied by using DLS, rheology, SANS, and differential scanning calorimetry (DSC) [56, 57]. A mechanism of macroscopic liquid–liquid phase separation was proposed to be responsible for the gelation. After the 33 wt% aqueous solution of PEG–PLGA–PEG (550–2,810–550) was subcutaneously injected into rats, a transparent hydrogels was formed in situ [58]. The gels exhibited good mechanical strength and the gels duration was over 1 month. The difference between the duration of the PEG–PLGA–PEG hydrogels and that of the Pluronic gels (1 day) was attributed to different erosion mechanisms. The latter disintegrated through surface erosion while the former through a degradation process. The PEG-rich components were lost preferentially during the degradation of the PEG–PLGA–PEG hydrogels. The in vitro drug release behavior of the PEG–PLGA–PEG hydrogels was evaluated by using ketoprofen and spironolactone as the hydrophilic and hydrophobic model drugs, respectively [59]. The release of ketoprofen showed a first-order release trace over 2 weeks, indicating a diffusion-controlled mechanism. In contrast, the release of spironolactone exhibited an S-shaped release profile over 2 months, suggesting a combined process composed of the initial diffusion-controlled process followed by the degradation-dominated process. The release rate was reduced with increasing the hydrophobic PLGA block length. The drug-contained PEG–PLGA–PEG aqueous solutions were directly instilled into the bladder of normal adult female Sprague–Dawley (SD) rats or cyclophosphamide-induced cystitis rats [60]. The in-situ formed hydrogels exhibited a sustained release profile, significant efficacy, and less systemic adverse effects. In addition, controlled gene delivery systems were also prepared by the PEG–PLGA–PEG hydrogels [61, 62]. The plasmid DNA (pDNA) was released in a zero-order profile for 12 days [61]. Most of the released pDNA were proven to maintain a supercoiled conformation at 12 days, even though the pH in the hydrogels markedly decreased from 7.4 to around 4 after incubation for 12 days caused by the degradation of the PLGA block. When the hydrogels loaded with luciferase pDNA were administered to the skin wounds of CD-1 mice, the expression of luciferase exhibited the maximum at 24 h and then decreased markedly. After administration to the

skin wounds of genetically diabetic mice, the hydrogels containing plasmid TGF-β1 showed significantly higher levels of reepithelialization, cell proliferation, and collagen organization, as compared either with the commercial wound dressing, Humatrix®, or with the plasmid TGF-β1-loaded Humatrix® [62].

The thermosensitive hydrogels based on the BAB-type PLGA–PEG–PLGA triblock copolymers was subsequently developed (Scheme 5) [63, 64]. The synthesis of PLGA–PEG–PLGA was simpler than that of PEG–PLGA–PEG, because the coupling procedure using HMDI could be avoided in the former synthesis. PLGA–PEG–PLGA also exhibited a reversible sol–gel–sol transition with increasing temperature, and the phase diagram was influenced by the block length and composition as well as by additives [63, 65]. PLGA–PEG–PLGA showed marked lower CGC and lower sol-to-gels transition temperature in comparison with PEG–PLGA–PEG, suggesting different gelation mechanisms for them. At a lower temperature, some interconnected micelles composed of PLGA–PEG–PLGA are formed in the aqueous solution, but no stable network is formed, due to the less hydrophobicity of the PLGA blocks at lower temperatures. As the temperature is increased to LCGT, an interconnected micelle network is formed and gelation occurs (Fig. 3). The release behaviors of proteins and several conventional drugs from the PLGA–PEG–PLGA (1,500–1,000–1,500) hydrogels (commercially available as ReGel®) were studied [64]. The in vivo degradation of ReGel® after subcutaneous injection into rats was over 4 weeks. ReGel® showed significant solubilization and stabilization to the hydrophobic drugs, such as paclitaxel and cyclosporin A. The in vitro release of paclitaxel from the ReGel® exhibited a diffusion-controlled release profile in the initial 2 weeks, followed by a combined diffusion/degradation process for about 5 weeks. In contrast, the in vitro release of paclitaxel from the Pluronic F127 hydrogels was complete in around 1 day. The in vivo distribution of paclitaxel was monitored after an intratumor injection of ReGel® containing paclitaxel and [^{14}C] paclitaxel. The C-14 levels in tumors were reduced slowly over 6 weeks. The elimination of C-14 was mainly through the feces and urine, with less than 0.1% being distributed to other organs. The ReGel®/paclitaxel showed higher antitumor efficacy and



PLGA-PEG-PLGA

Scheme 5. Chemical structure of PLGA–PEG–PLGA.

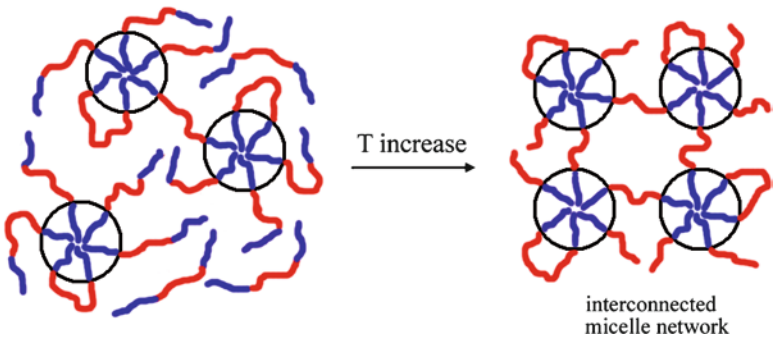


Fig. 3. Schematical illustration for the phase transition of PLGA–PEG–PLGA aqueous solution in response to temperature.

