Smart Polymeric Materials for Drug Delivery

Gheorghe Fundueanu, Sanda Bucatariu, and Marieta Constantin

Abstract Smart polymers or polymers sensitive to external stimuli are remarkable materials because they behave similarly to living organisms. Basically, these polymers as linear molecules or cross-linked networks modify their physico-chemical properties such as hydrophilic/ hydrophobic balance, solubility, degree of swelling in response to the action of external stimuli present in the human body. Among stimuli-sensitive polymers, those sensitive to pH and temperature are the most useful in biomedical area because in the human body, these physico-chemical parameters change in different body compartments. All of these changes were exploited to control the moment and release rates of drugs. Moreover, these systems have the ability of reacting to the presence of biomolecules released by the body under pathological conditions.

Keywords Poly(*N*-isopropylacrylamide) · Smart polymers · Drug delivery · Hydrogels

Abbreviations

AA	Acrylic acid
AAm	Acrylamide
BMA	Butyl methacrylate
CAB	Cellulose acetate butyrate
DF	Diclofenac

G. Fundueanu (✉) · S. Bucatariu · M. Constantin
Department of Natural Polymers, Bioactive and Biocompatible Materials, "Petru Poni" Institute of Macromolecular Chemistry, 700487 Iassy, Romania
e-mail: ghefun@icmpp.ro

S. Bucatariu
e-mail: sanda.bucatariu@icmpp.ro

M. Constantin
e-mail: marieta@icmpp.ro

M. J. M. Abadie et al. (eds.), *New Trends in Macromolecular and Supramolecular Chemistry for Biological Applications*, https://doi.org/10.1007/978-3-030-57456-7_14

MA	Methacrylic acid
MM	Methyl methacrylate
SEM	Scanning electron microscopy
LCST	Lower critical solution temperature
NIPAAm	*N*-Isopropylacrylamide
NIPMAAm	*N*-Isopropylmethacrylamide
NVP	*N*-Vinylpyrrolidone
PBS	Phosphate buffer solution
PVA	Poly(vinyl alcohol)
Tg	Glass transition temperature

1 Introduction

Controlled drug delivery systems have represented a huge improvement in the way of drug administration compared to conventional formulations (Noorian et al. 2020; Krause et al. 2019; Kim et al. 2019). These systems have the advantage that they can maintain the concentration of the drug in the blood and tissues in the therapeutic field, reduce the frequency of drug administration and increase patient comfort and compliance. However, there are several medical disorders (diabet, heart disorders, etc.) when the continuous release of drugs in shorter or longer periods is not appropriate. For these cases, the pharmaceutical formulations must release the drug when one of the parameters that regulates the body's functions changes. These systems are generally based on intelligent or stimuli sensitive polymers (Liu et al. 2019; Jamwal et al. 2019; Alsuraifi et al. 2018).

Smart polymers are a category of macromolecules that in aqueous solution suffer a phase transition when small changes of the external parameters occur. These parameters can be temperature (Lee and Bae 2020), pH (Sapre et al. 2020), ionic strength (Zhang et al. 2005), electric field (Shang et al. 2007), magnetic field (Manouras and Vamvakaki 2017), light (Sedlacek et al. 2019), redox potential (Singh et al. 2020), the presence of biomolecules (Fundueanu et al. 2013), etc. Among polymers sensitive to external stimuli, those sensitive to temperature and pH are the most used for biomedical applications because both parameters vary most frequently in the human body. Changes in temperature and pH cause modifications in the physico-chemical properties of polymeric pharmaceutical formulation that result in modulation of the release profiles of loaded drugs (Wang et al. 2020; Zheng et al. 2019).

2 Thermosensitive Drug Delivery Systems

Drug delivery systems sensitive to temperature have the ability to control the moment of drug delivery only when the human body temperature is above or below the physiological temperature.

Poly (N-isopropylacrylamide) (poly(NIPAAm)) is the most used thermoresponsive polymer in biomedical applications because it shows a sharp phase transition around 32 °C, which is close to the human body temperature (Fig. 1) (Heskins and Guillet 1968).

The temperature at which this transition takes place is called the lower critical solution temperature (LCST). Below the LCST the polymer chains are surrounded by water molecules bound by hydrogen bonds, exposing an extended coil conformation and making the polymer soluble in water. However, when the temperature exceeds the LCST, the polymer loses the hydration water, becomes hydrophobic and precipitates (Fig. 1). Consequently, the cross-linked hydrogels obtained from these polymers swell under the LCST and shrink above the LCST (Fig. 2) (Fundueanu et al. 2009a).

This swelling/shrinking process has been exploited for the development of thermosensitive drug delivery systems based on poly(NIPAAm-co-hydroxyethylacrylamide) microspheres (Fundueanu et al. 2013). The drug is usually released when the hydrogel is in the swollen state (below LCST). Above the LCST, the matrix shrinks and the release of drug is stopped. The pulsatile effect depends to the size of hydrogel microspheres. It was established that microspheres with the diameter ranging between 5 and 60 μm release the drug with almost the same rate below (in the swollen state) and above the LCST (in the collapsed state). On the contrary, microspheres with the diameter ranging between 125 and 220 μm release a significantly higher amount of indomethacin below than above the LCST. This difference was enough pronounced to ensure a pulsatile release mechanism when the temperature changes cyclically below and above the LCST.

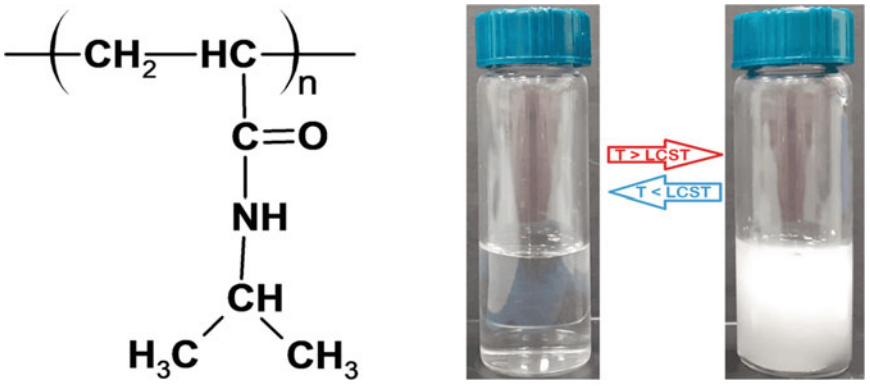

Fig. 1 Poly(N-isopropylacrylamide) aqueous solution below and above the LCST

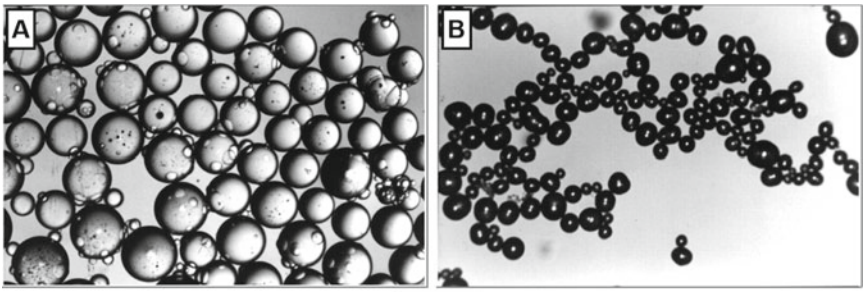

Fig. 2 Optical photomicrographs of poly(NIPAAm-co-acrylamide) microspheres in the swollen state in phosphate buffer below the LCST value at 22 °C (panel A) and above the LCST value at 45 °C (panel B). Reproduced with permission from Acta Biomaterialia (Fundueanu et al. 2009a)

In addition, the amount of entrapped drug influences the pulsatile release mechanism. Microspheres from poly(NIPAAm-co–N-vinylpyrrolidone) (poly(NIPAAm-co-NVP)) with a low loading degree (7.62%), display a substantially difference between the diclofenac (DF) release rate at temperatures situated below and above the LCST (Fig. 3A); this difference guarantees a pulsatile release mechanism (Fig. 3B). On the opposite, the release profiles of DF from microspheres with high loading degree (13.08%) at temperatures lower and higher the LCST are almost superimposable (Fig. 3C), therefore the cyclically variation of temperature does not ensure a pulsatile mechanism (Fig. 3D) (Fundueanu et al. 2020a).

