HHS Public Access

Author manuscript

ACS Appl Mater Interfaces. Author manuscript; available in PMC 2017 August 24.

Published in final edited form as:

ACS Appl Mater Interfaces. 2016 August 24; 8(33): 21107–21133. doi:10.1021/acsami.6b00371.

Temperature-Responsive Smart Nanocarriers for Delivery Of Therapeutic Agents: Applications and Recent Advances

Mahdi Karimi[‡,§,⊥,†], **Parham Sahandi Zangabad**[#,||,§,†], **Alireza Ghasemi**[||], **Mohammad Amiri**[||], **Mohsen Bahrami**[||], **Hedieh Malekzad**[§,¶], **Hadi Ghahramanzadeh Asl**[||], **Zahra Mahdieh**[□], **Mahnaz Bozorgomid**[○], **Amir Ghasemi**[||,§], **Mohammad Reza Rahmani Taji Boyuk**[||], and **Michael R. Hamblin**[*,⊥,△,▲]

[#]Research Center for Pharmaceutical Nanotechnology (RCPN), Tabriz University of Medical Science (TUOMS), Tabriz, Iran

[||]Department of Materials Science and Engineering, Sharif University of Technology, Azadi Avenue, 14588 Tehran, Iran

[‡]Department of Medical Nanotechnology, Faculty of Advanced Technologies in Medicine, Iran University of Medical Sciences, Tehran, Iran

[§]Advanced Nanobiotechnology and Nanomedicine Research Group (ANNRG), Iran University of Medical Sciences, Tehran, Iran

[⊥]Wellman Center for Photomedicine, Massachusetts General Hospital, Boston, Massachusetts 02114, United States

[¶]Department of Chemistry, Kharazmi University of Tehran, Tehran, Iran

[□]Department of Biomedical and Pharmaceutical Sciences, Material Science and Engineering, University of Montana, Missoula, Montana 59812, United States

[○]Department of Applied Chemistry, Central Branch of Islamic Azad University of Tehran, Tehran, Iran

[△]Department of Dermatology, Harvard Medical School, Boston, Massachusetts 02115, United States

[▲]Harvard-MIT Division of Health Sciences and Technology, Cambridge, Massachusetts 02139, United States

Abstract

Smart drug delivery systems (DDSs) have attracted the attention of many scientists, as carriers that can be stimulated by changes in environmental parameters such as temperature, pH, light, electromagnetic fields, mechanical forces, etc. These smart nanocarriers can release their cargo on demand when their target is reached and the stimulus is applied. Using the techniques of nanotechnology, these nanocarriers can be tailored to be target-specific, and exhibit delayed or

[*]Corresponding Author: hamblin@helix.mgh.harvard.edu.

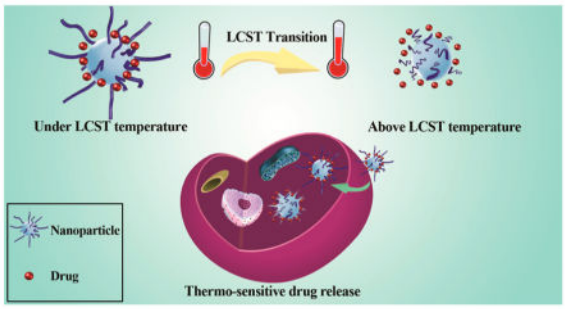
[†]**Author Contributions:** M.K. and P.S.Z. contributed equally.

Notes

The authors declare no competing financial interest.

controlled release of drugs. Temperature-responsive nanocarriers are one of most important groups of smart nanoparticles (NPs) that have been investigated during the past decades. Temperature can either act as an external stimulus when heat is applied from the outside, or can be internal when pathological lesions have a naturally elevated termperature. A low critical solution temperature (LCST) is a special feature of some polymeric materials, and most of the temperature-responsive nanocarriers have been designed based on this feature. In this review, we attempt to summarize recent efforts to prepare innovative temperature-responsive nanocarriers and discuss their novel applications.

Graphical Abstract



Keywords

smart drug delivery systems; temperature-responsive nanocarriers; LCST/UCST behavior; gene delivery; dual/multi responsive; synthesis; characterization; anticancer delivery

1. INTRODUCTION

The outstanding area of nanobiotechnology has led to many innovative breakthroughs, and has made tremendous impacts on various fields of basic and applied science ranging from chemistry and physics, to medicine and biomedical engineering. In the biomedical field several nanotechnology-based approaches have influenced the development of therapeutic drugs and pharmaceutical formulations. In particular, the increasing burden to human health caused by serious diseases (e.g., various types of cancers, coronary artery disease (CAD), etc.) has required ongoing innovations in drug delivery and drug eluting systems, many of which rely on nanotechnology. The main applications of these new drug-delivery and targeting systems have been focused on imaging agents for cancer diagnosis, and the development of new anticancer drugs and strategies.[1–9] The most utilized nanocarriers in recent years have been based on NPs including liposomes, dendrimers, polymeric NPs, metal NPs, carbon nanomaterials (e.g., fullerenes, carbon nanotubes (CNTs)), solid lipid NPs (SLNs), nanoshells and magnetic NPs, mesoporous silica NPs (MSNs), as well as novel nanostructures based on albumin, chitosan, etc.[5,10–17]

DDSs for targeted-delivery and controlled-release of drugs and other cargos can be designed to be responsive to a range of different stimuli. These stimuli can either be internal in nature (pH, redox, enzymatic activity, concentration of specific biomolecules) or external in nature

(magnetic fields, light of various wavelengths, electric fields, ultrasound, mechanical pressure, etc.).[18–23] Temperature is unique among this array of stimuli as it can be considered to be either internal (some tissues such as tumors and infections are naturally at elevated temperature) or to be external (heat is applied from outside the body). For example, hydrogels can respond to many of these stimuli by undergoing abrupt swelling of their structure leading to volume changes and consequent drug release.[24,25]

Recently, the introduction of biocompatible temperature-responsive polymeric materials that can be designed to make smart drug nanocarriers, has motivated many investigations.[26–31] Many studies have confirmed the hypothesis that drug release from these thermoresponsive polymers can be triggered by relatively small variations in temperature.[26,27,29–32] Various thermosensitive polymers can be used to prepare temperature-responsive hydrogels,[33,34] such as poly(*N*-isopro-pylacrylamide (PNIPAAm) derivatives,[35] poly(ethylene oxide)-poly(propylene oxide) (PEO–PPO) pluronic copolymers[36]), core–shell thermoresponsive NPs,[29,37] polymeric nanotubes,[28] polymeric micelles,[10,27,38] layer-by-layer (LBL)-assembled nanocapsules,[26] microbeads (MBs),[39] and elastin-like polypeptides (ELPs).[40] These nanostructures have all been utilized as thermoresponsive carriers in DDSs. Furthermore, other particles formed from polysaccharides (e.g., chitosan and hyaluronic acid (HA)) have been used to modulate the drug encapsulation and release efficiency of thermoresponsive polymeric NPs (e.g., hydrogels).[41,42] As a consequence of the temperature-sensitivity of these particles (that can respond to only very slight variations in temperature) leading to a subsequent phase transition, the encapsulated cargo can be released.[43] Thermoresponsive delivery systems can be designed according to the required application, to lead to increased cytotoxicity of the encapsulated drug,[33,38] or conversely, can be designed to limit the cytotoxicity[44] until it reaches the targeted site. Recently, some innovative multifunctional nanosystems such as thermoresponsive polymer-coated magnetic NPs (MNPs)[11] and temperature sensitive liposomes (TSLs) encapsulating MRI contrast agents (CAs)[3] have been developed for simultaneous diagnosis and therapeutic application (theranostics) for cancer treatment. In addition, thermoresponsive nanocarriers have been utilized in multi-stimuli-responsive delivery systems. For example, dual stimuli-responsive systems, e.g., temperature/pH-responsive polymeric NPs,[43] pH/thermosensitive microcontainers,[45] pH/thermosensitive core–shell nanostructures,[39] and triple-stimul-responsive systems, e.g., thermo-, pH-, and reduction-sensitive polymeric micelles[46] and glucose-, pH-, and thermoresponsive nanogels[47] have been reported so far.

Many different synthesis methods have been tested for preparing these thermoresponsive polymeric NPs, such as free radical polymerization followed by hydrolysis,[48] UV-irradiation-mediated graft copolymerization,[49] and phase separation, emulsion, and foaming.[50] Moreover, diverse methodologies have been used for characterization of the thermoresponsive polymeric NPs such as cryogenic transmission electron microscopy (cryo-TEM),[51] atomic force microscopy (AFM),[52] nuclear magnetic resonance (NMR) spectroscopy,[53,54] small-angle X-ray scattering (SAXS),[51,55,56] Fourier-transform infrared spectroscopy (FT-IR),[10] UV–vis,[46] and static and/or dynamic light scattering (SLS/DLS).[46,57]

Thermoresponsive nanocarriers have mostly been tested for the controlled delivery of drugs[58,59] and genes.[60,61] However, in recent years, thermoresponsive DDSs have been mainly used for anticancer drugs and imaging agents.[62,63] In cancer treatment, improving the efficient and specific targeting of the anticancer therapeutics to cancerous sites is a critical issue.[2] Cancerous cells and tumors have been reported to have particular physical and biological features in comparison with normal cells and tissues, including differences in morphology, permeability, elasticity, blood flow, interstitial pressure, mechanical microenvironment, expression of glycans, and metabolic autonomy. Furthermore, the pH of the cancerous microenvironment found in tumors is significantly lower (more acidic) than that of blood and normal tissues.[64–67] Targeted, smart stimulus-responsive multifunctional DDSs are of great importance due to their advantages such as lower systemic toxicity, prevention of overdosing, and the more controlled cancer therapy. Temperature, as external stimulus triggering, can be caused by a light irradiation from outside, applying magnetic fields, electric fields, or external heating.[1] Thermoresponsive intracellular release of the drugs can enhance their cytotoxicity inside the cancer tissue.[38] Furthermore, cancer cells are intrinsically susceptible to the heat, and can also be directly eradicated by means of external heating.[68]

The principal of the thermoresponsive polymer NPs (e.g., hydrogels) is based on their solubility behavior that changes in response to the alterations in the temperature. Most often, the thermoresponsive behavior is obtained by utilizing the LCST polymers by which the solubility behavior of polymeric particles is controlled by temperature changes. The solubility of thermosensitive NPs is increased by decreasing the temperature below the LCST. Subsequently, a volume phase transition and swelling of the hydrogel occurs by means of the formation of hydrogen bonds. Such polymeric NPs are called “negative thermosensitive” polymers.[34,69,70] Many studies have been conducted to develop controlled drug/gene delivery systems (DGDSs) (especially for anticancer applications) using the LCST features of polymeric particles.[62,63,71,72] However, only a relatively few studies have been done on the application of positive hydrogel polymers possessing an “upper critical solution temperature” (UCST) responsive transition.[73] In such polymers, through exploiting the changes that occur while solubilizing the hydrogels above the UCST, they start swelling.[34,69,70] Thus, by means of controlling and altering the LCST/UCST transition of the polymers, controlled delivery systems can be acquired

Thermoresponsive nanostructures are considered as useful drug carriers because of advantages such as low toxicity, being target specific, and prevention of overdoses. Polymers with LCST serve as triggers for these innovative NPs, which can be stimulated with external stimuli such as temperature and can help with release of drugs in cancerous tissues, and increase the cytotoxicity. These nanocarriers have been produced with different shapes for different applications. For example, magnetic nanocarrier-coated polymers can be carriers for MRI contrast agents, or microgels can not only be used as enzymes carriers, but also can be used to study the rheology and the crystallization of concentrated colloidal suspensions.

In this review, the most recent advances in temperature-responsive DDSs will be discussed.

2. LCST/UCST BEHAVIOR OF THERMORESPONSIVE POLYMERIC NPS

Smart polymers undergo fast and reversible structural changes in response to the specific biological stimulus, such as temperature, pH, enzyme activity, hypoxia, and reduction/oxidation (redox) potentials. In particular, the temperature-sensitive features of these intelligent polymeric systems, cause a phase transition that occurs above or below a specific temperature. Thermoresponsive polymers are generally divided into two types according to how the smart polymer reacts to changes in temperature; the first group has a lower critical solution temperature (LCST), whereas the second group has an UCST. As mentioned above, there are only a few reports of polymers that display a UCST, therefore this review mainly focuses on polymers, which display a LCST.[74–77]

Tunability, versatility in design and site specific phase transition are features of temperature responsive DDSs.[78] Among various temperature responsive polymers, PNIPAAm is the most studied polymer with a well-known reproducible LCST behavior.[79] The LCST of PNIPAAm can range from 30 to 35 °C depending on the precise solvent and chain modifications.[80] However, some researchers,[81] have reported an unusual UCST behavior. In the forthcoming section, PNIPAAm will be used to elucidate transitional behavior and delivery features of drug-loaded polymeric carriers.

Below the transition temperature of the LCST, polymers with transitional behavior are soluble and polymeric medium is swollen because of hydrogen bonds formed between molecules of water and the functional groups of incorporated polymer, making them ready to be loaded with drug molecules. When the temperature is increased beyond the LCST, a hydrophilic–hydrophobic transition occurs accompanied by a change in morphology from coil-to-globule. During this transition, the hydrogen bonds and the network collapses, and the polymer becomes insoluble, leading to volumetric shrinkage and squeezing-out of internal water molecules. This transition results in release of the encapsulated cargo from the medium.[82–87]

Shrinkage of a loaded polymer is shown in Figure 1 as a result of LCST transition. As it is obvious, the shrinkage is accompanied by release of loaded drug.[88] Although this scheme refers to a cross-linked PNIPAAm hydrogel network, but same scenario is applicable in the case of polymeric micelles and so on.[89] Originally, LCST transition is related to the nature of polymer. Regardless of carrier state, i.e., polymeric hydrogel or micelle and so on, below the LCST temperature, the polymeric medium is hydrophilic in nature, whereas above the LCST, it became hydrophobic. Increasing temperature beyond transitional temperature decreases solubility and volumetric features of carrier medium, leading to release of loaded drug.[90] On the other hand, an increase in the polymer solubility can occur when the temperature rises above the UCST, resulting in the swelling of the carrier medium.[34,70] It is worth noting that the volumetric change is reversible[79,82–84] and it is termed “swelling-shrinkage” behavior.

3. MISCELLANEOUS TEMPERATURE-RESPONSIVE NANOCARRIERS

3.1. Hydrogels

An important group of materials used in thermoresponsive carriers are hydrogels/nanogels. Thermosensitive nanogels have a volume phase transition at a specific temperature known as the volume phase transition temperature (VPTT), which causes an increase or decrease in particle size.[91] The VPTT and the phase transition temperature range are the key factors in thermoresponsive nanogels suitable for developing a smart DDS. Because the temperature at most disease sites within the body is higher than that at normal sites, the VPTT with slightly higher than biological temperature is preferred.

Hydrogels are physically or chemically cross-linked networks with a high water content.[92] After exposure to external stimulation, a variety of responses may occur in the hydrogel, such as sol–gel phase transition, swelling–shrinking behavior or hydrogel degradation. It is worth noting that these responses generally originate from the effects of specific functional groups. Because of the wide range of applications of thermoresponsive hydrogels, they have been studied more extensively than the other types. Drug/protein delivery, tissue engineering, and sensor films have been proposed as some of their applications.

Among thermally responsive hydrogels, PNIPAAm is the most often studied, because of its unique thermosensitive properties, i.e., a considerable swelling ratio and good thermal reversibility.[53] Copolymerization and grafting of PNIPAAm with other monomers, e.g., polyethyelene glycol (PEG)[81,83] and poly(ethylene oxide) (PEO),[85] are established routes to obtain new hydrogels. Cellulose derivatives,[93,94] polycaprolactone,[95] polylactic-*co*-glycolic acid (PLGA),[96] and polyurethane amide (PUA)[97,98] are other frequently studied polymers that form hydrogels.

Potential cytotoxicity and lack of biodegradability can be considered possible drawbacks of NIPAAm-based materials. Different approaches have been proposed to overcome these problems. The introduction of biodegradable segments into the polymer backbone or into the side chains, or using cleavable bonds as a cross-linking agent can all result in faster degradation and better biocompatibility.[99–101] In another study,[83] citric acid, PEG, and PNIPAAm were copolymerized. The incorporation of biocompatible polymers such as PEG facilitated degradability and biocompatibility. Additionally, citric acid could act as an antioxidant to reduce side effects in patients.

A biodegradable, specially designed PNIPAAm-*g*-PEO was synthesized using reversible addition–fragmentation chain transfer (RAFT) polymerization.[85] The hydrophilic constituent had a comb-type structure allowing facile diffusion of water molecules across the “tunnels”. This improved the responsivity to thermal conditions that was achieved in this PNIPAAm-*g*-PEO copolymer. Figure 2 shows these tunnels that provide facile water diffusion.

Some details concerning temperature-responsive hydrogels, such as LCST/UCST behavior, synthesis method, drug loading, and release profile are listed in Table 1.

3.1.1. Transition Temperature of Hydrogels—Hydrogels that have a transition temperature near to physiological temperature conditions can be used for in situ hydrogel-mediated drug delivery, with many advantages, i.e., they can be used as nonsurgical drug depots and for sustained delivery of drugs and proteins to susceptible living cells.[81,82,106]

The transition temperature can be readily tuned by the copolymerization conditions and by varying the content of repeating units in the copolymer, for instance, in a study the content of acrylic acid (AAc) in NIPAAm copolymer was indicated to be varied.[82] The LCST temperature could be changed by changing the AAc content, and at 1.8% AAc an appropriate hydrogel for physiological conditions (i.e., temperature in the range of 37–38 °C) was obtained. Because of the carboxylic groups in AAc, pH-sensitivity could also be obtained. Therefore, this dual-responsive hydrogel could be a candidate for a hydrophilic drug carrier. A favorable poly(*N*-acryloyl glycinamide) (PNAGA)-based hydrogel was obtained by Boustta et al.,[73] who used alteration in the molar mass and concentration of the polymer. This polymer, whose UCST transition could be adjusted, was suggested as a delivery system for both neutral and ionic drugs.

Ha et al.[108] proposed a PEG-based hydrogel as a codelivery system. The length of the PEG chains and cyclodextrin (αCDs) acted as transition temperature-determining factors. The transition temperature varied from 30 to 60 °C, thus showing the potential for a variety of different applications. The hydrogel could be loaded simultaneously with two different anticancer drugs, camptothecin (CPT) and 5-fluorouracil (5-FU), to form a smart codelivery system. These are hydrophobic and hydrophilic drugs, respectively. Cellulose derivatives such as methylcellulose (MC) and carboxymethyl cellulose (CMC) can form thermally responsive hydrogels with a LCST. To lower the transition temperature and impart better biocompatibility, researchers have investigated the incorporation of hyaluronic acid (HA),[93] poly(acrylic acid) (PAAc),[106] and PEG[106].

HA with different molecular weight ranges was introduced into MC.[93] The LCST was lowered to physiological temperature conditions. A hydrogel with the required rheological properties and good biocompatibility (cell viability assay with HFFF2 cells) was achieved with MC/LMW-HA blends. Lü et al.[94] developed a CMC-*g*-PNIPAAm hydrogel in which the PNIPAAm side chains and the CMC backbone provided thermosensitivity and biodegradability, respectively. This hydrogel showed a low gelation concentration, i.e., a concentration of 2 wt %, that was significantly lower than the normal gelation concentration of PNIPAAm-based hydrogels (Figure 3).

3.1.2. Encapsulation and Membranes—Burst drug-release kinetics may be sometimes observed in the case of hydrogels and porous capsules with low strength that are easily eroded. Additionally, hydrogels can suffer from poor encapsulation of hydrophobic drugs due to the hydrophilic nature of the hydrogel. To overcome these disadvantages, we must take special measures to prolong drug release and better encapsulate hydrophobic drugs.[86,92,96,98]

Shi et al.[98] designed hybrid membranes made from alginate/$CaCO_3$/PUA pH-temperature dual-responsive hydrogels, based on interaction of PUA and PAA and with LCST transition

of 55.3 °C. Here, sustained drug-release profile and lowered diffusion rate were observed because of the presence of hybrid $CaCO_3$ microparticles and alginate membranes.

In a similar approach, composites of alginate microspheres combined with PLGA–PEG–PLGA triple coblock hydrogel, were prepared as hybrid-membranes.[96] Figure 4 shows a schematic representation of these hydrogels loaded with alginate microspheres. A significantly reduced diffusion rate of water-soluble drugs, and more sustained release (the release time was enhanced approximately 4–6 times compared with control) were observed. This system could combine the properties of both barriers, thus achieving prolonged release of hydrophilic drug.

Poloxamer 407-based hydrogels were used to encapsulate etoposide (ETO).[86] Because of the high molecular weight of the polymer compared to the drug (almost 20 times greater), encapsulation was possible. The in vitro drug release profile showed sustained release, up to 48 h for poloxamer-based hydrogels.

3.2. Polymeric Micelles

The polymeric micelles with specific core/shell architecture generated by self-assembly of amphiphilic block copolymers, have been extensively studied as nanoscaled DDSs.[109] Polymeric micelles possess unique core–shell structure, in which an inner core entraps hydrophobic drugs as a nanocontainer and is surrounded by an outer shell of hydrophilic polymers, such as poly(ethylene glycol) (PEG), and have proved persistency in the bloodstream and effective tumor accumulation after their systemic administration.[110]

Polymeric micelles are promising nanocarriers for anticancer agents and are currently being employed in all three stages of clinical trials due to several advantages for drug delivery applications including high payload, ease of administration, reduced systemic toxicity, and improved therapeutic efficacy.[111]

Thermosensitivity of micelles depends on the LCST or cloud point (CP) of the thermosensitive block of the block copolymer forming micelle. Based on the LCST behavior, several PNIPAAm-containing block copolymers have been reported to prepare thermosensitive micelles for DDSs.[112]

The mechanism applied for controlled cargo release of thermoresponsive micelle polymers is related to reversible phase transition of PNIPAAm or other thermoresponsive contents in aqueous solution at around 33 °C (LCST); so that it is water-soluble and hydrophilic below its LCST and is insoluble, hydrophobic and aggregates above this point.

Micelles with a LCST of 38 °C would be mostly suitable to ensure constant temperature-dependent drug release in the bile duct. Because the temperature inside the liver is usually around 38 °C and because of the lower temperature of the intestine, drug release from micelles that reach the intestine through the bile duct would be decreased, leading to lower side effects. The bile duct is fully covered by liver tissue and its inner temperature is generally the same.[113]

As an alternative to PNIPAAm, the oligomers containing oligo(ethylene glycol) (OEG) with amphiphilic characteristics that integrate the biocompatibility of PEG with a versatile and controllable LCST behavior have been used to develop polymeric micelles, such as copolymers of 2-(2-methoxyethoxy) ethyl methacrylate (MEO2MA) and OEG methacrylate (OEGMA), (P(MEO2MA-*co*-OEGMA), which self-assemble into spherical nanomicelles in water and exhibit a LCST, which can be tuned between 26 to 90 °C by altering the ratio of MEO2MA and OEGMA.[114–116] The thermoresponsive biodegradable copolymer of poly(DL-lactide-*co*-glycolide)-poly-(ethylene glycol)-poly(DL-lactide-*co*-glycolide) (PLGA-PEG-PLGA) is proved to overcome shortcomings of the PNIPAAm copolymers, i.e., lack of biocompatibility and biodegradability, poor micellar stability.

3.3. Core–Shell Structures

Besides temperature-sensitive polymers such as hydrogels and micelles, core–shell nano-structures and even films have been also designed. Figure 5 illustrates a commonly employed core–shell structure. The main polymer network that has been of particular interest is formed from poly(*N*-isopropylacrylamide) (PNIPAAm). This polymeric network can undergo a volumetric phase transition after temperature elevation or by solvent uptake at low temperatures followed by swelling. After that phase transition, the swollen network then expels water and shrinks.

Another common structure that has been used in drug carriers, is that of thermoresponsive star block copolymers that can encapsulate the drug between their branches. Xiaojie Li and his group[117] developed a star-shaped block copolymer H40-PCL-*b*-P(OEGMA-*co*-AzPMA). The polymer micelle showed satisfactory drug-release properties, with controlled release at temperatures higher than the LCST temperature.

