10

Controlled Release and Nanotechnology

Tania Betancourt, Amber Doiron, Kimberly A. Homan,
and Lisa Brannon-Peppas

Introduction

Nanosized controlled release systems for drug delivery are segregated into several categories including polymeric nanoparticles, liposomes, solid lipid nanoparticles, polymeric micelles, and dendrimers. This topic is extensive and as such is only briefly reviewed here. More detailed information may be found in more focused chapters of this book. With this in mind, this chapter will provide an overview of nanoparticulate systems, followed by some of the more interesting opportunities and applications of nanotechnology in controlled release: metal–organic systems, nanotubes, responsive systems, and personal care products.

The use of a drug as a therapeutic agent is often a delicate balance between therapeutic efficacy and detrimental side effects including toxicity. The control of the amount of drug delivered over time and the spatial localization of that delivery are paramount in overcoming the challenges of providing optimal therapy. This challenge drives the design of various drug delivery strategies that strive to revolutionize the way drugs exert their actions. Much of this attention has focused on nanoparticles due to their small size, relatively high surface area, influence on biodistribution, ability to make drugs available for intravascular delivery, their stabilizing effect on therapeutic agents, and the capability of sustaining release of the agent (Mainardes and Silva 2004). All these elements ultimately lead to more effective delivery of the active agent to a desired physiological or pathophysiological location.

Modification of the nanocarrier composition largely controls the release of the active agent from the carrier. This can be accomplished by using various types of polymers or lipids, changing the molecular weight of those components, or changing the surface characteristics such as by crosslinking or adding a separate component like poly (ethylene glycol). In addition, more specific modifications can be made in order to achieve the optimal controlled drug release from the nanodevice. The following reviews the major classes of nanoscale drug delivery devices.

M.M. de Villiers et al. (eds.), *Nanotechnology in Drug Delivery*, DOI: 10.1007/978-0-387-77667-5_10,

Types of Nanoscale Drug Delivery Devices

Structure and Behavior of Polymeric Nanoparticles

Polymeric nanoparticles have been investigated as drug delivery devices for several decades due to their ability to carry a wide variety of drugs or genes and sustain delivery for an extended period of time. Nanoparticles are submicron-sized polymeric colloidal spheres that can entrap an active agent within the polymer matrix, or the active agent can be adsorbed or conjugated to the outside of the particle. The term nanoparticle encompasses both nanocapsules and nanospheres. Nanocapsules have a core–shell morphology with the active agent trapped within the core by the polymeric shell. The matrix structure of a nanosphere serves to entrap the drug molecules, or alternatively, the drug is conjugated at the surface of the particle (Brannon-Peppas 1995; Soppimath and Aminabhavi 2002; Mainardes and Silva 2004).

Many techniques have been used successfully to prepare nanoparticles and are generally stratified into (i) methods that use preformed polymer and (ii) methods involving the polymerization of monomers. These methods include but are not limited to the following: emulsion–solvent evaporation, salting out, production using supercritical fluid technology, phase separation, and in situ polymerization (Jain 2000; Soppimath, Aminabhavi et al. 2001).

Various classes of polymers have been used in drug delivery applications and are stratified into biodegradable polymers and non-biodegradable polymers. Biodegradable nanoparticles have received much attention because they do not require further intervention, i.e., removal, after being placed into the body. Depending on the formulation type, the drug is released by one or a combination of several mechanisms: desorption of adsorbed drug, diffusion through the polymer matrix, diffusion through the polymeric membrane shell in the case of nanocapsules, and polymer degradation and erosion (Uhrich et al. 1999; Jain 2000; Soppimath, Aminabhavi et al. 2001; Mainardes and Silva 2004). These mechanisms are influenced by the rate of degradation of the material, and the choice of polymer largely dictates the controlled release properties of the system (Uhrich Cannizzaro et al. 1999; Jain 2000; Soppimath, Aminabhavi et al. 2001). Many factors outside of the kinetics of degradation must be considered for a polymer used in a drug delivery device including the difficulty of preparation, biocompatibility, favorable interactions with the active agent, and mechanical properties (Uhrich, Cannizzaro et al. 1999; Jain 2000).

Nanocapsules and nanospheres differ in their release profiles due to the nature of the containment of the active agent. Nanospheres encapsulate the drug molecules within the matrix of polymer in a uniform distribution. The release of the drug from the matrix occurs through diffusion as well as erosion of the matrix itself. If diffusion occurs more quickly than degradation, then the process is diffusion dependent, otherwise the process of degradation is highly influential (Niwa, Takeuchi et al. 1993). An initial burst release is observed due to the presence of drug near or adsorbed to the large surface area of the nanoparticle. After the burst effect, diffusion

largely controls the release leading to an exponential delayed release rate. Matrix-type nanoparticles usually exhibit first-order kinetics (Fresta, Puglisi et al. 1995; Radwan 1995).

Conversely, nanocapsules have a reservoir-like morphology and exhibit release profiles as such. The drug is contained in the core and must diffuse through the polymer shell in order to be released. This morphology theoretically leads to zero-order kinetics of release. It has been shown experimentally that drug release from nanocapsules can occur by either partitioning of the drug or diffusion across the polymer coating (Calvo, VilaJato et al. 1996; Lu, Bei et al. 1999). Additionally, it has been shown that the method of drug incorporation, conjugation or adsorption, greatly affects the release profile with adsorption leading to higher burst release and a quicker overall release (Soppimath, Aminabhavi et al. 2001).

Polymers Used in Nanoscale Release Systems

Various synthetic polymers have been used in drug delivery devices including poly(esters), poly(ortho esters), poly(anhydrides), poly(amides), and phosphorus-containing polymers, and many naturally derived polymers such as chitosan, dextran, and gelatin have also been extensively researched. Several of the most common polymers used in nanoscale devices are reviewed here.

Poly(esters)

The most studied and best characterized class of polymers for controlled release is the poly(esters). One of the most common polymers used in nanoparticle drug delivery approaches is poly(lactic-*co*-glycolic acid) (PLGA) due to its degradation properties, biocompatibility, and the fact that it is very well characterized (Jain 2000). PLGA degrades in an aqueous environment through the hydrolysis of the backbone ester linkages (Brannon-Peppas 1995; Uhrich, Cannizzaro et al. 1999; Jain 2000). The polymeric device based on PLGA degrades through bulk erosion at a uniform rate throughout the matrix (Jain 2000). The degradation process is self-catalyzed as the number of terminal carboxylic acid groups rises with increasing chain scission, and the acids catalyze the hydrolysis. The degradation is highly dependent on the ratio of lactide to glycolide moieties as lactide is more hydrophobic and reduces the rate of degradation (Jain 2000; Mainardes and Silva 2004). Also, important factors in the degradation process are the degree of crystallinity, the molecular weight, and the glass transition temperature of the polymer (Jain 2000).

PLGA has been used to encapsulate a myriad of drugs and genes for controlled delivery applications for many diseases or other applications, and only a few are mentioned here. One popular area for the application of PLGA nanoparticles is in the treatment of cancer. Paclitaxel is a drug used in cancer treatment that causes cell death by inhibiting cell division (Brannon-Peppas and Blanchette 2004). Fonseca et al. loaded paclitaxel into nanoparticles (< 200 nm) with near 100% efficiency using an interfacial deposition method. The loaded PLGA nanoparticles released

approximately half of their payload within the first 24 hours and had a slowing release rate over the subsequent 4 days. Significant losses in viability were shown in the human small lung cancer cell line NCI-H69 with exposure to as little as 0.025 μg/ml paclitaxel-loaded nanoparticles (Fonseca, Simões et al. 2002). Doxorubicin is a widely used cancer drug that impedes nucleic acid synthesis, yet is also known to have various systemic side effects (Brannon-Peppas and Blanchette 2004). Nanoparticles prepared from PLGA–doxorubicin conjugates of about 200 nm in diameter suppressed tumor growth for 12 days after a single administration (Yoo, Lee et al. 2000). The hydrophilic cancer drug 5-fluorouracil has been encapsulated in PLGA/O-CMC (*O*-carboxmethyl-chitosan) nanoparticles along with antisense EGFR (epidermal growth factor receptor) plasmids by Hu and colleagues in a novel approach to combine chemotherapy and gene therapy for the treatment of cancer. Encapsulation efficiencies of both agents in the 90th percentile were achieved, and release of 5-fluorouracil was prolonged for up to 3 weeks. In glioma cells, the nanoparticles caused cytotoxicity upward of 90%, and decreased EGFR expression confirmed transfection of the cells (Hu, Chang et al. 2005).

Polymeric PLGA nanoparticles have also been used as a method to prolong release of and control distribution of antiproliferative drugs at the sight of balloon injury in a dog atherosclerosis model (Guzman, Labhasetwar et al. 1996; Labhasetwar, Song et al. 1998). Nanoparticles containing dexamethasone were delivered to the arterial wall and observed to penetrate the wall without additional modification. Within several days, systemic levels of the drug were undetectable, but nanoparticles were detected in the artery wall for up to 14 days. This is indicative of the ability of PLGA nanoparticles to control the release of drugs and be useful in sustaining release in a stent-like treatment without inducing systemic toxicity of these powerful drugs. Additional work has been accomplished in this area showing the promise of active targeting and the further utility of nanoparticles to prevent restenosis (Labhasetwar, Song et al. 1998; Lanza, Yu et al. 2002). These studies show the utility of nanoparticles to sustain and spatially concentrate the delivery of an active agent in treatment of restenosis (Caves and Chaikof 2006).

Poly(ortho esters)

Devices degrading through bulk erosion have an undesirable release profile for many applications, and the need for a device controlling release solely through hydrolysis of chains at the surface of the device effected the design of poly(ortho esters) (Uhrich, Cannizzaro et al. 1999). The release rates from devices composed of poly(ortho esters) can be controlled by including acidic or basic excipients into the matrix as its hydrolysis is acid catalyzed. This has been used in the release of 5-fluorouracil (Seymour, Duncan et al. 1994), tetracycline (Roskos, Fritzinger et al. 1995), and others (Uhrich, Cannizzaro et al. 1999). Additionally, the mechanical properties of these polymers can be tailored by choosing from the various diols available (Mainardes and Silva 2004).