As it was mentioned, the LCST of the poly(NIPAAm) in aqueous solution is around 32 ºC, which is slightly lower than the temperature of the human body. In order to increase the transition temperature, NIPAAm is copolymerized with hydrophilic co-monomers (Khan 2007; Liu et al. 2004). In fact, by copolymerization with hydrophilic monomers, the hydrophilic/hydrophobic balance is disturbed and higher energy (temperature) is necessary to break the hydrogen bonds between copolymer and water. In addition, the insertion of hydrophilic co-monomers along the main chain in a higher proportion alters the amide and isopropyl sequences of poly(NIPAAm) and can lead to a decrease or even loss of thermosensitive properties (Feil et al. 1933). On the opposite, the copolymerization of the NIPAAm with hydrophobic monomers leads to a decrease of the LCST below 32 °C (Luan et al. 2016). An atypical behaviour was observed for poly(N-isopropylmethacrylamide) (poly(NIPMAAm) with structure similar to that of NIPAAm with the exception of methyl groups. Although poly(NIPMAAm) is more hydrophobic than poly(NIPAAm), the phase transition temperature of poly(NIPMAAm) in aqueous solution is higher (≈46 °C) (Djokpé and Vogt 2001), reflecting the conformation of the monomeric structure in poly(NIPMAAm). The presence of methyl groups throughout the main polymeric chain hinders the hydrophobic interactions, therefore a higher temperature is necessary for polymer precipitation. The copolymerization of NIPAAm with increasing amount of NIPMAAm gives copolymers with increased values of LCST (Table 1) (Fundueanu et al. 2016).

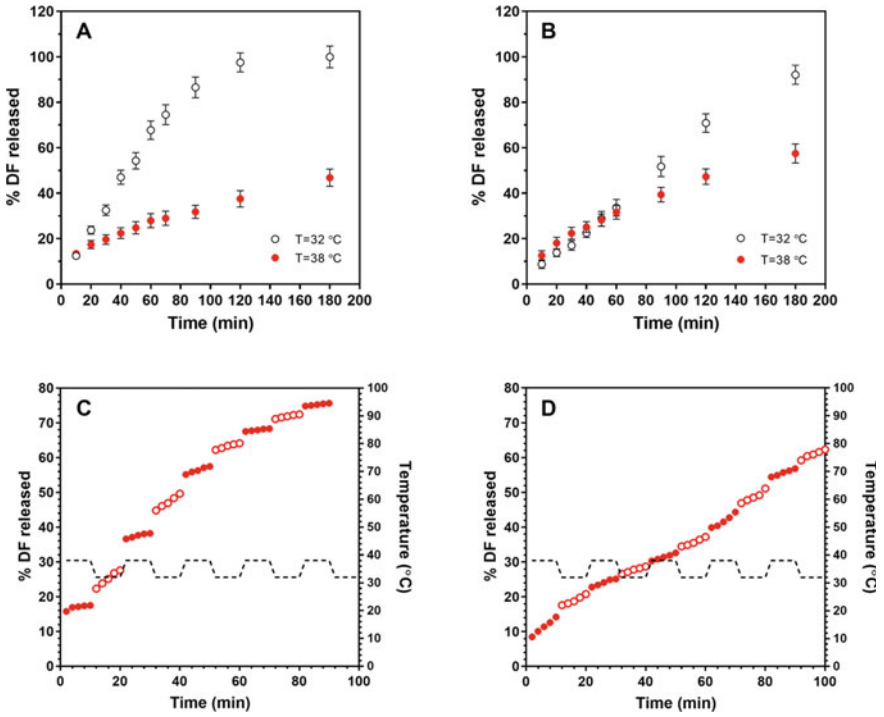

Fig. 3 Effect of temperature (panels A and B) and temperature cycling (panels C and D) (32 °C (empty symbols) and 38 °C (full symbols)) on DF release profiles from poly(NIPAAm-co-NVP) microspheres. The release studies were performed in PBS at pH = 7.4, using microspheres with 7.62% (w/w) DF (panel A and C) and with 13.08% (w/w) DF (panel B and D). Reproduced with permission from Express Polymer Letters (Fundueanu et al. 2020)

Table 1 Dependence of LCST on the co-monomer ratio in the feed and in the copolymer. The concentration of the copolymer solution was 1% (w/v). Reproduced with permission from Macromolecular Chemistry and Physics (Fundueanu et al. 2016)

Sample	Co-monomer composition (% mol ratio)				LCST (°C)		
	In the feed		In copolymer		pH 7.4	pH 1.2	H2O
	NIPAAm	NIPMAAm	NIPAAm	NIPMAAm			
S_0	100	0	100	0	29.9 ± 0.2	31.6 ± 0.2	32.6 ± 0.2
S_1	66.67	33.33	55.00	45.00	35.9 ± 0.3	– [a]	– [a]
S_2	60	40	51.00	49.00	36.8 ± 0.3	38.2 ± 0.3	38.8 ± 0.2
S_3	50	50	41.75	58.25	38.3 ± 0.3	– [a]	– [a]
S_4	33.33	66.67	28.75	71.25	40.9 ± 0.2	– [a]	– [a]
S_5	0	100	0	100	44.7 ± 0.3	45.0 ± 0.2	45.5 ± 0.3

Data are the results of two independent experiments.
[a] not done.

Practically, the LCST of poly(NIPAAm-co-NIPMAAm) increases almost linearly with the percentage of NIPMAAm in the copolymer; comparable results were found by Djokpé and Vogt (Djokpé and Vogt 2001).

In fact, the phase transition of the copolymer is situated between the LCST of poly(NIPAAm) homopolymer and that of poly(NIPMAAm). Under simulated physiological conditions (phosphate buffer at pH 7.4) poly(NIPAAm) displays a LCST of 29.9 °C while poly(NIPMAAm) has a LCST at 44.8 °C, which both are lower than that determined in aqueous solution. This decrease in the transition temperature is due to the high ionic strength of the phosphate buffer which weakens the interaction between the polymer chains and the water.

However, the most used pharmaceutical form for these polymers in biomedical applications are hydrogels (Andrei et al. 2016; Sung et al. 2015). They are obtained by copolymerization of corresponding co-monomers in the presence of small amounts of cross-linkers. The resulted hydrogels should preserve the thermosensitive properties of the linear polymer. Moreover, they should display fast swelling deswelling rates to small changes of the temperature to ensure a pulsatile drug delivery (Fig. 4) (Fundueanu et al. 2016).

Swelling/desweling rates of thermosensitive hydrogels depend mainly by the diffusion rate of water molecules into and out of the hydrogel. The diffusion rate

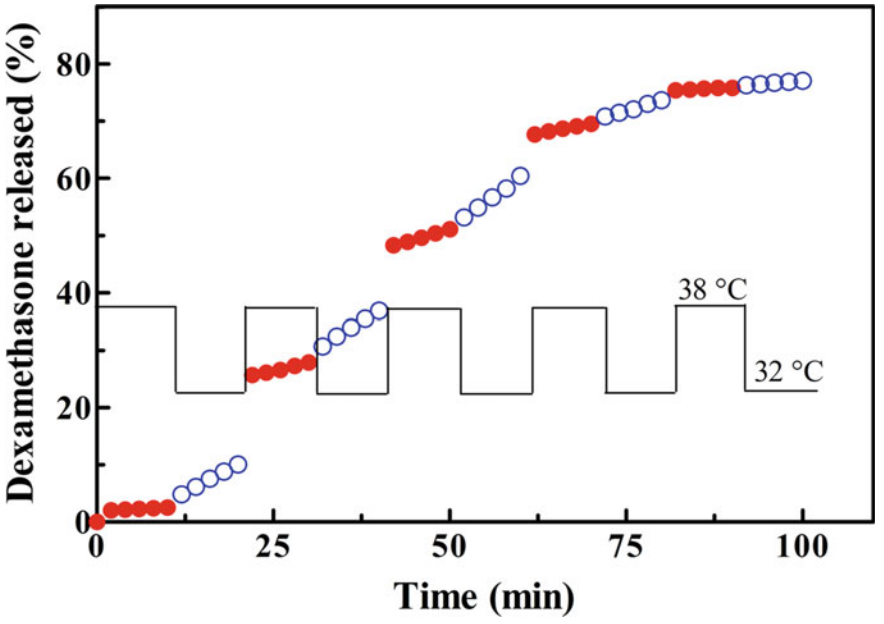

Fig. 4 Influence of cyclically temperature change on dexamethasone release from the poly(NIPAAm-*co*-NIPMAAm) microgels. Data were obtained in simulated physiological conditions (PBS at pH = 7.4) at 32 °C (empty symbols) and 38 °C (full symbols). Reproduced with permission from Macromolecular Chemistry and Physics (Fundueanu et al. 2016)