In another study, hydrogen-bonded layer-by-layer films were utilized. These films could serve as a robust platform that could be triggered by temperature changes to release drugs. These films were assembled with temperature-responsive block copolymer micelles and combined with tannic acid. The polymeric micelles were made by heating solutions of a neutral diblock copolymer, poly(*N*-vinylpyrrolidone)-*b*-poly(*N*-isopropylacrylamide) (PVPON-*b*-PNIPAAm), to a temperature above the LCST of PNIPAAm. Films and micelles made from PNIPAAm core–shell structure did not lose their structure after repeating the drug loading and releasing processes for 15 times.[118]

4. SYNTHESIS METHODS OF THERMOSENSITIVE NPs

4.1. Core–Shell Microgel Particles

Core–shell microgel particles that are thermosensitive can be synthesized by various methods, which depends on the nature and structure of the particles. Usually, the synthesize methods have a common path, that at first the core will be synthesized and then the shell is either synthesized by polymerization on the core or another step (usually surface modification) is required before the core will be covered with the shell. As follows, some cases has been mentioned to clear the synthesis method.

a. Two-step synthesis, that at first the core is synthesized and then the shell is prepared by polymerization on the core. One of the examples of this method is to prepare a core–shell particle with polystyrene (PS) as core and PNIPAAm as the shell. First, a seed latex component is synthesized by emulsion polymerization of PS. A thin shell of PNIPAAm is then generated around the core particles by copolymerization of NIPA monomer. Using seeded emulsion polymerization, the PNIPAAm network is polymerized onto the PS-cores (at $T >$ LCST). Gel-like PNIPAAm particles form when the polymerization of NIPA and the cross-linker starts in the aqueous phase. Finally, these particles are precipitated onto the surface of the core particle.[119]

Another example of two-step synthesis is when nonspherical systems or more accurately dumbbell-shape core–shell systems are made. For example, by using a precipitation polymerization method, a cross-linked PNIPAAm-shell is grafted onto the paired spheres each consisting of a PS core and a PMMA shell. Also, another example is when functional monomers have been used, and in these cases, it is possible to make either neutral shells with charged core or neutral core with charged shell. Richtering et al.[120] developed core–shell microgels, utilizing temperature-sensitive PNIPAAm. Two different types of microgels could be produced, (a) neutral core and charged shell, and (b) charged core and neutral shell. In the microgel core, polyelectrolytes could be encapsulated and would be protected by the shell.[119]

b. Three-step synthesis is is when there is another step between synthesizing the core and putting a shell on it, so-called photoemulsion polymerization. This synthetic approach is a way to overcome a problem with the polymerization rate of the cross-linker, N,N′-methylenebis(acrylamide) (MBAm), which is higher than that of NIPA.[121] Therefore, the growth of the microgel particles may not be uniform. As shown in Figure 6, this synthesis was accomplished in three steps: (1) Using a conventional emulsion-polymerization method, the PS-core particles containing 5 mol % NIPA were synthesized. (2) The photoinitiator 2-[p-(2-hydroxy-2-methylpropiophenone)]-ethylene-glycol-methacrylate (HMEM) is used to cover these core particles. (3) By means of light irradiation of a suspension of these particles, free radical generation occurs, leading to photoemulsion polymerization.[122]

The core can also be made with inorganic materials. Fe_3O_4 is one of the materials that can be used as the super paramagnetic core. In the study by Zeng and his group, a core–shell NP with Fe_3O_4 was provided as the core and the next layer was poly(methacrylic acid) (PMAA), which is dual-responsive, and the final layer was PNIPAAm, which is thermal-responisve and works as a “gatekeeper”. The Fe_3O_4 NPs has been made with the help of copercipitation of ferric and ferrous ions in alkali solution. After the core was made, the $Fe_3O_4^-$ PMAA NPs were made by

distillation precipitation polymerization. The latest layer was made by two-stage distillation precipitation polymerization with $Fe_3O_4^-$ PMAA as seeds.

c. Another method to synthesize core–shell particles is to graft copolymers. In this case, core and shell are created at the same time. In the study by Zhou and his group, the graft copolymer CS-*g*-PSBMA is synthesized under gamma-ray irradiation with by graft polymerization, and folic acid was loaded into the hydrophobic CS core with the help of self-assembly (Figure 7).[123]

In a research by Huang and his group, the same method was used to graft chitosan to N-(2-hydroxyethyl) prop-2-enamide (HEPE) and produce another core–shell particle that could be responsive both to pH and temperature. Here, pH and temperature equal to 5.4 and 37 °C, respectively, showed the best drug release rate.[124]

4.2. Hydrogels

Hydrogels are three-dimensional polymeric networks dispersed in water and make semi solid states containing more than 99% water in their composition. There are two main kinds of hydrogels: physical hydrogels formed by several physical entanglement between the polymer chains and/or micelle ordering in solution, and covalently (chemically) linked hydrogels that are synthesized by cross-linking of polymer chains with covalant bonds.[125]

Over the past two decades, both groups of cohydrogels have been under intense development for such applications as drug delivery and tissue engineering. The transition from a polymer solution to a gel resulting from exposure to the thermal environment like temperature are initiated in a certain temperature.[126] In other words, the hydrophilic groups in the polymeric network become hydrated in aqueous media and this prepares the mentioned transition from a polymer solution to a hydrogel.

Hydrogels may be explored by number of classical chemical methods including one-step procedures and multiple step ones. In the one-step method a hydrogel is synthesized by polymerization and parallel cross-linking of multifunctional monomers. However, in the multiple step method, a hydrogel can be prepared by reaction of polymers with suitable cross-linking agents, and possibly by synthesis of polymer molecules with reactive groups and the subsequent cross-linking of the polymers.[127]

Hydrogel-containing polymersomes can act as thermosensitive nanocarriers. Lee et al.[128] developed novel bilayer-enclosed nanostructures containing hydrosomes (a thermosensitive hydrogel). Such NPs were prepared by incorporation of PNIPAAm into the mPEG–PDLLA particles by injection of a THF solution into the water. The hydrogel-containing polymersomes were obtained at 37 °C. By using mPEG–PDLLA labeled with lissamine rhodamine B (RB) and fluorescein isothiocyanate (FITC)-labeled PNIPAAm dual fluorescent NP were prepared. Confocal microscopy showed the colocalization of both fluorophores in the particles. By using $CHCl_3$ as the organic phase, “giant” polymersomes with a diameter of 5–10 μm were formed that underwent phase separation of the internal FITC-NP above the LCST.

4.3. Thermoresponsive Composite Films

Thermoresponsive composite films can be prepared by grafting hydrogel layers (submicron thickness to several microns thick) onto a polycarbonate (PC) support layer in order to achieve controlled drug release. The grafting methods that can be used to prepare composite films include plasma modification,[129,130] ionizing irradiation,[131] redox polymerization,[132,133] UV photografting,[134] and atomic-transfer radical-polymerization (ATRP).[135] To adjust the LCSTs of the products, NIPAAm and AAc copolymer hydrogels were formulated in grafted films. At temperatures near their LCST, the films showed swelling or shrinking of the poly(NIPAAm-*co*-AAc), and the PC support exhibited straight cylindrical pores.

The PC-hydrogel composite films were characterized in terms of film thickness and grafting yield by employing X-ray photoelectron spectroscopy (XPS) and FTIR techniques, and the LCSTs were calculated from effective pore diameter, water permeability and drug permeability data. The LCST increased from 34.0 to 41.8 °C, and the grafting yield and film thickness increased, as the concentration of AAc in the preparative solution was increased. With on–off ratios between 1.4 and 3.2, the films showed a good permeability to drugs (3.22×10^{-7} to $4.64 \times 10^{-7} cm^2/s$). With 1.8% AAc, the film exhibited an LCST close to human body temperature.[133] In these composites citric acid, 4-acetamidophenol, KCl, and methyl orange could be used as model drugs. The larger molecules exhibited higher on–off ratio and lower permeability than the smaller model drugs (for neutral compounds). Lower permeability was found with ionic compounds with large hydration shells, such as the strong electrolyte (KCl). Increasing on–off ratio and drug permeability was found for the acidic compound (citric acid), promoting gel shrinkage. Figure 8 illustrates an on–off valve mechanism designed through grafting a P(NIPAAm-*co*-AAc) hydrogel onto microporous films. Thus, as the environmental temperature changes, drug permeation can be regulated.[136]

4.4. Smart Three Dimensionally Ordered Porous (3DOM) Materials

In recent years, three-dimensionally ordered macroporous (3DOM) structures have attracted the attention of great researchers because of their interconnected porous structures, well-defined surfaces, and precisely controlled pore sizes in submicrometer range.[137,138] Such materials have accessible macroporous networks, which can increase mass transport by reducing diffusion resistance. With this in mind, 3DOM materials are widely used as a drug/gene carrier in an efficient manner.[139,140]

Although some of 3DOMs are synthesized by synthetic polymer, inorganic oxides, and semiconductor materials, 3DOMs are generally synthesized by the colloidal crystal template method. This method is based on such templates including monodispersed as PMMA, PS, which can be assembled into colloidal crystals by several methods like vertical deposition, gravitational sedimentation, and centrifugal sedimentation. Then, the interstitial areas of the colloidal crystal template are filled with a precursor solution. In the next step, a subsequent transformation ocurres in the colloidal crystals, and the template is removed by roasting or dissolving. Through this process, 3DOM materials with a wide range of pore sizes, different components materials, and different properties can be prepared.[137,141,142]

Among the many prospective materials, chitosan is a promising candidate for developing a porous 3D chitosan (CHT)-matrices. These kind of 3DOM materials attracted abiding interest not only for their good film forming properties but also for sticking of polymer layers on the surface of the sacrificing spheres of chitosan. It is worth mentioning that the shrinkage and deformation took place in drying of polymer into the corresponding gel structures are the main challenges in 3D CHT matrices. So, many researchers increased the mechanical, thermal, and physical properties of polymers by such inorganic compounds as silica NPs.[143] These matrices can be produced through microscale technologies, and show dual pH-responsive as well as temperature-responsive behaviors. Such materials have also possible applications as switchable devices for release of bioactive molecules and uses in tissue engineering. For instance, using in situ synthesis of PNIPAAm within CHT micropores, CHT-scaffolds (the pH sensitive component) were coated/impregnated with PNIPAAm (the thermoresponsive component). A fine balance between hydrophobic and hydrophilic moieties in the molecular structure determined the LCST of this polymer (between 30 and 31 °C).[144]

To impregnate the polymeric devices, two different strategies were used: (1) bulk loading to impregnate a model protein (e.g., (bovine serum albumin (BSA)); (2) supercritical fluid impregnation with a model drug (i.e., ibuprofen(Ibu)) for scaffold uptake. BSA release was controlled by the PNIPAAm temperature-responsiveness (e.g., in temperature 20 and 37 °C), whereas the release of Ibu was responsive to changes in environmental pH (e.g., pH 5.4 and 7.4) conditions.[145]

4.5. Thermosensitive Micelles

According to their formation mechanism, it is possible to categorize the polymer micelles into three types of block copolymer micelles, graft copolymer micelles, polyelectrolyte micelles or hybrid polyion complex micelles.[146] Prior to predicting and controlling the properties of a micellar system, the copolymers utilized should be well-defined. Depending on the nature of the polymer to be synthesized, the most appropriate polymerization reaction to use, such as anionic, ring-opening polymerization (ROP), or radical polymerization is determined.[147] Currently, RAFT polymerization and ATRP are the most common controlled radical polymerization techniques used for building thermoresponsive block copolymers.[148]

Hu et al.[149] synthesized thermoresponsive brushlike amphiphilic poly[2-(2-methoxyethoxy) ethyl methacrylate-*co*-OEG methacrylate]-*b*-poly(L-lactide)-*b*-poly[2-(2-methoxye-thoxy) ethyl methacrylate-*co*-OEG methacrylate] [P-(MEO_2 MA- *co* -OEGMA)- *b* -PLLA- *b* -P(MEO_2 MA- *co* -OEGMA)] triblock copolymers by ATRP of MEO_2MA and OEGMA comonomers using a α,ω-bromopropionyl poly(L-lactide) (Br-PLLA-Br) macroinitiator and the polymeric micelles were generated by self-assembly of copolymers in aqueous medium.

In another study, thermosensitive polyelectrolyte complex (PEC) NPs assembled from two oppositely charged poly-electrolytes, chitosan-*graft*- PNIPAAm (CS-*g*-PNIPAAm) and sodium alginate-*graft*- PNIPAAm (SA-*g*-PNIPAAm) were prepared with size range of 130–180 for entrapment and release of 5-fluorouracil (5-FU). The CS-*g*- PNIPAAm and SA*g*-PNIPAAm graft copolymers were synthesized by employing free radical polymerization

using cerium ammonium nitrate (CAN) and potassium persulfate (KPS), sodium sulphite (SDS) initiators, respectively.[150]

Luo et al.[151] fabricated triarmed star-shaped s-P(NIPAM-*co*-DMAM) random copolymers with tunable CP values via one-pot ammonolysis reaction between triethyl 1,3,5-benzenetricarboxylate and P(NIPAM-*co*-DMAM)-NH2. And, copolymers formed thermosensitive micelle aggregates through hydrophobic interactions among the isopropyl groups of PNIPAAm chains and interstar association at a polymer concentration above critical aggregation concentrations from 4.06 to 6.55 mg L^{-1}, with a CP range from 36.6 to 52.1 °C, and micelle size below 200 nm.

The combination of ROP and ATRP was applied to synthesize a thermosensitive diblock polymer based on complex polymeric micelles of PNIPAAm and biotin, which is a cell-interacting ligand. The obtained nanocarrier demonstrated targeted delivery with enhanced cellular uptake properties and prolonged blood circulation time.[152]

4.6. Thermosensitive Nanogels

Temperature-responsive nanogels with a VPPT[91] can be prepared via different methods. The techniques of reverse microemulsion combined with thermally induced gelation were used to prepare cross-linked κ-carrageenan hydrogel NPs (i.e., nanogels). They were prepared via water-in-oil microemulsions with average diameters smaller than 100 nm. In a temperature range that was tolerable for living cells (37–45 °C), the nanogels were found to be thermosensitive with temperature-responsive reversible volume transitions. These nanogels could be applicable as smart thermosensitive drug carriers.[153]

Since 1986 when Pelton synthesized NIPAAm for the first time, it has become the most-reported water-based thermo-sensitive nanogel. Generally, the phase transition temperature of such nanogels is fitted by copolymerizing ionic monomers with NIPAAm.[154,155] Nanogels of NIPAAm are synthesized in a one-step polymerization procedure, in which a divinyl cross-linker copolymerizes enables polymer chains into a porous 3D nanospherical network.[156]

4.7. Thermosensitive Microcontainers (MCs)

In the recent decade, the synthesize of multifunctional MCs has been considerably increased thanks to their unique advantages in such applications as DGDSs, water treatment, and catalysis.[157] MCs have potential to encapsulate and release such biomolecules as drugs/genes, and peptides in a controlled manner. In addition, MCs have better chemical resistance, increased colloidal stability, and easier formation compared with other reviewed NPs in previous parts.[158] There are several strategies that have been reported to synthesize polymeric microcontainers including layer-by-layer techniques, emulsion/interfacial polymerization, and precipitation polymerization onto templates, which is the most widespread method.[159–161]

The fabrication of the MCs had two steps including the core synthesis, as well as the shell formation. For example, the shell consisted of PNIPAAm, PMMA, polydivinylbenzene (PDVB), and PAA, whereas the core consisted of PMMA. PMMA seeds were synthesized

using the emulsifier-free emulsion polymerization method.[162] Daunorubicin (DNR) was selected as a model drug for studying loading and release under different pH (i.e., physiological pH of 7.4 and an acidic pH of 5.4) and thermo-conditions (i.e., 25 and 45 °C). Two different cell lines (3T3 mouse embryonic fibroblast cells and MCF-7 breast cancer) were subjected to cytotoxicity studies. Hollow MCs possessed different temperature sensitivities and were suggested as a potential smart anticancer drug delivery system using traditional highly cytotoxic drugs to reduce side effects.[45]

5. CHARACTERIZATION OF THERMOSENSITIVE NPS

Temperature-responsive nanocarriers are characterized using several different techniques which are described as follows.

5.1. Transmission Electron Microscopy (TEM)

TEM analysis is utilized for characterizing the size and size distribution, structure, shape, morphology, and chemical composition of thermos-sensitive NPs.[163–170]

Besides, cryogenic-TEM is used for the characterization of colloidal DDSs. As well as being used for studying the overall colloidal composition of the corresponding dispersions, it can also be applied to study the internal structure, shape morphology, and size of the nanoparticulate carrier systems.[171] In an amorphous water phase, the particles are embedded and the aqueous phase is converted into hyper quenched glassy water.[172]

5.2. Scanning Electron Microscopy (SEM) and AFM

SEM and AFM are used to observe the surface morphology, average diameter, particle size, and pore volume of the NPs. Moreover, SEM observation is performed using a cold field-emission scanning electron microscope (FESEM) and energy-dispersive X-ray (EDX) analysis to demonstrate overall elemental composition.[173–176]

5.3. X-ray Diffraction (XRD) Spectroscopy

Purity and structural properties of samples are determined by XRD system. Specially, the peaks of XRD pattern are used to find the existence of crystalline structure. Moreover, from XRD data, the crystallite size (Dc) of the NPs is calculated using the Debye–Scherrer equation.[164,167,177]

5.4. Fourier Transforms Infrared (FTIR)

The chemical structure of the synthesized thermoresponsive nanocarriers, are examined by FTIR. According to absorption peaks which correspond to the frequencies of vibrations between the bonds, useful information can be obtained, such as component confirmation, stretch and vibration of bands, bonds interaction, and binding of coating on surface of NPs.[167,170,174,177–180] For instance, by means of FTIR spectra and calculation of Gibbs free energies for the hydrogen-bonded intermolecular complexes, the importance of hydrogen bonds in creating the NPs is realized.[181] In addition, cross-linked and non-cross-linked states of the polymers can be studied by FTIR.[182]

5.5. Gel Permeation Chromatography (GPC) and NMR Spectroscopy

GPC and hydrogen or carbon NMR spectroscopy implement to examine the chemical structure besides the molecular number and molecular weight (M_n, M_w), purity and polydispersity index (PDI, PDI = M_w/M_n) of synthesized thermosensitive NPs. Also, these analyses are utilized in order to understand the environmentally responsive mechanism at different temperatures.[166,173,180,183–185] The visible peaks of NMR spectroscopy and their movement in respect of temperature changes, indicating the thermal-responsive behavior of NPs such as breakdown of polymer–water hydrogen bonding (water solubility of the polymers).[183]

5.6. SAXS and small angle neutron scattering (SANS)

SAXS and SANS are both well-suited for studying microgels with sizes below 200 nm. The size of the particles and their interaction at low q (magnitude of scattering vector) can be determined from the scattering intensity $I(q)$.[172] Through using a range of large scattering angles, network fluctuations can be analyzed quantitatively and small-scale details can be probed. Also, the early stages of NPs creation and closer insight into the particles interior are realized via SAXS and SANS.[181]

5.7. Scanning X-ray Microscopy (STXM)

STXM has high chemical sensitivity, and it is possible to monitor wet samples directly using the osmotic deswelling of the particles, which results from high salt concentrations. Therefore, for in situ study of microgels, this technique is very well suited. Recent improvements have led to a resolution of 14 nm.[172]

5.8. DLS and Depolarized Dynamic Light Scattering (DDLS)

To investigate the hydrodynamic diameter (D_h) of the NPs, size distribution and PDI, samples are characterized by DLS.[163–170,178,185,186] It should be noted that the diameters are obtained from SEM/TEM measurement could be equal or different with the size measured by DLS method. This phenomenon could be due to morphological changes during preparation of samples. Furthermore, the data provided by DLS are related to the particles swollen in the solution, whereas SEM/TEM images are related to dried particles.[179,180,187,188]

Moreover, DLS and DDLS (characterization in dilute solution) can characterize microgels in dilute solution, and is used to measure the diffusion coefficient and the overall size of core–shell microgels [1]. Previously, DDLS techniques were developed for characterization of anisometric particles but recently they have been used to study microgels.[172] Also, by using the results of angle-dependent DLS, self-assembly behavior of the brush that could self-assemble into thermosensitive micelles in water is explored.[189] To evaluate the NPs' colloidal stability, researchers used the average diameter of TEM and zeta potential, which is obtained by zeta potential analyzer with a DLS detector.[163,190]

5.9. Thermal Gravimetric Analysis (TGA) and Differential Scanning Calorimetry (DSC)

Thermal properties of the nanocarriers are measured using TGA and DSC.[164,177,183,187,191] By means of TGA analysis, the rate of weight loss of the samples as a function of temperature assists to define the thermal degradation mechanism of polymeric particles, strength of intermolecular interactions, and finally the thermal stability are exploited.[177,192] Also, the chains flexibility and grafted polymer density (ρ_{graft}) is estimated from the weight loss of polymeric grafts which is measured by TGA. In addition, the LCST transitions of the synthesized copolymers and T_g of the dried samples are characterized by DSC. For instance, DSC thermograms of the hydrogel nanocarriers indicate the miscibility of them. Single T_g in the thermograms means that the samples have a good miscibility.[192]

For a brief look at the recent studies regarding various thermosensitive DDSs, several different synthesis and characterization methods are provided in Table 2.

6. TEMPERATURE RESPONSIVE NANOCARRIERS FOR CARGO DELIVERY

One of the challenges of DDSs is drug delivery and release at the right time and right concentration (i.e., spatiotemporal control). Temperature-responsive polymers NPs can be used as biocompatible nanocarriers for drugs and other cargos. Using these nanocarriers, it is possible to control drug release with the aid of changes in environmental parameters such as temperature. These kinds of nanocarriers were initially studied as drug-delivery systems to treat cancer, because they could reduce the side effects of chemotherapy drugs. Also, they are good candidates for gene delivery vehicles. There are also many common diseases like diabetes and hypercholesterolemia, which need repeated daily doses of the appropriate drugs. Therefore, another application that was proposed for these kinds of NPs was to carry the drugs capable to treat diseases requiring a regular repeated dose of the drug, because of their delayed drug-release properties. Therefore, such NPs not only are used for carrying genes or cancer drugs such as Paclitaxel, but also for carrying common drugs like Atorovastatin, a statin for cholesterol regulation, or antiflammatory drugs.

6.1. Anticancer Drug Delivery

Thermoresponsive DDSs can be aimed to strengthen the efficacy of anticancer drugs against cancerous sites, and obtaining controlled drug delivery/release. Enhancing cancer chemotherapy using thermoresponsive NPs such as hydrogels loaded with etoposide (ETO),[86] and with the combination of camptothecin and 5-fluorouracil (CPT/5-FU),[108] has been a field of interest. Another effort that was made using PNIPAAm was to prepare a graft copolymer from sodium alginate-*g*- PNIPAAm (ALG-*g*- PNIPAAm) with multivalent metal ions (Ca^{2+} and Al^{3+}) and 5-fluorouracil (5-FU) anticancer drug.[211] In this study, Guiying Li and his group prepared a hollow structure with a size of about 100 nm. The swollen polymer chains formed the neutral part, and the complex bonds between the sodium alginate chains and the metal ions imparted stability to the micelles. The polymer vesicles had a LCST temperature around 37 °C. It was observed that the hollow structure of the particles had large cavities that enabled chemical attraction between the surface of the cavities and the drug, thus helping the drug-loading efficiency.

A thermoresponsive polymer system with a LCST close to body temperature is Pluronic F127-poly(D,L-lactic acid) (F127-PLA, abbreviated as FP). The copolymer micelles were decorated with folate (FA) to impart active targeting. Xing Guo et al.[212] reported that using this polymeric micelle system, a small amount of the encapsulated DOX was released as soon as the temperature reached the LCST (39.2′C) and, at a temperature only just a little bit higher (40 °C), the polymer shells released the drug rapidly.