lower drug-related adverse effects, as compared with the maximum tolerated systemic dose of the commercial paclitaxel product (Taxol®). In addition, the sustained releases of some proteins, such as porcine growth hormone (pGH), glycosylated, insulin, and recombinant hepatitis B surface antigen (rHBsAg), were investigated and discussed. Separate studies on the release of insulin from ReGel® were reported [66, 67]. The in vitro release of insulin from ReGel® showed a zero-order release trace without initial burst [66]. The insulin with 0.2 wt% zinc obtained a higher release rate, and was almost released completely over 15 days. The in vivo insulin level was maintained over 15 days after a subcutaneous injection of ReGel®/0.2 wt% Zn-insulin into Sprague–Dawley (SD) rats (Fig. 5). The insulin released from Regel® was confirmed to be bioactive after being injected into Zucker Diabetic Fatty (ZDF) rats [67]. The blood glucose concentration in the ZDF rats was reduced to normal level during the insulin release period. Regel® also showed sustained release of the incretin hormone glucagons-like peptide-1 (GLP-1), which was a very useful drug for type 2 diabetes but exhibited extremely short plasm half-life due to rapid degradation [68]. The in vitro release of zinc-complexed GLP-1 from Regel® exhibited almost a linear release profile over 2 weeks without initial burst. After one subcutaneous injection of zinc-complexed GLP-1/Regel® into rats, the plasma GLP-1 level was maintained significantly higher than that of the control group for about 2 weeks, and the insulin level induced by GLP-1 stayed at a higher level over 2 weeks, leading to the obvious reduction of the blood glucose concentration during the same period.

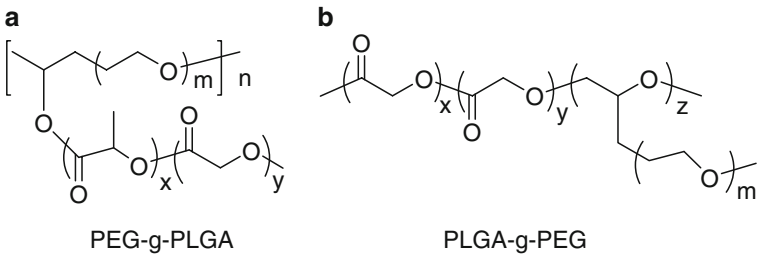
The effects of composition and sequential distribution on the phase-transition and drug release behavior of the PLGA/PEG block copolymers were investigated. A series of poly(D,L-3-methylglycolide)–PEG–poly(D,L-3-methylglycolide) triblock copolymers (PMG–PEG–PMG) was synthesized by using D,L-3-Methylglycolide (MG) as the cyclic monomer, leading to the well-defined alternating LA/GA sequence in the PMG blocks [69]. The sol–gel transition temperature of PMG–PEG–PMG was higher than that of ReGel®, due to less hydrophobicity of the PMG block with higher GA content (50 mol%). The effect of composition of PLGA–PEG–PLGA on the drug release behavior was investigated [70]. The results indicated that the LA/GA ratio exhibited less effect on the release of a diffusion-controlled stage, but markedly affected the release of a degradation-dominative stage. The phase-transition behavior of the PEG/PLGA copolymers can be also tuned by modifying the polymer structure. A series of PLGA–PEG–PLGA end-capped with small alkyl groups (acetate or propionate) was prepared [71, 72]. The CGC and CGT of the alkyl-capped triblock copolymers were markedly lower than those of the unmodified triblock copolymers, and decreased with increasing the length of the terminal alkyl groups [72]. It was also found that the CGC of the end-modified triblock copolymers was significantly affected by the degree of end-modification [73]. A micellar network caused by the micellar hydrophobic aggregation was proposed to be responsible for the gels formation [72].

Thermogelling Star-Shaped and Graft PEG/PLGA Amphiphilic Copolymers

The topology structure is known to affect the polymer properties and self-assembly behaviors; therefore, developing PEG-polyester amphiphilic copolymers with different structures may be interesting. In addition, because the MW of the PEG block is limited due to its nonbiodegradability, there is also a limitation of molecular weight for the PEG–PLGA–PEG triblock copolymers [74]. Accordingly, PEG–PLGA amphiphilic copolymers with nonlinear architectures, including star-shaped and graft structures, have been developed. Three-arm and four-arm star-shaped PLGA–PEG block copolymers were prepared by coupling 3-arm and 4-arm

PLGA with carboxyl terminated MPEG (M_n = 550) [75]. The concentrated PLGA–PEG star-shaped block copolymers aqueous solutions displayed sol–gel–sol transitions with increasing temperature. The CGCs of the star-shaped block copolymers were higher than those of the PEG–PLGA–PEG triblock copolymers, and the CGC and CMT decreased with increasing the PLGA block length. The in vitro and in vivo release of Doxorubicin (DOX) from the 4-arm star-shaped copolymer hydrogels was investigated [76]. The in vitro release of DOX showed sustained profiles. After the subcutaneous injections of the DOX-loaded polymer solutions into tumor-bearing mice, the growth of tumor was significantly suppressed. Additionally, the antitumor efficacy was also found to be affected by the PLGA block length.

PEG-*g*-PLGA graft copolymers with hydrophilic backbones were synthesized by the ring-opening polymerization of LA and GA using PEG with hydroxyl pendant groups [77]. The schematically chemical structure of PEG-*g*-PLGA is shown in Scheme 6a. The PEG-*g*-PLGA graft copolymer solutions exhibited sol–gel–sol transitions at concentrations above 16 wt%. The hydrogels remained a gels state for 1 week at physiological conditions. PLGA-*g*-PEG graft copolymers with hydrophobic backbones were also developed by the one-step ROP of LA, GA, and epoxy-terminated PEG (Scheme 6b) [74, 78]. After a subcutaneous injection of the PLGA-*g*-PEG aqueous solution into a rat, a round shaped gels was formed in situ [78]. In comparison with 1 week for the PEG-*g*-PLGA hydrogels, the PLGA-*g*-PEG hydrogels persisted more than 2 months in vivo. The sol–gel transition temperature could be adjusted within a range of 15–45°C by changing the number of PEG grafts and the composition of the copolymer [79], or by mixing two PLGA-*g*-PEG copolymers with different compositions [78]. Based on the IR, ^{13}C NMR, small-angle neutron scattering (SANS), and rheological studies, PEG dehydration is assumed to be the major driving force for the phase transition [78]. After one injection of the insulin-loaded PLGA-*g*-PEG solution into a diabetic SD rat, the blood glucose level was adjusted to maintain normal level for 16 days, caused by the sustained release of insulin from the hydrogels [80]. Interestingly, the normal blood glucose level could be controlled from 5 to 16 days by using the hydrogels based on the PEG-*g*-PLGA/PLGA-*g*-PEG blends. In addition, the biodegradable PLGA-*g*-PEG hydrogels containing chondrocyte cell showed a superior efficacy in cartilage defect repairing, as compared with the nonbiodegradable PNIPAM-*co*-PAA/hydroxyapatite collagen sponge.