Poly(anhydrides)

Poly(anhydrides) degrade by hydrolysis yet the polymer itself is hydrophobic in nature. These properties lead to surface erosion of the polymeric device and nearly zero-order release. The hydrolytic bond cleavage of poly(anhydrides) produces water-soluble products that in many cases are considered biocompatible. Poly(anhydrides) are most commonly produced through a melt-condensation polymerization. The most common polymers in this class are based on sebacic acid, *p*-(carboxyphenoxy)propane, and *p*–(carboxyphenoxy)hexane. Variations in monomer composition, such as hydrophobicity, influence the degradation rate of the polymeric device. The degradation can last from days to years depending on the composition (Uhrich, Cannizzaro et al. 1999).

The photosensitizer phthalocyanine was chemically incorporated into nanoparticles based on biodegradable poly(sebacic anhydride) by Fu and colleagues (Fu, Li et al. 2002) for cancer treatment through photodynamic therapy. The attachment of the phthalocyanines to the polymer in the nanoparticles impedes the tendency of the agent to aggregate and become less useful for photodynamic therapy. The average hydrodynamic radius of the nanoparticles was found to be 166 nm. The release of photosensitizer from the particles was degradation dependent, and the rate of degradation increased with pH and temperature. This colloidal system has the potential to be useful for the delivery and controlled release of photosensitizer for photodynamic therapy (Fu, Li et al. 2002). Many other types of poly(anhydrides) have been used in drug delivery applications in the nanoscale size range.

Chitosan

As opposed to the other materials mentioned above, chitosan is a naturally derived polysaccharide created by the deacetylation of chitin (Mainardes and Silva 2004). The advantageous properties of chitosan include its biocompatibility, positive charge, the abundance of amine groups available for crosslinking, ease of processing, mucoadhesiveness, and its degradation into amino sugars, which are all attractive for drug delivery applications (Agnihotri, Mallikarjuna et al. 2004; Mainardes and Silva 2004). Chitosan nanoparticles have been formulated by a variety of techniques including emulsion crosslinking, complex coacervation, emulsion droplet coalescence method, ionic gelation, ionotropic gelation, and the reverse micellar method. The molecular weight of the chitosan, its degree of deacetylation, the extent of crosslinking, and its interactions with the encapsulated molecule play a role in controlling the release of the therapeutic agent from the particle. Due to its charge, the pH of the release media also influences release from chitosan particles. Release from chitosan particles occurs through similar mechanisms as mentioned for other particles: desorption of surface-adhered drug, diffusion through a swollen rubbery polymer matrix, and release due to erosion. Release of drugs from surface layers of the matrix involves a large burst effect, but increasing the crosslinking density can reduce this effect (Agnihotri, Mallikarjuna et al. 2004). Diffusion out of the matrix occurs through a three-step process: diffusion of water into the matrix causing swelling, transition from glassy to rubbery polymer, and

diffusion of drug out of the matrix. The release follows a typical hydrogel release profile (Agnihotri, Mallikarjuna et al. 2004).

Chitosan nanoparticles of approximately 100 nm in diameter prepared by a microemulsion method have been used to encapsulate a doxorubicin–dextran conjugate. In a mouse model, tumor volume was reduced after four weekly injections of the nanoparticle formulation 40% more than in mice treated with the conjugate alone, and injection of drug alone had no effect over control conditions (Mitra, Gaur et al. 2001). As an adjuvant to another cancer therapy–neutron-capture therapy–gadopentetic acid (Gd-DTPA) has been loaded in chitosan nanoparticles formed by an emulsion droplet coalescence technique. Less than 2% of the Gd-DTPA was released over 7 days in PBS, but over 90% was released in plasma over 1 day. After an intratumoral injection in a mouse melanoma model, 92% of the Gd-DTPA was contained within the tumor site compared to only 1.2% of the Gd-DTPA injected in a non-nanoparticle formulation (Tokumitsu, Ichikawa et al. 1999). The Gd-DTPA chitosan nanoparticles have been shown to have a high affinity for uptake in several cell types, suggesting the mechanism for high retention in tumor (Shikata, Tokumitsu et al. 2002).

Gelatin

Gelatin is a naturally occurring biopolymer that is biocompatible and biodegradable. The polymer is obtained through heat-dissolution and partial hydrolysis of collagen obtained from animal connective tissues. It has been used for many years in pharmaceutical applications such as capsules and ointments as well as early nanoformulations (Zwiorek, Kloeckner et al. 2004; Verma, Sachin et al. 2005). Recently, gelatin nanoparticles made by a two-step desolvation process involving crosslinking of the polymer using gluteraldehyde have been used to entrap cycloheximide, a protein synthesis inhibitor used in cancer treatment. Cycloheximide was entrapped with 26% efficiency in nanoparticles of 168 nm diameter. The particles were stable in whole blood, and they showed anti-tumor activity in two breast cancer cell lines over a period of time. The release kinetics curve was interestingly biphasic, and release was relatively slow. The gelatin nanoparticles are reportedly a good candidate for biopharmaceutical delivery (Verma, Sachin et al. 2005). Zwiorek et al. produced gelatin nanoparticles by the same desolvation method as a carrier for plasmid DNA (Zwiorek, Kloeckner et al. 2004). The particles were cationized in order to have an electrostatic interaction with the DNA which bounds onto the surface of the particles. The nanoparticles showed little cytotoxic effect, and efficient gene transfection was exhibited by an exponential increase in gene expression in B16 F10 cells (Zwiorek, Kloeckner et al. 2004).

Other Structures for Nanoparticle Delivery Systems

Polymeric Micelles

Block copolymers have been used as the basis for drug delivery carriers due to their ampiphilic nature and ability to organize into concentric regions.

A polymeric micelle consists of a dense core region comprised of hydrophobic blocks and a region of more loosely packed hydrophilic blocks. Polymeric micelles are typically 20–100 nm in diameter, and polyethylene oxide (PEO) is often used as the hydrophilic block. Polymeric micelles have a low critical micelle concentration and as such have higher stability than low molecular weight surfactants and many liposome formulations. Micelles have a small size and small polydispersity due to their molecular organization. The hydrophilic shell has been shown to prevent immune recognition and increase circulation time in vivo, and many groups have investigated polymeric micelles for drug and gene delivery (Mainardes and Silva 2004). The stability has been increased even more by incorporating crosslinking into the preparation scheme. This additional step also affects the release of active agent from the carrier in a system-specific manner (O'Reilly, Hawker et al. 2006).

One polymer class popular for use as the dense core in polymer micelles is poly(ortho esters) due to their hydrophobic nature and favorable interactions with poorly soluble hydrophobic drugs. The value for the critical micelle concentration for these types of polymers is in the range of 10^{-4} g/l, which is low enough to insure stability upon injection in vivo. The entrapment efficiency of taxol has been shown to be approximately 40% using a PEG–poly(ortho ester)–PEG block copolymer micelle (Heller, Barr et al. 2002).

Liposomes and Lipid-Based Systems

Liposomes

Lipids are organic molecules that contain a hydrophilic head group and a hydrophobic chain region. Much like polymer micelles, lipids organize in water into aggregates called liposomes with the hydrophobic regions packed in a core and the hydrophilic heads freely interacting with the surrounding water. Although cationic lipids are the most predominant, anionic and neutral lipids are also investigated for use in drug and gene delivery. Lipid design is an increasingly important avenue of research as the controlled release applications of lipids grow (Bhattacharya and Bajaj 2005). Liposomes are classified on the basis of their size, which can range from several nanometers to microns, and the number of lipid bilayers. Both hydrophilic and hydrophobic drugs can be carried by liposomes depending on the lipid structure.

Liposomes have been under investigation for many decades and the number of drugs and genes investigated for controlled release with liposome formulations is very extensive. Liposomes have been investigated for use in cancer treatment (Brannon-Peppas and Blanchette 2004), have been shown to reduce systemic side effects (Mainardes and Silva 2004), and have been researched for the delivery of proteins and nucleic acids (Mainardes and Silva 2004; Bhattacharya and Bajaj 2005). Like all other colloidal systems, liposomes suffer from various shortcomings including interactions with lipoproteins, having a high critical micelle concentration that limits stability, and limited availability of inexpensive pharmaceutical grade lipid (Muller, Mader et al. 2000; Mainardes and Silva 2004; Bhattacharya and Bajaj 2005).

Solid Lipid Nanoparticles

Solid lipid nanoparticles are a matrix device composed of solid lipid in the size range of 50–1000 nm. They are prepared through a variety of techniques including hot or cold high-pressure homogenization, microemulsion, and precipitation. Drugs such as paclitaxel, gadolinium complexes, prednisone, and many others have been incorporated into solid lipid nanoparticles. The drug loading and subsequent release is dependent on the solubility of the drug in the melted lipid, the miscibility of drug melt with lipid melt, the structure of the solid lipid matrix, and the polymorphic state of lipid material. Solid lipid nanoparticles can be modified in order to provide either a large burst release or a slow uniform release rate for a period of several weeks. The production parameters are influential on the release profile but size is not a significant factor in release (Muller, Mader et al. 2000).

Dendrimers

Dendrimers are a newer class of polymeric drug delivery devices with a unique macromolecular structure. The three-dimensional complexes are produced in an iterative sequence of reaction steps leading to generations of branches organized around an inner core. The hierarchical synthesis of these complexes lends itself to finely controlled size, composition, and reactivity. Poly(amidoamine) dendrimers were the first constructed and characterized, but dozens of other dendrimer types have been investigated to date (Mainardes and Silva 2004). Dendrimers are formed either by divergent or convergent methods, each having its own advantages and disadvantages. The behavior and characteristics of dendrimers can differ greatly from their linear counterparts. Due to their step-wise synthesis, the polydispersity of dendrimers is quite low, contributing to their utility as drug delivery devices. The scaffold provides an ideal platform for drug molecules that does not depend on thermodynamics or physical factors. The choice of polymer used in the dendritic system plays heavily into its utility as a drug carrier owing to the association between the polymer and drug molecule.