Thermoresponsive micelle systems are promising for anticancer therapy, because they respond to the demands of active tumor targeting and considerations related to tumor tissues' abnormal temperature gradients as compared with those of normal ones. Micelles can be specifically taken up by folate receptor (FR)-overexpressed tumor cells through receptor-mediated endocytosis, and rapidly release the cargo inside cells at low hyperthermia (40 °C) because tumor tissues are much more sensitive to high temperatures and effectively terminating the tumor by hyperthermia is obtained in the range of 40–43 °C (Figure 9).[212]

The drug loading and release behavior for the (P(MEO2MA-*co*-OEGMA) copolymer was assessed using chlorambucil (CBL), as a model hydrophobic anticancer agent under various temperature conditions. Under the LCST (28 °C), the highly hydrated micellar polymer segments stabilized the hydrophobic–hydrophilic core–shell structure of micelles of dendrimer-star copolymer with Y-shaped arms, hence the drug release was slow and about 78% drug still remained in the core of the micelles after 40 h. But, after the temperature was raised above the LCST (40 °C), and because the polymers shell became hydrophobic, the temperature-induced structural changes of the micelles were led to deformation of the micellar core–shell structure and the drug release was accelerated.[116]

6-2. Gene Delivery via Thermoresponsive Nano-carriers

Gene therapy is a technique in which genetic materials (nucleic acids) are delivered to the nuclei of diseased cells or tissues. Different forms of exogenous nucleic acids (such as plasmid DNA, minivector DNA, siRNA, antisense oligonucleotides, etc.) are used in order to treat pathological conditions and remedy genetic disorders.[213–216] The issue of gene delivery into the cell nuclei is one of the critical steps and is the most important barrier to successful gene therapy. The existence of several obstacles that need to be overcome before genetic materials arrive in target cells has hindered efficient gene delivery so far.[214]

6.2.1. Nonviral Gene Delivery—Considering the fact that naked DNA does not enter cells efficiently, researchers have investigated two different broad types of gene delivery system, namely viral and nonviral vectors (natural or synthetic).[214,216,217] The pros and cons of the above-mentioned approaches can be considered in three areas: production, safety, and efficiency, which are described in Table 3.

The concerns over safety issues that have been reported to have occurred with viral vectors have limited their routine utilization in both basic research laboratories, as well as in clinical trial applications. In contrast, nonviral gene delivery methods are characterized by relatively favorable features such as a lack of safety concerns, and the potential ability to carry a wider variety of different nucleic acids and genetic materials. Furthermore, nonviral vectors can be selective targeted toward specific cell types by attaching molecular recognition ligands.[215]

In nonviral gene delivery systems, a synthetic vehicle is used to encapsulate the nucleic acid until it reaches its target cell. These vehicles can be mainly divided into two different broad groups. The first group is composed of organic nanostructures, which include cationic lipids and various polymers (the most common and efficient nonviral gene vectors), and also peptides or carbon nanotubes. The second group is based on NPs formed from inorganic components such as gold NPs, quantum dots and calcium phosphate particles.[213–215,219–222]

Among the above-mentioned groups, polymeric gene delivery systems have received particular attention thanks to their stimuli-responsive features. It is ideal for a polymeric nanocarrier with gene delivery purpose to exhibit (1) efficient protection of nucleic acids from degradation and clearance during circulation, (2) specific delivery of nucleic acids to target tissues/cells, and (3) readily nucleic acids release in the target intracellular compartments after overcoming multiple intra-cellular barriers. Those requirements are achievable by employing molecularly engineered nonviral vectors to transform their physicochemical properties in response to various extra and intracellular stimuli as well as external triggers.[223]

Here, in the same manner as for drug delivery, cargo release occurs as a result of their phase transition at certain temperatures (LCST or UCST). These polymers should have the potential to form strong complex binding with DNA while being transported to the target sites at a temperature below their LCST. On the other hand, the binding in these complexes should become loose enough at the target cells and tissue sites to release the DNA at temperatures above their LCST. In this way, this smart stimulus-responsive system enhances the efficiency of transfection.[74–77]

PNIPAAm is the most extensively investigated thermoresponsive polymer used in gene delivery systems. PNIPAAm is a water-soluble polymer that is hydrophilic below 32 °C (its critical solution temperature) due to the hydrogen bonding between its amide groups and water molecules. When the temperature reaches this critical point (LCST or “cloud point”), the hydrogen bonds are broken and the structure of the polymer chains changes from water-soluble coils (hydrophilic) to a globular structure (hydrophobic). It is considered that PNIPAAm may have some slight toxicity after long-term exposure, in that its accumulation in the body could be injurious because of its nonbiodegradability. Copolymerization of PNIPAAm with other biodegradable polymers (hydrophilic or hydrophobic blocks) would not only partially overcome this drawback but also could alter the LCST of PNIPAAm.[74,76,216,224–226] A short summary of examples of thermosensitive polymers as gene delivery systems including their features and applications is provided in Table 4.

Feng et al.[239] developed an efficient nonviral cationic block copolymer gene delivery system to deliver therapeutic plasmid DNA (pDNA) for nucleus pulposus regeneration toward Disc degeneration disease. The copolymer was prepared via complexation between the mixed cationic block copolymers and pDNA. The obtained mixed polyplex micelles (MPMs) containing thermoresponsive heterogeneous coronas with hydrophobic and hydrophilic microdomains coexisting could be achieved upon heating from 25 to 37 °C, which demonstrated high tolerability against nuclease, strong resistance toward protein adsorption, high gene transfection efficiency, and low cytotoxicity.

The polyplex micelles have also been investigated for antitumor gene delivery. The rod-shaped ternary polyplex micelles (TPMs) was synthesized via complexation between the mixed block copolymers of poly(ethylene glycol)-*b*-poly(N′-[N-(2-aminoethyl)-2-aminoethyl]aspartamide) (PEG-*b*-PAsp-(DET)) and PNIPAAm-*b*-PAsp(DET) (PNIPAAm-*b*-PAsp-(DET)) and plasmid DNA (pDNA) at room temperature. The formation of a hydrophobic intermediate layer between PEG shells and pDNA cores demonstrated thermoresponsive behaviors through facile temperature increase from room temperature to body temperature (~37 °C). The results revealed that incorporating the thermoresponsive PNIPAAm content as an intermediate barrier provided prolonged blood circulation and the TPMs loading therapeutic pDNA encoding an antiangiogenic protein remarkably suppressed tumor growth following intravenous injection into H22 tumor-bearing mice.[240]

The differences between the physicochemical structures of DNA and nucleic acids and those of small molecule drugs, makes it impossible to use similar nanocarriers to deliver both of them. Gaspar et al.[241] reported on the preparation of nanocarriers capable of efficient gene delivery. They synthesized amphiphilic triblock poly(2-ethyl-2-oxazoline)-PLA-g-PEI (PEOz-PLA-g-PEI) micelles for the delivery of minicircular DNA (mcDNA) vectors. In this copolymer, a hydrophilic shell was constructed by replacing the PEG blocks with PEOz. Additionally, the concept of codelivery of mcDNA and doxorubicin (DOX) was demonstrated by their simultaneous encapsulation in PEOz-PLA-g-PEI carriers with high efficiency.

6.2.2. Gene Transfection by Thermoresponsive Nano-carriers—Gene transfection is the main goal of nucleic acid delivery using polymeric nonviral carriers (Figure 10), which involves four stages: (1) complexation of plasmid DNA and the polymer that is usually performed at room temperature; (2) addition of the complex of DNA with the polymer (polyplex) into cell culture medium for a period of time (transfection time) which is carried out at 37 °C); (3) DNA release into the cytoplasm, when the polyplex is eliminated from the cell; and (4) the cells are left to incubate for a period of time (incubation time) in order to transfer the DNA into nucleus. One strategy to increase the transfection efficiency is changing the temperature during each of the steps (complexation, incubation or transfection period). Another method to enhance the transfection efficiency is increasing the weight ratio of polymer to DNA in the complexes.[216,224,242,243]

For instance, Wenguang Liu et al.[232] used PEG polymers with grafted PEI chains and tested changing the temperature of complexation as well as of transfection, and improved transfection efficiency. They could increase efficiency of transfection by increasing the weight ratio of PEIMH/DNA from 5:1 to 30:1. In another study, Zhang et al.[244] employed a cationic lipid as a nonviral vector for gene delivery and they could show that cationic lipids with a shorter chain length had a higher transfection efficiency. In an interesting study, Freitag et al.[245] found that a minimum molecular weight for the polymers was necessary for high transfection efficiency, and also with increased branching (e.g., arm number) better transfection could be observed. In this regard, they suggested star-shaped polymers, which not only had several arms (more than 5), but also possessed intermediate molecular weight to obtain the appropriate transfection efficiency.

6-2-3. Gene Expression by Thermoresponsive Nano-carriers—Gene expression is a complex process, which requires the nucleic acids to cross the cellular membrane, to undergo appropriate intracellular trafficking, and finally nuclear delivery of the cargo of genetic material. When the DNA is transcribed in the nucleus it produces mature mRNA, which is released and exported to the cytoplasm for translation. At this stage, mRNA engages with the ribosomes at initial step of translation, and after elongation and termination (as intermediate steps), the ribosome disengages from the mRNA, and then its subunits dissociate to repeat the cycle. In other words, at translation step, the genetic code is read, and the formation of the polypeptides builds up stepwise and finally the completed polypeptide chain is released.[246,247]

7. DUAL STIMULI IN TEMPERATURE-RESPONSIVE SYSTEMS

7.1. pH/Temperature

Multistimuli-responsive hydrogels (MSRHs) can be produced by taking advantage of the interaction between various functional groups that exist in macromolecules. MSRHs, particularly those that respond to changes in pH and temperature, have been reported extensively, focusing on their in situ hydrogel-forming abilities and site-specific applications.

Némethy et al.[53] reported a pH-and temperature-responsive poly(aspartic acid)-l-poly(*N*-isopropylacrylamide) conetwork hydrogel (PASP-l-PNIPAAm) in which PASP and PNIPAAm imparted pH- and thermoresponsivity, respectively. The hydrogel formed a conetwork and made it possible to absorb more water. The in vitro release rate of the loaded anti-inflammatory drug, diclofenac sodium (DFS), was reported to be slower at 37 °C and pH 7.6 after preconditioning in an acidic milieu (with pH 1.2) compared to the case of purely alkaline milieu. In another study, the introduction of carboxylated nanocrystalline cellulose (CNCC) into PNI-PAAm, resulted in generation of a pH-thermoresponsive PNIPAAm-based hydrogel.[84] The carboxyl groups of CNCC imparted hydrophilicity, improved swelling ability, better swelling rate, and pH responsiveness.

The urea groups present in poly(amino urea urethane) (PAUU) contained in PEG-*g*-PAUU, enhanced the hydrogel interaction with drugs, reduced the critical copolymer concentration needed for gelation, lowered toxicity, and imparted pH–temperature dual responsivity.[248] By studying the long-term release of FITC-BSA-loaded protein, the research team proposed PEG-*g*-PAUU hydrogel as a protein-release depot that could be injected.

Dual-responsive polymeric micellar NPs have been also reported. Herein, to develop more advanced micellar polymers that can respond both to temperature and pH variations, the pH responsive poly(L-histidine) was integrated to PLGA-PEG-PLGA forming dual-responsive copolymer for more efficient drug delivery. The lower pH amounts (e.g., 6.0 and 5.0) and higher temperatures (above LCST e.g. 41 °C) showed higher rates of DOX release (Figure 11a–c).[249]

7.1.1. pH/Temperature for Anticancer Drug Delivery—In a study using PNIPAAm, two new formulations were designed for dual temperature and pH-responsive nanogels.

PNIPAAm was used as the basis of the hydrogel structure. They introduced PNIPAAm to *N*-hydroxyethyl acrylamide (HEAA) and *tert*-butyl 2-acrylamidoethyl carbamate (2AAECM) to form a nanocarrier to deliver the common and well-known anticancer drug, Paclitaxel. They found out that at a pH value around 5, a faster drug-release was obtained. The thermo-induced changes were also investigated in these polymeric micelles by observing the optical absorbance as a function of temperature. The temperature of the solution was varied between 15 and 45 °C and it was observed that the absorbance increased when the temperature of solution was between 28 and 33 °C, which is the temperature range in which the phase change occurs in PNIPAAm, and it releases the water absorbed by PNIPAAm.[250]

In another effort to prepare polymeric NPs that were responsive to both temperature and pH, Yongkyun et al.[251] used a fluorescent cross-linked PNIPAAm-based probe. They studied the influence of the temperature and pH on the following the opening of the lactone ring of fluorescein and the lactam ring of rhodamine and the subsequent fluorescence emission at 514 and 586 nm. They showed influence of temperature changes (i.e., between 25 and 42 °C) and pH changes, (i.e., acidic (2.3–5.3) and basic (7.4.–12.4) conditions) on the turbidity of the microgels, above the LCST transition temperature, and the fluorescence intensity.

7.1.2. Stability of Particles and Controlled Drug Release—One of the challenging issues with polymeric micelles is the required balance between the stability needed during prolonged blood circulation, and active drug-release when the nanocarrier arrives at the tumor site. Stimuli-responsive materials including a pH-responsive element provide a mechanism for triggered drug-release in the acidic tumor and intracellular microenvironments. Ching-Yi Chen et al.[252] synthesized a series of dual pH-and temperature-responsive block copolymers, with the optimized LCST transition ~35 °C, containing a poly(*ε*-caprolactone) (PCL) hydrophobic block with a PEG block. The block copolymers formed micellar structures in aqueous solutions. The optimized polymer was stable at blood pH (~7.4), whereas they showed pH-responsive phase transitions at mildly acidic pH (~5.3) and at body temperature. These micelles showed better anticancer activity than free DOX. Yukun Wu et al.[253] tested a method to increase the stability of an amphiphilic block copolymer by developing novel core cross-linked micelles. They synthesized poly(ethylene glycol)-*block*-poly(*N*-isopropylacrylamide-*co*-N-(4-hydroxypHenethyl) acrylamide) diblock copolymer (PEG-*b*-P(NIPAAm-*co*-NHPAAm)). The diblock copolymer had a phase transition into non-cross-linked (NCL) micelles. When the temperature exceeded the LCST (about 32 °C), at the temperature about 37 °C, the NCL micelles exhibited a better performance for drug release in comparison with core cross-linked micelles (Figure 12).

Agut et al.[254] fabricated polypeptide-based double hydrophilic block copolymers (DHBCs), namely, poly[2-(dimethylamino)ethyl methacrylate]-*b*-poly(glutamic acid) (PDMAEMA-*b*-PGA) with dual temperature and pH sensitivity. At pH values close to the isoelectric point, the polymeric micelles had a spherical morphology. The thermo-responsivity of the double-hydrophilic block copolymers (DHBC) was evident around the LCST (about 40 °C). Around pH 11 and when the temperature was below the LCST, the DHBC polymer was present as free chains, whereas at temperatures higher than the LCST, the polymers changed their

conformation and assembled into spherical polymeric micelles. Moreover, by varying the length of the PGA blocks, a variety of different sizes, shapes, and morphologies could be produced.

Zhang et al.[255] synthesized double-block hydrophilic copolymers composed of PEG blocks and bicyclodextrin (*b*-CD) blocks to form PEG-*b*-PCD. Next a drug carrier was synthesized as a core–shell structure using poly(b-benzyl L-aspartate) (PBLA) as the core, and the hydrophilic copolymer PEG-*b*-PCD was the shell of this core–shell structure. The hydrophobic drug could interact with the *b*-CD and the hydrophobic benzyl groups to form stable assemblies. In vivo and in vitro drug release studies confirmed the ability of the polymeric core–shell nanostructure to mediate controlled drug delivery.

7.2. Magnetic/Temperature Dual Stimuli-Responsive Nanocarriers

Using magnetic properties to control and improve the drug-release profile is another common method that has been used. Sundaresan et al.[256] used PNIPAAm that was coated onto MNPs to achieve better and more precise control, over drug release. The core of the ~150 nm sized core–shell structure was Iron oxide with superparamagnetic properties. The dual-responsive functionality of these NPs was observed, and it was reported that at 40 °C and pH 6, the release of anticancer drug DOX was at a maximum level. The drug-release rate was increased when the NPs were exposed to a 1.3T magnetic field. In the study conducted by Pernia Leal et al.,[257] they produced thermosensitive polymer shells on the surface of superparamagnetic iron oxide particles that were embedded onto the surface of poly(maleic anhydride-*alt*-1-ocatadecene) polymeric nanobeads. The phase transition temperature of the PNIPAAm-magnetic nanobead NPs, which was initially reported to be (about 32 °C), can differ depending on the composition of the comonomers and their relative concentrations; however, in the range between 26 and 47 °C it was observed that nanobeads formed a stable suspension below the LCST, and magnetic field could not be easily applied to trap the beads. However, at temperatures higher than the CST, the nanobeads agglomerated and formed clusters, with magnetic moments high enough to be trapped by the magnetic field. At the same time, because the temperature was higher than the LCST, the swollen polymer started to release the drug. Another magnetic core structure that can be used in core–shell structures is Fe_3O_4. Haie Zhu and his group[258] developed a novel core–shell structure with Fe_3O_4 as the core and PNIPAAm as the shell. This new core–shell structure demonstrated unique triple features, fluorescence, superpara-magnetism and thermoresponsivity. They synthesized this core–shell structure using Fe_3O_4/poly(Styrene (St)-NIPAM) NPs, which acted as nucleation sites for polymerization of $Eu(AA)_3Phen$ with the addition of St and NIPAM to form an outer fluorescent layer. The fluorescent layer on the NP was employed as a tracer to follow the NP distribution.

7.3. Biomolecule/Temperature Dual Stimuli-Responsive Nanocarriers

Thermoresponsive polymeric nanocarriers can also be designed to be glucose-responsive, thus they can be used as insulin carriers, and could release insulin in response to varying glucose levels in the body. The fact that the cores in core–shell structures can be made from inorganic materials, gives the opportunity to produce a nanocarrier that is responsive not only to temperature but also to magnetic fields, and consequently the drug-release process

could be managed more effectively. Yuan Yao et al.[259] attempted to prepare a glucose-responsive carrier for controlled release of insulin by combining PNIPAAm with poly(ethylene glycol)-*block*-poly-(phenyl bromate ester). They prepared polymeric micelles with a core–shell–corona structure comprising glucose-responsive PPDEMA as the core and PNIPAAm as the shell components. They could control the structure by varying the temperature between 15 and 37 °C. As a result, they achieved much better stability for insulin encapsulation in the polymeric micelles by introducing PNIPAAm as the shell in the core–shell structures. In another study, thermo- and enzyme dual-responsive polymeric amphiphiles rendered by Kashyap et al.[260] In this approach, tailor-made polymer of 3-pentadecylphenol (PDP) copolymerized with oligoethylene glycol acrylate by both radical and reversible addition–fragmentation chain transfer (RAFT) synthesis methods and spherical core–shell nano-particles were self-assembled in aqueous medium and doxorubicin (DOX) was loaded. This assembly showed tunable LCST behavior at the tumor site and sensitivity to esterase enzyme at the intracellular environment.

7.4. Redox/Temperature Dual Stimuli-Responsive Nanocarriers

Cleavable diselenide bonds[87] as well as cleavable disulfide bonds[261,262] can be introduced into hydrogels to achieve better degradability. Diselenide bonds have been used as a cross-linker in NIPAAM and HEA hydrogels. In an environment containing oxidizing agents such as enzymes and ROS, Se–Se bonds can be cleaved, thus achieving an oxidation-responsive hydrogel. Huo and his group[263] developed a novel dual oxidation-thermoresponsive PNIPAAm-based hydrogel with enhanced degradability

7.5. Triple Stimuli-Responsive Nanocarriers

Temperature can be employed in triple stimuli-responsive DDSs in order to obtain highly controlled drug delivery and release.[46,264–266] Zeng et al.[267] report a strategy for formation of a multiresponsive Fe_3O_4@PMAA@PNIPAM magnetic core, inner shell (reduction/pH-responsive), and outer shell (thermal-responsive), respectively, core–shell NP. A model drug, DOX, showed efficiency of these NPs for controlled releasing purposes.

8. LIGHT-INDUCED THERMORESPONSIVE NANOCARRIERS

Light is an appreciable source to be exploited in nanomedicine and therapies and especially DDSs.[268–272] Here, an important group of the temperature-responsive nanocarriers includes the nanocarriers having photoinduced temperature-responsive drug release capability for controlled drug delivery. In the structure of these nanocarriers, photothermal inducing agents can be employed, which absorb the irradiated light and convert it into a localized heat. The induced heat can be employed to induce temperature-triggered drug/gene release inside the biological milieus.[273]

Noble metal nanomaterials (Au, Ag, Pt, and Ge) are one of the most studied agents for photothermally induced drug release.[274] Nobel metals exhibit remarkable optical-electronic properties because of their surface plasmon resonances (SPRs). SPR is the free electrons resonant oscillation at the interface of a negative and positive permittivity material stimulated by external electromagnetic fields. SPR in a material results in dipole oscillations

along the direction of electromagnetic field. At the SPR frequency, the amplitude of oscillation reaches a maximum, inducing high absorption of the electric field of the incident light. Noble metals exhibit strong SPR when stimulated by visible light, which could be used in photothermal therapy (PTT) applications.

Other groups of materials including CNTs; graphene, and GO; transition metal and semiconductor based NPs have been also suggested to be efficiently used for controlled release of therapeutic agents from thermoresponsive nanocarriers.[275–277] Transition-metal dichalcogenides (TMDCs), such as MoS_2 and WS_2, are new groups of materials being proposed as photothermal agents. These materials are analogous to the graphene in having layered structures.[278]

Another recent group of materials with photothermal capability are semiconductors nanomaterials (Cu_xS, Cu_xSe, and Cu_xTe).[279–281] Semiconductors have the advantages of high plasmonic absorption in a wide near-infrared (NIR) range. They have a relatively low cost, good biocompatibility, high photothermal efficiency, and high in vivo stability.[281]

The majority of mentioned photothermal agents are nonbiodegradable and would retain in the body for a long time. This could result in a consequent long-term toxicity of these materials. Therefore, concerns about the potential long-term toxicity of the inorganic photothermal agents have caused a delay in their future clinical transition. Hence, to overcome this concern, scientists have tried to explore the possibility of using organic nanomaterials as substitution photothermal agents. Consequently, in the recent years, various nanomaterials complexes, conjugated polymers, or even organic/inorganic nanocomposites have been successfully synthesized and administrated employing their photothermal capability. Comprehensive reviews over these new promising group of biodegradable materials exist in the literature, which readers are encouraged to refer for more detailed information.[282–284]

8.1. Applications of Photoinduced Thermoresponsive Nanocarriers

In photoinduced thermoresponsive nanocarriers, the released heat can be used to damage or destroy the nearby diseased cells, e.g., cancer cells.[285] This medical treatment, in which cancerous tissue is ablated by being exposed to a higher temperature than normal condition is called as hyperthermia.[286] Applying NIR light (wavelength = 700–1100) is the mostly preferred as the light source for PTT applications, because of noninvasive and relatively deep tissue penetration. Therefore, the main challenge in developing photothermal agents is to produce material with a high light absorbance and high photothermal conversion efficiency.[274,287]

Among metal NPs, gold NPs are the basic photothermal materials for the initial development of PTT, and hence, they are the most studied photothermal agents.[274] Gold NPs exhibit strong SPR and scatter the incident NIR light with a large extinction cross-section. The large absorption cross-section and high photothermal conversion efficiency of gold NPs allow for less invasive PTT treatment with higher NIR light wavelengths.[288] There are good reviews in the literature, discussing the development and progress of gold NPs in PTT applications.[288–292] In addition, PTT could be combined with chemotherapy, by means of

thermoresponsive polymers, as a highly effective cancer therapy. Combination of thermally responsive polymers with photothermal agents results in an intelligent NIR-controlled drug release. The produced heat by photothermal agents when stimulated by NIR light, can trigger the shrinkage of drug loaded thermoresponsive hydrogels and the consequent controlled drug release. This mechanism increases the efficiency of drug release at the cancer site in addition to hyperthermia of the cancerous cells. Ying Qu et al.[293] developed a combined hydrogel system by doping gold nanorods (GNRs), as the photothermal agents, into the DOX embedded hydrogels. They demonstrated that efficient release of DOX from hydrogel matrix, by means of stimulating GNRs photothermal agents, resulted in a remarkable decrease of the tumor recurrence in mice 4T1 breast cancer models. In a similar study, a conjugate of GNR-poly(ethylene glycol)-*block*-poly(caprolactone) (PEG-*b*-PCL)-DOX was utilized as a switch for controlled release of DOX via a 808 nm NIR irradiation induced thermoresponsiveness.[294] In other studies, photosensitizer dyes such as indocyanine green (ICG)[295] and also photothermal semiconductor nanocrystals such as $Cu_{1.75}S$[281] have been reported to show NIR triggered thermoresponsive controlled release of anticancer drugs from polymeric-based nanocarriers (Figure 13a, b).