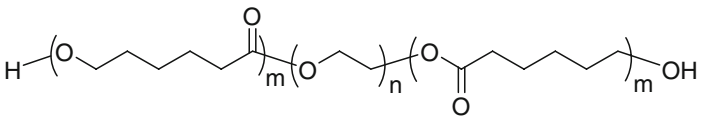


Scheme 6. Chemical structures of (**a**) PEG-*g*-PLGA and (**b**) PLGA-*g*-PEG.

Thermogelling PEG–PCL Amphiphilic Copolymers

Many amphilic copolymers composed of PEG and other biodegradable aliphatic polyesters also showed a thermoreversible sol–gel transition in the aqueous solutions. Different PEG-polyester diblock copolymers, including PEG–PCL, PEG–poly(δ-valerolactone) (PEG–PVL), PEG–PLLA, and PEG–PLGA, with well-defined structure were synthesized by living ring-

opening polymerization [81]. The concentrated aqueous solutions of the diblock copolymers (PEG≥2,000) showed a gels-to-sol transition with increasing temperature. The gels-to-sol transition was highly affected by the block length and the hydrophobic nature of the polyester block [81, 82]. When the 23 wt% PEG–PCL (2,000–2,300) aqueous solution was heated to 42°C and then injected into a mouse, it formed a gels immediately at normal body temperature [83]. The hydrogels maintained in vivo for 1 month, due to the slow degradation of the PCL block. In subsequent reports, PEG-polyester diblock copolymers with shorter PEG blocks (Mw = 750), including PEG–PCL, PEG–poly(ε-caprolactone-*co*-trimethylene carbonate) (PEG–P(ε-CL-*co*-TMC)), and PEG–poly(ε-CL-*co*-1,4-dioxan-2-one) (PEG–P(ε-CL-*co*-DO)), were prepared [84, 85]. It was claimed that the slow degradation of these copolymers did not lead to an acidic environment, which is often observed with the rapid degradation of PLLA and PLGA. In addition, these PEG-polyester diblock copolymers with shorter PEG length (Mw = 750) display a sol–gel–sol transition as temperatures increased; the gels window was highly dependent on the polyester block length. Contrary to the PEG/PLGA block copolymer systems, the lower sol-to-gels transition temperature of the PEG–PCL systems is markedly influenced by polymer concentration as compared with almost no concentration-dependence for the upper gels-to-sol transition temperature. As the PEG–PCL (750–2,440) solution loading rat bone marrow stromal cells (rBMSC) and dexamethasone was subcutaneously injected into SD rats, a gels formed in situ and maintained its integrity for over 4 weeks [86]. Histological analysis indicated that the in-situ formed scaffolds were biocompatible and accelerated the bone formation. The in vitro release of FITC-labeled bovine serum albumin (BSA–FITC) from the PEG–PCL (750–2,490) hydrogels exhibited a sustained profile over 20 days [87]. Even though an initial burst was observed, the release of BSA–FITC persisted over 30 days after the PEG–PCL (750–2,490) solutions containing BSA–FITC were subcutaneously injected into SD rats.



PCL-PEG-PCL

Scheme 7. Chemical structure of PCL–PEG–PCL.

Recently, a series of ABA- and BAB-type triblock copolymers consisting of PEG and PCL, i.e., PEG–PCL–PEG and PCL–PEG–PCL, was prepared (Scheme 7) [88, 89]. Both types of triblock copolymers display a clear sol-to-gels-to-turbid sol transition as the temperature increases from 10 to 60°C. Light scattering and ^{13}C NMR studies indicated that the sol-to-gels transition at lower temperature followed the micellar association mechanism; the upper gels-to-sol transition was due to the breakage of the micellar core-shell structure. Similar to the previous PEG–PCL diblock copolymer system, the polymer concentration only significantly influenced the lower sol–gel transition temperature of PCL–PEG–PCL system. Both types of triblock copolymers lyophilize into a powder form, and easily redissolve in water at lower temperatures. In contrast to PEG–PCL–PEG, the PCL–PEG–PCL has a wider gels domain and a higher gels modulus. However, the concentrated PCL–PEG–PCL aqueous solution (20 wt%) is not stable and turns into an opaque gels in 1 h at room temperature [90]. The Raman spectra, X-ray diffraction (XRD), DSC, and polarized optical microscope (POM) studies clearly indicate that the slow gelation at room temperature is attributed to the crystallization of the copolymer, which is quite different from the micellar aggregation mechanism

of the thermo-induced gelation at 37°C. The unstable PCL–PEG–PCL solution would be impractical for application. Therefore, the above PCL–PEG–PCL (1,000–1,000–1,000) was coupled, by terephthaloyl chloride, to fabricate PEG–PCL multiblock copolymers. The 20 wt% aqueous solution of the PEG/PCL multiblock copolymer has a sol–gel–sol transition as temperature increased and exists as a stable transparent solution at room temperature, which is convenient for drug delivery applications. Based on the ^{13}C NMR and DLS studies, the gelation of the multiblock copolymer aqueous solution was thought to be driven by the increase in polymer–polymer attraction.