The drug indomethacin was loaded (11 wt%) into dendritic micelles composed of a hydrophobic Fréchet-type dendrimer and a shell of hydrophilic poly(ethylene glycol) by Fréchet et al. The release of the drug from the complex was much slower than that of the same drug from a cellulose membrane: all drug was released over 25 hours as opposed to 4 hours with the cellulose (Liu, Kono et al. 2000). 5-Fluorouracil has been incorporated into poly(amidoamine) dendrimers augmented with mPEG-500. The complexation between the hydrophilic drug and dendrimer occurred with incubation. In vitro release from the PEGylated dendrimers occurred over 6 days, whereas the non-PEGylated formulations released all drug over 1 day. This same relationship was true in studies in albino rats with PEGylated formulations showing prolonged release, without producing any significant hematological instability (Bhadra, Bhadra et al. 2003). Instead of entrapment within the dendrimer, drugs may also be electrostatically or covalently bound to the surface of the dendrimer. Owing to the highly branched and functionalized structure of dendrimers,

oftentimes there are large numbers of ionizable groups at the surface that are available for complexation (D'Emanuele and Attwood 2005).

Viral Vectors

Viral vectors have been proposed as efficient gene delivery devices due to their evolutionary advantage over man-made colloidal systems for transfection of cells. Synthetic or modified viruses carry the therapeutic gene in their capsid, being able to protect it until it reaches its intended target. Many exciting strides have been made in this field, yet many hurdles remain to make the device safe and viable in vivo (Mainardes and Silva 2004).

In addition to the various examples listed above, other classes of nanoscale-controlled delivery devices exist including protein-based delivery devices, magnetic nanoparticles, inorganic nanoparticles, and others.

Controlled Release from Metal–Organic Nanoparticles and Complexes

Bioinorganic chemistry is an expanding field showing great promise for applications in medicine, both for novel drug formulations and drug delivery vehicles. As mentioned in the first section of this chapter, metallic and metal oxide nanostructures have gained significant attention in recent years. In fact, elucidating the mechanisms by which metals interact naturally in the body (Guo and Sadler 1999) has allowed researchers to devise new metallodrugs involving such metals as vanadium and zinc for insulin-mimetic solutions (Sakurai, Katoh et al. 2006), platinum for use in the very popular anti-tumor drug cisplatin (van Zutphen and Reedijk 2005; Bontha, Kabanov et al. 2006), and selenium for use in the anti-inflammatory and anti-viral drug ebselen and its derivatives (Wojtowicz, Kloc et al. 2004; Bhabak and Mugesh 2007), just to name a few.

Metallodrugs are clearly an area of growing research, but they are not the focus of our discussion here. Rather, this section reports on the budding field of metal–organic complexes as an alternative to strictly organic constructs used for controlled drug and gene delivery. The incorporation of metal nanoshells into organic frameworks to provide remote control release of drugs is also presented. Finally, where controlled discharge of metal ions is desired, their release through organic nanoconstructs is briefly considered.

Metal–Organic Hybrids in Controlled Release

Polymers represent the most extensively studied class of materials for controlled release. Since most of these organic systems operate to control drug release via diffusion, controlling pore size in these systems is critical and challenging (Horcajada, Serre et al. 2006). As a result of these challenges, many researchers searched for other materials with well-defined, tunable porous structures and found zeolites to be the answer. Zeolites are minerals consisting of metals or metalloid components in a crystalline

framework with nanopore sizes in the range of 2–20Å (Smaihi, Gavilan et al. 2004). When AlO_4 is part of the zeolite, it adds an ion-exchange component that can be used to increase the loading efficiency of charged compounds or drugs into the porous structure (Zhang, Kim et al. 2006). In a proof of concept study, Zhang et al. loaded zeolite Y with a common herbicide, paraquat, and then modified the pore size of the zeolite by functionalizing the surface with 1,1,3,3-tetramethyldisilazane (TMDS) (Zhang, Kim et al. 2006). Functionalizing the surface effectively reduced the pore size after paraquat loading, which allowed for maximum loading efficiencies to be conserved in conjunction with a subsequent controlled release of paraquat. Results proved that undamaged paraquat released in aqueous solution by Na^+ exchange from functionalized zeolite Y over a 7-day period, versus the 20-min release exhibited by the unfunctionalized zeolite Y (Zhang, Kim et al. 2006). Although this study used an herbicide, the concept of loading and exhibiting controlled release can be extrapolated to small, charged drugs and the use of zeolites for controlled release applications was clearly demonstrated.

Researchers have recently produced stable colloidal suspensions of zeolite nanoparticles. Functionalized nanocrystalline zeolite particles hold promise as future controlled release capsules or chemical sensors (MacLachlan, Manners et al. 2000). One group has developed a procedure for making template-free zeolite nanoparticles with the possibility of altering surface functional groups. Producing zeolites with different functionalities allows for a range of future interactions with targeting agents and other drugs (Smaihi, Gavilan et al. 2004).

A common zeolite used for controlled release applications is crystalline aluminosilicate. By changing the ratio of Si/Al in this zeolite framework, overall zeolite pore size and ion-exchange properties can be tuned (Horcajada, Marquez-Alvarez et al. 2006). Many claim that the uniform pore size and structure of zeolites lend to more even drug loading, and thus, more predictable controlled release behavior. A pictorial representation of a Y-type zeolite with cubo-octahedral sodalite cages is shown in Figure 10.1. One study proved using release of ibuprofen from an aluminosilicate Y-type zeolite, that after an initial period of diffusion-controlled release

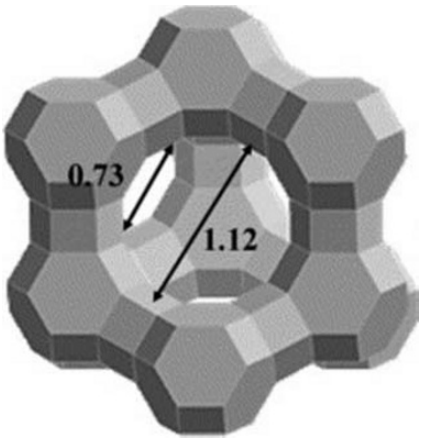

Figure 10.1 Building units of zeolite Y showing the dimensions of the supercage and windows (nm). Modified from Horcajada, Marquez-Alvarez et al. (2006).

which remained similar for all models, further discharge of ibuprofen from the zeolite could be controlled by varying the Al content in the zeolite framework (the hydrophobic nature of the zeolite increases as Al content decreases). They showed that dealuminating the zeolite structure increased the rate of hydrophobic drug release, like ibuprofen, but only up until a specified Si/Al ratio of 22. At higher ratios, van der Waals forces between the drug and the zeolite framework slowed release (Horcajada, Marquez-Alvarez et al. 2006). Controlled release through zeolite structures is gaining popularity for a host of applications in medical and agricultural sciences and is expected to continue populating the literature in coming years.

Metal Nanoshells for Remote Controlled Release

Metal nanoshells generally have dielectric core–shell morphologies where a silica core is surrounded by a thin layer of metal. The surrounding metal could be Ag, Au, Pt, or any other bulk metal, but the Au-layered shells are the most widely studied. The beauty of the core–shell design is that the plasmon optical resonance peak of elemental gold can be shifted from the visible to the near infrared region (NIR) by varying the core diameter and metal shell thickness (Lin, Lewinski et al. 2005). This shift to the NIR is critical considering that light in the NIR region can penetrate deep (2–3 cm) into biological tissue (Steinbrink, Wabnitz et al. 2001). Thus, metal nanoshells have the potential, once injected into the body, to respond thermally to light shone externally on the body. The numerous applications of these novel metal nanoshells in the realm of imaging are covered elsewhere in this book, so here we focus on the use of these nanoshells in triggering drug release.

One interesting method of effecting a pulsatile drug release involves incorporation of these metal nanoshells via entrapment into a temperature-sensitive polymer–drug matrix. One example includes the widely studied thermosensitive polymer *N*-isopropylacrylamide (NIPAAm). When NIPAAm is copolymerized with acrylamide and formed as a hydrogel, the hydrogel exhibits a lower critical solution temperature (LCST) slightly above body temperature (Hirsch, Gobin et al. 2006). Once this LCST is reached, the polymer matrix exhibits a drastic phase change and collapses, as shown in Figure 10.2. During the collapse, water and much of the encapsulated drug are expelled. Sershen et al. proved that these hydrogel systems with entrapped gold nanoshells can in fact be used for pulsatile protein release in response to a pulsed NIR laser light (Sershen, Westcott et al. 2000). Another example includes the work of Owens and Peppas who created temperature-sensitive inter-penetrating polymer networks (IPN) using acrylamides and acrylic acids formed as hydrogels which exhibited an upper critical solution temperature (UCST) (Owens III and Peppas 2006). In their case, the polymer remained compact under the UCST and exhibited a phase change by expanding above the UCST. Again, gold nanoshells were incorporated into these IPNs via entrapment, and pulsatile release of encapsulated drug was observed in correlation with pulsed external laser light excitation.

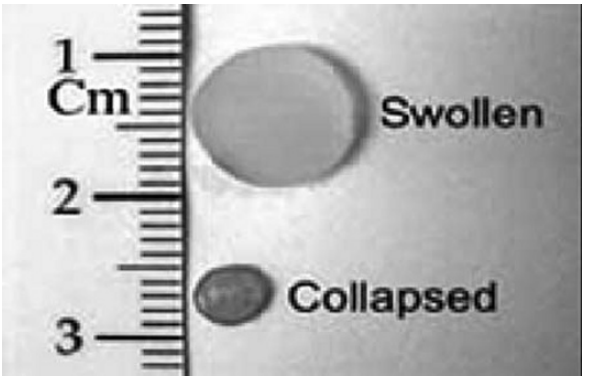

Figure 10.2 NIPAAm-co-acrylamide hydrogels shown in the swollen state (below LCST) and collapsed state (above LCST). Adopted from Hirsch, Gobin et al. (2006).