9. OTHER APPLICATIONS OF TEMPERATURE-RESPONSIVE NANOCARRIERS

Apart from the aforementioned applications, some other potential applications are also provided in the recent researches for the thermoresponsive DDSs. For instance, considering the flexibility of hydrogels, they can carry out a variety of applications. Protein delivery is the most frequently studied application. So far, various proteins, such as insulin,[79,296] lysozyme,[94,104] BSA,[93,103,248] as well as FITC as a protein label[248] have been studied and their release profiles have been monitored. Moreover, the delivery of human growth hormone (HGH)[297,298] and DFS[53] as an anti-inflammatory drug, have been the subject of other studies. Co-delivery systems also have been studied[104,248,296] in order to achieve enhanced therapeutic activity. In this regard, a study[296] on the combined delivery of both insulin and GSH was carried out. Boustta and coworkers[73] focused on simultaneous codelivery of model neutral proteins and cobalt acetate, an ionic drug. Delivery of therapeutic enzymes is a rather less studied area, but as mentioned earlier, encapsulated hydrogels are novel versatile ways for site-specific drug delivery. In a study,[87] calcium alginate (CaAlg) encapsulated hydrogels, were able to deliver not only hydrophilic and hydrophobic model drugs (FITC and piroxicam) but also could deliver horse radish peroxidase (HRP) as a model enzyme.

In a study,[299] PNIPAAm hydrogels were used in order to deliver colloidal drug carriers (CDCs). This route was investigated as a treatment for myocardial infarction (heart attack). Core–shell structures that are temperature-responsive can release their loaded drug with a programmed delay. Some common diseases like diabetes and hypercholesterolemia need regular doses of drugs delivered over a long time. Therefore, an application that was proposed for these temperature-responsive NPs was to carry drugs for these chronic diseases, because of their delayed drug release. Therefore, such NPs could be used for

carrying common drugs like the cholesterol-lowering Atorvastatin or antiflammatory drugs.[300]

Another group of temperature-responsive systems are microgel particles which have various applications as active carrier systems for the immobilization of catalytically active enzymes onto metal NPs. Manipulation of the properties of the embedded NPs depends on the volume transition of the microgel particles.[119]

Antimicrobial peptides (AMPs) are excellent nominees as antimicrobial agents rather than antibiotics that are less effective for treatment of bacterial infections because of their extensive use. Ping Dong et al.[301] innovated a drug delivery system against the main mankind pathogen of Grampositive bacteria– *Staphylococcus aureus* – by assembly of alamethicin, an AMP, to a thermosensitive polymeric carrier (PSBMA (poly(2-(adenine-9-yl) ethanol methacrylate-cosulfobetaine methacrylate) (poly(AEM-*co*-SBMA))). The copolymer had UCST in water that provided controlled release of alamethicin. This idea can be generalized for treatment of microbial strains by temperature-responsive delivery of AMPs.

It is demonstrated that, by incorporating magnetic resonance imaging (MRI) contrast agents into the structural design of thermoresponsive micellar copolymers with tumor targeting and controlled release abilities loaded with anticancer therapeutic agents, it is possible to acquire efficient theranostic systems, provided that (1) the unimolecular micelles have size range of 20–100 nm to minimize recognition by reticuloendothelial systems; (2) the hydrophilic corona is biologically inert and noncytotoxic; (3) hydrophobic core is capable of sufficient loading capacity for hydrophobic therapeutic drugs; (4) targeting moieties and T_1 contrast agents (DOTAeGd) were covalently attached to the hydrophilic periphery to achieve cancer-targeted delivery of chemotherapeutic drugs and contrast agents.[302]

10. CONCLUSION AND FUTURE PERSPECTIVE

In the present review paper, we have summarized the use of temperature-responsive nanocarriers for drug and gene delivery. The LCST and the UCST temperature transitions, which are defined by the properties of particular polymer functional groups are the defining feature of these smart delivery vehicles. Thermally responsive nanocarriers such as hydrogels, micelles, core–shells, etc., can be used for delivery of therapeutic drugs (particularly anticancer drugs) and for gene delivery purposes

In recent years, targeted smart DDSs due to using low doses of drugs, local targeting, decreasing side-effect and high efficiency for disease treatment are intensively taken into consideration by researchers. Temperature sensitive nano-carriers as one of the most important smart nanocrriers for drug or gene delivery for treatment of various diseases and cancer therapy were highlighted in this study. Solubility, phase transition behavior, release of drug or gene, synthesis, and characterizations methods of thermoresponsive NPs, membranes, in core–shell structures, micelles, etc., were discussed. Furthermore, the capability of thermoresponsive structures in combination with other stimuli including pH, light, redox, magnetic field, etc., forming dual/multi-responsive systems was discussed.

Apart from drug/gene delivery applications of thermoresponsive NPs, their other innovative applications for delivery of antimicrobial agents, anti-inflammatory drugs, enzymes as well as codelivery systems and theranostic nanosystems were mentioned.

Regarding the question, why temperature sensitive systems are considered significantly important besides other stimulus sensitive such as pH, light, enzyme, etc. One answer refers to the abnormal temperature of inflamed tissue (tumor), which has higher temperature in comparison with body temperature and can acts as a endogenous stimuli in order to nanocarrier activation, thus the therapeutic cargo can be released. Another reason is that external heating as an exogenous temperature can activate nanocarrier operation. Also drugs can be loaded in the nanovehicles in liquid state and without any surgery injected to the body. But despite many advantages, some challenges still exist. Although PNIPAAm due to the LCST about round 32 °C is very appealing for DGDSs, but its nonbiodegradable properties in body is a disadvantage; hence to overcome this disadvantage, PNIPAAm can copolymerized with PEG or polysaccharides. Encapsulating drugs and using membranes are some other appropriate solutions to decrease the initial burst release and have a continuous release without overdose. Toxicity of some drugs such as anticancer drugs limits their application, but temperature-sensitive nanocarriers have been presented to overcome this challenge. For reducing the problems with some hydrophobic drugs, core–shell structure and interpenetrating network are alternative strategies. Hydrophilic or hydrophobic polymers or photoresponsive groups such as azobenzene can conjugated to nanocarriers therefore inducing changes in the LCST or UCST for specific applications and increasing or decreasing the drug release rate. However, significant progresses that have been made in thermoresponsive DDSs suggest a hopeful future for exploring more practical targeted methods for cancer treatment. Another future direction for smart thermoresponsive stimuli-responsive NPs could be their application for treatment of chronic diseases, such as diabetes and hypercholesterolemia, which require regular doses of the drug to be administered. These nanocarriers by providing continuous and delayed release extend the specific period of drug effectiveness, so patients could have an injection only every 1 or 2 weeks, instead of each and every day. In addition to the temperature, other stimulus such as pH, enzyme, light irradiation, electricity and magnetic field, ultrasound, mechanical forces (e.g., shear stresses, etc.) can be added to temperature stimuli system through a logic gate algorithm and form the dual/triple and even multi-responsive nanocarriers for better efficiency. Also in the near future, codelivery systems for simultaneous gene and drug delivery, or dual therapy (chemotherapy and hyperthermia) plus MRI can be accomplished. Novel smart nanocarriers such as nanobombs, which can be swelled 800-fold by changing the temperature, because of inducing physical/mechanical forces can breakdown the diseased tissue, in addition to targeted delivery and release of drugs. Anyway, in answer to “How is our position for smart delivery of therapeutic agents, especially through administrating thermoresponsive nanosystems in the next ten years?”, it must be said that the future is bright.

References

1. Wang C, Xu H, Liang C, Liu Y, Li Z, Yang G, Cheng L, Li Y, Liu Z. Iron Oxide@ Polypyrrole Nanoparticles As A Multifunctional Drug Carrier For Remotely Controlled Cancer Therapy With Synergistic Antitumor Effect. ACS Nano. 2013; 7(8):6782–6795. [PubMed: 23822176]

2. Sahoo B, Devi KSP, Banerjee R, Maiti TK, Pramanik P, Dhara D. Thermal And pH Responsive Polymer-Tethered Multifunctional Magnetic Nanoparticles For Targeted Delivery Of Anticancer Drug. ACS Appl Mater Interfaces. 2013; 5(9):3884–3893. [PubMed: 23551195]
3. De Smet M, Langereis S, Van Den Bosch S, Bitter K, Hijnen NM, Heijman E, Grüll H. SPECT/CT Imaging Of Temperature-Sensitive Liposomes For MR-Image Guided Drug Delivery With High Intensity Focused Ultrasound. J Controlled Release. 2013; 169(1):82–90.
4. Wang AZ, Langer R, Farokhzad OC. Nanoparticle Delivery Of Cancer Drugs. Annu Rev Med. 2012; 63:185–198. [PubMed: 21888516]
5. Mudshinge SR, Deore AB, Patil S, Bhalgat CM. Nanoparticles: Emerging Carriers For Drug Delivery. Saudi Pharm J. 2011; 19(3):129–141. [PubMed: 23960751]
6. Mazaheri M, Eslahi N, Ordikhani F, Tamjid E, Simchi A. Nanomedicine Applications In Orthopedic Medicine: State Of The Art. Int J Nanomed. 2015; 10:6039.
7. Ordikhani F, Simchi A. Long-Term Antibiotic Delivery By Chitosan-Based Composite Coatings With Bone Regenerative Potential. Appl Surf Sci. 2014; 317:56–66.
8. Ordikhani F, Ramezani Farani M, Dehghani M, Tamjid E, Simchi A. Physicochemical And Biological Properties Of Electro-deposited Graphene Oxide/Chitosan Films With Drug-Eluting Capacity. Carbon. 2015; 84:91–102.
9. Karimi M, Zare H, Bakhshian Nik A, Yazdani N, Hamrang M, Mohamed E, Sahandi Zangabad P, Moosavi Basri SM, Bakhtiari L, Hamblin MR. Nanotechnology In Diagnosis And Treatment Of Coronary Artery Disease. Nanomedicine. 2016; 11(5):513–530. [PubMed: 26906471]
10. Yeh JC, Hsu YT, Su CM, Wang MC, Lee TH, Lou SL. Preparation And Characterization Of Biocompatible And Thermoresponsive Micelles Based On Poly (N-Isopropylacrylamide-Co-N, N-Dimethylacrylamide) Grafted On Polysuccinimide For Drug Delivery. J Biomater Appl. 2014; 29:442–453. [PubMed: 24825758]
11. Wadajkar AS, Menon JU, Tsai Y-S, Gore C, Dobin T, Gandee L, Kangasniemi K, Takahashi M, Manandhar B, Ahn J-M. Prostate Cancer-Specific Thermo-Responsive Polymer-Coated Iron Oxide Nanoparticles. Biomaterials. 2013; 34(14):3618–3625. [PubMed: 23419645]
12. Mofazzal Jahromi MA, Karimi M, Azadmanesh K, Naderi Manesh H, Hassan ZM, Moazzeni SM. The Effect Of Chitosan-Tripolyphosphate Nanoparticles On Maturation And Function Of Dendritic Cells. Comp Clin Pathol. 2014; 23(5):1421–1427.
13. Karimi M, Avci P, Mobasseri R, Hamblin MR, Naderi-Manesh H. The Novel Albumin–Chitosan Core–Shell Nanoparticles For Gene Delivery: Preparation, Optimization And Cell Uptake Investigation. J Nanopart Res. 2013; 15(5):1–14.
14. Karimi M, Solati N, Ghasemi A, Estiar MA, Hashemkhani M, Kiani P, Mohamed E, Saeidi A, Taheri M, Avci P, et al. Carbon Nanotubes Part II: A Remarkable Carrier For Drug And Gene Delivery. Expert Opin Drug Delivery. 2015; 12:1089–1105.
15. Karimi M, Solati N, Amiri M, Mirshekari H, Mohamed E, Taheri M, Hashemkhani M, Saeidi A, Estiar MA, Kiani P, et al. Carbon Nanotubes Part I: Preparation Of A Novel And Versatile Drug-Delivery Vehicle. Expert Opin Drug Delivery. 2015; 12:1071–1087.
16. Karimi M, Bahrami S, Ravari SB, Zangabad PS, Mirshekari H, Bozorgomid M, Shahreza S, Sori M, Hamblin MR. Albumin Nanostructures As Advanced Drug Delivery Systems. Expert Opin Drug Delivery. 2016; 1
17. Karimi M, Mirshekari H, Aliakbari M, Sahandi-Zangabad P, Hamblin MR. Smart Mesoporous Silica Nanoparticles For Controlled-Release Drug Delivery. Nanotechnol Rev. 2016; 5:195–297.
18. Mura S, Nicolas J, Couvreur P. Stimuli-Responsive Nanocarriers For Drug Delivery. Nat Mater. 2013; 12(11):991–1003. [PubMed: 24150417]
19. Cheng R, Meng F, Deng C, Klok H-A, Zhong Z. Dual And Multi-Stimuli Responsive Polymeric Nanoparticles For Programmed Site-Specific Drug Delivery. Biomaterials. 2013; 34(14):3647–3657. [PubMed: 23415642]
20. Karimi M, Eslami M, Sahandi-Zangabad P, Mirab F, Farajisafiloo N, Shafaei Z, Ghosh D, Bozorgomid M, Dashkhaneh F, Hamblin MR. pH-Sensitive Stimulus-Responsive Nanocarriers For Targeted Delivery Of Therapeutic Agents. WIREs Nanomed Nanobiotechnol. 2016; doi: 10.1002/wnan.1389

21. Karimi, M.; Zangabad, PS.; Ghasemi, A.; Hamblin, MR. Smart Internal Stimulus-Responsive Nanocarriers For Drug And Gene Delivery. IOP Concise Physics; Morgan & Claypool Publishers; San Rafael, CA: 2015.
22. Karimi, M.; Zangabad, PS.; Ghasemi, A.; Hamblin, MR. Smart External Stimulus-Responsive Nanocarriers For Drug And Gene Delivery. Morgan & Claypool Publishers; San Rafael, CA: 2015. Electrical And Mechanical-Responsive Nanocarriers.
23. Karimi, M.; Zangabad, PS.; Ghasemi, A.; Hamblin, MR. Smart External Stimulus-Responsive Nanocarriers For Drug And Gene Delivery. Morgan & Claypool Publishers; San Rafael, CA: 2015. Light-Sensitive Nanocarriers.
24. Bando A, Mayumi K, Yokoyama H, Ito K. Theory Of Volume Phase Transition Of Slide-Ring Gels. React Funct Polym. 2013; 73(7):904–910.
25. Jagur-Grodzinski J. Polymeric Gels And Hydrogels For Biomedical and Pharmaceutical Applications. Polym Adv Technol. 2010; 21(1):27–47.
26. Zhou J, Pishko MV, Lutkenhaus JL. Thermoresponsive Layer-By-Layer Assemblies For Nanoparticle-Based Drug Delivery. Langmuir. 2014; 30(20):5903–5910. [PubMed: 24787428]
27. Sun F, Wang Y, Wei Y, Cheng G, Ma G. Thermo-Triggered Drug Delivery From Polymeric Micelles Of Poly (N-Isopropylacrylamide-Co-Acrylamide)-B-Poly (N-Butyl Methacrylate) For Tumor Targeting. J Bioact Compat Polym. 2014; 29:301.
28. Chen G, Chen R, Zou C, Yang D, Chen Z-S. Fragmented Polymer Nanotubes From Sonication-Induced Scission With A Thermo-Responsive Gating System For Anti-Cancer Drug Delivery. J Mater Chem B. 2014; 2(10):1327–1334.
29. Kashyap S, Jayakannan M. Thermo-Responsive And Shape Transformable Amphiphilic Scaffolds For Loading And Delivering Anticancer Drugs. J Mater Chem B. 2014; 2(26):4142–4152.
30. Bae YH, Okano T, Kim SW. Insulin Permeation Through Thermo-Sensitive Hydrogels. J Controlled Release. 1989; 9(3):271–279.
31. Okano T, Bae Y, Jacobs H, Kim S. Thermally On-Off Switching Polymers For Drug Permeation And Release. J Controlled Release. 1990; 11(1):255–265.
32. Okahata Y, Noguchi H, Seki T. Thermoselective Permeation From A Polymer-Grafted Capsule Membrane. Macromolecules. 1986; 19(2):493–494.
33. Chen YY, Wu HC, Sun JS, Dong GC, Wang TW. Injectable And Thermoresponsive Self-Assembled Nanocomposite Hydrogel For Long-Term Anticancer Drug Delivery. Langmuir. 2013; 29(11):3721–3729. [PubMed: 23441993]
34. Schmaljohann D. Thermo-And pH-Responsive Polymers In Drug Delivery. Adv Drug Delivery Rev. 2006; 58(15):1655–1670.
35. Beija M, Marty J-D, Destarac M. Thermoresponsive Poly (N-Vinyl Caprolactam)-Coated Gold Nanoparticles: Sharp Reversible Response And Easy Tunability. Chem Commun. 2011; 47(10): 2826–2828.
36. Elluru M, Ma H, Hadjiargyrou M, Hsiao BS, Chu B. Synthesis And Characterization Of Biocompatible Hydrogel Using Pluronics-Based Block Copolymers. Polymer. 2013; 54(8):2088–2095.
37. Picos-Corrales LA, Licea-Claveríe A, Arndt KF. Bisensitive Core–Shell Nanohydrogels By E-Beam Irradiation Of Micelles. React Funct Polym. 2014; 75:31–40.
38. Nakayama M, Chung J, Miyazaki T, Yokoyama M, Sakai K, Okano T. Thermal Modulation Of Intracellular Drug Distribution Using Thermoresponsive Polymeric Micelles. React Funct Polym. 2007; 67(11):1398–1407.
39. Chiang WL, Lin TT, Sureshbabu R, Chia WT, Hsiao HC, Liu HY, Yang CM, Sung HW. A Rapid Drug Release System With A NIR Light-Activated Molecular Switch For Dual-Modality Photothermal/Antibiotic Treatments Of Subcutaneous Abscesses. J Controlled Release. 2015; 199:53–62.
40. Ryu JS, Raucher D. Anti-Tumor Efficacy Of A Therapeutic Peptide Based On Thermo-Responsive Elastin-Like Polypeptide In Combination With Gemcitabine. Cancer Lett. 2014; 348(1):177–184. [PubMed: 24680816]

41. Muzzarelli RA, Greco F, Busilacchi A, Sollazzo V, Gigante A. Chitosan, Hyaluronan And Chondroitin Sulfate In Tissue Engineering For Cartilage Regeneration: A Review. Carbohydr Polym. 2012; 89(3):723–739. [PubMed: 24750856]
42. Chen M-C, Mi F-L, Liao Z-X, Hsiao C-W, Sonaje K, Chung M-F, Hsu L-W, Sung H-W. Recent Advances In Chitosan-Based Nanoparticles For Oral Delivery Of Macromolecules. Adv Drug Delivery Rev. 2013; 65(6):865–879.
43. Yin H, Casey PS. Effects Of Iron Or Manganese Doping Of Zno Nanoparticles On Their Dissolution, ROS Generation And Cytotoxicity. RSC Adv. 2014; 4(50):26149–26157.
44. Akimoto J, Nakayama M, Sakai K, Okano T. Temperature-Induced Intracellular Uptake Of Thermoresponsive Polymeric Micelles. Biomacromolecules. 2009; 10(6):1331–1336. [PubMed: 19358525]
45. Efthimiadou EK, Tapeinos C, Tziveleka LA, Boukos N, Kordas G. pH-And Thermo-Responsive Microcontainers As Potential Drug Delivery Systems: Morphological Characteristic, Release And Cytotoxicity Studies. Mater Sci Eng, C. 2014; 37:271–277.
46. Huang X, Jiang X, Yang Q, Chu Y, Zhang G, Yang B, Zhuo R. Triple-Stimuli (pH/Thermo/Reduction) Sensitive Copolymers For Intracellular Drug Delivery. J Mater Chem B. 2013; 1(13):1860–1868.
47. Liu J, Detrembleur C, Debuigne A, De Pauw-Gillet M-C, Mornet S, Vander Elst L, Laurent S, Duguet E, Jérôme C. Glucose-, pH-And Thermo-Responsive Nanogels Crosslinked By Functional Superparamagnetic Maghemite Nanoparticles As Innovative Drug Delivery Systems. J Mater Chem B. 2014; 2(8):1009–1023.
48. Kulkarni RV, Boppana R, Krishna Mohan G, Mutalik S, Kalyane NV. pH-Responsive Interpenetrating Network Hydrogel Beads Of Poly (Acrylamide)-G-Carrageenan And Sodium Alginate For Intestinal Targeted Drug Delivery: Synthesis, In Vitro And In Vivo Evaluation. J Colloid Interface Sci. 2012; 367(1):509–517. [PubMed: 22047923]
49. Bardajee GR, Pourjavadi A, Ghavami S, Soleyman R, Jafarpour F. UV-Prepared Salep-Based Nanoporous Hydrogel For Controlled Release Of Tetracycline Hydrochloride In Colon. J Photochem Photobiol, B. 2011; 102(3):232–240. [PubMed: 21251847]
50. Motornov M, Roiter Y, Tokarev I, Minko S. Stimuli-Responsive Nanoparticles, Nanogels And Capsules For Integrated Multifunctional Intelligent Systems. Prog Polym Sci. 2010; 35(1):174–211.
51. Yom-Tov O, Frisman I, Seliktar D, Bianco-Peled H. A Novel Method For Hydrogel Nanostructuring. Eur Polym J. 2014; 52:137–145.
52. Bardajee GR, Pourjavadi A, Soleyman R. Novel Nano-Porous Hydrogel As A Carrier Matrix For Oral Delivery Of Tetracycline Hydrochloride. Colloids Surf, A. 2011; 392(1):16–24.
53. Némethy Á, Solti K, Kiss L, Gyarmati B, Deli MA, Csányi E, Szilágyi A. pH-And Temperature-Responsive Poly (Aspartic Acid)-L-Poly (N-Isopropylacrylamide) Conetwork Hydrogel. Eur Polym J. 2013; 49(9):2392–2403.
54. López-Cebral R, Martin-Pastor M, Paolicelli P, Casadei MA, Seijo B, Sanchez A. Application Of NMR Spectroscopy In The Development Of A Biomimetic Approach For Hydrophobic Drug Association With Physical Hydrogels. Colloids Surf, B. 2014; 115:391–399.
55. Gasser U, Lietor-Santos J-J, Scotti A, Bunk O, Menzel A, Fernandez-Nieves A. Transient Formation Of Bcc Crystals In Suspensions Of Poly (N-Isopropylacrylamide)-Based Microgels. Phys Rev E. 2013; 88(5):052308.
56. Behrens MA, Bergenholtz J, Pedersen JS. Temperature-Induced Attractive Interactions Of PEO-Containing Block Copolymer Micelles. Langmuir. 2014; 30(21):6021–6029. [PubMed: 24850568]
57. Khorsand Sourkohi B, Cunningham A, Zhang Q, Oh JK. Biodegradable Block Copolymer Micelles With Thiol-Responsive Sheddable Coronas. Biomacromolecules. 2011; 12(10):3819–3825. [PubMed: 21879701]
58. Ulasan M, Yavuz E, Bagriacik EU, Cengeloglu Y, Yavuz MS. Biocompatible Thermoresponsive PEGMA Nanoparticles Crosslinked With Cleavable Disulfide-Based Crosslinker For Dual Drug Release. J Biomed Mater Res, Part A. 2015; 103(1):243–251.
59. Bölgen N, Aguilar MR, Fernández MDM, Gonzalo-Flores S, Villar-Rodil S, San Román J, Piskin E. Thermoresponsive Biodegradable HEMA-Lactate-Dextran-Co-NIPA Cryogels For Controlled

