

Smart Polymer Based Delivery Systems for Peptides and Proteins

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Abstract: Biodegradable polymeric systems represent promising means for delivering many bioactive agents, including peptide and protein drugs. The importance of these systems grew with the advancement in the understanding of peptide and protein pharmacology as well as the ability to mass-produce these compounds. Some polymers undergo sol-gel transition once administered. *In situ* gel formation happens in response to one or a combination of two or more stimuli. These stimuli include UV-irradiation, pH change, temperature change, and solvent exchange. These smart polymeric systems have several advantages over conventional methods, such as ease of manufacturing, ease of administration, biodegradability, and the ability to alter release profiles of the incorporated agents. In the past few years, an increasing number of *in situ* gel-forming systems have been investigated and many patents for their use in various biomedical applications, including drug delivery, have been reported. In this article, we introduce the different strategies that have been developed and patented for the use of smart polymers in delivering peptide and protein drugs. The advantage, disadvantages, possibilities, and limitations of each of the smart polymer systems have been discussed.

Keywords: Drug delivery, controlled release, smart polymer, *in situ*, injectable, peptide, protein.

1. INTRODUCTION

Peptides and proteins play a vital role in all biological processes and have received a growing attention in recent years as drug candidates. The rapid advances in peptide and protein pharmacology along with the large-scale production of these compounds by recombinant DNA technology - among other techniques- have fueled enormous interest in these compounds. Unfortunately, peptide and protein development has far outpaced and advanced more rapidly than the ability to deliver these compounds systemically using convenient and effective delivery systems [1]. Oral route is the most convenient and popular but most peptide drugs show low oral activity. This is mainly due to degradation by gastrointestinal tract enzymes and poor permeability of the intestinal mucosa. Alternative routes, such as nasal, pulmonary, rectal, buccal, vaginal, and transdermal, have been investigated. It has been shown that protease activities in the homogenates of the nasal, buccal, rectal, and vaginal mucosa of rabbits are substantial and quite comparable to those in the intestinal mucosa [2]. In another study, degradation of luteinizing hormone releasing hormone (LHRH) was reported in buccal, liver, and nasal tissues [3]. Consequently, most of the new protein-based therapeutics are administered by frequent injections through the parenteral routes such as intravenous, intramuscular, and subcutaneous. With the exception of life-threatening conditions, this form of delivery has traditionally been poorly accepted by patients.

More than ever before, there has been a significant interest recently in developing novel delivery systems which will deliver peptide and protein drugs at a controlled rate

throughout extended lengths of time. Implantable drug delivery systems of biocompatible polymers, such as ethylene-vinyl acetate (EVAc) [4], cellulose acetate, and polymethylmethacrylate (PMMA) [5], have been studied. These implanted devices release drugs with zero order kinetics for an extended period which is desired. However, these devices must be surgically implanted and, in some cases, explanted. Consequent disadvantages of using these implants include patient discomfort, possibility of infection, and medical costs [6].

Smart polymers are macromolecules that display a dramatic physicochemical change in response to small changes in their environment. Smart polymer-based injectable formulations are easy to prepare and form implants at the site of injection upon administration. Smart polymers can be classified according to the external stimulus they respond to (i.e. temperature, pH, solvent, magnetic field, ions, and pressure). The development of smart polymer-based injectable drug delivery systems has gained attention over the past few years. This interest has been sparked by the advantages these delivery systems possess, which include ease of application, localized delivery for a site-specific action, prolonged delivery periods, decreased body drug dosage with concurrent reduction in possible undesirable effects common to most forms of systemic delivery, the non toxic degradability, and improved patient compliance and comfort.

Instability of proteins can be classified into two types, physical and chemical instability. Physical instability does not involve any covalent modification. It refers to changes in higher order structures (secondary, tertiary, and quaternary). This includes denaturation (unfolding of a protein molecule), adsorption to surfaces, aggregation, and precipitation. Chemical instability involves the formation or breaking of covalent bond, leading to new chemical entities. Examples on such pathways are chain cleavage (hydrolysis),

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deamidation, isomerization, racemization, and oxidation. Different polymers have been shown to stabilize proteins. Stabilization can be due to surface activity, preferential exclusion, steric hindrance of protein-protein interactions, and increased viscosity limiting protein structural movement. Hydroxypropyl- β -cyclodextrin has shown to prevent the thermal and interfacial denaturation of porcine growth hormone [7] and to stabilize bovine insulin [8]. Proteins can be stabilized by polymers through multiple electrostatic interactions like sulfated polymers. Also polymers may inhibit chemical degradation in proteins. For example, dextran was shown to inhibit the metal oxidization of human relaxin [9].

In this review, we will outline the different major strategies that have been developed and patented for the use of smart polymers in delivering peptide and protein drugs. The coverage of the patents in this review is not encyclopedic. A few select examples have been cited to emphasize certain points.