Remote control of drug release through polymeric systems can also be achieved using magnetically responsive metal particles. In the 1980s the first triggered release using magnetic particles was forged by Kost, Edelmen, and Langer at the Massachusetts Institute of Technology (Kost, Noecker et al. 1985; Edelman, Brown et al. 1987; Kost, Wolfrum et al. 1987). In these first works, cylindrical magnets (1.4 mm) were placed inside polymeric matrices with encapsulated bovine serum albumin (BSA). Upon induction of an oscillating magnetic field, the release rates of BSA significantly increased. Upon removal of the field, release rates returned to baseline (diffusion-controlled release). They later proved that externally triggered delivery of insulin to diabetic rats was possible using similar methods (Kost, Wolfrum et al. 1987). In these cases, the motion of the magnets induced a mechanical deformation of the matrix, which in turn allowed for the increased drug release (Edelman, Fiorino et al. 1992).

A more recent attempt to build a magnetically responsive construct was made by Gaponik et al. at the University of Munich. They created a multifunctional polymeric microcapsule which houses both CdTe semiconductor nanocrystals and magnetic Fe_3O_4 nanoparticles (Gaponik, Radtchenko et al. 2004). The CdTe nanocrystals are meant to serve as luminescent markers while the magnetic oxide nanoparticles aid in targeting of the microcapsule. The general nature of their method for building the construct allows for drug encapsulation in future studies. In that case, the role of magnetic oxide in the microcapsule could be used not just for targeting, but for triggered drug release by employing similar methods used by Langer and inducing oscillating magnetic fields to spark increased drug release.

Magnetic iron oxides nanoparticles are also being used to enhance gene delivery. Plank et al. from the Technical University Munich have coined the term magnetofection for their science of magnetically induced transfection (Plank, Schillinger et al. 2003). The general method is that superparamagnetic iron oxide particles are surface treated with polyelectrolytic coating. This coating allows for the nanoparticles' salt-induced colloidal aggregation with viral and non-viral gene vectors. Applied magnetic gradients during transfection experiments using these new constructs in vitro showed dramatically reduced transfection times (Plank, Scherer et al. 2003). Classical carriers act via diffusion to deliver genes and can take several hours to transfect what the magnetofection method attained in

10 min (Plank, Schillinger et al. 2003). Effectively, the use of these iron oxide nanoparticles and induced magnetic field is enhancing the interaction between target cells and gene vectors, effecting the delivery of the gene in a timely manner. In trying to elucidate the mechanism of magnetofection, some theorize that endocytic uptake of genes on the cell surface is not sped up by the magnetic forces, instead the magnetic forces simply serve to accelerate sedimentation of magnetofectins on the cell surface (Huth, Lausier et al. 2004).

Controlled Release of Metal Ions Through Organic Nanoconstructs

Some metals such as silver and copper have been known to exhibit antibacterial properties. Considering these biocidal properties, silver have been used in formulations to treat or prevent infection in postoperative scenarios or cases such as burn victims. In these instances, the mode of action for "drug" delivery is the controlled release of the silver ions into the wound which subsequently interacts with bacterial DNA, preventing replication. Several researchers have created intriguing conjugates of silver to polymeric materials and nanoconstructs (Balogh, Swanson et al. 2001; Bromberg, Buxton et al. 2001; Abo El Ola, Kotek et al. 2004; Kumar, Howdle et al. 2005; Isab and Wazeer 2006; Rhim, Hong et al. 2006). Others have validated the use of silver nanoparticle impregnation methods as a way to reduce postimplantation infection risk (Karlov, Khlusov et al. 2002; Sambhy, MacBride et al. 2006).

Dendrimer–silver complexes represent one example of these interesting silver–polymer conjugates produced on the nanoscale. In one study, poly (amidoamine) (PAMAM) dendrimers were surface-modified to contain immobilized silver ions in stabile, silver domains (Balogh, Swanson et al. 2001). Using standard agar overlay methods, Balogh and collaborator's silver–PAMAM dendrimers exhibited significant antimicrobial activity against three bacterial strains: *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and *Escherichia coli*. Most studies of silver ion release in biological tissue show decreased antimicrobial efficacy of the silver in the presence of chloride or sulfate ions, mostly because their interaction with silver forms insoluble complexes (Schierholz, Lucas et al. 1998; Brett 2006). Silver–PAMAM complexes, however, maintain their antimicrobial activity in the presence of sulfate or chloride ions. Balogh et al. attributed this apparent continued silver activity to that fact that "macroscopically, the silver remained conjugated to the dendrimer in the form of ions, stabile metallic silver clusters or silver compounds." Since the dendrimer itself is soluble and the silver ions remain active while attached to the polymer complex, sulfate and chloride ions in the media are no longer a factor, and the movement of silver ions which impart antibacterial properties is only limited by the diffusion of the dendrimer itself.

Nanotubes

Carbon nanotubes can be thought of as tubular nanoscale particles that can potentially be used as delivery systems for imaging and therapeutic agents through conjugation or other techniques. Numerous studies have

shown that nanotubes can readily enter cells for intracellular delivery of active agents (Kam and Dai 2005; Wu, Wieckowski et al. 2005). Although studies of in vivo biodistribution of carbon nanotubes have not observed toxicity (Singh, Pantarotto et al. 2006), the actual properties of nanotubes are highly dependent on their functionalization, water solubility, size, etc., and need to be further studied for the numerous nanotube-based drug delivery systems that have been developed to date.

Functionalization of carbon nanotube surfaces is necessary to confer water solubility to these systems and for conjugation of therapeutic agents. Georgakilas et al. have reported on the orthogonal functionalization of carbon nanotubes via 1,3-dipolar cycloaddition reaction that allows selective conjugation of N-protected amino-terminated tether molecules to the walls of the tubes (Georgakilas, Kordatos et al. 2002; Georgakilas, Tagmatarchis et al. 2002). Further derivatization of surface carboxylic acid groups formed by acid-mediated oxidation of nanotubes permits simultaneous conjugation of two different therapeutic molecules.

Pastorin et al. have recently reported on the use of various protection methods for selective conjugation of therapeutic and imaging agents to multifunctional carbon nanotubes derivatized by the above-mentioned methods (Pastorin, Wu et al. 2006). One scheme utilizes *tert*-butyloxycarbonyl (Boc) and benzyloxycarbonyl as protecting groups of the conjugated amino-terminated tether groups. These protecting groups require treatment with strong acids for removal of the protecting groups, and are consequently not appropriate for conjugation of labile molecules. The second method is based on the protection of the amino groups by Boc and mono-phthalimide (Pht), respectively. While the Boc group is still removed by acid treatment, Pht is removed with hydrazine in alcohol at room temperature. Utilizing the second protection scheme, carbon nanotubes were successfully conjugated to fluorescein isothiocyanate and methotrexate for fluorescence detection and in vitro therapeutic evaluation, respectively (Pastorin, Wu et al. 2006). This group was able to show that the functionalized carbon nanotubes were readily internalized by human Jurtak T lymphocytes in a dose-dependent manner with confocal microscopy (Pastorin, Wu et al. 2006).

Similar techniques were utilized for conjugation of fluorescein and the antifungal antibiotic amphotericin B (AmB) to carbon nanotubes (Wu, Wieckowski et al. 2005). Conjugation of AmB to carbon nanotubes was found to significantly reduce toxicity of this drug to human Jurtak T lymphoma cells even after 16 hours or exposure (Wu, Wieckowski et al. 2005). On the other hand, AmB was found to be more effective against three fungi species when conjugated to carbon nanotubes than in the free form, possibly because of higher drug solubility, higher payload, and the prevention of AmB aggregation which commonly occurs in solution. Additionally, this group observed maximum nanotube uptake into the cells after just 1 hour of incubation. Further studies suggested that endocytosis was not involved in the rapid uptake of the nanotubes (Wu, Wieckowski et al. 2005).

Nanotubes have emerged as nanomaterials with high potential for application in drug delivery and biosensing. Further research is needed for gaining full understanding of the benefits and limitations of these

systems for biomedical applications. The combination of mechanical, electrical, and structural properties of carbon nanotubes will surely be exploited in the near future for the development of complex systems with very specific functionalities.

Responsive Drug Delivery Systems

Responsive drug delivery systems, as their name implies, are those that are able to act in response to a trigger, be it an external signal or changes in the surrounding environment (Tirelli 2006). The triggered response could include dissolution, precipitation, degradation, swelling, collapsing, change in hydrophilic/hydrophobic balance, phase separation, and shape alteration, among other conformational changes (Schmaljohann 2006). Systems that respond to external trigger commonly combine metals and polymers, as was discussed earlier in this chapter. In this section we focus on polymeric drug delivery systems able to recognize and modulate the delivery of a drug based on localized changes in temperature, pH, or concentration of oxidizing molecules.

Temperature-Sensitive Nanoparticles

Poly(*N*-isopropylacrylamide) (PNIPAAm) has been extensively used for the formulation of temperature-sensitive drug delivery systems because it exhibits a lower critical solution temperature (LCST) of about 30–34°C (Schmaljohann 2006). Below the LCST the polymer is soluble in water, while above this temperature the polymer becomes insoluble. This behavior can be utilized for controlling the delivery of active agents from temperature-sensitive drug delivery systems such as core–shell micelles of copolymers containing PNIPAAm and a hydrophobic polymer. Above the LCST, the micelles deform as the PNIPAAm, initially present in the nanoparticle shell in contact with the aqueous environment, becomes insoluble and disrupts the equilibrium of the core–shell configuration. If the LCST of a given temperature-sensitive polymer is higher than physiological temperature of 37°C, micelles of this polymer will remain stable until the local temperature of the target pathological tissue is raised above the LCST by external heating. For materials that have an LCST lower than normal body temperature, their thermoresponsive behavior can also be exploited for delivery of drugs to regions of low temperature such as hypoxic tissue (Patton and Palmer 2005). It is important to note that the LCST of polymers such as PNIMPAAm can be increased or decreased upon conjugation to a hydrophobic or hydrophilic copolymer, respectively (Schmaljohann 2006). For a more extensive review on temperature-sensitive systems readers are referred to a review by Dirk Schmaljohann (Schmaljohann 2006).