Release Of Simvastatin. Artif Cells, Nanomed Biotechnol. 2015; 43(1):40–49. [PubMed: 24047541]

60. Cardoso AM, Calejo MT, Morais CM, Cardoso AL, Cruz R, Zhu K, Pedroso De Lima MC, Jurado ALS, NyströM B. Application Of Thermoresponsive PNIPAAM-B-PAMPTMA Di-block Copolymers In Sirna Delivery. Mol Pharmaceutics. 2014; 11(3):819–827.
61. Yuan Q, Yeudall WA, Yang H. Thermoresponsive Dendritic Facial Amphiphiles For Gene Delivery. Nanomedicine And Nanobiology. 2014; 1(1):64–69.
62. Luo YL, Yang XL, Xu F, Chen YS, Zhang B. Thermosensitive PNIPAM-B-HTPB Block Copolymer Micelles: Molecular Architectures And Camptothecin Drug Release. Colloids Surf, B. 2014; 114:150–157.
63. Men K, Liu W, Li L, Duan X, Wang P, Gou M, Wei X, Gao X, Wang B, Du Y, et al. Delivering Instilled Hydrophobic Drug To The Bladder By A Cationic Nanoparticle And Thermo-Sensitive Hydrogel Composite System. Nanoscale. 2012; 4(20):6425–6433. [PubMed: 22955255]
64. Vaupel P, Kallinowski F, Okunieff P. Blood Flow, Oxygen And Nutrient Supply, And Metabolic Microenvironment Of Human Tumors: A Review. Cancer Res. 1989; 49(23):6449–6465. [PubMed: 2684393]
65. Ghosh K, Thodeti CK, Dudley AC, Mammoto A, Klagsbrun M, Ingber DE. Tumor-Derived Endothelial Cells Exhibit Aberrant Rho-Mediated Mechanosensing And Abnormal Angiogenesis In Vitro. Proc Natl Acad Sci U S A. 2008; 105(32):11305–11310. [PubMed: 18685096]
66. Deberardinis RJ, Lum JJ, Hatzivassiliou G, Thompson CB. The Biology Of Cancer: Metabolic Reprogramming Fuels Cell Growth And Proliferation. Cell Metab. 2008; 7(1):11–20. [PubMed: 18177721]
67. Bies C, Lehr C-M, Woodley JF. Lectin-Mediated Drug Targeting: History And Applications. Adv Drug Delivery Rev. 2004; 56(4):425–435.
68. Wust P, Hildebrandt B, Sreenivasa G, Rau B, Gellermann J, Riess H, Felix R, Schlag P. Hyperthermia In Combined Treatment Of Cancer. Lancet Oncol. 2002; 3(8):487–497. [PubMed: 12147435]
69. Indiana I. Triggering In Drug Delivery Systems. Adv Drug Delivery Rev. 2001; 53:245.
70. Ward MA, Georgiou TK. Thermoresponsive Polymers For Biomedical Applications. Polymers. 2011; 3(3):1215–1242.
71. Park JS, Yang HN, Woo DG, Jeon SY, Park K-H. Poly (N-Isopropylacrylamide-Co-Acrylic Acid) Nanogels For Tracing And Delivering Genes To Human Mesenchymal Stem Cells. Biomaterials. 2013; 34(34):8819–8834. [PubMed: 23937912]
72. Shen Z, Shi B, Zhang H, Bi J, Dai S. Exploring Low-Positively Charged Thermosensitive Copolymers As Gene Delivery Vectors. Soft Matter. 2012; 8(5):1385–1394.
73. Boustta M, Colombo PE, Lenglet S, Poujol S, Vert M. Versatile UCST-Based Thermoresponsive Hydrogels For Loco-Regional Sustained Drug Delivery. J Controlled Release. 2014; 174:1–6.
74. Nirmal HB, Bakliwal SR, Pawar SP. In-Situ Gel: Newtrends In Controlled And Sustained Drug Delivery System. Int J Pharmtech Res. 2010; 2(2):1398–1408.
75. Caldorera-Moore ME, Liechty WB, Peppas NA. Responsive Theranostic Systems: Integration Of Diagnostic Imaging Agents And Responsive Controlled Release Drug Delivery Carriers. Acc Chem Res. 2011; 44(10):1061–1070. [PubMed: 21932809]
76. Chang Y, Yandi W, Chen WY, Shih YJ, Yang CC, Chang Y, Ling QD, Higuchi A. Tunable Bioadhesive Copolymer Hydrogels Of Thermoresponsive Poly(N-Isopropyl Acrylamide) Containing Zwitterionic Polysulfobetaine. Biomacromolecules. 2010; 11:1101–1110. [PubMed: 20201492]
77. Saravanakumar G, Kim WJ. Stimuli-Responsive Polymeric Nanocarriers As Promising Drug And Gene Delivery Systems. Fundamental Biomedical Technologies. 2014; 7:55–91.
78. Pippa N, Meristoudi A, Pispas S, Demetzos C. Temperature-Dependent Drug Release From DPPC: C 12 H 25-PNIPAM-COOH Liposomes: Control Of The Drug Loading/Release By Modulation Of The Nanocarriers' Components. Int J Pharm. 2015; 485(1):374–382. [PubMed: 25776453]
79. Gao X, Cao Y, Song X, Zhang Z, Xiao C, He C, Chen X. pH-And Thermo-Responsive Poly (N-Isopropylacrylamide-Co-Acrylic Acid Derivative) Copolymers And Hydrogels With LCST Dependent On pH And Alkyl Side Groups. J Mater Chem B. 2013; 1(41):5578–5587.

80. Yoo M, Sung Y, Lee Y, Cho C. Effect Of Polyelectrolyte On The Lower Critical Solution Temperature Of Poly (N-Isopropyl Acrylamide) In The Poly (Nipaam-Co-Acrylic Acid) Hydrogel. Polymer. 2000; 41(15):5713–5719.
81. Guo P, Li H, Ren W, Zhu J, Xiao F, Xu S, Wang J. Unusual Thermo-Responsive Behaviors Of Poly (NIPAM-Co-AM)/PEG/PTA Composite Hydrogels. Mater Lett. 2015; 143:24–26.
82. Lue SJ, Chen C-H, Shih C-M. Tuning Of Lower Critical Solution Temperature (LCST) Of Poly (N-Isopropylacrylamide-Co-Acrylic Acid) Hydrogels. J Macromol Sci, Part B: Phys. 2011; 50(3): 563–579.
83. Yang J, Van Lith R, Baler K, Hoshi RA, Ameer GA. A Thermoresponsive Biodegradable Polymer With Intrinsic Antioxidant Properties. Biomacromolecules. 2014; 15(11):3942–3952. [PubMed: 25295411]
84. Cha R, He Z, Ni Y. Preparation And Characterization Of Thermal/pH-Sensitive Hydrogel From Carboxylated Nanocrystalline Cellulose. Carbohydr Polym. 2012; 88(2):713–718.
85. Zheng Y, Zheng S. Poly (Ethylene Oxide)-Grafted Poly (N-Isopropylacrylamide) Networks: Preparation, Characterization And Rapid Deswelling And Reswelling Behavior Of Hydrogels. React Funct Polym. 2012; 72(3):176–184.
86. Soni G, Yadav KS. High Encapsulation Efficiency Of Poloxamer-Based Injectable Thermoresponsive Hydrogels Of Etoposide. Pharm Dev Technol. 2014; 19(6):651–661. [PubMed: 23879721]
87. Oddo L, Masci G, Di Meo C, Capitani D, Mannina L, Lamanna R, De Santis S, Alhaique F, Coviello T, Matricardi P. Novel Thermosensitive Calcium Alginate Microspheres: Physico-Chemical Characterization And Delivery Properties. Acta Biomater. 2010; 6(9):3657–3664. [PubMed: 20226887]
88. Cheng X, Jin Y, Sun T, Qi R, Fan B, Li H. Oxidation- And Thermo-Responsive Poly(N-Isopropylacrylamide-Co-2-Hydroxyethyl Acrylate) Hydrogels Cross-Linked Via Diselenides For Controlled Drug Delivery. RSC Adv. 2015; 5(6):4162–4170.
89. Bastakoti BP, Guragain S, Nakashima K, Yamauchi Y. Stimuli-Induced Core–Corona Inversion Of Micelle Of Poly (Acrylic Acid)-Block-Poly (N-Isopropylacrylamide) And Its Application In Drug Delivery. Macromol Chem Phys. 2015; 216(3):287–291.
90. Pennadam SS, Firman K, Alexander C, Górecki DC. Protein-Polymer Nano-Machines. Towards Synthetic Control Of Biological Processes. J Nanobiotechnol. 2004; 2(8):8.
91. Zha L, Banik B, Alexis F. Stimulus Responsive Nanogels For Drug Delivery. Soft Matter. 2011; 7(13):5908–5916.
92. Cheng X, Jin Y, Sun T, Qi R, Fan B, Li H. Oxidation-And Thermo-Responsive Poly (N-Isopropylacrylamide-Co-2-Hydroxyethyl Acrylate) Hydrogels Cross-Linked Via Diselenides For Controlled Drug Delivery. RSC Adv. 2015; 5(6):4162–4170.
93. Mayol L, De Stefano D, De Falco F, Carnuccio R, Maiuri MC, De Rosa G. Effect Of Hyaluronic Acid On The Thermogelation And Biocompatibility Of Its Blends With Methyl Cellulose. Carbohydr Polym. 2014; 112:480–485. [PubMed: 25129771]
94. Lü S, Liu M, Ni B. Degradable, Injectable Poly (N-Isopropylacrylamide)-Based Hydrogels With Low Gelation Concentrations For Protein Delivery Application. Chem Eng J. 2011; 173(1):241–250.
95. Kim DY, Lee BN, Seo HW, Kwon JS, Lee B, Han DK, Kim JH, Min BH, Park K, Kim MS. Injectable In Situ-Forming Hydrogels For A Suppression Of Drug Burst From Drug-Loaded Microcapsules. Soft Matter. 2012; 8(29):7638–7648.
96. Zhao J, Guo B, Ma PX. Injectable Alginate Microsphere/PLGA–PEG–PLGA Composite Hydrogels For Sustained Drug Release. RSC Adv. 2014; 4(34):17736–17742.
97. Manokruang K, Lym JS, Lee DS. Injectable Hydrogels Based On Poly (Amino Urethane) Conjugated Bovine Serum Albumin. Mater Lett. 2014; 124:105–109.
98. Shi J, Zhang Z, Li G, Cao S. Biomimetic Fabrication Of Alginate/Caco 3 Hybrid Beads For Dual-Responsive Drug Delivery Under Compressed CO 2. J Mater Chem. 2011; 21(40):16028–16034.
99. Wu HQ, Wang C. Biodegradable Smart Nanogels: A New Platform for Targeting Drug Delivery and Biomedical Diagnostics. Langmuir. 2016; 32:6211. [PubMed: 27255455]

100. Zhang X, Malhotra S, Molina M, Haag R. Micro-and nanogels with labile crosslinks–from synthesis to biomedical applications. Chem Soc Rev. 2015; 44(7):1948–1973. [PubMed: 25620415]
101. Fathi M, Entezami AA, Arami S, Rashidi MR. Preparation of N-isopropylacrylamide/itaconic acid magnetic nano-hydrogels by modified starch as a crosslinker for anticancer drug carriers. Int J Polym Mater. 2015; 64(10):541–549.
102. Tang Y, Zhang S, Wang M, Zhu J, Sun T, Jiang G. A Glucose-Based Diblock Copolymer: Synthesis, Characterization And Its Injectable/Temperature-Sensitive Behaviors. J Polym Res. 2014; 21(4):1–8.
103. Peña B, Shandas R, Park D. A Heparin-Mimicking Reverse Thermal Gel For Controlled Delivery Of Positively Charged Proteins. J Biomed Mater Res, Part A. 2015; 103(6):2102–2108.
104. Chee PL, Prasad A, Fang X, Owh C, Yeo VJJ, Loh XJ. Supramolecular Cyclodextrin Pseudorotaxane Hydrogels: A Candidate For Sustained Release? Mater Sci Eng, C. 2014; 39:6–12.
105. Liu F, Seuring J, Agarwal S. A Non-Ionic Thermophilic Hydrogel With Positive Thermosensitivity In Water And Electrolyte Solution. Macromol Chem Phys. 2014; 215(15): 1466–1472.
106. Yu J, Xu X, Yao F, Luo Z, Jin L, Xie B, Shi S, Ma H, Li X, Chen H. In Situ Covalently Cross-Linked PEG Hydrogel For Ocular Drug Delivery Applications. Int J Pharm. 2014; 470(1):151–157. [PubMed: 24768405]
107. Mishra GP, Kinser R, Wierzbicki IH, Alany RG, Alani AW. In Situ Gelling Polyvalerolactone-Based Thermosensitive Hydrogel For Sustained Drug Delivery. Eur J Pharm Biopharm. 2014; 88(2):397–405. [PubMed: 24931340]
108. Ha W, Yu J, Song XY, Chen J, Shi YP. Tunable Temperature-Responsive Supramolecular Hydrogels Formed By Prodrugs As A Codelivery System. ACS Appl Mater Interfaces. 2014; 6(13):10623–10630. [PubMed: 24919142]
109. Wei H, Zhuo R-X, Zhang X-Z. Design And Development Of Polymeric Micelles With Cleavable Links For Intracellular Drug Delivery. Prog Polym Sci. 2013; 38(3):503–535.
110. Nishiyama N, Kataoka K. Current State, Achievements, And Future Prospects Of Polymeric Micelles As Nanocarriers For Drug And Gene Delivery. Pharmacol Ther. 2006; 112(3):630–648. [PubMed: 16815554]
111. Chen C-Y, Kim TH, Wu W-C, Huang C-M, Wei H, Mount CW, Tian Y, Jang S-H, Pun SH, Jen AKY. pH-Dependent, Thermosensitive Polymeric Nanocarriers For Drug Delivery To Solid Tumors. Biomaterials. 2013; 34(18):4501–4509. [PubMed: 23498892]
112. Massoumi B, Abdollahi M, Fathi M, Entezami AA, Hamidi S. Synthesis of novel thermoresponsive micelles by graft copolymerization of *N*-isopropylacrylamide on poly (*ε*-caprolactone-co-*α*-bromo-*ε*-caprolactone) as macroinitiator via ATRP. J Polym Res. 2013; 20(2): 1–8.
113. Wang X, Li S, Wan Z, Quan Z, Tan Q. Investigation Of Thermo-Sensitive Amphiphilic Micelles As Drug Carriers For Chemotherapy In Cholangiocarcinoma In Vitro And In Vivo. Int J Pharm. 2014; 463(1):81–88. [PubMed: 24406676]
114. Wang F, Klaikherd A, Thayumanavan S. Temperature Sensitivity Trends And Multi-Stimuli Sensitive Behavior In Amphiphilic Oligomers. J Am Chem Soc. 2011; 133(34):13496–13503. [PubMed: 21739959]
115. Vancoillie G, Frank D, Hoogenboom R. Thermoresponsive Poly(Oligo Ethylene Glycol Acrylates). Prog Polym Sci. 2014; 39(6):1074–1095.
116. Yuan W, Zhang J, Wei J, Yuan H, Ren J. Synthesis, Characterization, And Properties Of Tunable Thermosensitive Amphiphilic Dendrimer-Star Copolymers With Y-Shaped Arms. J Polym Sci, Part A: Polym Chem. 2011; 49(18):4071–4080.
117. Li X, Qian Y, Liu T, Hu X, Zhang G, You Y, Liu S. Amphiphilic Multiarm Star Block Copolymer-Based Multifunctional Unimolecular Micelles For Cancer Targeted Drug Delivery And MR Imaging. Biomaterials. 2011; 32(27):6595–6605. [PubMed: 21663960]

118. Zhu Z, Gao N, Wang H, Sukhishvili SA. Temperature-Triggered On-Demand Drug Release Enabled By Hydrogen-Bonded Multilayers Of Block Copolymer Micelles. J Controlled Release. 2013; 171(1):73–80.
119. Lu Y, Ballauff M. Thermosensitive Core–Shell Microgels: From Colloidal Model Systems To Nanoreactors. Prog Polym Sci. 2011; 36(6):767–792.
120. Kleinen J, Klee A, Richtering W. Influence Of Architecture On The Interaction Of Negatively Charged Multisensitive Poly (N-Isopropylacrylamide)-Co-Methacrylic Acid Microgels With Oppositely Charged Polyelectrolyte: Absorption Vs Adsorption. Langmuir. 2010; 26(13):11258–11265. [PubMed: 20377221]
121. Sun Y-N, Gao G-R, Du G-L, Cheng Y-J, Fu J. Super Tough, Ultrastretchable, And Thermoresponsive Hydrogels With Functionalized Triblock Copolymer Micelles As Macro-Cross-Linkers. ACS Macro Lett. 2014; 3(5):496–500.
122. Lu Y, Wittemann A, Ballauff M, Drechsler M. Preparation Of Polystyrene-Poly(N-Isopropylacrylamide) (PS-PNIPA) Core-Shell Particles By Photoemulsion Polymerization. Macromol Rapid Commun. 2006; 27(14):1137–1141.
123. Zhou Y, Hua S, Yu J, Dong P, Liu F, Hua D. A Strategy For Effective Radioprotection By Chitosan-Based Long-Circulating Nanocarriers. J Mater Chem B. 2015; 3(15):2931–2934.
124. Huang W, Wang Y, Zhang S, Huang L, Hua D, Zhu X. A Facile Approach For Controlled Modification Of Chitosan Under Γ-Ray Irradiation For Drug Delivery. Macromolecules. 2013; 46(3):814–818.
125. Tsitsilianis C. Responsive Reversible Hydrogels From Associative “Smart” Macromolecules. Soft Matter. 2010; 6(11):2372–2388.
126. Klouda L. Thermoresponsive Hydrogels In Biomedical Applications: A Seven-Year Update. Eur J Pharm Biopharm. 2015; 97:338–349. [PubMed: 26614556]
127. Akhtar MF, Hanif M, Ranjha NM. Methods Of Synthesis Of Hydrogels… A Review. Saudi Pharm J. 2015; doi: 10.1016/j.jsps.2015.03.022
128. Lee JS, Zhou W, Meng F, Zhang D, Otto C, Feijen J. Thermosensitive Hydrogel-Containing Polymersomes For Controlled Drug Delivery. J Controlled Release. 2010; 146(3):400–408.
129. Wang W, Tian X, Feng Y, Cao B, Yang W, Zhang L. Thermally On– Off Switching Membranes Prepared By Pore-Filling Poly (N-Isopropylacrylamide) Hydrogels. Ind Eng Chem Res. 2010; 49(4):1684–1690.
130. Kuroki H, Ohashi H, Ito T, Tamaki T, Yamaguchi T. Isolation And Analysis Of A Grafted Polymer Onto A Straight Cylindrical Pore In A Thermal-Responsive Gating Membrane And Elucidation Of Its Permeation Behavior. J Membr Sci. 2010; 352(1):22–31.
131. Ikram S, Kumari M, Gupta B. Thermosensitive Membranes By Radiation-Induced Graft Polymerization Of N-Isopropyl Acryl-amide/Acrylic Acid On Polypropylene Nonwoven Fabric. Radiat Phys Chem. 2011; 80(1):50–56.
132. Evingür GA, Pekcan Ö. Effect Of LCST On The Swelling Of Paam-NIPA Copolymers: A Fluorescence Study. Polym Bull. 2012; 68(1):223–238.
133. Lue SJ, Chen C-H, Shih C-M, Tsai M-C, Kuo C-Y, Lai J-Y. Grafting Of Poly (N-Isopropylacrylamide-Co-Acrylic Acid) On Micro-Porous Polycarbonate Films: Regulating Lower Critical Solution Temperatures For Drug Controlled Release. J Membr Sci. 2011; 379(1): 330–340.
134. Yang B, Yang W. Thermo-Sensitive Switching Membranes Regulated By Pore-Covering Polymer Brushes. J Membr Sci. 2003; 218:247–255.
135. Pan K, Zhang X, Ren R, Cao B. Double Stimuli-Responsive Membranes Grafted With Block Copolymer By ATRP Method. J Membr Sci. 2010; 356(1):133–137.
136. Tsai M-C, Shih C-M, Lue SJ. Drug Permeation Behavior Through Thermo-And pH-Responsive Polycarbonate-G-Poly (N-Isopropylacrylamide-Co-Acrylic Acid) Composites. Polym Bull. 2013; 70(3):1003–1017.
137. Fang J, Xuan Y, Li Q. Preparation Of Three-Dimensionally Ordered Macroporous Perovskite Materials. Chin Sci Bull. 2011; 56(20):2156–2161.

138. Woo S-W, Dokko K, Kanamura K. Preparation And Characterization Of Three Dimensionally Ordered Macroporous Li 4 Ti 5 O 12 Anode For Lithium Batteries. Electrochim Acta. 2007; 53(1):79–82.
139. Park SH, Xia Y. Macroporous Membranes With Highly Ordered And Three-Dimensionally Interconnected Spherical Pores. Adv Mater. 1998; 10(13):1045–1048.
140. Stein A, Schroden RC. Colloidal Crystal Templating Of Three-Dimensionally Ordered Macroporous Solids: Materials For Photonics And Beyond. Curr Opin Solid State Mater Sci. 2001; 5(6):553–564.
141. Rong J-H, Ji L-J, Yang Z-Z. Some Key Ordered Macroporous Composites. Chin J Polym Sci. 2013; 31(9):1204–1217.
142. Li H, Zhang L, Dai H, He H. Facile Synthesis And Unique Physicochemical Properties Of Three-Dimensionally Ordered Macro-porous Magnesium Oxide, Gamma-Alumina, And Ceria–Zirconia Solid Solutions With Crystalline Mesoporous Walls. Inorg Chem. 2009; 48(10):4421–4434. [PubMed: 19348445]
143. Kierys A, Dziadosz M, Goworek J. Polymer/Silica Composite Of Core–Shell Type By Polymer Swelling In TEOS. J Colloid Interface Sci. 2010; 349(1):361–365. [PubMed: 20573353]
144. Costa E, De-Carvalho J, Casimiro T, Da Silva CL, Cidade MT, Aguiar-Ricardo A. Tailoring Thermoresponsive Microbeads In Supercritical Carbon Dioxide For Biomedical Applications. J Supercrit Fluids. 2011; 56(3):292–298.
145. Temtem M, Barroso T, Casimiro T, Mano JF, Aguiar-Ricardo A. Dual Stimuli Responsive Poly (N-Isopropylacrylamide) Coated Chitosan Scaffolds For Controlled Release Prepared From A Non Residue Technology. J Supercrit Fluids. 2012; 66:398–404.
146. Yu N, Li G, Gao Y, Jiang H, Tao Q. Thermo-Sensitive Complex Micelles From Sodium Alginate-Graft-Poly (N-Isopropyla-crylamide) For Drug Release. Int J Biol Macromol. 2016; 86:296. [PubMed: 26806647]
147. Gaucher G, Dufresne M-H, Sant VP, Kang N, Maysinger D, Leroux J-C. Block Copolymer Micelles: Preparation, Characterization And Application In Drug Delivery. J Controlled Release. 2005; 109(1–3):169–188.
148. Strandman S, Zhu X. Thermo-Responsive Block Copolymers With Multiple Phase Transition Temperatures In Aqueous Solutions. Prog Polym Sci. 2015; 42:154–176.
149. Croissant JG, Mongin O, Hugues V, Blanchard-Desce M, Cattoen X, Wong Chi Man M, Stojanovic V, Charnay C, Maynadier M, Gary-Bobo M, Garcia M, Raehm L, Durand J-O. Influence Of The Synthetic Method On The Properties Of Two-Photon-Sensitive Mesoporous Silica Nanoparticles. J Mater Chem B. 2015; 3(26):5182–5188.
150. Qi M, Li G, Yu N, Meng Y, Liu X. Synthesis Of Thermo-Sensitive Polyelectrolyte Complex Nanoparticles From CS-G-PNIPAM And SA-G-PNIPAM For Controlled Drug Release. Macromol Res. 2014; 22(9):1004–1011.
151. Luo Y-L, Wang Y, Wang X, Xu F, Chen Y-S. Thermosensitive Tribrachia Star-Shaped Sp (NIPAM-Co-DMAM) Random Copolymer Micelle Aggregates: Preparation, Characterization, And Drug Release Applications. J Biomater Appl. 2016; 30(6):662–676. [PubMed: 25926671]
152. Wu Y, Yang C, Lai Q, Zhang Q, Wang W, Yuan Z. Fabrication Of Thermo-Sensitive Complex Micelles For Reversible Cell Targeting. J Mater Sci: Mater Med. 2015; 26(11):1–13.
153. Daniel-Da-Silva AL, Ferreira L, Gil AM, Trindade T. Synthesis And Swelling Behavior Of Temperature Responsive K-Carrageenan Nanogels. J Colloid Interface Sci. 2011; 355(2):512–517. [PubMed: 21251667]
154. Amamoto Y, Otsuka H, Takahara A. Synthesis And Characterization Of Polymeric Nanogels. Nanotechnologies For The Life Sciences. 2012; doi: 10.1002/9783527610419.ntls0246
155. Pelton R, Chibante P. Preparation Of Aqueous Latices With N-Isopropylacrylamide. Colloids Surf. 1986; 20(3):247–256.
156. Maya S, Sarmento B, Nair A, Rejinold NS, Nair SV, Jayakumar R. Smart Stimuli Sensitive Nanogels In Cancer Drug Delivery And Imaging: A Review. Curr Pharm Des. 2013; 19(41): 7203–7218. [PubMed: 23489200]

157. Wyss A, Boucher J, Montero A, Marison I. Micro-Encapsulated Organic Phase For Enhanced Bioremediation Of Hydrophobic Organic Pollutants. Enzyme Microb Technol. 2006; 40(1):25–31.