of water molecules is governed by the dimension and porosity of the hydrogel. Certainly, the smaller is the size the faster is the diffusion rate; in addition a high porosity allows a rapid diffusion of water (Sato-Matsuo and Tanaka 1988; Tanaka and Fillmore 1979). The creation of a porous structure in a three-dimensional network is one of the best procedure used to avoid the „skin effect" that hinders the diffusion of water and promotes a rapid volume change of hydrogel in response to temperature modifications (Strachotova et al. 2007). In hydrogels with porous structures, the water is ejected or absorbed by convection, a much faster process than by conventional diffusion. Usually, porous poly(NIPAAm) hydrogels were prepared in the presence of a template molecule. Tokuyama and Kanehara (2007) have synthesized hydrogels by free radical copolymerization of NIPAAm and N,N-methylene-bis-acrylamide, as cross-linker, in an oil-in-water emulsion (Tokuyama and Kanehara 2007). At the end of polymerization, the dispersed oil minidroplets used as template are removed by washing with appropriate solvents resulting a porous hydrogel. Thermally reversible macroporous poly(NIPAAm) hydrogel has also been synthesized in aqueous solution at a temperature above the LCST. As long as the reaction takes place at a temperature above the LCST, the polymer collapses as it reaches a certain degree of polymerization. The hydrophobic interactions that take place later between the aggregates create bridges that generate a heterogenous macroporous structure (Yan and Hoffman 1995). Hydrogels with large pores were also obtained in the presence of hydroxypropyl cellulose as a pore-forming agent. The response rate to the temperature changes was much faster for these hydrogels than those obtained without porogens. However, since the polymerization occurred above LCST, the macroporous hydrogel displayed a heterogeneous structure (Wu et al. 1992). Microgels with homogeneous pore size and size distribution were obtained by chemical cross-linking of preformed poly(NIPAAm-co-AAm) in the presence of poly(NIPAAm) thermosensitive oligomers, taken as porogen (Fundueanu et al. 2009b). The porous microspheres were obtained both below and above the LCST of the poly(NIPAAm) oligomer. The size of the pores is obviously influenced by the amount of the porogen in the reaction mixture. In the absence of the porogen no pores are formed and the microspheres show a compact internal structure and a smooth surface. The best size distribution and homogeneity of the pores were obtained under the LCST of porogen because the synthesis process takes place in a homogeneous solution of the polymer and oligomer (Fig. 5).

Due to the porous structure and hydrophilic character of the co-monomers, the microgels display relatively high values of water regain and swelling degree. Also, they are characterized by a very rapid response rate when the temperature changes below and above the body temperature. The porous microspheres have proven to be suitable matrix for loading and temperature-controlled release of the high molecular weight model drug blue dextran (Fig. 6).

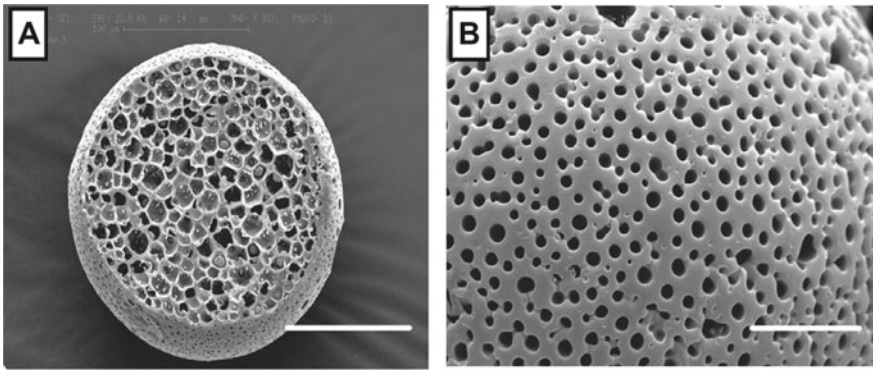

Fig. 5 SEM micrographs of porous poly(NIPAAm-co-AAm) microspheres obtained using poly(NIPAAm) oligomers as porogens: cross-section (panel A) and surface detail (panel B). The bars correspond to 100 and 20 μm in panels A and B, respectively. Reproduced with permission from International Journal of Pharmaceutics (Fundueanu et al. 2009b)

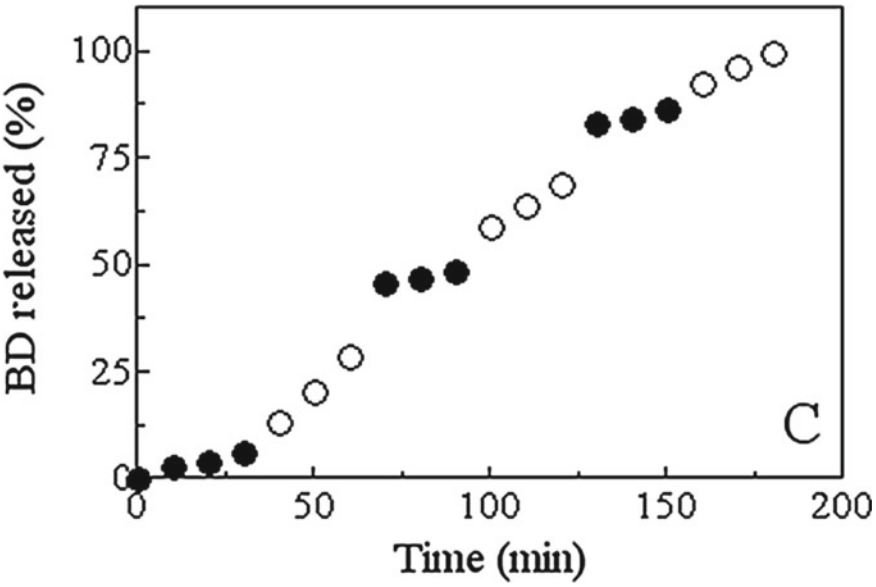

Fig. 6 Effect of temperature cycling (33 °C (empty symbols) and 38 °C (full symbols)) on Blue Dextran release from poly(NIPAAm-co-AAm) microspheres in phosphate buffer at pH 7.4. Reproduced with permission from International Journal of Pharmaceutics (Fundueanu et al. 2009b)

3 PH-Sensitive Drug Delivery Systems

Drug delivery systems sensitive to pH were designed and developed to exploit the pH variations in the human body. For example, the pH varies from 1–2 in the stomach

to 7.4 and even 8 in the blood and duodenum, respectively. Additionally, the pH-sensitive systems have appealed an enormous interest because the differences in pH between normal tissue and tumoral tissues can be used to design pH-sensitive drug delivery carriers that can target tumors and release loaded drugs at the tumor site (Xu et al. 2020; Men et al. 2020; Sim et al. 2019).