One example of temperature-responsive drug delivery systems based on PNIPAAm consists of self-assembled micelles of amphiphilic Y-shaped copolymers of poly(undecylenic acid) and PNIPAAm, or P(UA-Y-NIPAAm), as shown in Figure 10.3 (Li, Zhang et al. 2006). These micelles presented a very low critical micelle concentration of 20 μg/ml and a LCST

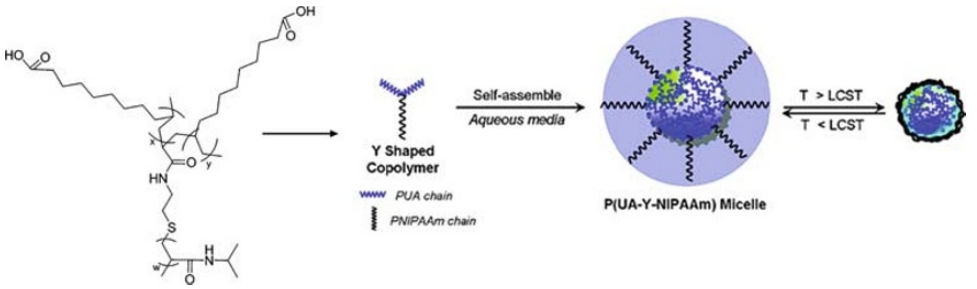

Figure 10.3 Schematic of temperature-sensitive micelles self-assembled in aqueous solutions from Y-shaped copolymers of poly(undecylenic acid) (PUA) and poly(*N*-isopropylacrylamide) (PNIPAAm). Reproduced with permission from Li, Zhang, et al. (2006). (*See* Color Plate 15)

of 31°C. Above this temperature the PNIPAAm shell of the micelles becomes hydrophobic and deforms, thus leading to the rapid release of encapsulated drugs, as shown for the anti-inflammatory drug prednisone acetate. In vitro tests revealed that this novel copolymer was biocompatible at concentrations as high as 1 mg/ml in 3T3 fibroblasts.

Another example of a thermoresponsive drug delivery system was reported by Nakayama et al. and consists of biodegradable polymeric micelles of the hydrophilic copolymer of PNIPAAm-poly(dimethylacrylamide) conjugated to the hydrophobic polymers poly(D,L-lactic acid) (PLA), poly(ε-caprolactone) (PCL), or PLA–PCL (Nakayama, Okano et al. 2006). These systems presented a LCST of about 40°C, which would enable external triggering of drug release. Interestingly, only micelles that had PLA–PCL copolymer in the hydrophobic core resulted in thermoresponsive character. Evaluation of this system with the drug doxorubicin proved a highly thermoresponsive release behavior, with the drug slowly diffusing out of the micelles at body temperature but exhibiting a high release rate 5°C above this temperature (Nakayama, Okano et al. 2006).

pH-Responsive Nanosystems

Polymers and drug delivery systems able to undergo conformation changes depending on the acidity of the surrounding environment have a number of important applications in nanomedicine. For example, these nanocarriers can selectively deliver chemotherapeutic agents at the site of a tumor as a result of the lower pH found in the tumor interstitium (Wike-Hooley, Haveman et al. 1984; Vaupel, Kallinowski et al. 1989), while sparing the rest of the body from the toxic drug (Schmaljohann 2006). Additionally, these systems can be designed to delay release of a specific drug until after the nanocarrier is endocytosed by a target cell and exposed to the lower pH of the endolysosomal compartments (Schmaljohann 2006). Some systems by means of their pH responsiveness are able to escape lysosomes upon configuration changes that allow them to interact with the organelle membrane (Panyam, Zhou et al. 2002). Finally, pH-responsive systems have also been extensively studied for applications in oral drug delivery (Schmaljohann 2006). Responsive nanocarriers can protect labile drugs

from the acid environment of the stomach while promoting their absorption in the more neutral small intestine. Examples of recently developed pH-responsive nanocarriers are reviewed here.

Nanoparticles of poly(beta-amino ester) (PbAE) modified with poloxamers, triblock copolymers of poly(ethylene oxide)–poly(propylene oxide)–poly(ethylene oxide), were designed for the delivery of hydrophobic drugs to the acidic environment of tumors and intracellular acidic organelles (Potineni, Lynn et al. 2003; Shenoy, Little et al. 2005). Studies demonstrated that the pH-responsive PbAE nanoparticles successfully delivered the chemotherapeutic drug paclitaxel to SKOV-3 ovarian cancer cells in vitro, and led to higher paclitaxel accumulation at the tumor than when administered as free drug in solution or in pH-insensitive pluronic-modified poly(ε-caprolactone) (PCL) nanoparticles in vivo (Shenoy, Little et al. 2005; Shenoy, Little et al. 2005). Further in vivo studies revealed that paclitaxel-loaded PbAE nanoparticles resulted in greater therapeutic efficacy than the free drug and paclitaxel PCL in mice xenografts of ovarian cancer, and did not result in systemic toxicity as judged from body weight losses or blood cell counts (Devalapally, Shenoy et al. 2007).

Another pH-responsive nanoparticle design is based on block copolymers of poly[2-(*N*,*N*-diethylamino)ethyl methacrylate] (PDEA) and poly (ethylene glycol) (PEG) (Xu, Van Kirk et al. 2006). This copolymer formed approximately 80 nm core–shell nanoparticles with a pH-responsive core and a PEG-dense shell. These pH-responsive nanoparticles were designed in such a way that they would release the drug in a very short period of time upon being endocytosed by target cancer cells and consequently being exposed to the acidic conditions of the lysosomal compartments. Such rapid release is achieved by the ability of the PDEA–PEG nanoparticles to become soluble in the aqueous biological environment when the pH drops below about 6. PDEA–PEG nanoparticles loaded with cisplatin were used to evaluate their potential to overcome multidrug resistance in SKOV-3 ovarian cancer cells in vitro and in vivo in nude mice xenografts (Xu, Van Kirk et al. 2006). Results showed that PDEA–PEG nanoparticles were in fact internalized by cells into lysosomes and caused significantly higher cellular growth inhibition than free cisplatin. In vivo, this system was observed to lower the number of blood vessels and increase the number of apoptotic cells in tumors.

Block ionic complexes (BIC) formed by ionic interactions between hydrophilic block copolymers containing ionic and nonionic regions with an oppositely charged molecule have been recently proposed as responsive drug delivery systems because of their ability to undergo changes in response to the environmental conditions (Oh, Bronich et al. 2006). The core of BIC is formed by electrostatically bound polyion–counterion complexes. Active agents can be encapsulated within these systems via hydrophobic or electrostatic interactions. The composition and structure of these systems can be customized by the choice of block copolymer and counterion. One specific BIC system recently reported consists of copolymers of Pluronic grafted with poly(acrylic acid) (PAA) complexed with the cationic surfactant hexadecyltrimethylamonium bromide (HTAB) (Oh, Bronich et al. 2006). Formation of the complexes occurs upon mixing aqueous solutions of the copolymer and of the surfactant, and results in

particles of about 50–100 nm depending on the relative amounts of copolymer and surfactant. Similar to the size, the surface charge of the particles is highly dependent on the ratio of the constituents. These BICs were observed to respond to changes in salt concentration, pH, and temperature (Oh, Bronich et al. 2006). Specifically, increased salt concentration resulted in up to an eightfold increase in particle size up to a ceiling salt concentration above which the complexes completely dissociated. Increased pH led to decreased zeta potential, with a charge inversion from positive to negative at pH 6.0. This pH sensitivity can be effectively exploited for intracellular drug delivery through the endocytic pathway since the acidification of the endosomes would reverse the charge of the BIC and permit its interaction with the endosomal membrane.

An interesting drug delivery platform that utilizes pH-cleavable bonds to modify the surface of micelles and liposomes was recently proposed by Sawant et al. (Sawant, Hurley et al. 2006). By blending copolymers containing and lacking pH-cleavable bonds, this system is able to control the presentation or masking of functional moieties, such as targeting agents, according to the environmental pH, as can be seen in Figure 10.4. pH-cleavable polymers were made by linking poly(ethylene glycol) and phosphatidylethanolamine via a hydrazone bond (PEG–Hz–PE) (Sawant, Hurley et al. 2006). Nanocarriers were formulated with blends of low molecular weight pH-insensitive PEG–PE polymers conjugated to cell-penetrating peptide (TATp), high molecular weight pH-insensitive PEG–PE polymer conjugated to antibodies, and high molecular weight pH-sensitive PEG–Hz–PE. In such systems, the targeting antibody would be freely accessible on the surface of the nanoparticle shell and actively participate in targeting the nanocarriers to the diseased tissue, while the TATp moiety would be shielded by longer pH-sensitive PEG–Hz–PE chains. Once localized in the more-acidic tumor or inflamed interstitium, the medium-sized PEG chains would be cleaved off as a result of destabilization of the hydrazone bond, thus exposing the cell-penetrating agents and promoting internalization of the nanocarriers into the target cells. In vitro results showed that nanocarriers functionalized with monoclonal antimyosin antibody 2G4 bound specifically to the antigen myosin at all pH tested as expected since the antibody was present at the surface and was conjugated to the nanocarriers via a non-pH-sensitive polymer. However, internalization of nanocarriers with the cell-penetrating peptide bound to hidden PEG–PE molecules was increased only after the carriers had been exposed to acidic conditions which could remove the shielding PEG coat from the PEG–Hz–PE polymer (Sawant, Hurley et al. 2006).