Thermogelling PEG-Based Amphiphilic Multiblock Copolymers

In addition to PEG–PCL multiblock block copolymers above, the thermo-dependent phase behavior is also observed in other PEG-based poly(ether ester) multiblock copolymer systems. A series of PEG/PLLA multiblock copolymers, synthesized by the coupling reaction between dicarboxylated PLLA and PEG [91], has sharp LCST transitions in aqueous solution. This copolymer matrix loaded with basic fibroblast growth factor has significant wound healing activity [91, 92]. A range of PEG/PLLA multiblock copolymers with lower PEG Mw were prepared by coupling PEG (Mw = 600) and PLLA (Mw = 1,100–1,500) using succinic anhydride [93]. The PEG/PLLA multiblock copolymer solutions exhibit a sol–gel–sol transition with increasing temperature and the maximum gels modulus was displayed at around body temperature. The transition temperature and gels window were affected by the PLLA block length and PEG/PLLA ratio. Compared with the PEG/poly(D,L-lactide) (PEG/PLA) multiblock copolymer, the PEG/PLLA multiblock copolymer exhibited a lower CGC, a lower sol-to-gels transition temperature, and a broader gels window [94]. The isotactic arrangement of the methyl groups within the PLLA blocks developed a strong aggregate of the polyester blocks. It is noteworthy that the phase-transition of the above PEG/PCL (or PEG/PLA) multiblock copolymers could be changed to a UCST manner (gels-to-sol) by increasing the PCL (or PEG) block length, probably due to the increase in crystallizing ability of the PCL block [95, 96] (or the increase of interactions between PEG blocks [97]). A soft thermosensitive hydrogels made of PEG–sebacate (PEG–SA) multiblock copolymers was prepared by simple condensation polymerization [98]. When the 25 wt% aqueous solution of the PEG–SA multiblock copolymer was heated from room temperature to 37°C, a very soft gels was formed with a gels modulus less than 5 Pa. However, the gels retained its integrity for over 3 weeks. The release of hydrophilic FITC-dextran from the poly(PEG–SA) hydrogels indicated a constant rate during 5–24 h without an initial burst. A series of biodegradable poly(ether ester urethane) multiblock copolymers consisting of poly[(R)-3-hydroxybutyrate] (PHB), PEG, and poly(propylene glycol) (PPG) was reported very recently [99]. The poly(ether ester urethane) aqueous solutions underwent a sol–gel–sol transition with increasing temperature from 4 to 80°C. Notably, the poly(ether ester urethane) solutions showed a very low CGC ranging from 2 to 5 wt% depending on the composition and block lengths. An associated micellar packing was assumed to be responsible for the gelation.

pH- and Thermo-Sensitive PEG–Polyester Amphiphilic Copolymer Hydrogels

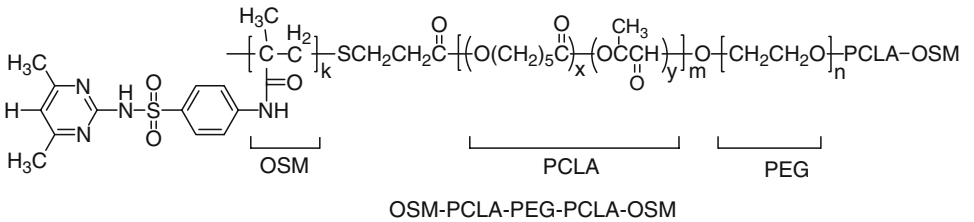
The in-situ gelling PEG/polyester amphiphilic copolymers have shown potential applications in the injectable drug and cell delivery systems. There are some limitations in the PEG–polyester systems. For example, when a thermosensitive polymer solution is

injected into the body, the increase in temperature by the body can cause a sol–gel transition within the needle and can plug the needle. However, the PEG–polyester amphiphilic copolymers, such as PLGA–PEG–PLGA, are nonionic systems, in which the drugs and proteins are loaded by hydrophobic association and/or simple physical encapsulation. The release of hydrophilic drugs or proteins from these systems is expected to be a diffusion process, and so difficult to obtain desirable profiles. Additionally, the reconstitution problem of the PEG–PLGA systems is a disadvantage. The PEG–PLGA copolymers need to be stored in solution, and the reversible gels-to-sol transition with decreasing temperature is rather slow. To overcome the above drawbacks, the pH-sensitive moieties are introduced into the PEG-polyester amphiphilic systems.

PEG-Based Amphiphilic Copolymers Modified by Anionic Weak Polyelectrolytes

The pH-sensitive polymers are classified as acidic weak polyelectrolytes and basic weak polyelectrolytes. Corresponding to the pH variation range in vivo, weak polyelectrolytes with the pK_a between 3 and 10 are suitable candidates for biomedical applications [100]. Representative acidic pH-sensitive polymers are based on the polymers containing pendant carboxylic groups, such as poly(acrylic acid) (PAA) [101] and poly(L-glutamic acid) (PLG) [102], and polymers containing sulfonamide groups [103]. Both PAA and PLG show continuous transitions rather than sharp transition in response to the pH change. The PAA and PLG systems were modified by hydrophobic groups, such as propyl [104] and benzyl groups [105], to obtain sharper transition behavior and smaller pH-responsive ranges. Compared with the PAA and PLG systems, the sulfamethazine oligomers exhibit very sharp pH-dependent transitions within a narrow pH range around pH 7.4 [103]; therefore, sulfamethazine oligomers were introduced into the PEG-polyester amphiphilic systems.