2. TEMPERATURE SENSITIVE POLYMERS

Many polymers show abrupt changes in their solubility as a function of environmental temperature. This property was employed to develop aqueous solutions of these polymers which undergo sol-gel transition in response to temperature changes. At lower critical solution temperature (LCST), the interaction forces (hydrogen bonding) between water molecules and polymer become unfavorable compared to polymer-polymer and water-water interaction and phase separation occurs as the polymer dehydrates. Consequently, aqueous polymer solutions display low viscosity at ambient temperature but exhibit a sharp increase in viscosity following a small temperature rise, forming a semi-solid gel at body temperature. One major advantage these formulations offer is the absence of organic solvents. However, these systems show high initial burst effect which has been attributed to the shrinkage in the volume which exudes a large amount of the encapsulated drug [10]. Among the polymers that show thermosensitive characters are poly (*N*-isopropylacrylamide) (PNIPAAm), poly (ethylene oxide)-poly (propylene oxide)-poly (ethylene oxide) triblock copolymers (PEO-PPO-PEO), poly (ethylene glycol)-poly (lactic acid)-poly (ethylene glycol) triblocks (PEG-PLA-PEG).

Triblock PEO-PPO-PEO copolymers (Pluronics[®] or Poxamers[®]) show gelation at body temperature at concentrations greater than 15% (w/w) [11]. However, such concentrations led to notable toxicity and increased cholesterol and triglycerides levels in plasma after intraperitoneal injection in rats [12]. Pluronic[®] F127 has been reported to be the least toxic and therefore it has been the most widely used in drug delivery studies [13]. Pluronic[®] F127 has been employed for the sustained release of insulin from insulin-PLGA nanoparticles following subcutaneous administration in rats [14]. Pluronics[®] gels release drugs slower than solutions but the release period rarely exceeds few days. Therefore, Pluronics[®] have been mainly studied for short-term therapies like fertility control [15], pain management [16], and infection treatment [17].

Biodegradable polymers of ABA and BAB triblock copolymers were introduced by MacroMed, where A denotes the hydrophobic polyester block and B denotes the hydrophilic poly (ethylene glycol) block. The leading product of MacroMed, ReGel[®], employs 23% (w/w) ABA copolymer of poly(lactide-co-glycolide)-poly(ethylene glycol)-poly(lactide-co-glycolide) (PLGA-PEG-PLGA) in phosphate buffer saline. OncoGel[®], which is composed of paclitaxel at a concentration of 6 mg/g ReGel[®], is used for injection into tumors and releases paclitaxel over a period of six weeks. ReGel[®] was investigated for the delivery of protein-based drugs such as insulin and rh-GH [18, 19].

In 1997, Cha *et al.* [20] introduced a patent that describes an injectable block copolymer drug delivery system. The copolymer is made up of a hydrophobic block (A) selected from the group consisting of poly (hydroxy acids) and poly (ethylene carbonate), and a hydrophilic block (B) comprising polyethylene glycol. These formulations were studied for the controlled release of human calcitonin, platelet-derived growth factor (PDGF-B), and insulin. In a series of patents, Rathi and Zentner [21-23] described the use of water soluble ABA or BAB type triblock polymers for drug delivery. The polymer is made up of a major amount of a hydrophobic polymer (PLGA or PLA) as the A block and a minor amount of a hydrophilic PEG block. The release rate of the drug may be altered by changing various parameters such as adjusting hydrophobic/hydrophilic content, molecular weight, polymer concentration, and polydispersity of the triblock copolymer. Insulin showed a controlled release over a week from 28% PLGA-PEG-PLGA formulation. The sustained delivery of leptin from diblock AB and triblock ABA thermosensitive polymers was described by Shah [24]. In this patent, The AB diblock was made of A polymer block comprising 74% by weight and B polymer block comprising 26% by weight of the diblock copolymer. It was shown that released leptin had increased stability and solubility under physiologic conditions. This can be explained by the amphiphilic nature of the used polymers.

Another diblock thermogelling biodegradable polymer was described by Jeong *et al.* [25]. The blocks are linked of a general structure (A.sub.n.B), star-shaped copolymer, where n is greater than 2; A and B blocks are selected from a group consisting of a polyethylene glycol block and a biodegradable polyester block. Aqueous solutions of 21-25% (w/w) PEG-g-PLGA copolymers exhibited sol-to-gel transition in response to an increase in temperature. A preliminary *in vivo* study in rats showed that the PLGA-g-PEG gel was still present at the injection site after 2 months. In a recent patent, Jeong and Gutowska [26] prepared a thermogelling biodegradable aqueous polymer solution that comprises a biocompatible block (PEG) and a biodegradable polypeptide block. The enzymatically biodegradable polypeptide block was disclosed since peptide bonds are more stable against hydrolysis than ester bonds, thus, providing superior storage stability. The degradation of the polypeptide system is accelerated only by proteolytic enzymes, so degradation occurs only after *in vivo* administration. Furthermore, PLGA/PLA polymer systems generate lactic acid and glycolic acid during degradation, which may result in the degradation of acid sensitive drugs. The degradation products of polypeptides are neutral amino

acids which means that there is no significant pH drop during polymer degradation.