Sajeesh and Sharma recently reported on the preparation of pH-responsive nanoparticles for oral delivery of proteins (Sajeesh and Sharma 2005). The nanoparticles were created spontaneously by ionic complexation of poly(methacrylic acid)–chitosan–poly(ethylene glycol) prepared by free radical polymerization in an aqueous environment. Loading of active agents was achieved by a diffusion filling method in which the nanoparticles are equilibrated in solutions of the agent, thus allowing the agent to partition into the polymeric system. Despite their irregular morphology, these nanoparticles displayed properties potentially useful for oral delivery of labile molecules. Release of model protein bovine serum albumin and

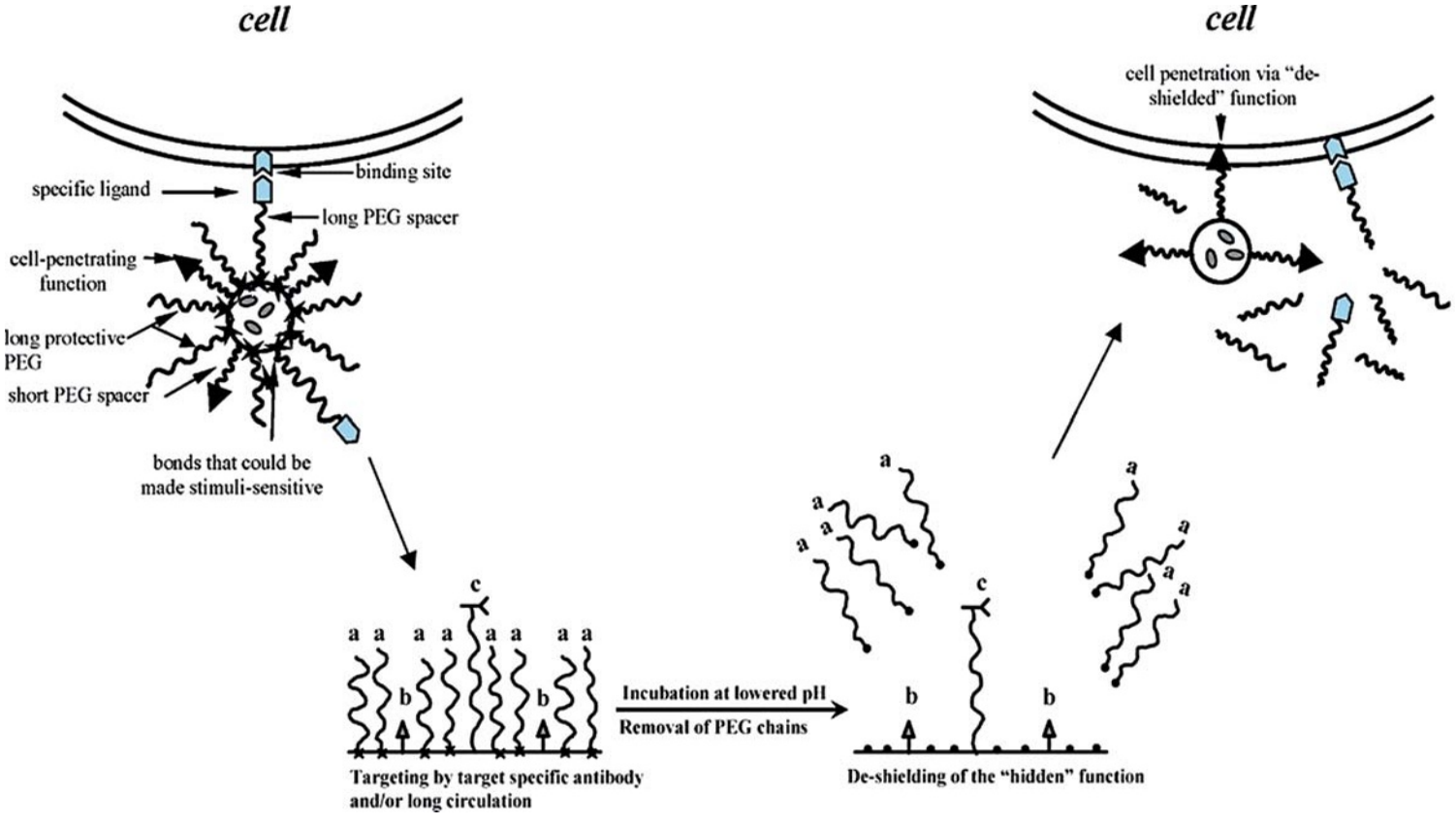

Figure 10.4 Diagram of pH-responsive nanocarriers that include permanent and cleavable surface polymeric chains that are used to modify the surface functionality at specific pathological conditions. Reproduced with permission from Sawant, Hurley, et al. (2006). (*See* Color Plate 16)

insulin was significantly delayed at a pH of 1.2, representative of the stomach, compared to a pH of 7.4. For example, only about 10% of loaded insulin was released from the nanoparticles at the acidic pH compared to about 90% at pH 7.4 (Sajeesh and Sharma 2005). The release of the protein at pH 7.4 was mostly complete by 4 hours, which is relevant to the residence time of the particles in the small intestine. The pH-dependent release behavior was attributed to the ability of the PMAA polymer to swell and shrink as the pH increases or decreases because of the protonation and deprotonation of carboxylic acid groups, respectively. When used for oral delivery, the nanoparticles would protect the active agent from degradation in the acidic environment of the stomach, but would release it at the small intestine as a result of nanoparticle swelling in the more neutral environment.

Nanocarriers with Combined Temperature- and pH-Responsive Properties

As described earlier, temperature-sensitive polymers such as PNIPAAm are most useful for drug delivery purposes if their LCST is higher than body temperature and their destabilization is triggered by artificially heating the target tissue to a temperature above the LCST with external sources. However, this limits their clinical applicability to the treatment of superficial malignancies due to low penetration of common heating sources. To overcome this problem, drug delivery systems incorporating both pH and temperature sensitivity have been investigated.

One such system consists of core–shell nanoparticles of poly(*N*-isopropylacrylamide-*co*-*N*,*N*-dimethylacrylamide-*co*-10-undecenoic acid) (PNIPAAm-co-DMAAm-co-UA) (Soppimath, Tan et al. 2005). In this system, UA makes up the hydrophobic and pH-sensitive core while PNIPAAm is present at the surface in contact with the aqueous environment. DMMAm, being a hydrophilic polymer, shifts the LCST of PNIPAAm to a higher temperature depending on the molar composition of the copolymers: the higher the molecular weight of the DMMAm section, the higher the LCST of the copolymer. This group was able to prepare a copolymer that had an LCST greater than body temperature (38.6°C) at physiological pH 7.4, and lower than body temperature (35.5°C) at a pH of 6.6 (Soppimath, Tan et al. 2005). Consequently, nanoparticles from this copolymer could easily deliver drugs selectively to the acidic interstitium of tumors while remaining stable and preventing toxicity to non-diseased tissue. The pH dependence of the LCST was attributed to the protonation and consequent decrease of hydrophobicity of the UA core with increasing pH, which resulted in increasing LCST for the copolymer. This system was evaluated with the chemotherapeutic drug doxorubicin and demonstrated significantly increased release rate, deformation, and precipitation at acidic pH and physiological temperature.

Wei et al reported on the preparation of temperature- and pH-responsive core–shell micelles that remain stable below the low critical solution temperature (LCST) of the polymer despite changes in pH, but become unstable and release the loaded drug in a pH-dependent manner above the LCST (Wei, Zhang et al. 2006). These micelles, which presented a critical micelle

concentration of 174 μg/ml, an LCST of 31°C, were prepared from the amphiphilic copolymer poly(10-undecenoic acid-*b*-*N*-isopropylacrylamide) (PUA-b-PNIPAAm), in which PUA represents the hydrophilic block while PNIPAAm represents the hydrophobic pH- and temperature-sensitive block (Wei, Zhang et al. 2006). Micelles loaded with the anti-inflammatory drug prednisone acetate displayed significantly increased release rates above the LCST due to deformation and precipitation at the higher temperature. In addition, lower environmental pH led to an initial faster release rate compared to that at normal physiological pH (7.4) when the temperature was maintained at 37°C which is above the LCST of the micelles. At 13°C, the drug release profile was not different at the two solution-pH tested.

Oxidation-Responsive Systems

Nanocarriers based on oxidation-sensitive materials have gained interest for application in drug delivery to inflamed tissue rich in oxidizing substances (Tirelli 2006). Polymeric vesicles of amphiphilic copolymer PEG–poly(propylene sulfide)–PEG (PEG–PPS–PEG) have been prepared for this application (Napoli, Valentini et al. 2005). PPS makes up the hydrophobic and oxidation-sensitive block. Upon exposure to an oxidative environment, PPS in the vesicles is transformed into hydrophilic poly (propylene sulfoxide) and poly(propylene sulfone), transforming the vesicles into worm-like and spherical micelles of progressively decreasing size that can ultimately be removed by glomerular filtration.

Another similar oxidation-responsive nanocarrier design consists of crosslinked poly(propylene sulfide) nanoparticles prepared by living emulsion polymerization of propylene sulfide in an aqueous phase containing Pluronic F-127 followed by curing by air exposure or by reaction with a bifunctional molecule (Rehor, Hubbell et al. 2005). Pluronic is presumed to cover the surface of the nanoparticles, thus imparting stability in aqueous environments. Nanoparticle size was easily controlled in the range of 25–250 nm by the ratio of Pluronic to PPS. As for the system previously described, exposure to oxidizing conditions leads to the transformation of the hydrophobic PPS onto hydrophilic poly(sulfoxides) and poly(sulfone), and in this case to the swelling and dissolution of the nanoparticles.

Personal Care Products

Although there is tremendous potential for nanotechnology in drug delivery, nanotechnology and nanoparticles, in particular, have already made a significant impact in consumer products. This mirrors the development of microparticle-based systems which appeared on supermarket shelves and cosmetic counters long before the pharmacy window.

In this section we will describe the use of nanotechnology in consumer products, primarily personal care products. Since these products are for external use and do not usually make substantiated medical claims, they are not often subject to FDA approval. Hence, the time from invention to market is much faster than for drug delivery systems. The research and development in the area is significant. For example, the company L'Oreal

ranks number six in their number of nanotechnology patents in the US with more than 190 patents using nanotechnology. For example, they hold patents on photoprotective and sunscreen products (Boutelet & Candau, 2006; Hansenne & Rick, 2002), nanocapsules based on poly(alkylene adipate) (Simonnet, Richart & Biatry, 2003), nanocapsules based on dendritic polymers (Simonnet & Richart, 2002), and nanoparticles containing oils for delivery to the upper layers of the epidermis based on poly(alkyl cyanoacrylates) (Handjani & Ribier, 2001).