pH-sensitive sulfamethazine oligomers (OSM) were introduced to both ends of poly(ε-CL-*co*-LA)–PEG–poly(ε-CL-*co*-LA) (PCLA–PEG–PCLA) to create a pH- and temperature-sensitive pentablock copolymers (OSM–PCLA–PEG–PCLA–OSM, Scheme 8) [106, 107]. The thermo-induced sol–gel–sol transition of the parent PCLA–PEG–PCLA is not affected by the pH changes between 7.2 and 8.0 (Fig. 4a). In contrast, the concentrated (15 wt%) OSM–PCLA–PEG–PCLA–OSM aqueous solution exhibited a thermoreversible sol–gel–sol transition only at a pH below 8.0, and the gels window became broader from pH 7.8 to 7.2 (Fig. 4b). The gels window can also be tuned wider by increasing the PCLA/PEG ratio. The sol–gel transition of the pentablock copolymer solutions can be tailored by varying the PEG block length, PCLA/PEG ratio, the OSM molecular weight, and the polymer concentration. An interconnected-micelle association mechanism was proposed for the gelation of the pentablock copolymer solution, as schematically illustrated in Fig. 5. At lower temperature (15°C) or/and



Scheme 8. Chemical structure of OSM–PCLA–PEG–PCLA–OSM.

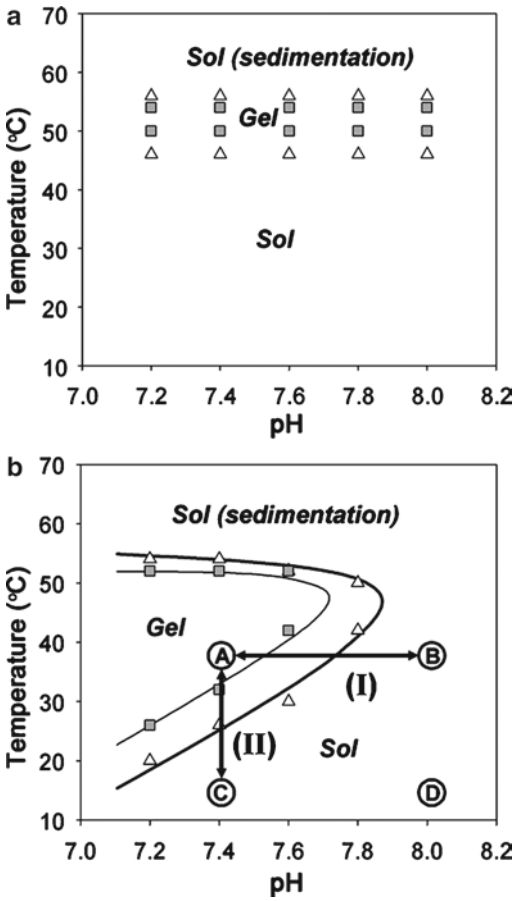


Fig. 4. Phase diagram of block copolymers in buffer solution. M_n of PEG = 1,750; concentration, 15 wt%; PEG/PCLA weight ratio, 1/1.89 (■), 1/2.08 (Δ). (**a**) PCLA–PEG–PCLA solution (**b**) OSM–PCLA–PEG–PCLA–OSM solution. (A) pH 7.4, 37°C; (B) pH 8.0, 37°C; (C) pH 7.4, 15°C; (D) pH 8.0, 15°C. Reproduced with permission from [106]. Copyright 2005 American Chemical Society.

higher pH (pH 8.0), the block copolymer solution exists as a sol state, due to less hydrophobicity of the PCLA–OSM block. In contrast, at 37°C and pH 7.4, a macrolattice composed of interconnected micelles was formed by the strong hydrophobic associations between the PCLA–OSM blocks. The 15 wt% OSM–PCLA–PEG–PCLA–OSM aqueous solution at pH 8.0 can be easily injected into the buffer solution at 37°C even with a long guide catheter. The state of the polymer solution after injection is highly sensitive to a small change in pH. A stable and compact gels is formed immediately in the pH 7.4 buffer solution, whereas the polymer dispersed quickly in the pH 8.0 buffer solution. In addition, the pentablock copolymer hydrogels exhibited a rapid gels-to-sol transition after temperature is lowered. The pentablock copolymer hydrogels maintains its integrity in the pH 7.4 PBS buffer solution at 37°C for over 2 weeks and has a slower degradation rate in comparison with the hydrogels made of the parent PCLA–PEG–PCLA triblock copolymer. Notably, in contrast to the marked pH decrease from 7.4 to 2.2 in the PCLA–PEG–PCLA triblock copolymer hydrogels, the pH maintained at round 5.5 in the pentablock copolymer hydrogels after incubation at 37°C for 1 month [108]. This was believed to be attributed to the proton sponge effect of the OSM block.