The use of polymeric hollow particles for peptide delivery has been described [27]. The particles have a shell made of an amphiphilic triblock ABA or BAB copolymer. These low permeability particles with a reversibly permeable shell expand and permeability increases in response to temperature. Surfaces of the particles can be modified with specific ligands allowing for molecular recognition which directs particles to specific targets. The use of a mixture of at least two types of triblock copolymers has also been described [28]. The triblock copolymers are made of a hydrophobic biodegradable polyester block and a hydrophilic polyethylene glycol block. The drug release can be modulated by various parameters such as the hydrophobic/hydrophilic component contents, polymer concentration, molecular weights, and weight ratios of the triblock copolymer contents in the mixture. It was shown that the release of insulin was modulated by mixing two different triblock copolymer solutions.

Multiblock biodegradable macromers including at least four polymeric blocks were introduced in a series of patents [29-32]. At least two polymeric blocks are hydrophobic and at least one is hydrophilic, and the macromer should include a crosslinkable group. The block copolymers may be linear, star-shaped, or branched. The macromers include one or more regions that have thermo-responsive properties such as poloxamers. The composition of the macromers can be altered to produce hydrogels with desired drug delivery properties. Macromers were synthesized with a wide range of hydrophobicity. Formulations prepared with PPO (polypropylene oxide) content of more than 60% showed a significant ability to retard water permeation and drug release.

A dual phase polymeric delivery system for releasing bioactive agents was patented [33]. This patent comprises a continuous biocompatible gel phase and a discontinuous particulate phase comprising defined microparticles. The microparticles are entrained within a polymeric gel matrix. The bioactive agent may be contained in the microparticle phase alone or in both the microparticle and the gel matrix. In addition, a second bioactive agent may be loaded in the microparticle phase and/or the gel matrix. It was shown that the burst release (>80%) of the growth hormone loaded microparticles was reduced to less than 20% when those microparticles were suspended in thermosensitive polyester-based systems. Similar results were obtained in the *in vivo* studies in rats. Dual phase formulations showed lower initial burst release and the plasma hGH levels were maintained above the therapeutic level for more than 4 weeks. Moreover, the bioavailability of the dual phase group was 50% higher than that of the microparticles group.

Self-assembling colloidal carriers for use in protein delivery has been introduced [34]. This invention comprises an active agent and a polyol ester having a linear polyol containing six or more hydroxyl groups as a central backbone and biodegradable hydroxy carboxylic ester groups attached to the central backbone. Preferred backbones include vinylpyrrolidones, vinylamines, vinylimidazoles, vinylpyridines, vinyl sulfonic acids, and vinyl phosphonic

acids. The compositions exhibit temperature dependent self-assembly properties. A pH-dependent controlled release of bovine serum albumin from this formulation was shown. Another patent that describes *in situ* gelation of pectic substances was introduced by Ni *et al.* [35]. Pectin and its derivatives can be isolated from Aloe vera leaves. The liquid pectin undergoes gelation at the site of administration. It was shown that the addition of other polymers such as hydroxyethyl cellulose (0.45% w/v), carboxymethyl cellulose (0.45% w/v), or sodium alginate (0.45% w/v) enhances the gelation properties of pectin. This formulation was used to deliver basic fibroblast growth factor (bFGF). The protein was mixed with Aloe pectin in physiological saline and injected subcutaneously into the abdominal region of mice. The results showed that the cell number was more than 2 times higher in the formulation group than in the control. An increase in blood vessel formation surrounding the gel was also observed.

3. PHASE SENSITIVE POLYMERS

The phase sensitive injectable polymeric systems have many advantages such as ease of manufacture, less stressful manufacturing conditions for sensitive drug molecules, and high loading capacity [1,36]. This approach was first introduced by Dunn [37] and it employs a water insoluble biodegradable polymer, such as poly(D,L-lactide), poly(D,L-lactide-co-glycolide) and poly(D,L-lactide-co-ε-caprolactone), dissolved in a pharmaceutically acceptable solvent to which a drug is added forming a solution or suspension. After injection of the formulation into the body, the water-miscible organic solvent dissipates and water penetrates into the organic phase. This causes phase separation and precipitation of the polymer forming a depot at the site of injection [38,39]. Organic solvents used include hydrophobic solvents, such as triacetin, ethyl acetate, and benzyl benzoate; and hydrophilic solvents, such as *N*-methyl-2-pyrrolidone (NMP), tetraglycol, and glycofurool.