Generally, the pH-sensitive polymers comprise weakly acidic (carboxylic) or weakly basic (amine) functional groups (Riyajan 2019; Liu et al. 2018). The most important properties of these polymers result from the protonation/deprotonation of the pH-sensitive groups to small pH change in the range of physiological conditions. The linear polymers change their hydrophilic / hydrophobic character and consequently their solubility. The cross-linked hydrogels synthesized from these polymers swell and shrink as a result of protonation/deprotonation of functional groups (Vaghani and Patel 2011; Sabzi et al. 2020). Basically, hydrogels possessing carboxylic pH-sensitive groups become protonated at low pH, below the pKa and therefore is relatively not swollen (Fig. 7A). As the pH rises above the pKa value,

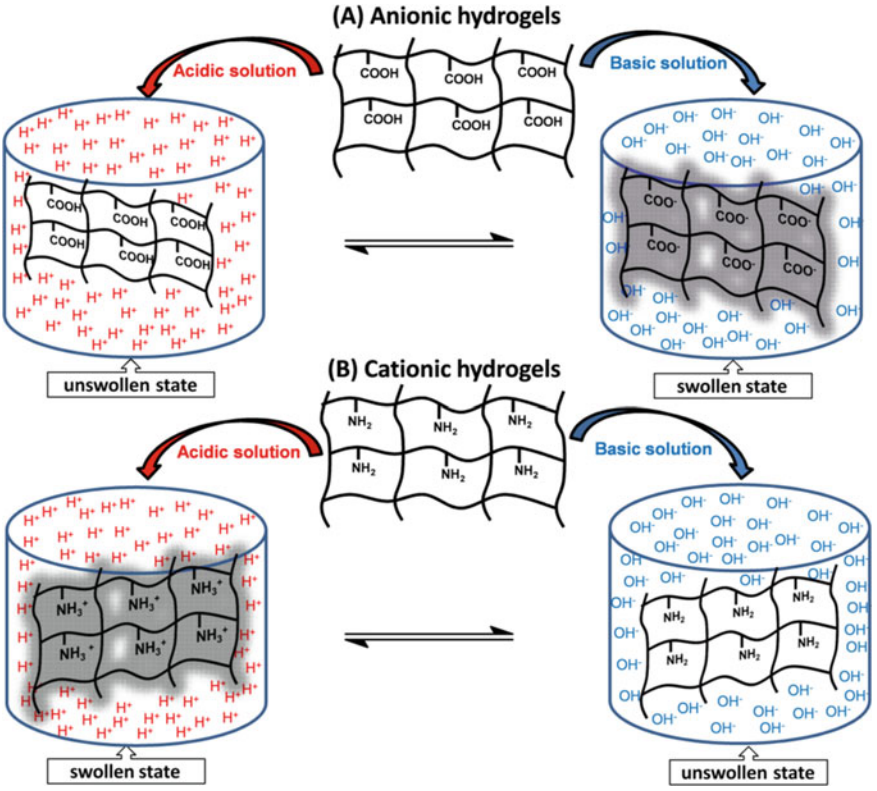

Fig. 7 Swelling behavior of an anionic (panel A) and cationic hydrogel (panel B) in acidic and basic solution

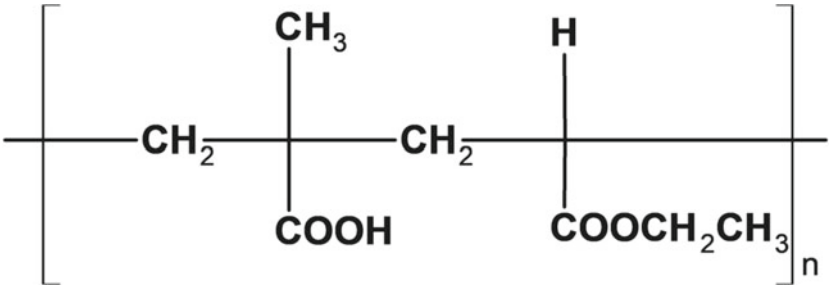

Fig. 8 Chemical structure of Eudragit L 100–55

the carboxylic groups ionize, become hydrophilic and the hydrogel swells. Hydrogels with amino pH-sensitive groups have a completely opposite behavior. At low pH, under the pKa, the amino groups are protonated (ionized) and the hydrogel swells while at higher pH values, above pKa, the amino groups are non-ionized, less hydrophilic and the hydrogel is in a collapsed state (Fig. 7B).

The most commonly used pH-sensitive polymers in the linear form are methacrylic acid copolymers, known as Eudragit R (a registered trademark of Rohm Pharmaceuticals; Darmstadt, Germany). For example, Eudragit L 100-55 T forms salts and dissolves above pH 5.5 and is insoluble in gastric fluids (Fig. 8). This property has been exploited to deliver drugs to the large intestine by way of pH sensitive enteric coatings or as micro- and nano-particles (Hao et al. 2013).

Besides Eudragits, other pH-sensitive polymers such as derivatives of cellulose are largely used for drug targeting to the small intestine. Among these, cellulose acetate phthalate (pKa ~ 5.5), also known as cellacefate is a cellulose derivative where about half of the hydroxyl groups are esterified with acetyls and a quarter is esterified with one or two carboxyls of the phthalic acid (Fig. 9). It is a commonly used polymer in the formulation of pharmaceuticals, such as the enteric coating of tablets or capsules for controlled release formulations (Jagdale and Chandekar 2017).

However, the most important drawback of these polymers is the lack of biodegradability. This inconvenience is not so severe for pharmaceutical formulations with oral application, instead, for other types of administrations such as parenteral ones, the biodegradability becomes a very acute problem. Therefore, natural polymers with native pH-sensitive units were frequently used for delivery of drugs modulated by the pH. Among these, alginic acid (Boi et al. 2020) and chitosan (Ata et al. 2020) are the most representative anionic and cationic natural polymers, respectively, used in biomedical applications. Concerning the chemical structure, alginic acid is described as a linear block copolymer composed of sequences with consecutive β-1,4-D-mannuronic acid residues (M-blocks), α-L-guluronic acid residues (G-blocks) and alternating M and G residues (MG-blocks) (Fig. 10).

The pKa of mannuronic and guluronic acid in 0.1 M NaCl are known to be 3.38 and 3.65, respectively. Owing to its pH-sensitivity, biocompatibility, low toxicity and muco-adhesive properties, alginic acid is considered a noticeable polymer

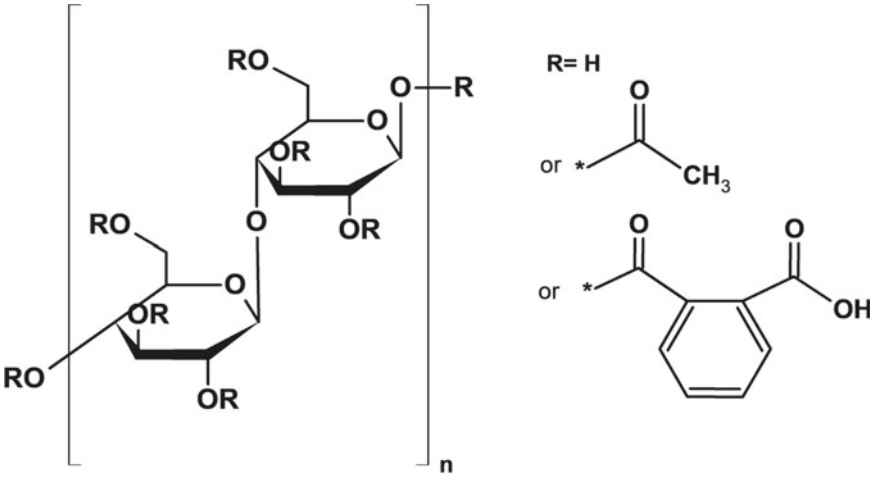

Fig. 9 Chemical structure of cellulose acetate phthalate

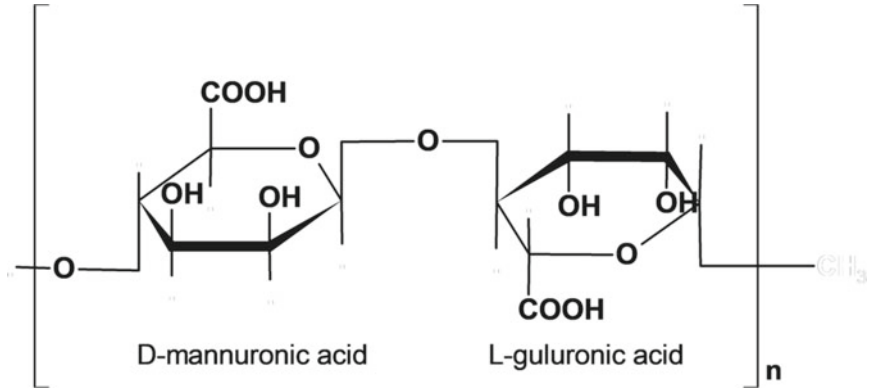

Fig. 10 Chemical structure of alginic acid

for biomedical applications. Therefore, George and Abraham (2007) developed a pH-sensitive alginate–guar gum hydrogel cross-linked with glutaraldehyde for oral administration of protein drugs (George and Abraham 2007). It is well-known that protein instability mainly in gastric fluids is one of the major motives by which proteins are administered usually through injection rather than taken by oral route. Due to the protonation of alginate, the amount of released protein from test hydrogels was minimal at pH 1.2 (∼20%), but it was found to be significantly higher (∼90%) at pH 7.4., when the carboxylic groups are in ionized form and the hydrogel swells.