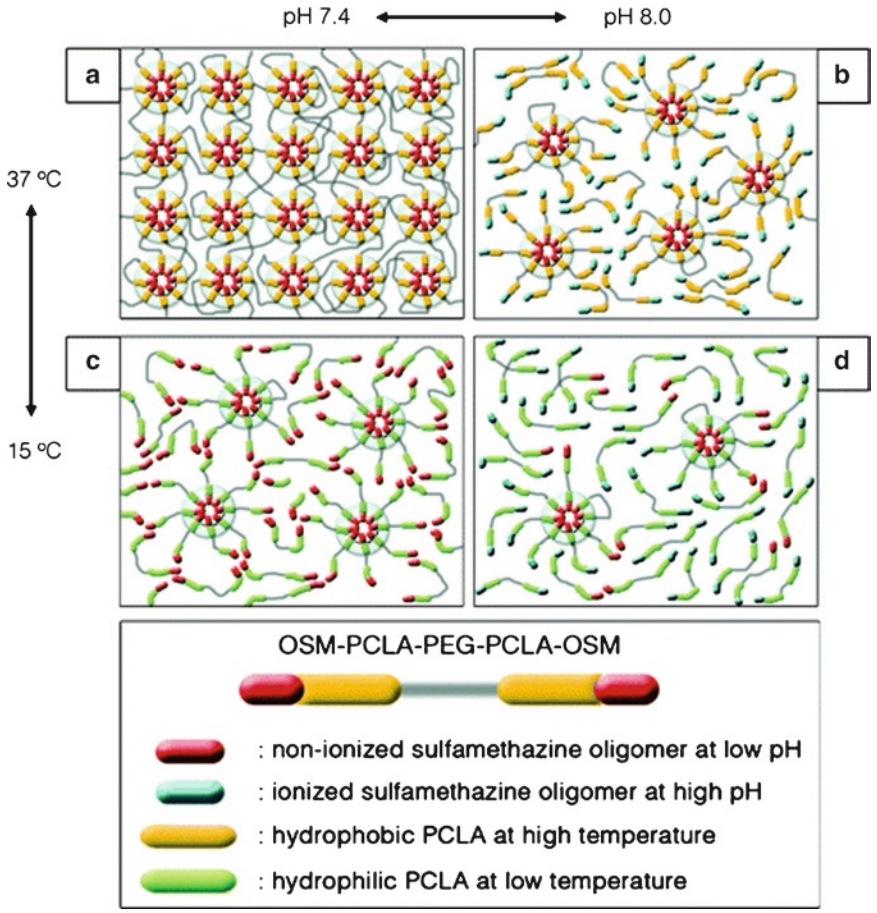


Fig. 5. Schematic diagram of the sol–gel mechanism of the pH and temperature sensitive block copolymer solution. (**a**) pH 7.4, 37°C; (**b**) pH 8.0, 37°C; (**c**) pH 7.4, 15°C; (**d**) pH 8.0, 15°C. Reproduced with permission from [106]. Copyright 2005 American Chemical Society.

A gels immediately forms in vivo after the OSM–PCLA–PEG–PCLA–OSM aqueous solution (20 wt% in PBS at pH 8.0) is subcutaneously injected into rats (Fig. 6) [108]. OSM–PCLA–PEG–PCLA–OSM has good in vitro biocompatibility. Although the pentablock copolymer hydrogels brought a typical acute inflammation within 2 weeks in vivo, chronic inflammation was not observed during the first 6 weeks. The in vitro release of paclitaxel (PTX) from the OSM–PCLA–PEG–PCLA–OSM hydrogels indicated a zero-order release profile over 20 days, which was independent of the initial loading amount [109]. The subcutaneously injected pentablock copolymer hydrogels containing PTX showed significant antitumor efficacy. OSM–PCLA–PEG–PCLA–OSM copolymers, as well-defined pentablock copolymers, were prepared by the polymerization of sulfamethazine methacrylate monomer using Br–PCLA–PEG–PCLA–Br as the ATRP macroinitiator. A series of OSM–poly(ε-CL-*co*-GA)–PEG–poly(ε-CL-*co*-GA)–OSM pentablock copolymers (OSM–PCGA–PEG–PCGA–OSM) was subsequently reported [110, 111]. The OSM–PCGA–PEG–PCGA–OSM aqueous solutions also exhibited a thermoreversible sol–gel–sol transition with the gels window depending

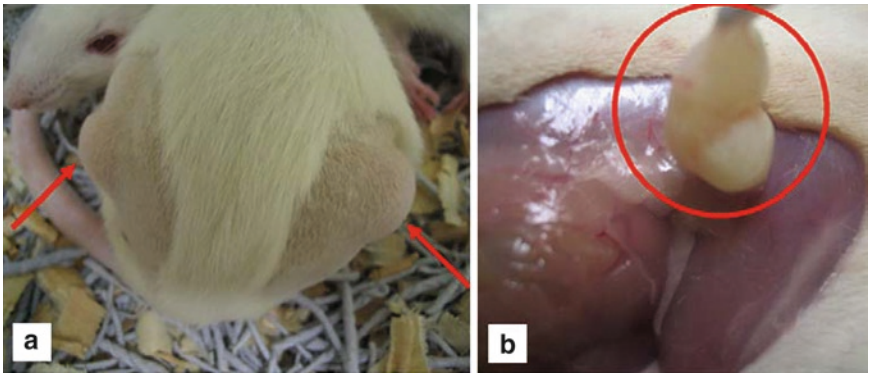


Fig. 6. Spontaneous forming hydrogels of a OSM–PCLA–PEG–PCLA–OSM block copolymer solution (20 wt% in PBS at pH 8.0). About 200 μl of the block copolymer solution was subcutaneously injected into SD rats with a syringe needle (**a**), and the resulting hydrogels was isolated by tweezers after only 10 min (**b**). Reproduced with permission from [108].

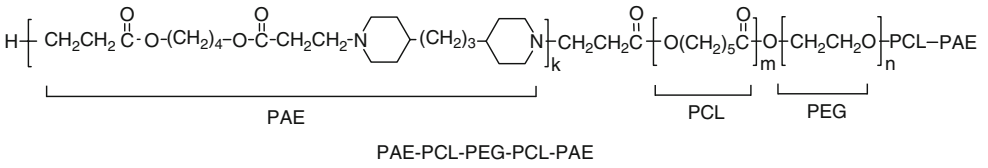
on pH. The sol–gel transition phase diagram can be controlled by changing the block length and PEG/PCGA ratio. The in vitro release of PTX from the OSM–PCGA–PEG–PCGA–OSM hydrogels also followed near zero-order kinetics without an initial burst. Compared with the OSM–PCLA–PEG–PCLA–OSM hydrogels, the OSM–PCGA–PEG–PCGA–OSM hydrogels have a faster release rate, due to the faster degradation of the PCGA block.

PEG-Based Amphiphilic Copolymers Modified by Cationic Weak Polyelectrolytes

In addition to the anionic weak polyelectrolytes, the cationic weak polyelectrolytes are of interest due to their pH-sensitivity and the ability to form electrostatic interaction with ionic DNA and proteins. Some therapeutic proteins, such as insulin and human growth hormone (hGH), are ampholytes and have net negative charges in aqueous solutions at physiological pH (7.4) because of their relatively low isoelectric points (~5.3) [112]. The cationic segments within the hydrogels network are expected to form electrostatic interactions with the negatively charged proteins at pH 7.4, and may lead to more sustained and controllable release profiles.