An example of phase sensitive polymer-based product is Eligard[®], which employs Atrigel[®] as a drug carrier, and it is used for management of advanced prostate cancer. It contains an LHRH agonist leuprolide acetate and PLGA 75/25 dissolved in *N*-methyl-2-pyrrolidone (NMP) [40-42]. This system led to suppression of testosterone levels in dogs for approximately 91 days [43]. Clinical studies demonstrated that a depot containing 22.5 mg leuprolide maintained an effective suppression of testosterone below the medical castration level of 50 ng/dl [44]. Another product that utilizes Atrigel[®] technology is Atridox[®] (8.5% doxycycline), which is used for treatment of chronic periodontitis. Atridox[®] is a subgingival controlled-release formulation that releases doxycycline over a week.

Finding non-toxic and biocompatible solvents is a major challenge in developing phase sensitive formulations. The solvents used must be biocompatible to avoid severe tissue irritation or necrosis at site of administration. There is a controversy about the use of dimethyl sulfoxide (DMSO) and NMP in these systems. There are extensive toxicity data for oral, intraperitoneal, and intravenous administration of these solvents, but not for subcutaneous or intramuscular use [45]. Interestingly, it has been shown that NMP and DMSO are myotoxic when injected intramuscularly into Spargue-

Dawley rats [46]. Phase sensitive polymer systems are also known to display a high initial release of the drug followed by a more sustained release profile. The initial high burst release follows the Higuchi square root of time relationship [46, 47]. However, some investigators were able to totally eliminate the initial burst for at least 1 week, using low molecular weight PLGA in DMSO or NMP [48].

Phase sensitive polymeric formulations have been investigated for the controlled release of several proteins. One of the problems is the burst release during the few hours following injection into the body. This could be due to the lag time between the injection of the delivery system and the formation of the gel depot. Burst release of lysozyme and insulin was modulated in benzyl benzoate/benzyl alcohol (BB/BA) solvent systems by controlling the composition of the solvent system and polymer concentration [36, 49]. Increasing the proportion of the hydrophilic solvent led to an increase in the burst release. Addition of a hydrophilic solvent increases the affinity between the water and polymer solution and increases water influx rate. Another study showed that the release of proteins can be modulated by using different polymer concentrations, altering the solvent composition, and using proteins of different molecular weights [50]. In addition, protein stability is a major problem with these formulations. Proteins undergo denaturation in the presence of organic solvents. As a result, proteins should be added as dry powders containing different stabilizers just before administrations.

There are several patents which use biodegradable polymers dissolved in suitable organic solvents for drug delivery [37, 51-57]. Examples of biodegradable polymers are polylactides, polyglycolides, polycaprolactones, polyanhydrides, polyamides, poly(amino acids), polyvinylpyrrolidone, polyethylene glycol, polyhydroxy cellulose, chitin, and chitosan. The solubility of the polymers in the different solvents will vary depending on their crystallinity, hydrophilicity, hydrogen bonding, and molecular weight. Therefore, the concentration of a polymer dissolved in the various solvents will differ depending on polymer type and its molecular weight. Normally, the high molecular weight polymers will tend to solidify faster and give higher solution viscosities than the low molecular weight polymers.

One concept of using emulsifying agents in phase sensitive formulations has been explored [58]. When the emulsifying agent is mixed with the viscous gel, the emulsifying agent forms a separate phase of microscopic-size dispersed droplets. The continuous phase is formed of the polymer and the solvent. The bioactive agent can be dissolved or dispersed in either the continuous phase or the droplet phase. In the latter case, the bioactive agent can be mixed with the emulsifying agent just prior to the time of use. This can be advantageous in terms of stability for many drugs. In addition, since the drug will remain in the droplet phase, it is possible to select an emulsifying agent that provides further stability than the gel formulation alone. Preferred emulsifying agents include alcohols, propylene glycol, ethylene glycol, glycerol, and water. A recent patent [59] describes the use of these phase sensitive systems for the controlled delivery of drugs. It was shown that the release of lysozyme and hGH were controlled over different

periods of time. Similar results were observed with interferon -2a, interferon -2b, consensus interferon, methionine human growth hormone, des-phenylalanine human growth hormone, carboplatin, and insulin-like growth factor.

4. pH-SENSITIVE POLYMERS

A macromolecule that dissociates to give polymeric ions after dissolving in water or another ionizing solvent is termed polyelectrolyte. Because of the repulsion between charges on the polymeric chains, the chains are expanded when ionized in a suitable solvent. However, if the solvent prevents ionization of the polyelectrolyte, the chains exist in a compact, folded state. If the polyelectrolyte's chains are hydrophobic when unionized, in a poor solvent they collapse into globules and precipitate from solution. The interplay between hydrophobic surface energy and electrostatic repulsion between charges dictates the behavior of the polyelectrolytes [60]. Since the degree of ionization of weak polyelectrolytes is controlled by the pH value and ionic composition of an aqueous medium, smart polymers dramatically change conformation in response to minute changes in the pH of aqueous environment. All the pH-sensitive polymers contain pendant acidic or basic groups that either accept or donate protons in response to environmental pH [61]. Swelling of hydrogel increases as the external pH increases in the case of weakly acidic (anionic) groups, but decreases if polymer contains weakly basic (cationic) groups.