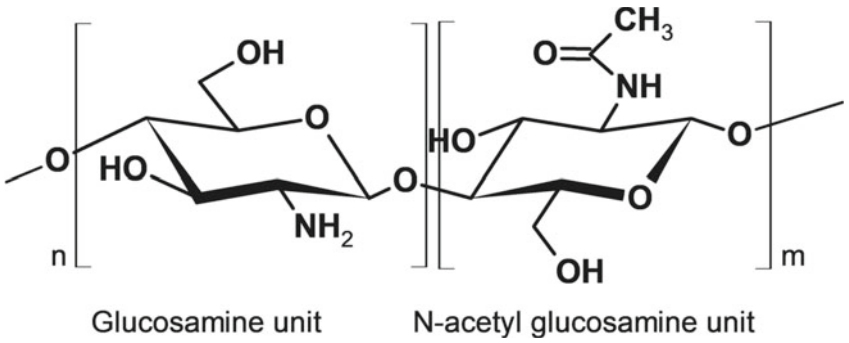

Fig. 11 Chemical structure of chitosan showing the repeating subunits

With the aim to enhance the drug entrapment efficiency and control the swelling properties and stability of drug delivery system in different pH media, Ca^{2+} cross-linking and freeze-thawing cycle techniques were used to prepare sodium alginate/poly(vinyl alcohol) (PVA) hydrogel beads (Hua et al. 2010). The alginate/PVA mixture was firstly cross-linked with Ca^{2+} to form beads and then subjected to freezing–thawing cycles for additional crosslinking. It was proved that the swelling, drug release profiles, and degradation of the developed beads were influenced by pH of the testing medium and PVA content.

Chitosan is another linear polysaccharide composed of randomly distributed β–linked D-glucosamine and N-acetyl-D-glucosamine (Fig. 11).

Like alginate, it is biocompatible, biodegradable and the degradation by-products are not toxic. It is the second abundant biomass-derived polysaccharide and is obtained by treating the chitin shells of shrimp and other crustaceans with sodium hydroxide. If alginate-based hydrogels collapse in an acidic environment and swell in a weakly basic environment, chitosan hydrogels have an opposite behaviour; they swell in acidic pH and collapse in basic media. This comportment is given by the presence of abundant amino groups with a pKa of around 6.5. Since the pKa value is situated in the pH range of tumor environment, it is widely used for controlled delivery of anticancer drugs (Ghaffari et al. 2020; Wang et al. 2019). As follows, an interesting hydrogel was designed and developed from the cross-linking of chitosan, graphene, and cellulose nanowhisker via Schiff base reaction by a synthetic dialdehyde (Omidi et al. 2020). The hydrogel was loaded with two antitumoral drugs, doxorubicin and curcumin.

The drug release profiles demonstrated that the rate of release is dependent on switching pHs from neutral pH to the acidic conditions. By decreasing of pH from 7.4–5.4, the strength of cross-linking imine bonds decreases and as well the repulsive ionic interactions between the protonated amino groups increase which leads to an expanded structure.

Chitosan was also used for the preparation of microcapsules for cell encapsulation and cell culture (Fundueanu et al. 2020b). Since the encapsulation process must take place in mild conditions and the neutral chitosan is soluble in just in acidic conditions,

harmful to cells, the amino groups were protonated and the chitosan was transformed in the hydrochloride form. Thus, the chitosan derivative was easily solubilized in isotonic solutions and cell encapsulation was successfully performed with no cell damage. Moreover, the protonated chitosan microcapsules allow the cells to adhere to their surface.

One of the most important drawbacks of hydrogels prepared from simple alginate or chitosan is their low stability in physiological fluids. Therefore, core–shell nanoparticles from polyelectrolytes complexes between chitosan and alginate were prepared for oral insulin administration (Mukhopadhyay et al. 2015).

The nanoparticles displayed an almost spherical shape, an average diameter of 100–200 nm determined by dynamic light scattering, and high insulin encapsulation efficiency. In simulated gastric fluid, the nanoparticles retained almost entire amount of encapsulated insulin while in simulated intestinal conditions a sustained release was achieved. Additionally, in vivo experiments presented substantial intestinal absorption of insulin, showing noticeable hypoglycaemic effects with enhanced insulin bioavailability in diabetic mice. After oral administration, the core–shell nanoparticles displayed no systemic toxicity.

4 pH/Temperature-Sensitive Drug Delivery Systems

pH/temperature sensitive drug delivery systems were designed and developed to exploit the variation of both pH and temperature of the human body. They are obtained either in the form of linear or cross-linked polymers by copolymerization of NIPAAm with pH-sensitive monomers. The pH-sensitive co-monomer can increase or decrease the LCST if they are in the un-protonated or protonated state (Yoo et al. 2000). Also, the modification of pH and temperature can modulate the solubility, if the polymer is in the linear form, or swelling behavior for cross-linked hydrogels.

Thus, stimuli-sensitive statistical terpolymers of N-isopropylacrylamide (temperature-sensitive), butyl methacrylate (BMA) and acrylic acid (AA) (pH-sensitive) of various molecular weights were used to prepare microcapsules for modulating the release of insulin (Ramkissoon-Ganorkar et al. 1999). This system has the advantage that only physical interactions polymer/polymer and polymer/protein were used to prepare insulin–loaded microcapsules in mild conditions.

The release rate of insulin was controlled by the pH, temperature and molecular weights of polymers. In gastric fluid at pH = 1.2 and body temperature, the beads were not soluble, and therefore no drug is released in the stomach. At pH 7.4 and body temperature, the low molecular weight polymeric beads shown a dump-like profile and dissolved within 2 h (release mechanism controlled by bead dissolution), while the high molecular weight polymeric beads swelled only and released insulin gradually over a period of 8 h (release mechanism controlled by bead swelling and insulin diffusion).

In another work, the escape of DNA loaded poly(vinyl alcohol) microspheres from multinucleated microcapsules took place through the holes induced by dissolution

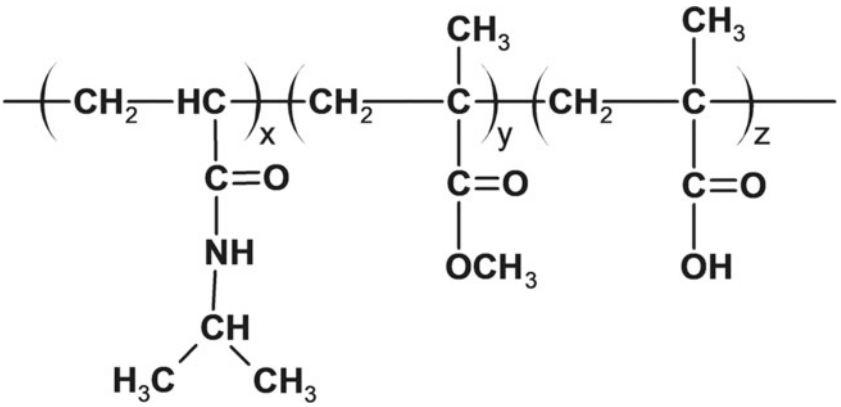

Fig. 12. Chemical structure of poly(NIPAAM-co-MM-MA)

of the pH/temperature sensitive polymer at colonic pH and temperature (Fundueanu et al. 2007).

Fundueanu et al. (2009) developed a pH/ temperature sensitive copolymer based on NIPAAm, methyl methacrylate (MM) and methacrilic acid (MA) (Fig. 12) (Fundueanu et al. 2009).

Poly (NIPAAm-co-MM-co-MA) with a co-monomer molar ratio of 79:13:8 is insoluble in the gastric fluid (pH = 1.2), but soluble in the intestinal fluid (pH = 6.8 and 7.4), at the body temperature (37 °C). This copolymer was mixed with the hydrophobic cellulose acetate butyrate (CAB) and vitamin B12 (taken as a water soluble drug model system) and transformed in spherical microparticles by oil-in-water solvent evaporation method. With the aim to determine the compatibility between the two components of the microspheres (CAB with pH/temperature sensitive copolymer), the glass transition temperatures (Tg) of separated polymers and of microspheres were determined.