Typical examples of basic polyelectrolytes include poly(tertiary amine methacrylate), such as poly(2-(dimethylamino)ethyl methacrylate) (PDMAEMA) and poly(2-(diethylamino) ethyl methacrylate) (PDEAEMA) [113], poly(2-vinylpyridine) (P2VP) [114] and poly(β-amino ester) (PAE) [115]. Some poly(tertiary amine methacrylate)s and P2VP exhibit sharp hydrophilic–hydrophobic transitions at around physiological pH; however, they are nonbiodegradable and, therefore, have limited applications as in-situ forming hydrogels. In contrast, a series of biodegradable cationic polyelectrolytes, poly(β-amino ester) (PAE), was developed recently for gene delivery [115]. PAE exhibits a sharp hydrophilic–hydrophobic transition in aqueous solution at pH around 6.5 [115]. It was established that PAE was noncytotoxic and could be degraded into nontoxic small molecular byproducts. In addition, the partially positively charged PAE forms electrostatic complexes with negatively charged pDNA at physiological pH (pH 7.2) [116]. A series of PEG–PCL–PAE triblock copolymers was synthesized

by Michael addition of piperazine, MPEG–PCL acrylate, and 1,6-hexanediol diacrylate [117]. When the pH was below 6.0, the 30 wt% triblock copolymer aqueous solution existed as a sol state within the temperature range of 0–60°C, whereas it showed a gels-to-sol transition upon heating at a pH above 6.0. The gels–sol phase-diagram could be tailored by varying the block length and composition. A PAE–PCL–PEG–PCL–PAE pentablock copolymer was subsequently prepared by the Michael addition of 4,4′-trimethylene dipiperidine, PCL–PEG–PCL diacrylate, and 1,4-butandiol diacrylate (Scheme 9) [118]. These pentablock copolymers have sol-to-gels-to-sol (sedimentation) transitions with increasing temperature (Fig. 7). In contrast to the OSM-based system, the gels window of PAE–PCL–PEG–PCL–PAE is observed in the pH region above the pKa of PAE. The cytotoxicity was ~100% cell viability even when the polymer concentration was increased to 100 μg/mL, indicating a good cellular compatibility. The degradation of the copolymer is a two-step process; a fast degradation of the PAE segments within about 10 days and a slower degradation of the PCL segments. The in vitro release of insulin exhibited sustained release profiles at constant rates. On loading insulin, the sol–gel phase diagram slightly shifts to lower temperature range. After a subcutaneous injection of the polymer solution containing insulin into SD rats, the elevated plasma insulin level maintained at constant level over 15 days without an initial burst for the PAE–PCL–PEG–PCL–PAE/insulin group. A marked initial bursts and shorter durations of the elevated insulin levels were observed for the PCL–PEG–PCL/insulin group and insulin solution group (Fig. 8). The sustained release behavior of the pentablock copolymer hydrogels is thought



Scheme 9. Chemical structure of PAE–PCL–PEG–PCL–PAE.

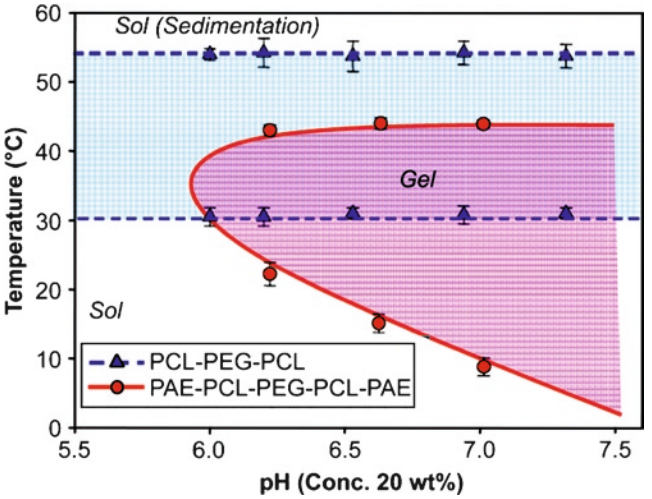


Fig. 7. Sol–gel phase diagram of triblock and pentablock copolymer solutions at 20 wt%. Reproduced with permission from [118].

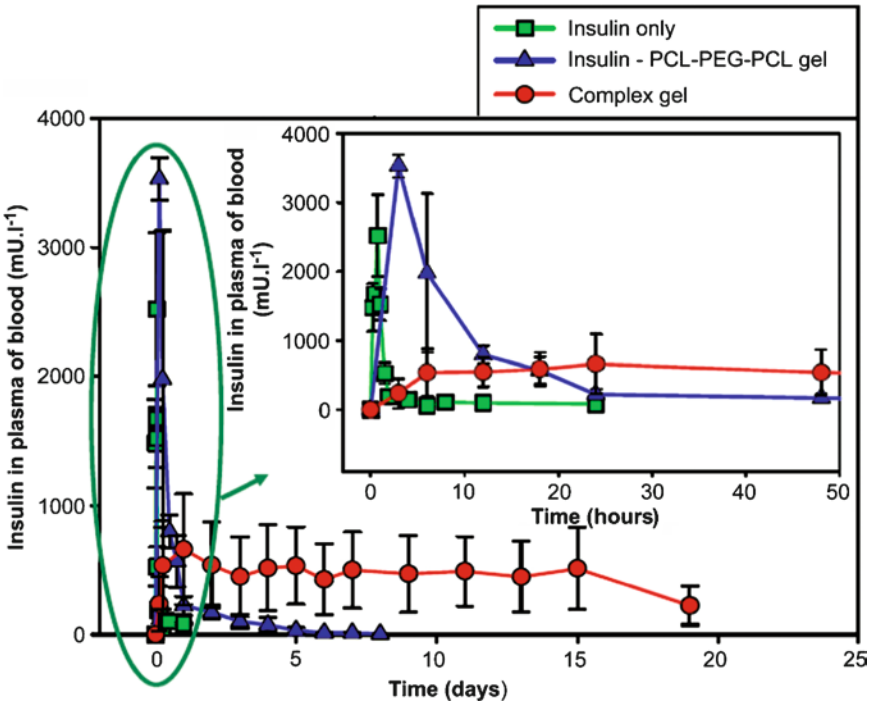
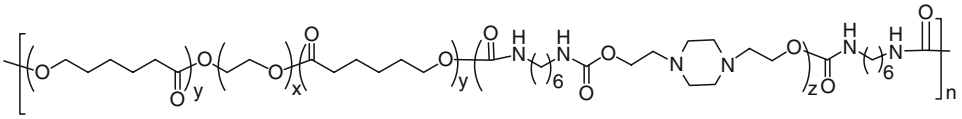