Along with so many products containing nanoparticles, nanocapsules, fullerenes, nanosomes, and other nanoencapsulated ingredients comes public concern over the safety of such products. This concern rises primarily from the enhanced effectiveness which is often seen in nanosized formulations over conventional or even micron-sized formulations. In Table 1 there

Table 10.1 Examples of personal care products currently on the market containing nano-scale ingredients, nanoparticles, nanocapsules and nano-delivery systems.

Company	Product	Potentially nano-sized ingredient or relevant label term
Acne treatment		
Celazome	O-Plex Target Acne Spot Treatment	Lyphazome nanospheres
Wilma Schumann	Acne Kit	Vitamin E (nanoparticles)
DS Laboratories	Trioxil Anti-Acne Gel	Wheat-germ, barley and arnica nanosomes
After sun product		
L'Oreal , Lancome	Soleil Instant Cooling Sun Spritz	Vitamin nanocapsules
Anti-aging		
Sircuit Skin Cosmeceuticals Inc.	Skin Addict Firming Anti-Oxidant Serum	Fullerenes
Sircuit Skin Cosmeceuticals Inc.	Skin O.M.G. Anti-Aging Skin Resusc Serum	Fullerenes
Can Do Spirit, Inc., TYK	TYK Revelation	Nanodelivery system
Cosmetic Dermatology, Inc.	Dr. Brandt Laser Tight	Nanoencapsulated ingredients
ProCyte Corporation	Neova Therapy Dual Action Lotion	Nanoencapsulated retinol
emerginC	emerginC Hyper-Vitalizer	Nanoliposome delivery system
DS Laboratories, Inc.	Viterol.A (viatrozene gel) 29%	Nanosomes of sodium lactate, calendula, witch hazel, ginseng, urea, Vitamin A, Vitamin E, Pro-vitamin B5, alpha-bisabolol and germal II
DS Laboratories, Inc.	Viterol.A (viatrozene gel) 16%	Nanosomes of Vitamin A
L'Oreal, Lancome	Soleil Soft-Touch Anti-Wrinkle Sun Cream SPF 15	Vitamin nanocapsules
Anti-itch/rash cream		
Cosmetic Dermatology, Inc.	Dr. Brandt Laser Relief	Nanoencapsulated ingredients
Around-eye cream		
Sircuit Skin Cosmeceuticals Inc.	White Out Daily Under Eye Care	Fullerenes
Celazome New Zealand Limited	Eye Treat with Lyphazome Technology	Lyphazome nanospheres
PerfectRx	Eye Perfect Serum	Nanodelivery system

(*Continued*)

Table 10.1 (*Continued*).

Company	Product	Potentially nano-sized ingredient or relevant label term
Body firming lotion		
Nutra Luxé M.D.	Lipo Reduction Anti-Cellulite Creme	Nanodelivery system
DS Laboratories, Inc.	Oligo.DX Cellulite Reducing Gel	Nanosomes of centella asiatica
DS Laboratories, Inc.	Oligo DX Cellulite Treatment	Nanosomes of centella asiatica
Osmotics Corporation	Lipoduction Body Perfecting Complex	Nano-technology delivery system
Body wash/cleanser		
DS Laboratories, Inc.	Hydroviton.CR Liquid Normalizing Soap	Nanosomes of Vitamin A
Concealer		
DERMAdoctor	Faux Fillment	Nanospheres of hyaluronic acid and fulvic acid
Facial cleanser		
Fancl International, Inc.	Boscia MakeUp BreakUp Cool Cleansing Oil	Nanotechnology ingredients
Facial moisturizer/treatment		
Ferndale Laboratories, Inc.	Nouriva Repair Moisturizing Cream	Nanoparticle delivery system
Hair-loss treatment		
DS Laboratories, Inc.	Spectral DNC Hair Loss Treatment	Nanosomes
Mask		
Celazome New Zealand Limited	Spoil Me Body Lotion	Lyphazome nanospheres
Nail treatment		
Celazome New Zealand Limited	Tip Treat Cuticle Exfoliator	Lyphazome nanospheres
Pain/wound treatment		
Marlyn Nutraceuticals, Inc.	Naturally Vitamins Wobenzyme N Pain Relieving Creme	Nanosphere technology
Sunscreen/tanning oil		
Korres Natural Products Ltd.	Red Vine Year Round Hair Sun Protection	Nanoparticles
Korres Natural Products Ltd.	Red Vine Year Round Hair Sun Protection Spray	Nanoparticles
DERMAdoctor	Fun in the Sun Kit	Nanotechnology ingredients
L'Oreal, Lancome	Soleil Soft-Touch Moisturising Sun Lotion SPF 15	Vitamin nanocapsules

is a summary of some, but certainly not all, personal care products that contain nanoscale materials. Much of this information has been compiled by the Environmental Working Group who prepared an analysis of 25,000 personal care product labels and found that approximately 250 of them specifically mentioned containing nanoscale or micron-sized ingredients. Some examples are shown in Table 1 (Environmental Working Group, 2006). Not all of these certainly would have controlled-release-based nanoscale components, but a great many do. A large number of these products contain nanosomes, or nanoscale liposomes, which provide stability to the encapsulated product, better skin coverage of the product, and subsequent controlled release of the active agent. Although the term nanosome has been

trademarked by Elsom Research, it also appears in the research literature and product descriptions by DS Laboratories, Min New York, Lipoxidil, L'Oreal, and Cosmosome to name just a few. Enhanced penetration of these nansomes can also occur since the gaps between dead skin cells on the surface of the epidermis are approximately 100 nm wide. This allows products which can easily penetrate the lipid matrix between dead skin cells, such as nanosomes, to deliver not only cosmetics and personal care products but also therapeutic agents directly to the living cells of the epidermis.

These companies are not just utilizing nanotechnology, they are marketing it as well. A look at the website for Beyond Skin Science (Beyond Skin Science, 2007) reveals the logos of the National Nanotechnology Initiative and the Nano Science Technology Institute, although there are no links to their websites and it is confusing what this Institute actually is. Their own technology is called "Nanochem" and their products are labeled as "Nanochem certified." Duprey cosmetics is using NanoDulcine™, nanosized particles of a low glycemic, human grade edible fruit glycoside, to deliver L-arginine and vitamin C "deep within the cells" of the epidermis. Their website carries a logo stating "Certified Nano Technology" (Duprey Cosmetics, 2007).

The greatest challenge in determining what actually goes into nanoscale skin care products is that more are proprietary and may or may not actually be patented and very few are described in the scientific literature. Most work described in the scientific literature addresses solid lipid nanoparticles (SLN), a system which combines the solubility and stability-enhancing advantages of liposomes for many compounds with the additional stability of solid formulations (Müller, Radtke & Wissing, 2002; Wissing & Müller, 2003). This form is achieved by exchanging the liquid lipid (oil) in liposome formulations with a lipid which is solid at room and body temperatures. The structure of these SLNs can be a matrix, a compound-enriched shell, or a compound-enriched core. For epidermal delivery, these formulations may show improved skin penetration and targeting, but those results may be dependent on the interaction of the drug and particle (Borgia, Regehly, Sivaramakrishnan, Mehnert, Korting, Danker, Röder, Kramer & Schäfer-Korting, 2005; Chen, Chang, Du, Liu, Liu, Weng, Yang, Xu & X, 2006). The most commonly reported studies involve delivery of retinol-based active agents for cosmetic use and dermatological disease (Jee, Lim, Park & Kim, 2006; Liu, Hu, Chen, Ni, Xu & Yang, 2007; Taha, Samy, Kassem & Khan, 2005), sunblock formulations (Cengiz, Wissing, Müller & Yazan, 2006; Song & Liu, 2005), and vitamin A delivery (Jenning, Gysler, Schäfer-Korting & Gohla, 2000; Jenning, Schäfer-Korting & Gohla, 2000).

References

Abo El Ola, S. M., R. Kotek, et al. (2004). "Studies on poly(trimethylene terephthalate) filaments containing silver." *J Biomater Sci Polym Ed* **15**(12): 1545–59.

Agnihotri, S. A., N. N. Mallikarjuna, et al. (2004). "Recent advances on chitosan-based micro- and nanoparticles in drug delivery." *J Control Release* **100**(1): 5–28.

Balogh, L., D. R. Swanson, et al. (2001). "Dendrimer-silver complexes and nanocomposites as antimicrobial agents." *Nano Lett* **1**(1): 18–21.

Beyond Skin Science (2007). Corona, CA (March 20, 2007); http://www.beyondskinscience.com

Bhabak, K. P. and G. Mugesh (2007). "Synthesis, characterization, and antioxidant activity of some ebselen analogues." *Chem A Eur J* **13**(16): 4594–4601.

Bhadra, D., S. Bhadra, et al. (2003). "A PEGylated dendritic nanoparticulate carrier of fluorouracil." *Int J Pharm* **257**: 111–124.

Bhattacharya, S. and A. Bajaj (2005). "Recent advances in lipid molecular design." *Curr Opin Chem Biol* **9**(6): 647–655.

Bontha, S., A. V. Kabanov, et al. (2006). "Polymer micelles with cross-linked ionic cores for delivery of anticancer drugs." *J Control Release* **114**(2): 163–74.

Borgia, S. L., Regehly, M., Sivaramakrishnan, R., Mehnert, W., Korting, H. C., Danker, K., Röder, B., Kramer, K. D. and Schäfer-Korting, M. (2005). "Lipid nanoparticles for skin penetration enhancement-correlation to drug localization within the particle matrix as determined by fluorescence and parelectric spectroscopy." *J Control Release*, **110**: 151–163.

Brannon-Peppas, L. (1995). "Recent advances on the use of biodegradable microparticles and nanoparticles in controlled drug delivery." *Int J Pharm* **116**: 1–9.