158. Jain RA. The Manufacturing Techniques Of Various Drug Loaded Biodegradable Poly (Lactide-Co-Glycolide) (PLGA) Devices. Biomaterials. 2000; 21(23):2475–2490. [PubMed: 11055295]

159. Bilalis P, Efthimiadou EK, Chatzipavlidis A, Boukos N, Kordas GC. Multi-Responsive Polymeric Microcontainers For Potential Biomedical Applications: Synthesis And Functionality Evaluation. Polym Int. 2012; 61(6):888–894.

160. Efthimiadou E, Tziveleka L-A, Bilalis P, Kordas G. Novel PLA Modification Of Organic Microcontainers Based On Ring Opening Polymerization: Synthesis, Characterization, Biocompatibility And Drug Loading/Release Properties. Int J Pharm. 2012; 428(1):134–142. [PubMed: 22402473]

161. Pardeshi S, Singh SK. Precipitation Polymerization: A Versatile Tool For Preparing Molecularly Imprinted Polymer Beads For Chromatography Applications. RSC Adv. 2016; 6(28):23525–23536.

162. Tapeinos C, Efthimiadou E, Boukos N, Charitidis C, Koklioti M, Kordas G. Microspheres As Therapeutic Delivery Agents: Synthesis And Biological Evaluation Of pH Responsiveness. J Mater Chem B. 2013; 1(2):194–203.

163. Wadajkar AS, Menon JU, Tsai YS, Gore C, Dobin T, Gandee L, Kangasniemi K, Takahashi M, Manandhar B, Ahn JM, Hsieh JT, Nguyen KT. Prostate Cancer-Specific Thermo-Responsive Polymer-Coated Iron Oxide Nanoparticles. Biomaterials. 2013; 34(14):3618–3625. [PubMed: 23419645]

164. Zhu H, Tao J, Wang W, Zhou Y, Li P, Li Z, Yan K, Wu S, Yeung KW, Xu Z, Xu H, Chu PK. Magnetic, Fluorescent, And Thermo-Responsive Fe 3 O 4/Rare Earth Incorporated Poly (St-NIPAM) Core–Shell Colloidal Nanoparticles In Multimodal Optical/Magnetic Resonance Imaging Probes. Biomaterials. 2013; 34(9):2296–2306. [PubMed: 23274069]

165. Sundaresan V, Menon JU, Rahimi M, Nguyen KT, Wadajkar AS. Dual-Responsive Polymer-Coated Iron Oxide Nanoparticles For Drug Delivery And Imaging Applications. Int J Pharm. 2014; 466(1):1–7. [PubMed: 24607216]

166. Zhang J, Liu J, Zuo Y, Wang R, Xiong Y. Preparation Of Thermo-Responsive Poly (Ionic Liquid) S-Based Nanogels Via OneStep Cross-Linking Copolymerization. Molecules. 2015; 20(9): 17378–17392. [PubMed: 26393567]

167. Jaber J, Mohsen E. Synthesis Of Fe 3 O 4@ Silica/Poly (N-Isopropylacrylamide) As A Novel Thermo-Responsive System For Controlled Release Of H 3 Pmo 12 O 40 Nano Drug In AC Magnetic Field. Colloids Surf, B. 2013; 102:265–272.

168. Liu G, Zhu C, Xu J, Xin Y, Yang T, Li J, Shi L, Guo Z, Liu W. Thermo-Responsive Hollow Silica Microgels With Controlled Drug Release Properties. Colloids Surf, B. 2013; 111:7–14.

169. Yuan Z, Wang Y, Chen D. Preparation And Characterization Of Thermo-, pH-, And Magnetic-Field-Responsive Organic/Inorganic Hybrid Microgels Based On Poly (Ethylene Glycol). J Mater Sci. 2014; 49(8):3287–3296.

170. Ding J, Zhao L, Li D, Xiao C, Zhuang X, Chen X. Thermo-Responsive “Hairy-Rod” Polypeptides For Smart Antitumor Drug Delivery. Polym Chem. 2013; 4(11):3345–3356.

171. Kuntsche JHJ, Bunjes H. Cryogenic Transmission Electron Microscopy (Cryo-TEM) For Studying The Morphology Of Colloidal Drug Delivery Systems. Int J Pharm. 2011; 417(1):120–137. [PubMed: 21310225]

172. Lu Y, Ballauff M. Thermosensitive Core–Shell Microgels: From Colloidal Model Systems To Nanoreactors. Prog Polym Sci. 2011; 36(6):767–792.

173. Bielska D, Karewicz A, Kamiński K, Kiełkowicz I, Lachowicz T, Szczubiałka K, Nowakowska M. Self-Organized Thermo-Responsive Hydroxypropyl Cellulose Nanoparticles For Curcumin Delivery. Eur Polym J. 2013; 49(9):2485–2494.

174. Salehi R, Rasouli S, Hamishehkar H. Smart Thermo/pH Responsive Magnetic Nanogels For The Simultaneous Delivery Of Doxorubicin And Methotrexate. Int J Pharm. 2015; 487(1):274–284. [PubMed: 25895723]

175. Borlido L, Moura L, Azevedo AM, Roque AC, Aires-Barros MR, Farinha JP. Stimuli-Responsive Magnetic Nano-particles For Monoclonal Antibody Purification. Biotechnol J. 2013; 8(6):709–717. [PubMed: 23420794]

176. McInnes SJ, Szili E, Al-Bataineh SA, Vasani RB, Xu J, Alf ME, Gleason KK, Short RD, Voelcker NH. Fabrication And Characterization Of A Porous Silicon Drug Delivery System With An Icvd Temperature-Responsive Coating. Langmuir. 2016; 32(1):301–308. [PubMed: 26654169]

177. Li J, Wu W, Han C, Zhang S, Zhou H, Guo J. Aggregation Behavior Of pH-And Thermo-Responsive Block Copolymer Protected Gold Nanoparticles. Colloid Polym Sci. 2014; 292(7): 1657–1664.

178. Wang Y, Wu G, Li X, Wang Y, Gao H, Ma J. Synthesis, Characterization And Controlled Drug Release From Temperature-Responsive Poly (Ether-Urethane) Particles Based On PEG-Diisocyanates And Aliphatic Diols. J Biomater Sci, Polym Ed. 2013; 24(14):1676–1691. [PubMed: 23627737]

179. Huang Y, Luo W, Ye G. Synthesis, Characterization, Conformation And Self-Assembly Behavior Of Polypeptide-Based Brush With Oligo (Ethylene Glycol) Side Chains. J Mol Struct. 2015; 1081:274–280.

180. Pan G, Guo Q, Cao C, Yang H, Li B. Thermo-Responsive Molecularly Imprinted Nanogels For Specific Recognition And Controlled Release Of Proteins. Soft Matter. 2013; 9(14):3840–3850.

181. Lee RS, Lin CH, Aljuffali IA, Hu KY, Fang JY. Passive Targeting Of Thermosensitive Diblock Copolymer Micelles To The Lungs: Synthesis And Characterization Of Poly (N-Isopropylacrylamide)-Block-Poly (E-Caprolactone). J Nanobiotechnol. 2015; 13(1):42.

182. Uto K, Ebara M, Aoyagi T. Temperature-Responsive Poly (E-Caprolactone) Cell Culture Platform With Dynamically Tunable Nano-Roughness And Elasticity For Control Of Myoblast Morphology. Int J Mol Sci. 2014; 15(1):1511–1524. [PubMed: 24451135]

183. Wang A, Gao H, Sun Y, Sun YL, Yang YW, Wu G, Wang Y, Fan Y, Ma J. Temperature-And pH-Responsive Nano-particles Of Biocompatible Polyurethanes For Doxorubicin Delivery. Int J Pharm. 2013; 441(1):30–39. [PubMed: 23262421]

184. Chen CY, Kim TH, Wu WC, Huang CM, Wei H, Mount CW, Tian Y, Jang SH, Pun SH, Jen AK. pH-Dependent, Thermosensitive Polymeric Nanocarriers For Drug Delivery To Solid Tumors. Biomaterials. 2013; 34(18):4501–4509. [PubMed: 23498892]

185. Lee RS, Wang SW, Li YC, Fang JY. Synthesis And Characterization Of Thermo-Responsive And Photo-Cleavable Block Copolymers As Nanocarriers. RSC Adv. 2015; 5(1):497–512.

186. Hu X, Hao X, Wu Y, Zhang J, Zhang X, Wang PC, Zou G, Liang XJ. Multifunctional Hybrid Silica Nanoparticles For Controlled Doxorubicin Loading And Release With Thermal And pH Dual Response. J Mater Chem B. 2013; 1(8):1109–1118.

187. Shi S, Wang Q, Wang T, Ren S, Gao Y, Wang N. Thermo-, pH-, And Light-Responsive Poly (N-Isopropylacrylamide-Co-Methacrylic Acid)–Au Hybrid Microgels Prepared By The In Situ Reduction Method Based On Au-Thiol Chemistry. J Phys Chem B. 2014; 118(25):7177–7186. [PubMed: 24897339]

188. Li Q, He X, Cui Y, Shi P, Li S, Zhang W. Doubly Thermo-Responsive Nanoparticles Constructed With Two Diblock Copolymers Prepared Through The Two Macro-RAFT Agents CoMediated Dispersion RAFT Polymerization. Polym Chem. 2015; 6(1):70–78.

189. Sedlák M. A Novel Approach To Controlled Self-Assembly Of pH-Responsive Thermosensitive Homopolymer Polyelectrolytes Into Stable Nanoparticles. Adv Colloid Interface Sci. 2016; 232:57. [PubMed: 26792020]

190. Ueno K, Fukai T, Watanabe M. Thermosensitive Soft Glassy Colloidal Arrays Of Block-Copolymer-Grafted Silica Nano-particles In An Ionic Liquid. Polym J. 2016; 48:289.

191. Begum R, Naseem K, Farooqi ZH. A Review Of Responsive Hybrid Microgels Fabricated With Silver Nanoparticles: Synthesis, Classification, Characterization and Applications. J Sol-Gel Sci Technol. 2016; 77:497.

192. Liu Y, Cui Y, Wu G, Liao M. Preparation And Properties Of Fast Temperature-Responsive Soy Protein/Pnipaam IPN Hydrogels. J Serb Chem Soc. 2014; 79(2):211–224.

193. Mohd Amin MCI, Ahmad N, Halib N, Ahmad I. Synthesis And Characterization Of Thermo-And pH-Responsive Bacterial Cellulose/Acrylic Acid Hydrogels For Drug Delivery. Carbohydr Polym. 2012; 88(2):465–473.

194. Kang X, Cheng Z, Yang D, Ma PA, Shang M, Peng C, Dai Y, Lin J. Design And Synthesis Of Multifunctional Drug Carriers Based On Luminescent Rattle-Type Mesoporous Silica Microspheres With A Thermosensitive Hydrogel As A Controlled Switch. Adv Funct Mater. 2012; 22(7):1470–1481.

195. Sanoj Rejinold N, Sreerekha P, Chennazhi K, Nair S, Jayakumar R. Biocompatible, Biodegradable And Thermo-Sensitive Chitosan-G-Poly (N-Isopropylacrylamide) Nanocarrier For Curcumin Drug Delivery. Int J Biol Macromol. 2011; 49(2):161–172. [PubMed: 21536066]

196. Koppolu B, Bhavsar Z, Wadajkar AS, Nattama S, Rahimi M, Nwariaku F, Nguyen KT. Temperature-Sensitive Polymer-Coated Magnetic Nanoparticles As A Potential Drug Delivery System For Targeted Therapy Of Thyroid Cancer. J Biomed Nanotechnol. 2012; 8(6):983–990. [PubMed: 23030006]

197. Lima AC, Song W, Blanco-Fernandez B, Alvarez-Lorenzo C, Mano JF. Synthesis Of Temperature-Responsive Dextran-MA/Pnipaam Particles For Controlled Drug Delivery Using Super-hydrophobic Surfaces. Pharm Res. 2011; 28(6):1294–1305. [PubMed: 21298327]

198. Ranjan A, Jacobs GC, Woods DL, Negussie AH, Partanen A, Yarmolenko PS, Gacchina CE, Sharma KV, Frenkel V, Wood BJ, Dreher MR. Image-Guided Drug Delivery With Magnetic Resonance Guided High Intensity Focused Ultrasound And Temperature Sensitive Liposomes In A Rabbit Vx2 Tumor Model. J Controlled Release. 2012; 158(3):487–494.

199. Akbarzadeh A, Zarghami N, Mikaeili H, Asgari D, Goganian AM, Khiabani HK, Samiei M, Davaran S. Synthesis, Characterization, And In Vitro Evaluation Of Novel Polymer-Coated Magnetic Nanoparticles For Controlled Delivery Of Doxorubicin. Nanotechnol, Sci Appl. 2012; 5:13. [PubMed: 24198493]

200. Nazar H, Fatouros D, Van Der Merwe S, Bouropoulos N, Avgouropoulos G, Tsibouklis J, Roldo M. Thermosensitive Hydrogels For Nasal Drug Delivery: The Formulation And Characterisation Of Systems Based On N-Trimethyl Chitosan Chloride. Eur J Pharm Biopharm. 2011; 77(2):225–232. [PubMed: 21130876]

201. Wang H, Sun D, Zhao N, Yang X, Shi Y, Li J, Su Z, Wei G. Thermo-Sensitive Graphene Oxide–Polymer Nanoparticle Hybrids: Synthesis, Characterization, Biocompatibility And Drug Delivery. J Mater Chem B. 2014; 2(10):1362–1370.

202. Li G, Meng Y, Guo L, Zhang T, Liu J. Formation Of Thermo-Sensitive Polyelectrolyte Complex Micelles From Two Biocompatible Graft Copolymers For Drug Delivery. J Biomed Mater Res, Part A. 2014; 102(7):2163–2172.

203. Lin Z, Gao W, Hu H, Ma K, He B, Dai W, Wang X, Wang J, Zhang X, Zhang Q. Novel Thermo-Sensitive Hydrogel System With Paclitaxel Nanocrystals: High Drug-Loading, Sustained Drug Release And Extended Local Retention Guaranteeing Better Efficacy And Lower Toxicity. J Controlled Release. 2014; 174:161–170.

204. Ninomiya K, Yamashita T, Kawabata S, Shimizu N. Targeted And Ultrasound-Triggered Drug Delivery Using Liposomes Co-Modified With Cancer Cell-Targeting Aptamers And A Thermosensitive Polymer. Ultrason Sonochem. 2014; 21(4):1482–1488. [PubMed: 24418100]

205. Pradhan L, Srivastava R, Bahadur D. pH-And Thermo-sensitive Thin Lipid Layer Coated Mesoporous Magnetic Nano-assemblies As A Dual Drug Delivery System Towards Thermochemotherapy Of Cancer. Acta Biomater. 2014; 10(7):2976–2987. [PubMed: 24747086]

206. Karavelidis V, Bikiaris D, Avgoustakis K. New Thermosensitive Nanoparticles Prepared By Biocompatible Pegylated Aliphatic Polyester Block Copolymers For Local Cancer Treatment. J Pharm Pharmacol. 2015; 67(2):215–230. [PubMed: 25616209]

207. Cirillo G, Spataro T, Curcio M, Spizzirri UG, Nicoletta FP, Picci N, Iemma F. Tunable Thermo-Responsive Hydrogels: Synthesis, Structural Analysis And Drug Release Studies. Mater Sci Eng, C. 2015; 48:499–510.

208. Liu M, Su H, Tan T. Synthesis And Properties Of Thermo-And pH-Sensitive Poly (N-Isopropylacrylamide)/Polyaspartic Acid IPN Hydrogels. Carbohydr Polym. 2012; 87(4):2425–2431.

209. Zhao J, Zhao X, Guo B, Ma PX. Multifunctional Interpenetrating Polymer Network Hydrogels Based On Methacrylated Alginate For The Delivery Of Small Molecule Drugs And Sustained Release Of Protein. Biomacromolecules. 2014; 15(9):3246–3252. [PubMed: 25102223]

210. Rahman MM, Elaissari A. A Versatile Method For The Preparation Of Rigid Submicron Hollow Capsules Containing A Temperature Responsive Shell. J Mater Chem. 2012; 22(3):1173–1179.

211. Li G, Qi M, Yu N, Liu X. Hybrid Vesicles Co-Assembled From Anionic Graft Copolymer And Metal Ions For Controlled Drug Release. Chem Eng J. 2015; 262:710–715.

212. Guo X, Li D, Yang G, Shi C, Tang Z, Wang J, Zhou S. Thermo-Triggered Drug Release From Actively Targeting Polymer Micelles. ACS Appl Mater Interfaces. 2014; 6(11):8549–8559. [PubMed: 24804870]

213. Foldvari M. Nanopharmaceutics Innovations In Gene Therapy: Moving Towards Nonviral And Non-Invasive Delivery Methods. Nanomedicine & Biotherapeutic Discovery. 2014; 4(2):129.

214. Shan Y, Luo T, Peng C, Sheng R, Cao A, Cao X, Shen M, Guo R, Tomás H, Shi X. Gene Delivery Using Dendrimer-Entrapped Gold Nanoparticles As Nonviral Vectors. Biomaterials. 2012; 33:3025–3035. [PubMed: 22248990]

215. Zhou Y, Tang Z, Shi C, Shi S, Qian Z, Zhou S. Polyethylenimine Functionalized Magnetic Nanoparticles As A Potential Non-Viral Vector For Gene Delivery. J Mater Sci: Mater Med. 2012; 23:2697–2708. [PubMed: 22826003]

216. Calejo MT, Cardoso AMS, Kjøniksen A, Zhu K, Morais CM, Sande SA, Cardoso AL, Pedroso De Lima MC, Jurado A, Nyström B. Temperature-Responsive Cationic Block Copolymers As Nanocarriers For Gene Delivery. Int J Pharm. 2013; 448:105–114. [PubMed: 23524085]

217. Guo X, Huang L. Recent Advances In Nonviral Vectors For Gene Delivery. Acc Chem Res. 2012; 45(7):971–979. [PubMed: 21870813]

218. Méndezardoy A, Urbiola K, Aranda C, Ortiz-Mellet C, Garcíafernández JM, Tros de Ilarduya C. Polycationic Amphiphilic Cyclodextrin-Based Nanoparticles For Therapeutic Gene Delivery. Nanomedicine. 2011; 6(10):1697–1707. [PubMed: 22122582]

219. Curtin CM, Cunniffe GM, Lyons FG, Bessho K, Dickson GR, Duffy GP, O'Brien FJ. Innovative Collagen Nano-Hydroxyapatite Scaffolds Offer A Highly Efficient Non-Viral Gene Delivery Platform For Stem Cell-Mediated Bone Formation. Adv Mater. 2012; 24:749–754. [PubMed: 22213347]

220. Dean DA, Gasiorowski JZ. Nonviral Gene Delivery. Cold Spring Harb Protoc. 2011; 2011:top101. [PubMed: 21363958]

221. Rehman ZU, Sjollema KA, Kuipers J, Hoekstra D, Zuhorn IS. Nonviral Gene Delivery Vectors Use Syndecan-Dependent Transport Mechanisms In Filopodia To Reach The Cell Surface. ACS Nano. 2012; 6(8):7521–7532. [PubMed: 22857607]

222. Karimi M, Avci P, Ahi M, Gazori T, Hamblin MR, Naderi-Manesh H. Evaluation Of Chitosan-Tripolyphosphate Nanoparticles As A P-Shrna Delivery Vector: Formulation, Optimization And Cellular Uptake Study. Journal Of Nanopharmaceutics And Drug Delivery. 2013; 1(3):266–278. [PubMed: 26989641]

223. Shim MS, Kwon YJ. Stimuli-Responsive Polymers And Nanomaterials For Gene Delivery And Imaging Applications. Adv Drug Delivery Rev. 2012; 64(11):1046–1059.

224. Chen THH, Bae Y, Furgeson DY, Kwon GS. Biodegradable Hybrid Recombinant Block Copolymers For Non-Viral Gene Transfection. Int J Pharm. 2012; 427:105–112. [PubMed: 21983093]

225. Lee HN, Rosen BM, Fenyvesi G, Sunkara HB. UCST And LCST Phase Behavior Of Poly(Trimethylene Ether) Glycol In Water. J Polym Sci, Part A: Polym Chem. 2012; 50:4311.

226. Shepherd J, Sarker P, Swindells K, Douglas I, Macneil S, Swanson L, Rimmer S. Binding Bacteria To Highly Branched Poly(N-Isopropyl Acrylamide) Modified With Vancomycin Induces The Coil-To-Globule Transition. J Am Chem Soc. 2010; 132:1736–1737. [PubMed: 20099898]

227. Zhang R, Wang Y, Du FS, Wang YL, Tan YX, Ji SP, Li ZC. Thermoresponsive Gene Carriers Based On Poly-ethylenimine-Graft-Poly[Oligo(Ethyleneglycol) Methacrylate]. Macromol Biosci. 2011; 11:1393–1406. [PubMed: 21830296]

228. Tran NTD, Truong NP, Gu W, Jia Z, Cooper MA, Monteiro MJ. Timed-Release Polymer Nanoparticles. Biomacromolecules. 2013; 14:495–502. [PubMed: 23298322]

229. Soliman M, Allen S, Davies MC, Alexander C. Responsive Polyelectrolyte Complexes For Triggered Release Of Nucleic Acid Therapeutics. Chem Commun. 2010; 46:5421–5433.

230. Akimoto J, Nakayama M, Sakai K, Okano T. Thermally Controlled Intracellular Uptake System Of Polymeric Micelles Possessing Poly(N-Isopropylacrylamide)-Based Outer Coronas. Mol Pharmaceutics. 2010; 7(4):926–935.

231. Petrov PD, Ivanova NI, Apostolova MD, Tsvetanov CB. Biodegradable Polymer Network Encapsulated Polyplex For DNA Delivery. RSC Adv. 2013; 3:3508–3511.

232. Liu W, Yang J, Zhang P, Tang L, Sun P, Sun P, Zuo A, Liang D. Temperature-Tuned DNA Condensation And Gene Transfection By PEI-G-(PMEO2MA-B-PHEMA) Copolymer-Based Nonviral Vectors. Biomaterials. 2010; 31:144–155. [PubMed: 19783298]

233. Iwai R, Kusakabe S, Nemoto Y, Nakayama Y. Deposition Gene Transfection Using Bioconjugates Of DNA And Thermoresponsive Cationic Homopolymer. Bioconjugate Chem. 2012; 23:751–757.