The occurrence of two Tg values validated that there is no interaction between the two components; they precipitated separately during microsphere preparation, the smart polymer forming small micro-domains on the surface and within the microspheres (Fig. 13A).

At pH = 1.2 and 37 °C, poly(NIPAAm-co-MM-co-MA) is not soluble, therefore the amount of vitamin B12 released is very low. When the temperature is lowered to 20 °C, below LCST, the copolymer solubilizes and an increased percentage of vitamin B12 is released. In phosphate buffer at pH = 6.8, the copolymer is less soluble than at pH = 7.4, therefore the release rate is lower but is much higher than at pH = 1.2. Obviously, the highest amount of vitamin is released at pH = 7.4, when the copolymeric domains solubilize totally creating pores within three-dimensional network (Fig. 13B).

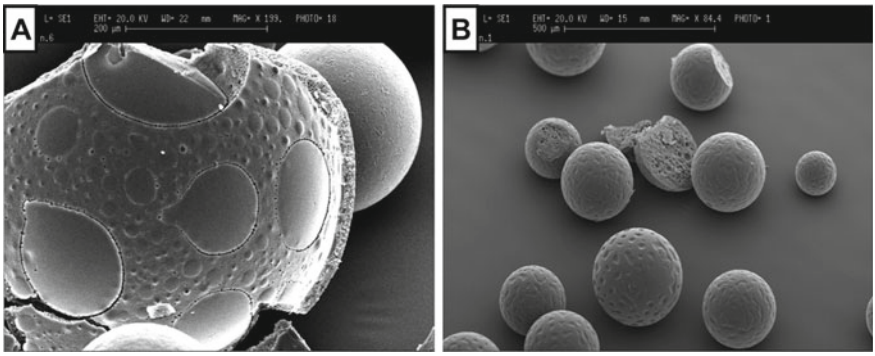

Fig. 13 SEM micrographs of CAB/poly(NIPAAm-co-MM-co-MA) microspheres before (panel A) and after (panel B) release studies in PBS at pH = 7.4. Reproduced with permission from Journal of Materials Science-Materials in Medicine (Fundueanu et al. 2009)

The copolymerization of NIPAAm with ionic co-monomers can increase or decrease the LCST if the charged co-monomers are in the protonated/unprotonated state (Yoo et al. 2000). However, the charged co-monomers, due to their high hydrophilicity in the ionized state, diminish dramatically or even annihilate the thermosensitivity of the copolymer (Constantin et al. 2014).

Remarkably, when the pH-sensitive units interact electrostatically with hydrophobic opposite charged bioactive compounds like diclofenac (Fundueanu et al. 2013), propranolol (Constantin et al. 2014), diphenhydramine (Fundueanu et al. 2017), indomethacin (Constantin et al. 2020), the copolymers restore the thermosensitive properties. Correspondingly, the hydrogels synthesized from these copolymers are in the "inactivated" state in the absence of specific biomolecules at normal physiological pH and temperature (pH = 7.4 and T = 36 °C). However, in the presence of bioactive biomolecules, hydrogels undergo "activation", shrink and expel mechanically a certain amount of drug (Fig. 14).

It must be mentioned that the pH-sensitive component plays the role of a biosensor, the biomolecule acts as a triggering agent, and the poly(NIPAAm) represents the delivery component (actuator).

The proposed hydrogels represent the support of the next generation of self-regulated drug delivery systems based on a sensor (pH-sensitive units) able to detect the perturbation of physiological conditions and a delivery component (thermosensitive units) able to push the necessary dose of drug.

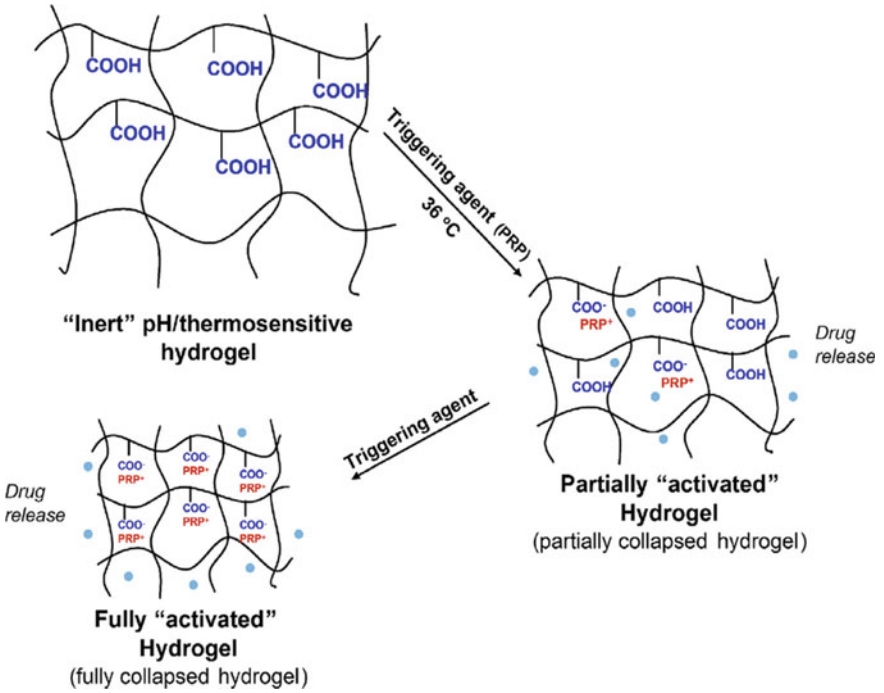

Fig. 14 Schematic representation of the principle of operation of pH/thermosensitive poly(NIPAAm-co-MA) microgels in the presence of the triggering agent, under simulated physiological conditions. Reproduced with permission from European Journal of Pharmaceutical Sciences (Constantin et al. 2014)

5 Conclusions

While the basic concepts of the stimuli-sensitive hydrogels are sound, the practical applications necessitate significant improvements in the hydrogel properties. For example, hydrogels do not have the mechanical strength indispensable in many applications. In addition, the response rate to the input signal is low for large-scale hydrogels. Difficulties to achieve pH/thermo-sensitivity within a narrow pH or temperature range (from pH 7.4 to pH = 7.0 or 6.5; from T = 36.5 °C to 37 °C) often appear since the transition temperature of the cross-linked hydrogels is not as abrupt as for linear polymers. Drug loading and drug retention within microgels for a long period of time could be a major drawback. Also, the selectivity of these systems must be substantially improved.

Acknowledgements The project leading to this application has received funding from the H2020 ERA Chairs Project no 667387: SupraChem Lab Laboratory of Supramolecular Chemistry for Adaptive Delivery Systems ERA Chair initiative. This work was supported also by a grant of the Romanian Ministry of Research and Innovation, CCDI-UEFISCDI, project number PN-III-P1-1.2.-PCCDI-2017-0697/contract nr. 13 PCCDI/2018 within PNCDI III.