Most of anionic pH-sensitive polymers are based on polyacrylic acid (PAA) (Carbopol[®]) or its derivatives. In addition to PAA, polymethacrylic acid (PMAA), poly(ethylene imine), poly(L-lysine), and poly(*N,N*-dimethyl aminoethyl methacrylamide) have also been explored for use in drug delivery [62]. Microparticles prepared of poly(methacrylic acid-*g*-ethylene glycol) P(MAA-*g*-EG) loaded with insulin exhibited unique pH-responsive characteristics in which interpolymer complexes were formed in acidic media and dissociated in neutral / basic environments [63]. Consequently, insulin release from the gel was significantly retarded in acidic media while rapid release occurred under neutral/ basic conditions. Copolymer networks of poly(methacrylic acid) grafted with poly(ethylene glycol) with reversible pH-dependent swelling behavior, due to the formation of interpolymer complexes between protonated pendant acid groups and the etheric groups on the graft chains, has been developed [64]. Gels containing equimolar amounts of MAA/EG exhibited lesser swelling at lower pH. The pH of the swelling solution affected the average network mesh size. *In vitro* release of insulin from P(MAA-*g*-EG) gels containing PEG grafts indicated a significant release of insulin as the gel decomplexed.

Composite membranes from nanoparticles of poly(*N*-isopropylacrylamide-co-methacrylic acid) of various NIPAAm:MAA ratios dispersed in a matrix of a hydrophobic polymer have been investigated [65]. *N*-Benzoyl-L-tyrosine ethyl ester HCl, momany peptide, Leuprolide, vitamin B₁₂, insulin, and lysozyme were used as model solutes. Permeability of the solutes across the membranes increased with increasing temperature or particle concentration, while decreased with increasing pH and molecular

size of the solutes. Membranes containing higher MAA content showed greater pH responsiveness.

A pH-responsive polymeric system was introduced by Himmelstein and Baustian [66]. The system exhibits sol-gel transition over physiologically compatible pH ranges. The pH sensitive polyacids used include carboxylic acid-containing polymers such as monomers of acrylic acid and methacrylic acid. A related drug delivery system, that employs a copolymer hydrogel made of 90% methyl methacrylate and 10% acrylic acid, was describe [67, 68]. The polymer matrix swells when the environment reaches pH 8.5, releasing the active drug. Another invention that is based on acrylic acid derivatives as pH-responsive polymers was introduced by Palasis [69]. The polymers used are derivatized to contain moieties that are cationically charged at a pH below their pKa values. Thus, they attract negatively charged therapeutic agents. At pH values above their pKa values, the polymers become predominantly uncharged and substantially release the therapeutic agents. Bae and Park described pH-sensitive polymers containing sulfonamide groups, which show changes in swellability and solubility depending on pH [70]. The pH-sensitive polymer may be linear, grafted copolymer, or hydrogel.

A different type of pH-responsive polymeric systems relates to sustained release formulations using alginate gel beads or particles [71]. This approach involves the formation of sustained-release gels by the co-precipitation of alginate gel beads with a biologically active agent. This can be advantageous as it provides high loading of the drug while achieving better protein stability. Precipitating agents include but are not limited to polyvalent metal ions. *In vivo* studies showed that rats injected with leptin bead formulations maintained a plasma concentration of over 50 ng/ml for over 112 h in contrast to that of 12-18 h for the control animals.

Some polymers are known to respond to a combination of two or more stimuli. This provides an obvious advantage since it introduces more means of release profile control. Shah and Dai [72] described the use of pH/thermosensitive biodegradable hydrogels for the sustained delivery of biologically active agents. The hydrogels consist of a A-B diblock or ABA triblock copolymer of PLA or PLGA (block A) and PEG (block B), with ionizable functional groups on one or both ends of the polymer chains. *In vitro* release studies from 30% (w/w) solution mixture of hydroxyl-terminated and carboxylic acid-terminated PLGA-PEG-PLGA showed sustained release of Fc-leptin over a 7-10 days period of time. In a related patent, block and grafted copolymers that contain both a temperature sensitive polymer component and a pH-sensitive polymer component were introduced [73]. Temperature sensitive polymers include poly(*N*-isopropylacrylamide), PEO-PPO-PEO, and cellulose derivatives. The pH-sensitive polymer is a synthetic carboxylic acid-containing polymer and may be derived from polymerizable carboxylic acids, including acrylic acid, methacrylic acid and ethacrylic acid. Systems containing PVA-g-PAAc grafted copolymers showed significantly greater lysozyme release at pH 7.4 (PBS buffer) than at pH 5.0 (2-[*N*-morpholino]-ethanesulfonic acid (MES) buffer). The difference in release rate suggests that drug

diffusion from the gel plays an important role. In general, the greater the gel swelling, the greater the diffusion of the drug from the gel.