Fig. 8. Insulin release experiment in vivo. In insulin-only group, 200 mL insulin solution 0.25 mg mL^{-1} (in PBS buffer (pH 7.4) is administered by subcutaneous injection (0.05 mg insulin for each rat)). In insulin-PCL-PEG-PVL gels group, 200 mL of solution (5 mg mL^{-1} insulin in PCL-PEG-PCL solutions 25 wt%) at pH 7.0 and 10°C is subcutaneously injected (1 mg insulin for each rat). In complex gels group, 200 mL of the complexation insulin solution (5 mg mL^{-1} in PAE-PCL-PEG-PCL-PAE solutions 25 wt%) at pH 7.0 and 10°C is subcutaneously injected (1 mg insulin for each rat). (Male SD rats, error bars represent the standard deviation ($n=5$).) Reproduced with permission from [118].

to be related to the electrostatic interactions between the partially positively charged PAE blocks and the negatively charged insulin at physiological pH. The release of insulin is mainly based on the breakage of the electrostatic complex caused by the degradation of the PAE segments, indicating that a degradation-controlled process is the major mechanism for the insulin release.

A series of the novel pH- and temperature-sensitive multiblock poly(ester amino urethane) $(PCL–PEG–PCL–PAU)_n$, was synthesized by reacting together hexamethylene diisocyanate (HDI), hydroxyl-terminated PCL–PEG–PCL, and bis-1,4-(hydroxyethyl)piperazine (HEP) with the OH/NCO molar ratio of 1:1 (Scheme 10) [119]. The tertiary amino groups of the poly(amino urethane) segments act as pH-responsive moieties, while the PCL–PEG–PCL



$[PCL\text{-}PEG\text{-}PCL\text{-}PAU]_n$

Scheme 10. Chemical structure of $[PCL–PEG–PCL–PAU]_n$.

blocks act as biodegradable and temperature-sensitive segments. At a relatively high pH (7.0 or above), the multiblock copolymer aqueous solution shows a sol-to-gels-to-aggregation transition with increasing temperature. In contrast, at a lower pH (<7.0), the polymer solution exists as a sol state within the experimental temperature range. The gelation was attributed to the formation of strong micellar interactions and aggregation. After a subcutaneous injection of a 20 wt% multiblock copolymer solution into mice, polymeric hydrogels are quickly formed in situ. The in vitro release of an anticancer drug, paclitaxel, persists over a month under physiological conditions.

Summary

Poly(ethylene glycol)s with relatively low molecular weight (<5,000) are widely used in biomedical applications because they are hydrophilic, nontoxic, absent of antigenicity and immunogenicity, and can be directly excreted by the kidneys. PEG-based amphiphilic copolymers are of interest for their unique self-assembly and biocompatibility. Additionally, the PEG-based amphiphilic copolymers exhibit unique changes in micellar architecture and aggregation number in response to changes near physiological temperature as this unique amphiphilic system may retain water molecules in the network during the sol–gel phase transition. Therefore, in-situ gelling systems made of the PEG-based amphiphilic copolymers are extensively used. Aqueous solutions of these block copolymers exhibit a sol–gel transition without syneresis in response to the changes in temperature or/and pH. These systems have many advantages, such as: simple drug formulation and administration procedure, no organic solvent, site-specificity, a sustained release behavior, less systemic toxicity, and ability to deliver both hydrophilic and hydrophobic drugs. The gelation of the block copolymer systems is due to the formation of the transient (or reversible) polymer network with absorbing a large amount of water caused by the stimuli-induced physical interactions, such as micellar aggregation and packing, hydrophobic association, phase-separation, and crystallization.

The main challenges for the application of in-situ forming hydrogels include a short gelation time, appropriate gelation temperature and/or pH, appropriate mechanical strength, biocompatibility, proper persistent time, convenient practical procedure, no significant syneresis, and desirable drug release behavior. These properties depend on copolymer composition, hydrophilic/hydrophobic balance, hydrophilic/hydrophobic block length, molecular weight, and polymer architecture. In addition, drug release from the block copolymer hydrogels is based on drug diffusion and gels erosion mechanisms; therefore, the biodegradability or bio-eliminability of these block copolymers is important for in vivo applications.

Amphiphilic copolymer hydrogels with multifunctionality and multisensitivity are attractive because of their unique advantages. In contrast to the thermosensitive systems, the pH- and temperature-sensitive amphiphilic copolymer hydrogels have some practical advantages, such as no premature gelation, electrostatic interaction with some biomolecules, and easy to store. The double-responsive system is easily injected into sites deep in the body. Some in-situ gelling systems, which contain cationic groups, may form electrostatic interactions with ionic proteins and DNA under physiological conditions, leading to sustained and constant release profiles as well as environments that benefit maintaining protein and DNA stability.

Acknowledgments

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