234. Qin SH, Geng Y, Discher DE, Yang S. Temperature-Controlled Assembly And Release From Polymer Vesicles Of Poly(Ethylene Oxide)-Block-Poly(N-Isopropylacrylamide). Adv Mater. 2006; 18:2905–2909.

235. Ivanova ED, Ivanova NI, Apostolova MD, Turmanova SC, Dimitrov IV. Polymer Gene Delivery Vectors Encapsulated In Thermally Sensitive Bioreducible Shell. Bioorg Med Chem Lett. 2013; 23(14):4080–4084. [PubMed: 23768908]

236. Shen Z, Shi B, Zhang H, Bi J, Dai S. Exploring Low-Positively Charged Thermosensitive Copolymers As Gene Delivery Vectors. Soft Matter. 2012; 8:1385–1394.

237. Seo HI, Cho AN, Jang J, Kim DW, Cho SW, Chung BG. Thermo-Responsive Polymeric Nanoparticles For Enhancing Neuronal Differentiation Of Human Induced Pluripotent Stem Cells. Nanomedicine. 2015; 11:1861–1869. [PubMed: 26093056]

238. Shen J, Chen C, Fu W, Shi L, Li Z. Conformation-Specific Self-Assembly Of Thermo-Responsive Poly (Ethylene Glycol)-B-Polypeptide Diblock Copolymer. Langmuir. 2013; 29(21):6271–6278. [PubMed: 23634643]

239. Feng G, Chen H, Li J, Huang Q, Gupte MJ, Liu H, Song Y, Ge Z. Gene Therapy For Nucleus Pulposus Regeneration By Heme Oxygenase-1 Plasmid DNA Carried By Mixed Polyplex Micelles With Thermo-Responsive Heterogeneous Coronas. Biomaterials. 2015; 52:1–13. [PubMed: 25818409]

240. Li J, Chen Q, Zha Z, Li H, Toh K, Dirisala A, Matsumoto Y, Osada K, Kataoka K, Ge Z. Ternary Polyplex Micelles With PEG Shells And Intermediate Barrier To Complexed DNA Cores For Efficient Systemic Gene Delivery. J Controlled Release. 2015; 209:77–87.

241. Gaspar VM, Gonçalves C, De Melo-Diogo D, Costa EC, Queiroz JA, Pichon C, Sousa F, Correia IJ. Poly (2-Ethyl-2-Oxazoline)–PLA-G–PEI Amphiphilic Triblock Micelles For Co-Delivery Of Minicircle DNA And Chemotherapeutics. J Controlled Release. 2014; 189:90–104.

242. Agarwal S, Zhang Y, Maji S, Greiner A. PDMAEMA Based Gene Delivery Materials. Mater Today. 2012; 15(9):388.

243. Ward MA, Georgiou TK. Thermoresponsive Polymers For Biomedical Applications. Polymers. 2011; 3:1215–1242.

244. Zhi D, Zhang S, Wang B, Zhao Y, Yang B, Yu S. Transfection Efficiency Of Cationic Lipids With Different Hydrophobic Domains In Gene Delivery. Bioconjugate Chem. 2010; 21:563–577.

245. Freitag R, Synatschke CV, Schallon A, Jé rôme V, Müller AHE. Influence Of Polymer Architecture And Molecular Weight Of Poly(2-(Dimethylamino)Ethyl Methacrylate) Polycations On Trans-fection Efficiency And Cell Viability In Gene Delivery. Biomacromolecules. 2011; 12:4247–4255. [PubMed: 22007721]

246. Schwanha Usser B, Busse D, Li Na, Dittmar G, Schuchhardt J, Wolf J, Chen W, Selbach M. Global Quantification Of Mammalian Gene Expression Control. Nature. 2011; 473:337–342. [PubMed: 21593866]

247. Gibney ER, Nolan C. Epigenetics And Gene Expression. Heredity. 2010; 105:4–13. [PubMed: 20461105]

248. Huynh CT, Nguyen QV, Kang SW, Lee DS. Synthesis And Characterization Of Poly (Amino Urea Urethane)-Based Block Copolymer And Its Potential Application As Injectable pH/Temperature-Sensitive Hydrogel For Protein Carrier. Polymer. 2012; 53(19):4069–4075.

249. Hong W, Chen D, Jia L, Gu J, Hu H, Zhao X, Qiao M. Thermo- And pH-Responsive Copolymers Based On PLGA-PEG-PLGA And Poly(L-Histidine): Synthesis And In Vitro Characterization Of Copolymer Micelles. Acta Biomater. 2014; 10(3):1259–1271. [PubMed: 24365708]

250. Pérez E, Martínez A, Teijón C, Teijón JM, Blanco MD. Bioresponsive Nanohydrogels Based On HEAA And NIPA For Poorly Soluble Drugs Delivery. Int J Pharm. 2014; 470(1):107–119. [PubMed: 24813784]

251. Kim Y, Kim D, Jang G, Kim J, Lee TS. Fluorescent, Stimuli-Responsive, Crosslinked PNIPAM-Based Microgel. Sens Actuators, B. 2015; 207:623–630.

252. Chen C-Y, Kim TH, Wu W-C, Huang C-M, Wei H, Mount CW, Tian Y, Jang S-H, Pun SH, Jen AK-Y. pH-Dependent, Thermosensitive Polymeric Nanocarriers For Drug Delivery To Solid Tumors. Biomaterials. 2013; 34(18):4501–4509. [PubMed: 23498892]

253. Wu Y, Lai Q, Lai S, Wu J, Wang W, Yuan Z. Facile Fabrication Of Core Cross-Linked Micelles By RAFT Polymerization And Enzyme-Mediated Reaction. Colloids Surf, B. 2014; 118:298–305.

254. Agut W, Brûlet A, Schatz C, Taton D, Lecommandoux S. pH And Temperature Responsive Polymeric Micelles And Polymer-somes By Self-Assembly Of Poly [2-(Dimethylamino) Ethyl Methacrylate]-B-Poly (Glutamic Acid) Double Hydrophilic Block Copolymers. Langmuir. 2010; 26(13):10546–10554. [PubMed: 20491497]

255. Hasegawa G, Kanamori K, Nakanishi K, Yamago S. Fabrication of highly crosslinked methacrylate-based polymer monoliths with well-defined macropores via living radical polymerization. Polymer. 2011; 52(21):4644–4647.

256. Sundaresan V, Menon JU, Rahimi M, Nguyen KT, Wadajkar AS. Dual-Responsive Polymer-Coated Iron Oxide Nanoparticles For Drug Delivery And Imaging Applications. Int J Pharm. 2014; 466(1):1–7. [PubMed: 24607216]

257. Pernia Leal M, Torti A, Riedinger A, La Fleur R, Petti D, Cingolani R, Bertacco R, Pellegrino T. Controlled Release Of Doxorubicin Loaded Within Magnetic Thermo-Responsive Nano-carriers Under Magnetic And Thermal Actuation In A Microfluidic Channel. ACS Nano. 2012; 6(12): 10535–10545. [PubMed: 23116285]

258. Zhu H, Tao J, Wang W, Zhou Y, Li P, Li Z, Yan K, Wu S, Yeung KW, Xu Z, et al. Magnetic, Fluorescent, And Thermo-Responsive Fe 3 O 4/Rare Earth Incorporated Poly (St-NIPAM) Core–Shell Colloidal Nanoparticles In Multimodal Optical/Magnetic Resonance Imaging Probes. Biomaterials. 2013; 34(9):2296–2306. [PubMed: 23274069]

259. Yao Y, Shen H, Zhang G, Yang J, Jin X. Synthesis Of Poly (N-Isopropylacrylamide)-Co-Poly (Phenylboronate Ester) Acrylate And Study On Their Glucose-Responsive Behavior. J Colloid Interface Sci. 2014; 431:216–222. [PubMed: 25014171]

260. Kashyap S, Singh N, Surnar B, Jayakannan M. Enzyme And Thermal Dual Responsive Amphiphilic Polymer Core-Shell Nanoparticle For Doxorubicin Delivery To Cancer Cells. Biomacromolecules. 2016; 17:384. [PubMed: 26652038]

261. Zhang BY, He WD, Li LY, Sun XL, Li WT, Zhang KR. Reducibly Degradable Hydrogels Of PNIPAM And PDMAEMA: Synthesis, Stimulus-Response And Drug Release. J Polym Sci, Part A: Polym Chem. 2010; 48(16):3604–3612.

262. Aleksanian S, Wen Y, Chan N, Oh JK. Thiol-Responsive Hydrogel Scaffolds For Rapid Change In Thermoresponsiveness. RSC Adv. 2014; 4(8):3713–3721.

263. Huo M, Yuan J, Tao L, Wei Y. Redox-Responsive Polymers For Drug Delivery: From Molecular Design To Applications. Polym Chem. 2014; 5(5):1519–1528.

264. Chen S, Jiang F, Cao Z, Wang G, Dang Z-M. Photo, pH, And Thermo Triple-Responsive Spiropyran-Based Copolymer Nano-particles For Controlled Release. Chem Commun. 2015; 51(63):12633–12636.

265. Karimi M, Ghasemi A, Zangabad PS, Rahighi R, Basri SMM, Mirshekari H, Amiri M, Pishabad ZS, Aslani A, Bozorgomid M. Smart Micro/Nanoparticles In Stimulus-Responsive Drug/Gene Delivery Systems. Chem Soc Rev. 2016; 45:1457–1501. [PubMed: 26776487]

266. Zhan Y, Gonçalves M, Yi P, Capelo D, Zhang Y, Rodrigues J, Liu C, Tomás H, Li Y, He P. Thermo/Redox/pH- Triple Sensitive Poly (N-Isopropylacrylamide-Co-Acrylic Acid) Nanogels For Anticancer Drug Delivery. J Mater Chem B. 2015; 3(20):4221–4230.

267. Zeng J, Du P, Liu L, Li J, Tian K, Jia X, Zhao X, Liu P. Superparamagnetic Reduction/pH/Temperature Multistimuli-Responsive Nanoparticles For Targeted And Controlled Antitumor Drug Delivery. Mol Pharmaceutics. 2015; 12(12):4188–4199.

268. Freire F, Ferraresi C, Jorge AOC, Hamblin MR. Photodynamic Therapy Of Oral Candida Infection In A Mouse Model. J Photochem Photobiol, B. 2016; 159:161–168. [PubMed: 27074245]

269. Wang Y, Wu X, Chen J, Amin R, Lu M, Bhayana B, Zhao J, Murray CK, Hamblin MR, Hooper DC. Antimicrobial Blue Light Inactivation Of Gram-Negative Pathogens In Biofilms: In Vitro And In Vivo Studies. J Infect Dis. 2016; 213(9):1380–1387. [PubMed: 26908743]

270. De Freitas, LF.; Hamblin, MR. Antimicrobial Photodynamic Inactivation And Antitumor Photodynamic Therapy With Fullerenes. Morgan & Claypool Publishers; San Rafael, CA: 2016. Photodynamic Therapy.

271. Broekgaarden M, Weijer R, Van Gulik TM, Hamblin MR, Heger M. Tumor Cell Survival Pathways Activated By Photodynamic Therapy: A Molecular Basis For Pharmacological Inhibition Strategies. Cancer Metastasis Rev. 2015; 34(4):643–690. [PubMed: 26516076]

272. Xuan W, Agrawal T, Huang L, Gupta GK, Hamblin MR. Low-Level Laser Therapy For Traumatic Brain Injury In Mice Increases Brain Derived Neurotrophic Factor (BDNF) And Synaptogenesis. Journal Of Biophotonics. 2015; 8(6):502–511. [PubMed: 25196192]

273. Kim J, Kim J, Jeong C, Kim WJ. Synergistic Nanomedicine By Combined Gene And Photothermal Therapy. Adv Drug Delivery Rev. 2016; 98:99.

274. Zhao W, Karp JM. Tumour Targeting Nanoantennas Heat Up. Nat Mater. 2009; 8(6):453–454. [PubMed: 19458645]

275. Levi-Polyachenko NH, Merkel EJ, Jones BT, Carroll DL, Stewart JH IV. Rapid Photothermal Intracellular Drug Delivery Using Multiwalled Carbon Nanotubes. Mol Pharmaceutics. 2009; 6(4):1092–1099.

276. Liao KH, Lin YS, Macosko CW, Haynes CL. Cytotoxicity Of Graphene Oxide And Graphene In Human Erythrocytes And Skin Fibroblasts. ACS Appl Mater Interfaces. 2011; 3(7):2607–2615. [PubMed: 21650218]

277. Yang K, Zhang S, Zhang G, Sun X, Lee S-T, Liu Z. Graphene In Mice: Ultrahigh In Vivo Tumor Uptake And Efficient Photothermal Therapy. Nano Lett. 2010; 10(9):3318–3323. [PubMed: 20684528]

278. Cheng L, Liu J, Gu X, Gong H, Shi X, Liu T, Wang C, Wang X, Liu G, Xing H, et al. Pegylated WS2 Nanosheets As A Multifunctional Theranostic Agent For In Vivo Dual-Modal CT/Photoacoustic Imaging Guided Photothermal Therapy. Adv Mater. 2014; 26(12):1886–1893. [PubMed: 24375758]

279. Hessel CM, Pattani VP, Rasch M, Panthani MG, Koo B, Tunnell JW, Korgel BA. Copper Selenide Nanocrystals For Photothermal Therapy. Nano Lett. 2011; 11(6):2560–2566. [PubMed: 21553924]

280. Li W, Zamani R, Rivera Gil P, Pelaz B, Ibáñez M, Cadavid D, Shavel A, Alvarez-Puebla RA, Parak WJ, Arbiol J, Cabot A. Cute Nanocrystals: Shape And Size Control, Plasmonic Properties, And Use As SERS Probes And Photothermal Agents. J Am Chem Soc. 2013; 135(19):7098–7101. [PubMed: 23647089]

281. Huang S, Liu J, He Q, Chen H, Cui J, Xu S, Zhao Y, Chen C, Wang L. Smart Cu1. 75S Nanocapsules With High And Stable Photothermal Efficiency For NIR Photo-Triggered Drug Release. Nano Res. 2015; 8(12):4038–4047.

282. Song X, Chen Q, Liu Z. Recent Advances In The Development Of Organic Photothermal Nano-Agents. Nano Res. 2015; 8(2):340–354.

283. Xu L, Cheng L, Wang C, Peng R, Liu Z. Conjugated Polymers For Photothermal Therapy Of Cancer. Polym Chem. 2014; 5(5):1573–1580.

284. Biju V. Chemical Modifications And Bioconjugate Reactions Of Nanomaterials For Sensing, Imaging, Drug Delivery And Therapy. Chem Soc Rev. 2014; 43(3):744–764. [PubMed: 24220322]
285. Peer D, Karp JM, Hong S, Farokhzad OC, Margalit R, Langer R. Nanocarriers As An Emerging Platform For Cancer Therapy. Nat Nanotechnol. 2007; 2(12):751–760. [PubMed: 18654426]
286. Song CW. Effect Of Local Hyperthermia On Blood Flow And Microenvironment: A Review. Cancer Res. 1984; 44(10 Supplement):4721s–4730s. [PubMed: 6467226]
287. Feng W, Zhou X, Nie W, Chen L, Qiu K, Zhang Y, He C. Au/Polypyrrole@ Fe3O4 Nanocomposites For MR/CT Dual-Modal Imaging Guided-Photothermal Therapy: An In Vitro Study. ACS Appl Mater Interfaces. 2015; 7(7):4354–4367. [PubMed: 25664659]
288. Hwang S, Nam J, Jung S, Song J, Doh H, Kim S. Gold Nanoparticle-Mediated Photothermal Therapy: Current Status And Future Perspective. Nanomedicine. 2014; 9(13):2003–2022. [PubMed: 25343350]
289. Abadeer NS, Murphy CJ. Recent Progress In Cancer Thermal Therapy Using Gold Nanoparticles. J Phys Chem C. 2016; 120(9):4691–4716.
290. Jabeen F, Najam-Ul-Haq M, Javeed R, Huck CW, Bonn GK. Au-Nanomaterials As A Superior Choice For Near-Infrared Photothermal Therapy. Molecules. 2014; 19(12):20580–20593. [PubMed: 25501919]
291. Xia K, Zhang L, Huang Y, Lu Z. Preparation Of Gold Nanorods And Their Applications In Photothermal Therapy. J Nanosci Nanotechnol. 2015; 15(1):63–73. [PubMed: 26328306]
292. Ahmad R, Fu J, He N, Li S. Advanced Gold Nanomaterials For Photothermal Therapy Of Cancer. J Nanosci Nanotechnol. 2016; 16(1):67–80. [PubMed: 27398434]
293. Qu Y, Chu BY, Peng JR, Liao JF, Qi TT, Shi K, Zhang XN, Wei YQ, Qian ZY. A Biodegradable Thermo-Responsive Hybrid Hydrogel: Therapeutic Applications In Preventing The Post-Operative Recurrence Of Breast Cancer. NPG Asia Mater. 2015; 7(8):E207.
294. Ko H, Son S, Bae S, Kim J-H, Yi G-R, Park JH. Near-Infrared Light-Triggered Thermochemotherapy Of Cancer Using A Polymer–Gold Nanorod Conjugate. Nanotechnology. 2016; 27(17):175102. [PubMed: 26987360]
295. Zan M, Li J, Huang M, Lin S, Luo D, Luo S, Ge Z. Near-Infrared Light-Triggered Drug Release Nanogels For Combined Photothermal-Chemotherapy Of Cancer. Biomater Sci. 2015; 3(7): 1147–1156. [PubMed: 26221947]
296. L'Azou B, Passagne I, Mounicou S, Tréguer-Delapierre M, Puljalté I, Szpunar J, Lobinski R, Ohayon-Courtès C. Comparative Cytotoxicity Of Cadmium Forms (Cdcl 2, Cdo, Cds Micro-And Nanoparticles) In Renal Cells. Toxicol Res. 2014; 3(1):32–41.
297. Singh NK, Nguyen QV, Kim BS, Lee DS. Nanostructure Controlled Sustained Delivery Of Human Growth Hormone Using Injectable, Biodegradable, pH/Temperature Responsive Nanobiohybrid Hydrogel. Nanoscale. 2015; 7(7):3043–3054. [PubMed: 25603888]
298. Huynh CT, Nguyen MK, Lee DS. Dually Cationic And Anionic pH/Temperature-Sensitive Injectable Hydrogels And Potential Application As A Protein Carrier. Chem Commun. 2012; 48(89):10951–10953.
299. Li Z, Guo X, Matsushita S, Guan J. Differentiation Of Cardiosphere-Derived Cells Into A Mature Cardiac Lineage Using Biodegradable Poly (N-Isopropylacrylamide) Hydrogels. Biomaterials. 2011; 32(12):3220–3232. [PubMed: 21296413]
300. Lapeyre V, Ancla C, Catargi B, Ravaine V. Glucose-Responsive Microgels With A Core–Shell Structure. J Colloid Interface Sci. 2008; 327(2):316–323. [PubMed: 18804779]
301. Dong P, Zhou Y, He W, Hua D. A Strategy For Enhanced Antibacterial Activity Against Staphylococcus Aureus By The Assembly Of Alamethicin With A Thermo-Sensitive Polymeric Carrier. Chem Commun. 2016; 52(5):896–899.
302. Oerlemans C, Bult W, Bos M, Storm G, Nijsen JFW, Hennink WE. Polymeric Micelles In Anticancer Therapy: Targeting, Imaging And Triggered Release. Pharm Res. 2010; 27(12):2569–2589. [PubMed: 20725771]

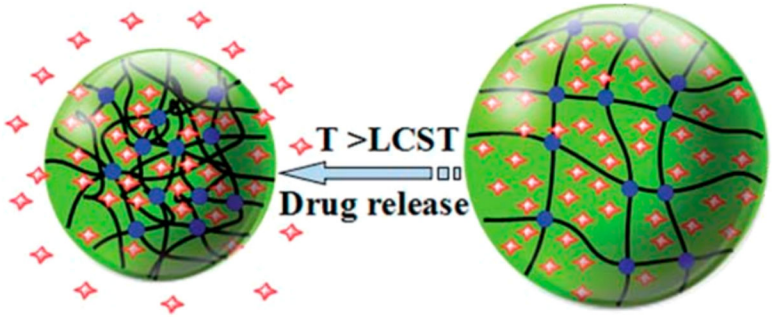


Figure 1.
Shrinkage of a loaded hydrogel and release of drug during LCST behavior. Reprinted with permission from ref 88. Copyright 2015 Royal Society of Chemistry (RSC).

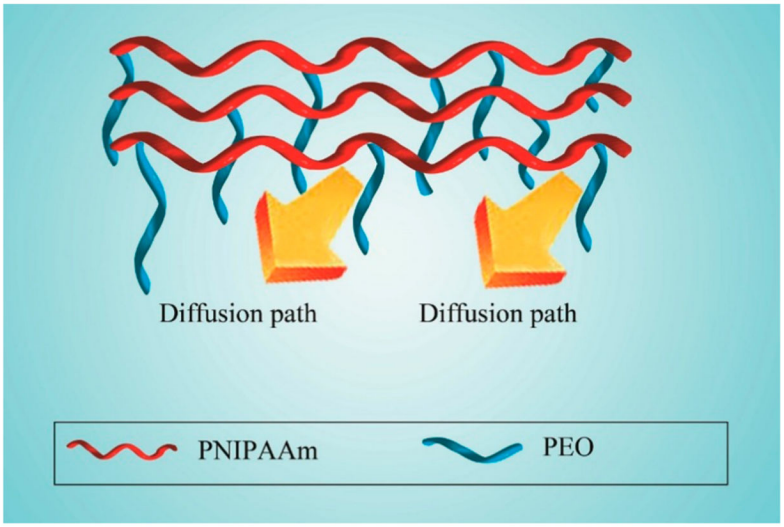


Figure 2.
Diffusion paths of water molecules in PNIPAAm-*g*-PEO.

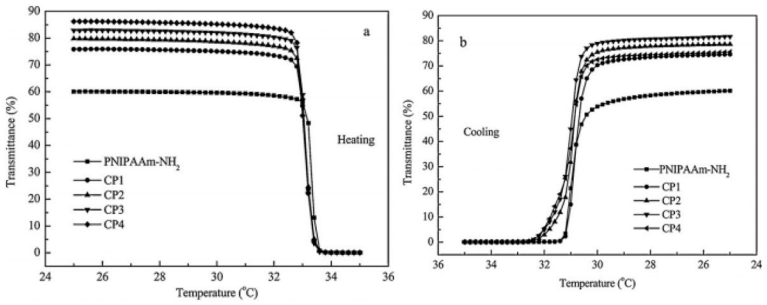


Figure 3.

Phase transition behavior of amino-terminated PNIPAAm (PNIPAAm-NH2) (2 wt %) and CMC-*g*-PNIPAAm (2 wt %) in PBS: (a) heating cycle from 25 to 35 °C and (b) cooling cycle from 35 to 25 °C. Reprinted with permission from ref 94. Copyright 2011 Elsevier.

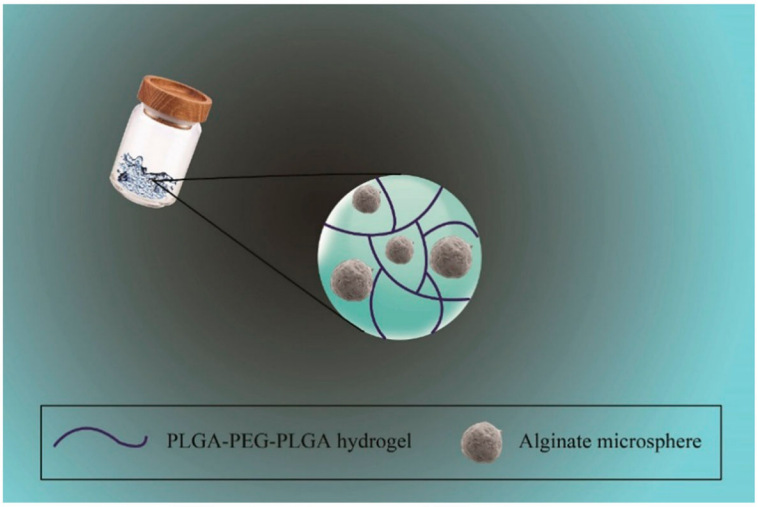


Figure 4.
Schematic representation of hydrogels loaded with alginate microspheres.