5. PHOTSENSITIVE POLYMERS

These systems are biocompatible, biodegradable, polymerizable, and at least partially water soluble macromers. The macromers include at least one water soluble region, at least one region which is biodegradable, and at least two free radical-polymerizable regions [74-77]. Macromers are polymerized by free radical initiators under ultraviolet light, visible light excitation, or thermal energy. The core water soluble region can consist of PEG, poly (vinyl alcohol), PEO-PPO, polysaccharides such as hyaluronic acid, or proteins such as albumin. The biodegradable region may be polymers made up from polylactic acid, polyglycolic acid, poly(anhydrides), poly(amino acids), and polylactones. Preferred polymerizable regions include acrylates, diacrylates, methacrylates, or other biologically accepted photopolymerizable groups. Initiators that can be used for generation of free radicals include ethyl eosin, acetophenone derivatives, or camphorquinone. *In vitro* study showed that the release of bovine serum albumin was relatively steady over more than a month from photosensitive formulations. In another study, the monomer PEG-dl-lactic acid-diacrylate was dissolved in PBS. Lysozyme was released from the polymer over 8 days, with the maximum rate of release occurring within the first two days. Other potential applications of these systems include temporary protection of tissue surfaces, sealing tissues together, and preventing the attachment of cells to tissue surfaces.

CURRENT & FUTURE DEVELOPMENTS

Over the last two decades, many strategies dealing with the controlled release of drugs from polymeric systems have been investigated. Many polymer-based delivery systems have progressed to the clinic and in some cases to the commercial production. Nevertheless, most of these delivery systems are still in the development stage and they are still far from ideal. For example, these polymeric systems are insensitive to the changing metabolic state in patients. Other considerations include biocompatibility, response time to stimuli, burst release, optimum release rate simulation, and formulation issues and challenges. We reviewed several patents which describe the use of smart polymers for the controlled delivery of peptide and protein drugs. These systems emerged as a potential approach for the controlled release of bioactive agents. The huge variation in these systems compositions and responsiveness render them ideal for delivery of different compounds with different requirements. However, tremendous research opportunities remain to be explored to find ideal delivery systems, which are biodegradable, biocompatible, easy to administer, and release the incorporated agent in a chemically and conformationally stable form for longer duration.

REFERENCES

- [1] Cleland JL, Daugherty A, Mrsny R. Emerging protein delivery methods. *Curr Opin Biotech* 2001; 12: 212-219.
- [2] Lee VH. Enzymatic barriers to peptide and protein absorption. *Crit Rev Ther Drug Carrier Sys* 1988; 5: 69-97.