References

Alsuraifi A, Curtis A, Lamprou DA, Hoskins C (2018) Stimuli responsive polymeric systems for cancer therapy. Pharmaceutics 10(3):136. https://doi.org/10.3390/pharmaceutics10030136

Andrei M, Turturica G, Stanescu PO, Teodorescu M (2016) Thermosensitive injectable hydrogels from poly(N-isopropylacrylamide)-dextran aqueous solutions: thermogelation and drug release properties. Soft Mater 14(3):162–169. https://doi.org/10.1080/1539445X.2016.1172317

Ata S, Rasool A, Islam A, Bibi I, Rizwan M, Azeem MK, Qureshi AUR, Iqbal M (2020) Loading of Cefixime to pH sensitive chitosan based hydrogel and investigation of controlled release kinetics. Int J Biol Macromol 155:1236–1244. https://doi.org/10.1016/j.ijbiomac.2019.11.091

Boi S, Rouatbi N, Dellacasa E, Di Lisa D, Bianchini P, Monticelli O, Pastorino L (2020) Alginate microbeads with internal microvoids for the sustained release of drugs. Int J Biol Macromol 156:454–461. https://doi.org/10.1016/j.ijbiomac.2020.04.083

Constantin M, Bucatariu S, Harabagiu V, Popescu I, Ascenzi P, Fundueanu G (2014) Poly(N-isopropylacrylamide-co-methacrylic acid) pH/thermo-responsive porous hydrogels as self-regulated drug delivery system. Eur J Pharm Sci 62:86–95. https://doi.org/10.1016/j.ejps.2014.05.005

Constantin M, Bucatariu S, Ascenzi P, Butnaru M, Fundueanu G (2020) Smart drug delivery system activated by specific biomolecules. Mat Sci Eng C 108:110466. https://doi.org/10.1016/j.msec.2019.110466

Djokpé E, Vogt W (2001) N-Isopropylacrylamide and N-Isopropylmethacrylamide: cloud points of mixtures and copolymers. Macromol Chem Phys 202(5):750–757. https://doi.org/10.1002/1521-3935(20010301)202:5%3c750::AID-MACP750%3e3.0.CO;2-8

Feil H, Bae YH, Feijen J, Kim SW (1933) Effect of comonomer hydrophilicity and ionization on the lower critical solution temperature of N-isopropylacrylamide copolymers. Macromolecules 26:2496–2500. https://doi.org/10.1021/ma00062a016

Fundueanu G, Constantin M, Bucatariu S, Ascenzi P (2016) Poly(N-isopropylacrylamide-co-N-isopropylmethacrylamide) thermo-responsive microgels as self-regulated drug delivery system. Macromol Chem Phys 217 (22):2525–2533. https://doi.org/10.1002/macp.201600324

Fundueanu G, Constantin M, Asmarandei I, Harabagiu V, Ascenzi P, Simionescu BC (2013) The thermosensitivity of pH/thermoresponsive microspheres activated by the electrostatic interaction of pH-sensitive units with a bioactive compound. J Biomed Mater Res A 101(6):1661–1669.https://doi.org/10.1002/jbm.a.34469

Fundueanu G, Constantin M, Ascenzi P (2009) Poly(N-isopropylacrylamide-co-acrylamide) cross-linked thermoresponsive microspheres obtained from preformed polymers: Influence of the physico-chemical characteristics of drugs on their release profiles. Acta Biomater 5(1):363–373.https://doi.org/10.1016/j.actbio.2008.07.011

Fundueanu G, Constantin M, Asmarandei I, Bucatariu S, Harabagiu V, Ascenzi P, Simionescu BC (2013) Poly(N-isopropylacrylamide-co-hydroxyethylacrylamide) thermosensitive microspheres: The size of microgels dictates the pulsatile release mechanism. Eur J Pharm Biopharm Part A 85(3):614-623. https://doi.org/10.1016/j.ejpb.2013.03.023

Fundueanu G, Constantin M, Bucatariu S, Ascenzi P (2020a) Poly(N-isopropylacrylamide-co-N-vinylpyrrolidone) thermoresponsive microspheres: the low drug loading ensures the pulsatile release mechanism. EXPRESS Polym Lett 14(1):63–76. https://doi.org/10.3144/expresspolymlett.2020.6

Fundueanu G, Constantin M, Ascenzi P (2009) Fast-responsive porous thermoresponsive microspheres for controlled delivery of macromolecules. Int J Pharm 379(1):9–17. https://doi.org/10.1016/j.ijpharm.2009.05.064

Fundueanu G, Constantin M, Bucatariu S, Nicolescu A, Ascenzi P, Moise LG, Tudor L, Trusca VG, Gafencu AV, Ficai D, Ficai A, Andronescu E (2020b) Simple and dual cross-linked chitosan millicapsules as a particulate support for cell culture. Int J Biol Macromol 143:200–212. https://doi.org/10.1016/j.ijbiomac.2019.12.045

Fundueanu G, Constantin M, Bortolotti F, Cortesi R, Ascenzi P, Menegatti E (2007) Cellulose acetate butyrate-pH/thermosensitive polymer microcapsules containing aminated poly(vinyl alcohol) microspheres for oral administration of DNA. Eur J Pharm Biopharm 66(1):11–20. https://doi.org/10.1016/S0939-6411(07)00176-2

Fundueanu G, Constantin M, Stanciu C, Theodoridis G, Ascenzi P (2009) pH- and temperature-sensitive polymeric microspheres for drug delivery: the dissolution of copolymers modulates drug release. J Mater Sci Mater Med 20(12):2465–2475. https://doi.org/10.1007/s10856-009-3807-0

Fundueanu G, Constantin M, Bucatariu S, Ascenzi P (2017) pH/thermo-responsive poly(N-isopropylacrylamide-co-maleic acid) hydrogel with a sensor and an actuator for biomedical applications. Polymer 110:177–186. https://doi.org/10.1016/j.polymer.2017.01.003

George M, Abraham TE (2007) pH sensitive alginate–guar gum hydrogel for the controlled delivery of protein drugs. Int J Pharm 335:123–129. https://doi.org/10.1016/j.ijpharm.2006.11.009

Ghaffari SB, Sarrafzadeh MH, Salami M, Khorramizadeh MR (2020) A pH-sensitive delivery system based on N-succinyl chitosan-ZnO nanoparticles for improving antibacterial and anti-cancer activities of curcumin. Int J Biol Macromol 151:428–440. https://doi.org/10.1016/j.ijbiomac.2020.02.141

Hao S, Wang Y, Wang B, Deng J, Liu X, Liu J (2013) Rapid preparation of pH-sensitive polymeric nanoparticle with high loading capacity using electrospray for oral drug delivery. Mater Sci Eng C 33:4562–4567. https://doi.org/10.1016/j.msec.2013.07.009

Heskins M, Guillet JE (1968) Solution properties of poly(N-isopropylacrylamide). J Macromol Sci Chem 2:1441–1455. https://doi.org/10.1080/10601326808051910

Hua S, Ma H, Li X, Yang H, Wang A (2010) pH-sensitive sodium alginate/poly(vinyl alcohol) hydrogel beads prepared by combined Ca2+crosslinking and freeze-thawing cycles for controlled release of diclofenac sodium. Int J Biol Macromol 46:517–523. https://doi.org/10.1016/j.ijbiomac.2010.03.004

Jagdale S, Chandekar A (2017) Optimization of chitosan and cellulose acetate phthalate controlled delivery of methylprednisolone for treatment of inflammatory bowel disease. Adv Pharm Bull 7(2):203–213. https://doi.org/10.15171/apb.2017.025

Jamwal S, Ram B, Ranote S, Dharela R, Chauhan GS (2019) New glucose oxidase-immobilized stimuli-responsive dextran nanoparticles for insulin delivery. Int J Biol Macromol 123:968–978. https://doi.org/10.1016/j.ijbiomac.2018.11.147

Krause J, Bogdahn M, Schneider F, Koziolek M, Weitschies W (2019) Design and characterization of a novel 3D printed pressure-controlled drug delivery system. Eur J Pharm Sci 140:105060. https://doi.org/10.1016/j.ejps.2019.105060

Khan A (2007) Preparation and characterization of N-isopropylacrylamide/acrylic acid copolymer core–shell microgel particles. J Colloid Interf Sci 313:697–704. https://doi.org/10.1016/j.jcis.2007.05.027

Kim C, Jeong D, Kim S, Kim Y, Jung S (2019) Cyclodextrin functionalized agarose gel with low gelling temperature for controlled drug delivery systems. Carbohydr Polym 22215:115011. https://doi.org/10.1016/j.carbpol.2019.115011

Lee CH, Bae YC (2020) Thermodynamic framework for switching the lower critical solution temperature of thermo-sensitive particle gels in aqueous solvent. Polymer 195:122428. https://doi.org/10.1016/j.polymer.2020.122428

Liu F, Huang PL, Huang D, Liu SF, Cao QY, Dong XM, Zhang HW, Ko R, Zhou WY (2019) Smart "on-off" responsive drug delivery nanosystems for potential imaging diagnosis and targeted tumor therapy. Chem Eng J 365:358–368. https://doi.org/10.1016/j.cej.2019.02.037

Liu XM, Wang LS, Wang L, Huang J, He C (2004) The effect of salt and pH on the phase-transition behaviors of temperature-sensitive copolymers based on N-isopropylacrylamide. Biomaterials 25(25):5659–5666. https://doi.org/10.1016/j.biomaterials.2004.01.019