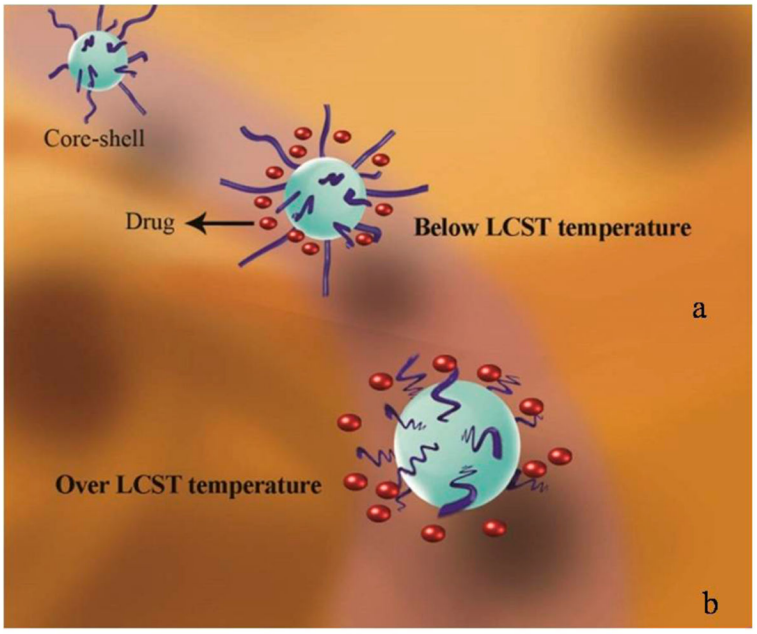


Figure 5.
Schematic of drug release from a temperature responsive core–shell nanocarrier: (a) below LCST temperature, (b) above LCST temperature.

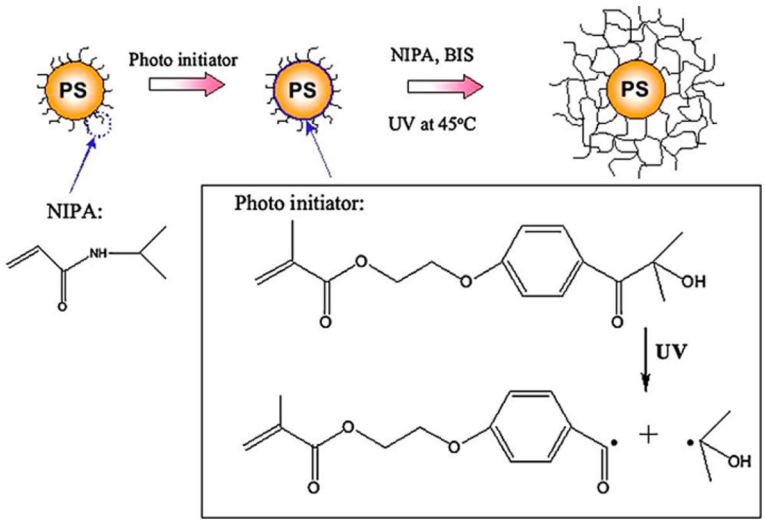


Figure 6.

Photoemulsion polymerization of PS-NIPA core–shell particles. Reprinted with permission from ref 122. Copyright 2006 John Wiley & Sons.

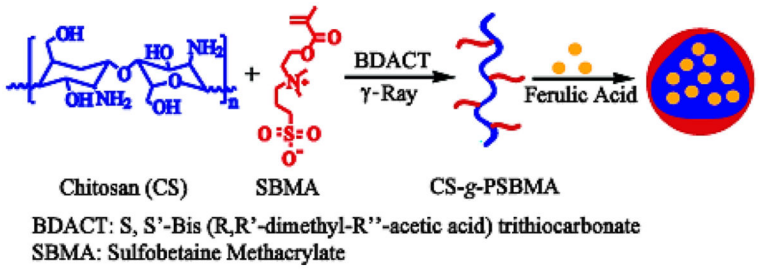


Figure 7.

Schematic of synthesis of CS-*g*-PSBMA. Reprinted with permission from ref 123. Copyright 2015 Royal Society of Chemistry (RSC).

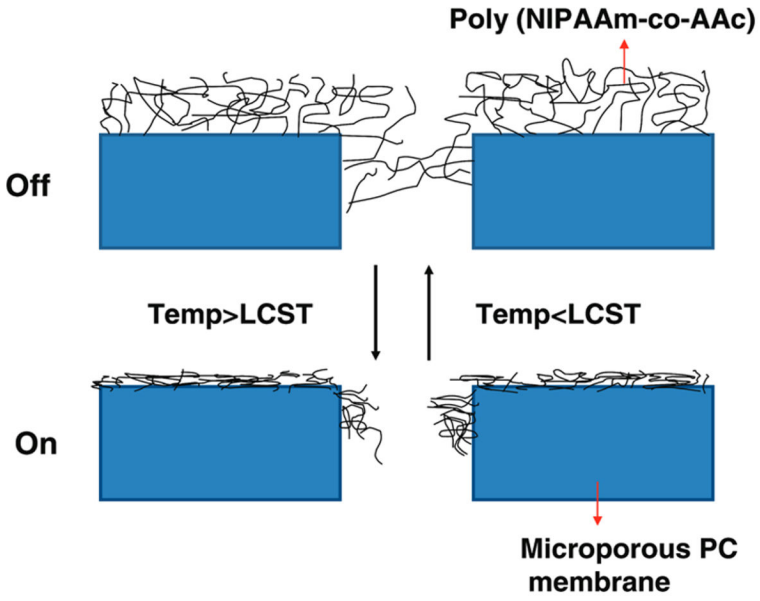


Figure 8.
On–off valve mechanism represented by PC-*g*-P(NIPAAm-*co*-AAc) grafted onto microporous composite films. Reprinted with permission from ref 136. Copyright 2012 Springer.

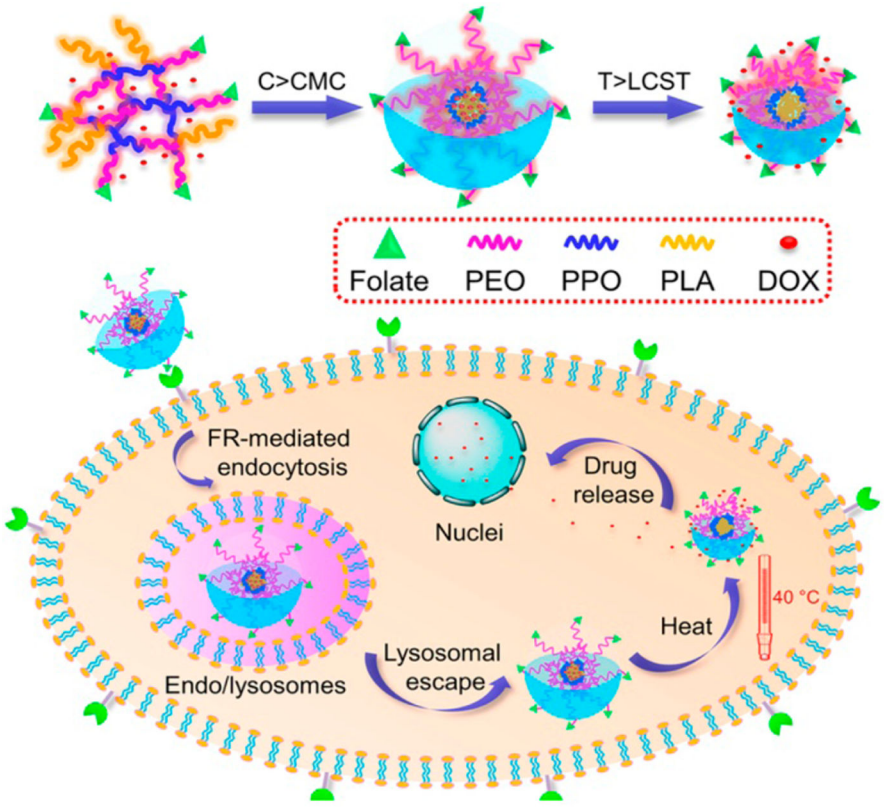


Figure 9.

Micelle formation occurs if the concentration of polymer is greater than CMC. Above the LCST, the thermosensitive block shrinks, inducing the release of incorporated agents. The nanocarrier can target tumor cells overexpressing FR, and rapidly intracellular drug release will be triggered by heating (40 °C) upon LCST on the tumor tissue. Reprinted with permission from ref 212. Copyright 2014 American Chemical Society.

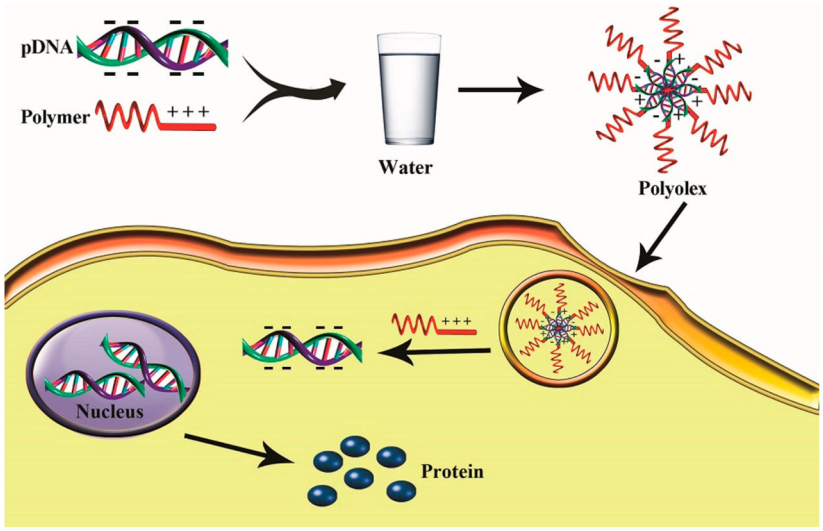


Figure 10.
Stages of gene transfection by cationic thermoresponsive polymers.

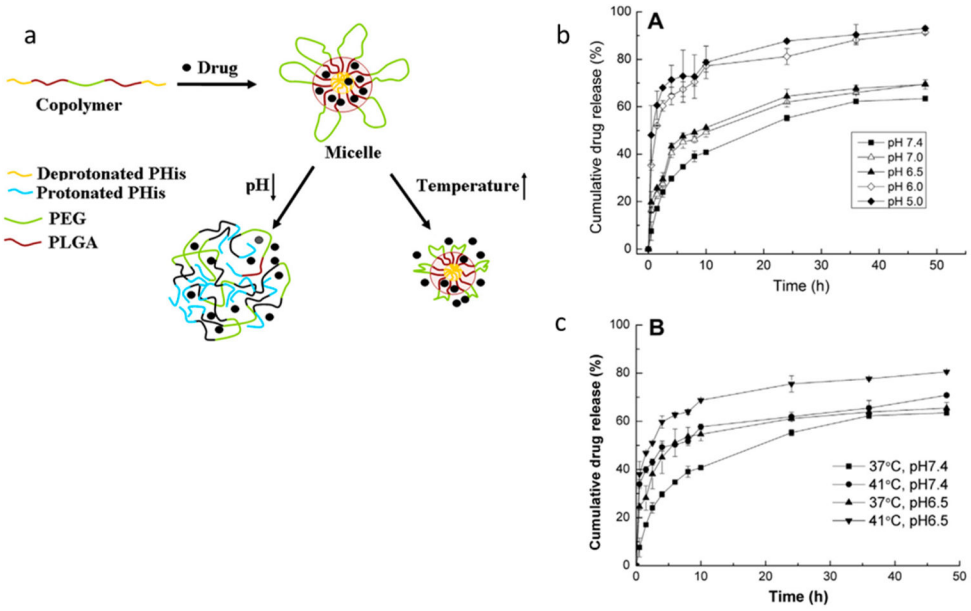


Figure 11.

(a) Schematic representing the self-assembly of PHis-PLGA-PEG-PLGA-Phis copolymer micellar nanocarrier and its dual responsiveness regarding temperature and pH stimulus, (b, c) in vitro DOX release from the nanocarrier in different pH amounts and temperatures. Reprinted with permission from ref 249. Copyright 2014 Elsevier.

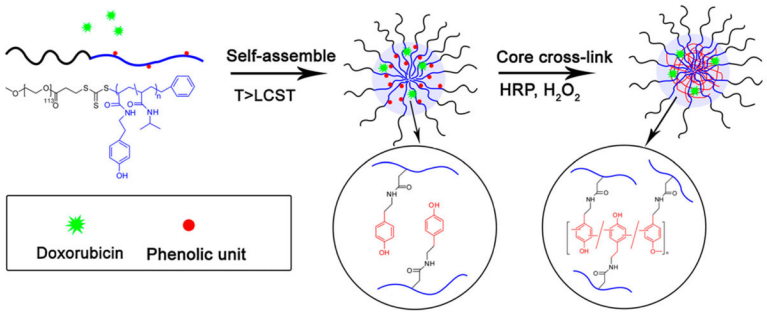


Figure 12.

In situ fabrication of core cross-linked (CCL) drug-loaded micelles. Reprinted with permission from ref 253. Copyright 2014 Elsevier.

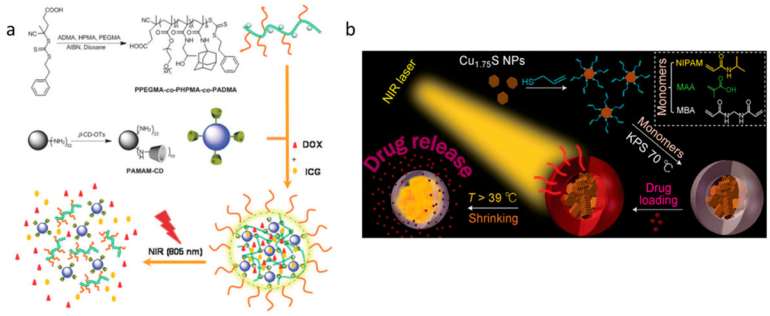


Figure 13.

Schematic of (a) NIR triggered drug release of encapsulated DOX and ICG from polymeric nanogels via photoinduced thermoresponsive relaxation of β-CD and AD-based host–guest interactions. Reprinted with permission from ref 295. Copyright 2015 Royal Society of Chemistry (RSC). (b) NIR-triggered thermoresponsive drug release from $Cu_{1.75}S$@p(NIPAM-MAA) NPs. Reprinted with permission from ref 281. Copyright 2015 Springer.

Table 1

Recent Reports on Thermoresponsive Hydrogels

functional material	synthesis method	drug loading and release characteristics	CST behavior	ref
poly(NIPAM-*co*-AM)/PEG/PTA composite hydrogels	free radical copolymerization with BIS and PTA acting as cross-linkers		UCST behavior at 59.6 °C	81
poly(polyethylene glycol (PEG) citrate-*co*-*N*-isopropylacrylamide)	polycondensation and radical polymerization,	loaded chemokine SDF-1α while retaining its bioactivity	LCST at 26 °C	83
poly(ethylene oxide)-grafted poly(*N*-isopropylacrylamide) (PNIPAAm) networks	RAFT polymerization synthesis		variation in LCST temperature	85
poly(*N*-isopropylacrylamide-*co*-2-hydroxyethyl acrylate (HEA)) hydrogels cross-linked via diselenides	free radical polymerization with diselenide bonds acting as cross-linker	sensitive to oxidizing environment	showing LCST behavior	92
glucose-based diblock copolymer	RAFT polymerization with in situ gel formation	MB showed sustained release at 37 °C up to 120 h.	LCST around 32 °C	102
PSHU-*co*-NIPAAm	sulfonate PSHU-*co*-NIPAAm	sulfonation led to sustained release of bovine serum albumin (BSA)	gel transition temperature around 32 °C	103
cyclodextrin-pseudorotaxane hydrogels	PEO-α-CD pseudorotaxane hydrogels formed by adding α-cyclodextrin to PEO solution	kinetics of loaded protein release (BSA and lysozyme), surface area of the gels were determinant factors		104
N-acryloylglycinamide	free radical polymerization		UCST-type thermo-sensitivity	105
PEG hydrogel	thiol-maleimide reaction used to synthesize PEG hydrogels	in vitro sustained release of Avastin over a period of 2 weeks		106
polyvalerolactone hydrogel	ring-opening polymerization and subsequently coupling with HDI	DEX and 5-FU. release profile showed only 14% of doxorubicin (DOX) released within 24 h	Variation of LCST temperature	107

Table 2

Examples of Temperature-Responsive Nanocarriers for Drug Delivery

stimuli-responsive nano-carrier	nanocarrier type	materials	drug	synthesis	characterization	refs
BC/AA hydrogels	thermo-and pH-responsive	BC, proportions of AA	BSA	mixing	FTIR, TGA, DTG, DSC, SEM	193
multifunctional composite microspheres	thermoresponsive	HMS, Gd_2O_3:Eu^{3+} luminescent	IMC	IWI method, photoinduced polymerization	UV irradiation, flow cytometry, CLSM, MRI	194
TRC-NPs	thermoresponsive	chitosan, NIPAAm, AIBN, EDC, NHS, TPP	curcumin	ionic cross-linking method	FTIR, flow cytometry, XRD, DLS, SEM, UV spectrophotometer	195
TPMNPs	thermoresponsive	silane-coupled iron oxide NPs	DOX	free radical polymerization of monomers on the surface	in vitro studies	196
smart hydrogel beads (Dextran-MA/ PNI-PAAm particles)	thermoresponsive	superhydrophobic surfaces, including PS, Al and Cu, photocross-linked dextran methacrylate, PNIPAAm, protein (insulin or albumin)		polymeric solutions dropped onto superhydrophobic surfaces in a dry environment under UV light	goniometer, SEM, FTIR	197
LTSLs	thermoresponsive	$ZnSO_4$, PBS, KH_2PO_4, NBT, $MgCl_2$, NADPH, trifluoroacetic acid	clinical-grade DOX		PK, HPLC, fluorescence microscopy	198
PNIPAAm-MAA-coated Fe_3O_4 NPs	thermo-and pH-responsive	$FeCl_3 \cdot 6H_2O$), $FeCl_2 \cdot 4H_2O$, ammonium hydroxide, ammonium persulfate, 1,4 dioxan, AIBN, NIPAAm, MAA, BIS, VTEO, acetic acid and ethanol	DOX	improved chemical CPT method, acid catalyst hydrolysis	XRD, SEM, FTIR, vibrating sample magnetometer (VSM)	199
hydrogels derived from TMC	thermoresponsive	PEG, chitosan, PAA		reductive methylation of chitosan	TGA, XRD, FT-Raman, rheometer, SEM	200
GO–polymer NPs (PNP) hybrids	thermoresponsive	GO, PNP	ADR	free radical polymerization, covalent interaction mediated assembly of PNPs on graphene oxide (GO) nanosheets	FTIR spectra, 1 H NMR, DSC, XRD, Raman spectra, XPS, TGA, AFM, TEM, fluorescence microscope image	201
PEC micelle	thermo-and pH-responsive	CS-*g*-PNIPAAm, CMC-*g*-PNIPAAm	5-FU	free radical polymerization, mixing	UV–vis spectrophotometer, luminescence spectrometer, DLS, TEM	202
PTX-NCs/F127 hydrogel	thermoresponsive	Pluronic F127, Taxol	PTX	dissolution, evaporation, stirring	DLS, TEM, DSC, rheometer	203
APT/TSP liposomes	thermoresponsive	ssDNA-APT, poly(NIPMAM-*co*-NIPAM)	DOX	vortexing, incubation	ultrasound irradiation, microplate spectrofluorometer	204
LMMNA	thermo-and pH-responsive	DPPC, Chol, DSPE-PEG_{2000}, MMNA	DOXTXL	solvothermal method, thin film hydration method	XRD, ZPA, TEM, AFM, VSM	205

stimuli-responsive nano-carrier	nanocarrier type	materials	drug	synthesis	characterization	refs
core–shell NPs (mPEG–PPAd NPs)	thermoresponsive	PPAd, mPEG		two-stage melt poly condensation method, oil-in-water emulsification and solvent evaporation technique	Ubbelohde capillary viscometer, GPC, DSC, WAXD, 1 H NMR, SEM, TEM	206
Hydrogel film	thermoresponsive	NIPAAm, EBA	DC, NPX	UV-initiated radical polymerization	FTIR, SEM, DSC	207
IPN hydrogels	thermo-and pH-responsive	PASP hydrogel(pH-sensitive, PNIPAAm) (temperature-sensitive)		redox radical polymerization in the presence of cross-linkers	SEM, DSC	208
injectable IPN	thermo-and pH-responsive	PEGMA, NIPAm, ALG, MA, BIS	DCS, BSA	esterification of hydroxyl groups, copolymerizing PEGMA and NIPAm	FTIR, rheometer	209
hollow polymer micro-capsules	thermoresponsive	DVB, KPS, NIPAm,		emulsion polymerization	TEM, DLS	210

Table 3

Comparison of Viral and Nonviral Vectors for Gene Delivery[213–219]

parameter	viral	nonviral
production	limited capacity for DNA delivery; small production scale; dificult production and expensive	easier and cheaper production
safety	On the basis of their residual viral elements, safety problems raised by:	
	potential toxicity; immune response and inflammatory reactions; insertional mutagenesis	ability to safely transport gene cargo in vivo (even into specific subcellular ingredients; limited toxicological implications; randomly integrate DNA into the host cells
efficiency	high gene transfection efficiency	transfection efficiencies are not as high as viral

Table 4

Examples of Temperature-Responsive Polymers Utilized for Nonviral Gene Delivery

polymer	advantages/properties	application	refs
PNIPAAm-*co*-PEI	LCST increases up to 37 °C; good gene expression; low toxicity	DNA complexation; preventing the adsorption of serum proteins	227
P(NIPAAm-co MMA)	switch from hydrophilic surface (below 29.18°C as a transition temperature) to hydrophobic surface (above this temperature) occurs in this polymer	modulate cell adhesion and cell sheet retrieval	228
P(NIPAM-*co*-DMAEMA-*co*-BMA)	temperature changes modulate polyplex formation/dissociation	delivery of therapeutic agents and nucleic acids	229
P(NIPAAm-*co*-DMAAm)-*b*-PLA	interaction between the hydrated NIPAAm and the cells; leads to internalization above the LCST	delivery of nucleic acids and peptides/protein	230
PDMAEMA–PPO-PDMAEMA	forms rodlike polyplexes; DNA is encapsulated into biodegradable polymer shell; effective transfection	DNA delivery to HEK 293 cells	231
$PMEO_2MA$-*b*-PHEMA diblock copolymers	condensation of DNA more efficiently by PEIMH. More exposure of surface positive charges of PEIMH/pDNA complexes due to collapse of $PMEO_2MA$ chains at temperature above LCST	DNA complexation. Gene expression level in HEK293 cells	232
PDMAPAAm-PNIPAAm	high transgene expression and god cell viability	plasmid DNA complexation	233
PEG-*b*-PNIPAAm	encapsulation of both hydrophilic drugs within the interior and hydrophobic molecules in the membrane	tissues such as tumors	234
PNIPAAm-*b*-PLLys	DNA is encapsulated into biodegradable polymer shell decorated with targeting functions; effective transfection	DNA delivery into cells	235
(PNIPAAm -*g*-PEG)-*b*-PLLys	high potential for condensing DNA; low cytotoxicity; effective transfection	DNA delivery into cells	235
PNIPAAm-*b*-PLLys	DNA is encapsulated into biodegradable polymer shell decorated with targeting function; effective transfection	DNA delivery into cells	235
P(NIPAAm-*co*-HEMA-*co*-DMAEMA)	positively charged copolymers as vectors for gene delivery	deliver green fluorescent protein (GFP) gene into HEK293T cells	236
PNIPAAm -*co*-Am	by increasing from room to body temperature, the size of PCANs decreases because of phase transition from hydrophilic to hydrophobic; this enhances cellular uptake	RA delivery to direct hiPSC	237
PEG-*b*-poly-L-EG_2Glu	this diblock copolymer displayed two levels of self-assembly behavior, which caused thermoinduced solubility and conformation transition		238