- [3] Mingda B, Singh J. Degradation of luteinizing hormone releasing hormone in buccal, liver, nasal and skin tissues. *Int J Pharm* 1998; 175: 269-273.
- [4] Hsieh DS, Smith N, Chien YW. Subcutaneous controlled delivery of estradiol by Compudose implants: *In vitro* and *in vivo* evaluations. *Drug Dev Ind Pharm* 1987; 13: 2651-2666.
- [5] Kirkpatrick DK, Trachtenberg LS, Mangino PD, Von Fraunhofer JA, Seligson D. *In vitro* characteristics of tobramycin-PMMA beads: compressive strength and leaching. *Orthopedics* 1985; 8: 1130-33.
- [6] Hsieh DS, Rhine WD, Langer R. Zero-order controlled-release matrices for Micro- and macromolecules. *J Pharm Sci* 1983; 72: 17-22.
- [7] Charman SA, Mason ML, Charman WN. Techniques for assessing the effects of pharmaceutical excipients on the aggregation of porcine growth hormone. *Pharm Res* 1993; 10: 954-962.
- [8] Zhang J, Peng X, Jonas A, Jonas J. NMR study of the cold, heat, and pressure unfolding of ribonuclease A. *Biochemistry* 1995; 34:8631-8641.
- [9] Li S, Patapoff TW, Nguyen TH, Borchardt RT. Inhibitory effects of sugars and polyols on the metal-catalyzed oxidation of human relaxin. *J Pharm Sci* 1996; 85: 868-872.
- [10] Hatefi A, Amsden B. Biodegradable injectable *in situ* forming drug delivery systems. *J Control Release* 2002; 80: 9-28.
- [11] Bochot A, Fattel E, Gulik A, Couarraze G, Couvreur P. Liposomes dispersed within a thermosensitive gel: a new dosage form for ocular delivery of oligonucleotides. *Pharm Res* 1998; 15: 1364-69.
- [12] Wasan K, Subramanian R, Kwong M, Goldberg I, Wright T, Johnston T. Poloxamer 407-mediated alterations in the activities of enzymes regulating lipid metabolism in rats. *J Pharm Sci* 2003; 6: 189-197.
- [13] Laughlin R. The aqueous phase behavior of surfactants. London, Academic Press 1994.
- [14] Barichello JM, Morishita M, Takayama K, Nagai T. Absorption of insulin from Pluronic F-127 gels following subcutaneous administration in rats. *Int J Phama* 1999; 184: 189-198.
- [15] Wenzel J, Balaji K, Koushik K, et al. Pluronic F127 gel formulations of deslorelin and GnRH reduce drug degradation and sustain drug release and effect in cattle. *J Control Release* 2002; 85: 51-59.
- [16] Paaola A, Kilpelainen I, Yliruusi J, Rosenberg P. Controlled release injectable liposomal gel of ibuprofen for epidural analgesia. *Int J Pharm* 2000; 199: 85-93.
- [17] Zhang L, Parsons D, Navarre C, Kompella U. Development and *in-vitro* evaluation of sustained release poloxamer 407 (P407) gel formulations of ceftiofur. *J Control. Release* 2002; 85: 73-81.
- [18] Zentner G, Rathi R, Shih C, et al. Biodegradable block copolymers for drug delivery of proteins and water-insoluble drugs. *J Control Release* 2001; 72: 203-215.
- [19] Kim YJ, Choi S, Koh JJ, Lee M, Ko KS, Kim SW. Controlled release of insulin from injectable biodegradable triblock copolymer. *Pharm Res* 2001; 18: 548-50.
- *[20] Cha, Y., Choi, Y.K., Bae, Y.H.: US5702717 (1997).
- *[21] Rathi, R.C., Zentner, G.M.: US6004573 (1999).
- [22] Rathi, R.C., Zentner, G.M., Jeong, B.: US20006117949 (2000).
- [23] Rathi, R.C., Zentner, G.M., Jeong, B.: US20016201072 (2001).
- [24] Shah, S.: US20036541033 (2003).
- *[25] Jeong, B.M., Gutowska, A.: US20056841617 (2005).
- [26] Jeong, B.M., Gutowska, A.: US20067087244 (2006).
- [27] Meier, W., Sauer, M.: US20036616946 (2003).
- [28] Piao, A.Z., Shih, C.: US20067018645 (2006).
- [29] Pathak, C.P., Barman, S., Philbrook, C.M., Sawhney, A.S., Coury, A.J., Avila, L.Z., Kieras, M.T.: US20016201065 (2001).
- [30] Pathak, C.P., Barman, S., Philbrook, C.M., Sawhney, A.S., Coury, A.J., Avila, L.Z., Kieras, M.T.: US20026410645 (2002).
- [31] Pathak, C.P., Barman, S., Philbrook, C.M., Sawhney, A.S., Coury, A.J., Avila, L.Z., Kieras, M.T.: US20036639014 (2003).
- [32] Pathak, C.P., Barman, S., Philbrook, C.M., Sawhney, A.S., Coury, A.J., Avila, L.Z., Kieras, M.T.: US20056923986 (2005).
- *[33] Shih, C., Zentner, G.M.: US20016287588 (2001).
- [34] Kissel, T., Breitenbach, A., Jung, T., Kamm, W.: US20036616944 (2003).
- [35] Ni, Y., Yates, K.M.: US20046777000 (2004).
- [36] Singh S, Singh J. Controlled release of a model protein lysozyme from phase sensitive smart polymer systems. *Intl J Phama* 2004; 271:189-96.
- *[37] Dunn, R.L., English, J.P., Cowsar, D.R., Vanderbilt, D.P.: US4938763 (1990).
- [38] Ravivarapu HB, Moyer KL, Dunn RL. Sustained activity and release of leuprolide acetate from an *in situ* forming polymeric implant. *AAPS PharmSciTech* 2000 1(1).