Liu YX, Liu KY, Zhao MN, Wang SH, Zhou ZX, Shen YQ, Jiang LM (2018) A pH-responsive fragrance release system based on pseudopeptide polymeric micelles. React Funct Polym 132:138–144. https://doi.org/10.1016/j.reactfunctpolym.2018.09.021

Luan B, Muir BW, Zhu J, Hao X (2016) A RAFT copolymerization of NIPAM and HPMA and evaluation of thermo-responsive properties of poly(NIPAM-co-HPMA). RSC Adv 6:89925–89933. https://doi.org/10.1039/C6RA22722H

Manouras T, Vamvakaki M (2017) Field responsive materials: photo-, electro-, magnetic- and ultrasound-sensitive polymers. Polym Chem 8(1):74–96. https://doi.org/10.1039/C6PY01455K

Men WF, Zhu PY, Dong SY, Liu WK, Zhou K, Bai Y, Liu XL, Gong SL, Zhang SG (2020) Layer-by-layer pH-sensitive nanoparticles for drug delivery and controlled release with improved therapeutic efficacy in vivo. Drug Deliv 27(1):180–190. https://doi.org/10.1080/10717544.2019.1709922

Mukhopadhyay P, Chakraborty S, Bhattacharya S, Mishra R, Kundu PP (2015) pH-sensitive chitosan/alginate core-shell nanoparticles for efficient and safe oral insulin delivery. Int J Biol Macromol 72:640–648. https://doi.org/10.1016/j.ijbiomac.2014.08.040

Noorian SA, Hemmatinejad N, Navarro JAR (2020) Bioactive molecule encapsulation on metal-organic framework via simple mechanochemical method for controlled topical drug delivery systems. Micropor Mesopor Mater 302:110199. https://doi.org/10.1016/j.micromeso.2020.110199

Omidi S, Pirhayati M, Kakanejadifard A (2020) Co-delivery of doxorubicin and curcumin by a pH-sensitive, injectable, and in situ hydrogel composed of chitosan, graphene, and cellulose nanowhisker. Carbohydr Polym 231:115745. https://doi.org/10.1016/j.carbpol.2019.115745

Ramkissoon-Ganorkar C, Liu F, Baudys M, Kim SW (1999) Modulating insulin-release profile from pH/thermosensitive polymeric beads through polymer molecular weight. J Control Release 59:287–298. https://doi.org/10.1016/S0168-3659(99)00006-1

Riyajan SA (2019) Development of a novel pH-sensitive polymer matrix for drug encapsulation from maleated poly(vinyl alcohol) grafted with polyacrylamide. Polym Bull 76(9):4585–4611. https://doi.org/10.1007/s00289-018-2615-4

Sabzi M, Afshari MJ, Babaahmadi M, Shafagh N (2020) pH-dependent swelling and antibiotic release from citric acid crosslinked poly(vinyl alcohol) (PVA)/nano silver hydrogels. Colloids Surf B 188:110757. https://doi.org/10.1016/j.colsurfb.2019.110757

Sapre N, Chakraborty R, Purohit P, Bhat S, Das G, Bajpe SR (2020) Enteric pH responsive cargo release from PDA and PEG coated mesoporous silica nanoparticles: a comparative study in Drosophila melanogaster. RSC Adv 10:11716–11726. https://doi.org/10.1039/C9RA11019D

Sato-Matsuo E, Tanaka T (1988) Kinetics of discontinuous volume phase transition of gels. J Chem Phys 89:1695–1703. https://doi.org/10.1063/1.455115

Sedlacek O, Filippov SK, Svec P, Hruby M (2019) SET-LRP synthesis of well-defined light-responsible block copolymer micelles. Macromol Chem Phys 220(19):1900238. https://doi.org/10.1002/macp.201900238

Shang J, Chen X, Shao ZZ (2007) the Electric-Field-Sensitive Hydrogels. Prog Chem 19(9):1393–1399

Sim T, Han SM, Lim C, Won WR, Lee ES, Youn YS, Oh KT (2019) A pH-sensitive polymer for cancer targeting prepared by one-step modulation of functional side groups. Macromol Res 27(8):795–802. https://doi.org/10.1007/s13233-019-7112-6

Singh S, Kumar V, Kumar B, Priyadarshi R, Deeba F, Kulshreshtha A, Kumar A, Agrawal G, Gopinath P, Negi YS (2020) Redox responsive xylan-SS-curcumin prodrug nanoparticles for dual drug delivery in cancer therapy. Mater Sci Eng C- Mater 107:110356. https://doi.org/10.1016/j.msec.2019.110356

Sung B, Kim C, Kim MH (2015) Biodegradable colloidal microgels with tunable thermosensitive volume phase transitions for controllable drug delivery. J Colloid Interf Sci 450:26–33. https://doi.org/10.1016/j.jcis.2015.02.068

Strachotova B, Strachota A, Uchman M, Slouf M, Brus J, Plestil J, Matejka L (2007) Super porous organic-inorganic poly(N-isopropylacrylamide)-based hydrogel with a very fast temperature response. Polymer 48:1471–1482. https://doi.org/10.1016/j.polymer.2007.01.042

Tanaka T, Fillmore DJ (1979) Kinetics of swelling of gels. J Chem Phys 70:1214–1218. https://doi.org/10.1063/1.437602

Tokuyama H, Kanehara A (2007) Novel synthesis of macroporous poly(Nisopropylacrylamide) hydrogels using oil-in water emulsions. Langmuir 23:11246–11251. https://doi.org/10.1021/la701492u

Vaghani SS, Patel MM (2011) pH-sensitive hydrogels based on semi-interpenetrating network (semi-IPN) of chitosan and polyvinyl pyrrolidone for clarithromycin release. Drug Dev Ind Pharm 37(10):1160–1169. https://doi.org/10.3109/03639045.2011.563422

Wang X, Xu JX, Xu XX, Fang Q, Tang RP (2020) pH-sensitive bromelain nanoparticles by ortho ester crosslinkage for enhanced doxorubicin penetration in solid tumor. Mater Sci Eng C- Mater 113:111004. https://doi.org/10.1016/j.msec.2020.111004

Wang YL, Khan A, Liu YX, Feng J, DaiL WGH, Alam N, Tong L, Ni YH (2019) Chitosan oligosaccharide-based dual pH responsive nano-micelles for targeted delivery of hydrophobic drugs. Carbohydr Polym 223:115061. https://doi.org/10.1016/j.carbpol.2019.115061

Wu XS, Hoffman AS, Yager P (1992) Synthesis and characterization of thermally reversible macroporous poly(N-isopropylacrylamide) hydrogels. J Polym Sci Part A Polym Chem 30:2121–2129. https://doi.org/10.1002/pola.1992.080301005

Xu YQ, Liu DX, Hu J, Ding PR, Chen MW (2020) Hyaluronic acid-coated pH sensitive poly (beta-amino ester) nanoparticles for co-delivery of embelin and TRAIL plasmid for triple negative breast cancer treatment. Int J Pharm 573:118637. https://doi.org/10.1016/j.ijpharm.2019.118637

Yan Q, Hoffman AS (1995) Synthesis of macroporous hydrogels with rapid swelling and deswelling properties for delivery of macromolecules. Polymer 36:887–889. https://doi.org/10.1016/0032-3861(95)93123-4

Yoo MK, Sung YK, Lee YM, Cho CS (2000) Effect of polyelectrolyte on the lower critical solution temperature of poly(Nisopropylacrylamide) in the poly(NIPAAm-co-acrylic acid) hydrogel. Polymer 41:5713–5719. https://doi.org/10.1016/S0032-3861(99)00779-X

Zhang RS, Tang MG, Bowyer A, Eisenthal R, Hubble J (2005) A novel pH- and ionic-strength-sensitive carboxy methyl dextran hydrogel. Biomaterials 26(22):4677–4683. https://doi.org/10.1016/j.biomaterials.2004.11.048

Zheng YT, Wang WF, Zhao JL, Wu CY, Ye CQ, Huang MX, Wang SG (2019) Preparation of injectable temperature-sensitive chitosan-based hydrogel for combined hyperthermia and chemotherapy of colon cancer. Carbohydr Polym 222:115039. https://doi.org/10.1016/j.carbpol.2019.115039