- [39] Eliaz RE, Kost J. Characterization of a polymeric PLGA-injectable implant delivery system for the controlled release of proteins. *J Biomed Mat Res* 2000; 50: 388-396.
- [40] Dunn, R.L., Garrett, J.S., Ravivarapu, H., Chandershekar, B.L.: US20036565874 (2003).
- [41] Dunn, R.L., Garrett, J.S., Ravivarapu, H., Chandershekar, B.L.: US20046773714 (2004).
- [42] Sartor O. Leuprolide acetate in a novel sustained-release delivery system. *Urology* 2003; 61(2 Suppl 1): 25-31.
- [43] Ravivarapu HB, Moyer KL, Dunn RL. Sustained suppression of pituitary- gonadal axis with an injectable, *in situ* forming implant of leuprolide acetate. *J Pharm Sci* 2000; 89: 732-741.
- [44] Chu FM, Jayson M, Dineen MK, Perez R, Harkaway R, Tyler RC. A clinical study of 22.5 mg La-2550: a new subcutaneous depot delivery system for leuprolide acetate for the treatment of prostate cancer. *J Urology* 2002; 168: 1199-1203.
- [45] Royals MA, Fujita SM, Yewey GL, Rodriguez J, Schultheiss PC, Dunn RL. Biocompatibility of a biodegradable *in situ* forming implant system in rhesus monkeys. *J Biomed Mater Res* 1999; 45: 231-239.
- [46] Kranz H, Brazeau GA, Napaporn J, Martin RL, Millard W, Bodmeier R. Myotoxicity studies of injectable biodegradable *in situ* forming drug delivery systems. *Int J Pharm* 2001; 212: 11-18.
- [47] Higuchi T. Mechanism of sustained-action medication. *J Pharm Sci* 1963; 52: 1145-49.
- [48] Lambert WJ, Peck KD. Development of an *in situ* forming biodegradable poly-lactide-coglycolide system for the controlled release of proteins. *J Control Release* 1995; 33: 189-95.
- [49] Kang F, Singh J. *In vitro* release of insulin and biocompatibility of *in situ* forming gel systems. *Int J Pharm* 2005; 304: 83-90.
- [50] Al-Tahami K, Meyer A, Singh J. Poly lactic acid based injectable delivery systems for controlled release of a model protein, lysozyme. *Pharm Dev Technol* 2006; 11: 79-86.
- [51] Dunn, R.L., English, J.P., Cowsar, D.R., Vanderbilt, D.P.: US5278201 (1994).
- [52] Dunn, R.L., English, J.P., Cowsar, D.R., Vanderbilt, D.P.: US5278202 (1994).
- [53] Dunn, R.L., English, J.P., Cowsar, D.R., Vanderbilt, D.P.: US5340849 (1994).
- [54] Dunn, R.L., English, J.P., Cowsar, D.R., Vanderbilt, D.P.: US5733950 (1998).
- [55] Dunn, R.L., English, J.P., Cowsar, D.R., Vanderbilt, D.P.: US5739176 (1998).
- [56] Dunn, R.L., English, J.P., Cowsar, D.R., Vanderbilt, D.P.: US5990194 (1999).
- [57] Dunn, R.L., English, J.P., Cowsar, D.R., Vanderbilt, D.P.: USRE200237950 (2002).
- *[58] Brodbeck, K.J., Shen, T.T.: US20016331311 (2001).
- [59] Brodbeck, K.J., Gaynor-Duarte, A.T., Shen, T.T.: US20046673767 (2004).
- [60] Bromberg L. Intelligent polyelectrolytes and gels in oral drug delivery. *Curr Pharm Biotechnol* 2003; 4: 339-49.
- [61] Qiu Y, Park K. Environment-sensitive hydrogels for drug delivery. *Adv Drug Del Rev* 2001; 53: 321-39.
- [62] Jeong B, Gutowska A. Lessons from nature: stimuli-responsive polymers and their biomedical applications. *Trends Biotechnol* 2002; 20: 305-11.
- [63] Morishita M, Lowman AM, Takayama K, Nagai T, Peppas NA. Elucidation of the mechanism of incorporation of insulin in controlled release systems based on complexation polymers. *J Cont Rel* 2002; 81: 25-32.
- [64] Peppas NA. Devices based on intelligent biopolymers for oral protein delivery. *Int J Pharm* 2004; 277: 11-17.
- [65] Zhang K, Wu XY. Temperature and pH-responsive polymeric composite membranes for controlled delivery of proteins and peptides. *Biomaterials* 2004; 22: 5281-91.
- [66] Himmelstein, K.J., Baustian, C.L.: US5599534 (1997).

- *[67] Batich, C.D., Cohen, M.S., Foster, K., Toreki, I.W.: US5788687 (1998).
- [68] Batich, C.D., Cohen, M.S., Foster, K., Toreki, I.W.: US20016306422 (2001).
- [69] Palasis, M.: US20036506408 (2003).
- [70] Bae, Y.H., Park, S.Y.: US20006103865 (2000).
- [71] Goldenberg, M.S., Beekman, A.C.: US20036656508 (2003).
- [72] Shah, S., Dai, W.: US20026451346 (2002).
- [73] Chen, G., Hoffman, A.S.: US20026486213 (2002).

- [74] Hubbell, J.A., Pathak, C.P., Sawhney, A.S., Desai, N.P., Hill-West, J.L.: US5986043 (1999).
- [75] Hubbell, J.A., Pathak, C.P., Sawhney, A.S., Desai, N.P., Hill-West, J.L.: US20006060582 (2000).
- [76] Hubbell, J.A., Pathak, C.P., Sawhney, A.S., Desai, N.P., Hill-West, J.L.: US20016306922 (2001).
- [77] Hubbell, J.A., Pathak, C.P., Sawhney, A.S., Desai, N.P., Hill-West, J.L.: US20036602975 (2003).