Brannon-Peppas, L. and J. O. Blanchette (2004). "Nanoparticle and targeted systems for cancer therapy." *Adv Drug Del Rev* **56**: 1649–1659.

Brett, D. W. (2006). "A discussion of silver as an antimicrobial agent: alleviating the confusion." *Ostomy Wound Manage* **52**(1): 34–41.

Bromberg, L. E., D. K. Buxton, et al. (2001). "Novel periodontal drug delivery system for treatment of periodontitis." *J Control Release* **71**(3): 251–9.

Boutelet, K. and D. Candau (2006). Photoprotective/cosmetic compositions comprising sulfonic/hydrophobic amphiphilic polymers, USA, L' Oreal, Patent 7,045,120.

Calvo, P., J. L. VilaJato, et al. (1996). "Comparative in vitro evaluation of several colloidal systems, nanoparticles, nanocapsules, and nanoemulsions, as ocular drug carriers." *J Pharm Sci* **85**(5): 530–536.

Caves, J. M. and E. L. Chaikof (2006). "The evolving impact of microfabrication and nanotechnology on stent design." *J Vasc Surg* **44**(6): 1363–1368.

Cengiz, E., Wissing, S. A., Müller, R. H. and Yazan, Y. (2006). Sunblocking efficiency of various ti)2-loaded solid lipid nanoparticle formulations. *Int J Cosm Sci* **28**: 371–378.

Chen, H., Chang, X., Du, D., Liu, W., Liu, J., Weng, T., Yang, Y., Xu, H. and Yang, X. (2006). Podophyllotoxin-loaded solid lipid nanoparticles for epidermal targeting. *J Control Release*, **110**: 296–306.

D'Emanuele, A. and D. Attwood (2005). "Dendrimer-drug interactions." *Adv Drug Del Rev* **57**(15): 2147–2162.

Devalapally, H., D. Shenoy, et al. (2007). "Poly(ethylene oxide)-modified poly (beta-amino ester) nanoparticles as a pH-sensitive system for tumor-targeted delivery of hydrophobic drugs: part 3. Therapeutic efficacy and safety studies in ovarian cancer xenograft model." *Cancer Chemo Pharmacol* **59**(4): 477–484.

Duprey Cosmetics (2007). (March 10, 2007); St. Pete Beach, Fl.; http://www.dupreycosmetics.com/ nano_dulcine.html

Edelman, E. R., L. Brown, et al. (1987). "In vitro and in vivo kinetics of regulated drug release from polymer matrices by oscillating magnetic fields." *J Biomed Mater Res* **21**(3): 339–53.

Edelman, E. R., A. Fiorino, et al. (1992). "Mechanical deformation of polymer matrix controlled release devices modulates drug release." *J Biomed Mater Res* **26**(12): 1619–31.

Environmental Working Group (2006). Washington, DC (October 10, 2006); http://www.ewg.org/issues/cosmetics/20061010/table2.php

Fonseca, C., S. Simões, et al. (2002). "Paclitaxel-loaded PLGA nanoparticles: preparation, physiochemical characterization and in vitro anti-tumoral activity." *J Control Release* **83**: 273–286.

Fresta, M., G. Puglisi, et al. (1995). "Pefloxacine mesilate-loaded and ofloxacin-loaded polyethylcyanoacrylate nanoparticles- Characterization of the colloidal drug carrier formulations." *J Pharm Sci* **84**(7): 895–902.

Fu, J., X. Li, et al. (2002). "Encapsulation of phthalocyanines in biodegradable poly(sebacic anhydride) nanoparticles." *Langmuir* **18**(10): 3843–3847.

Gaponik, N., I. L. Radtchenko, et al. (2004). "Luminescent polymer microcapsules addressable by a magnetic field." *Langmuir* **20**(4): 1449–52.

Georgakilas, V., K. Kordatos, et al. (2002). "Organic functionalization of carbon nanotubes." *J Am Chem Soc* **124**(5): 760–761.

Georgakilas, V., N. Tagmatarchis, et al. (2002). "Amino acid functionalization of water soluble carbon nanotubes." *Chem Communs* **24**: 3050–4051.

Guo, Z. and P. Sadler (1999). "Metals in medicine." *Angewandte Chemie-International Edition* **38**(11): 1513–1531.

Guzman, L. A., V. Labhasetwar, et al. (1996). "Local intraluminal infusion of biodegradable polymeric nanoparticles – A novel approach for prolonged drug delivery after balloon angioplasty." *Circulation* **94**(6): 1441–1448.

Handjani, R.-M. and Ribier, A. (2001). Compositions for the cosmetic and/or pharmaceutical treatment of the epidermis by topical application to the skin, and corresponding preparation process, USA, L' Oreal, Patent 6,203,802.

Hansenne, I. and Rick, D. W. (2002). High SPF nontacky/nongreasy uv-photoprotecting compositions comprising particulates of MMA crosspolymers, USA, L' Oreal, Patent 6,432,389.

Heller, J., J. Barr, et al. (2002). "Poly(ortho esters): synthesis, characterization, properties and uses." *Adv Drug Del Rev* **54**(7): 1015–1039.

Hirsch, L. R., A. M. Gobin, et al. (2006). "Metal nanoshells." *Ann Biomed Eng* **34**(1): 15–22.

Horcajada, P., C. Marquez-Alvarez, et al. (2006). "Controlled release of ibuprofen from dealuminated faujasites " *Solid State Sciences* **8**(12): 1459–1465

Horcajada, P., C. Serre, et al. (2006). "Metal-organic frameworks as efficient materials for drug delivery." *Angew Chem Int Ed Engl* **45**(36): 5974–8.

Hu, Y. X., J. Chang, et al. (2005). Preparation and evaluation of 5-FU/PLGA/gene nanoparticles. *Asbm6: Advanced Biomaterials Vi*. Zurich-Uetikon, Trans Tech Publications Ltd: 147–150.

Huth, S., J. Lausier, et al. (2004). "Insights into the mechanism of magnetofection using PEI-based magnetofectins for gene transfer." *J Gene Med* **6**(8): 923–36.

Isab, A. A. and M. I. Wazeer (2007). "Synthesis and characterization of thiolate-Ag(I) complexes by solid-state and solution NMR and their antimicrobial activity." *Spectrochim Acta A Mol Biomol Spectrosc* **66**(2): 364–370.

Jain, R. A. (2000). "The manufacturing techniques of various drug loaded biodegradable poly(lactide-co-glycolide) (PLGA) devices." *Biomaterials* **21**(23): 2475–2490.

Jee, J.-P., Lim, S.-J., Park, J.-S. and Kim, C.-K. (2006). "Stabilization of all-trans retinol by loading lipophilic antioxidants in solid lipid nanoparticles." *Eur J Pharm Biopharm*, **63**: 134–139.

Jenning, V., Gysler, A., Schäfer-Korting, M. and Gohla, S. H. (2000). "Vitamin A loaded solid lipid nanoparticles for topical use: Occlusive properties and drug targeting in the upper skin." *Eur J Pharm Biopharm*, **49**: 211–218.

Kam, N. W. and H. Dai (2005). "Carbon nanotubes as intracellular protein transporters: generally and biological functionality." *J Am Chem Soc* **127**(16): 6021–6026.

Karlov, A. V., I. A. Khlusov, et al. (2002). "Adhesion of Staphylococcus aureus to implants with different physicochemical characteristics." *Bull Exp Biol Med* **134**(3): 277–80.

Kost, J., R. Noecker, et al. (1985). "Magnetically controlled release systems: effect of polymer composition." *J Biomed Mater Res* **19**(8): 935–40.

Kost, J., J. Wolfrum, et al. (1987). "Magnetically enhanced insulin release in diabetic rats." *J Biomed Mater Res* **21**(12): 1367–73.

Kumar, R., S. Howdle, et al. (2005). "Polyamide/silver antimicrobials: effect of filler types on the silver ion release." *J Biomed Mater Res B Appl Biomater* **75**(2): 311–9.

Labhasetwar, V., C. X. Song, et al. (1998). "Arterial uptake of biodegradable nanoparticles: Effect of surface modifications." *J Pharm Sci* **87**(10): 1229–1234.

Lanza, G. M., X. Yu, et al. (2002). "Targeted antiproliferative drug delivery to vascular smooth muscle cells with a magnetic resonance imaging nanoparticle contrast agent implications for rational therapy of restenosis." *Circulation* **106**(22): 2842–2847.

Li, Y.-Y., X.-Z. Zhang, et al. (2006). "Novel stimuli-responsive micelle self-assembled from Y-shaped P(UA-Y-NIPAAm) copolymer for drug delivery." *Biomacromolecules* **7**(11): 2956–2960.

Lin, A. W., N. A. Lewinski, et al. (2005). "Optically tunable nanoparticle contrast agents for early cancer detection: model-based analysis of gold nanoshells." *J Biomed Opt* **10**(6): 064035.

Liu, J., Hu, W., Chen, H., Ni, Q., Xu, H. and Yang, X. (2007). "Isotretinoin-loaded solid lipid nanoparticles with skin targeting for topical delivery." *Int J Pharm*, **328**: 191–195.

Liu, M. J., K. Kono, et al. (2000). "Water-soluble dendritic unimolecular micelles: Their potential as drug delivery agents." *J Control Release* **65**(1–2): 121–131.

Lu, Z., J. Z. Bei, et al. (1999). "A method for the preparation of polymeric nanocapsules without stabilizer." *J Control Release* **61**(1–2): 107–112.

MacLachlan, M., I. Manners, et al. (2000). "New (inter)faces: Polymers and inorganic materials " *Adv Materials* **12**(9): 675–681.

Mainardes, R. M. and L. P. Silva (2004). "Drug delivery systems: Past, present, and future." *Curr Drug Targets* **5**(5): 449–455.

Mitra, S., U. Gaur, et al. (2001). "Tumour targeted delivery of encapsulated dextran-doxorubicin conjugate using chitosan nanoparticles as carrier." *J Control Release* **74**: 317–323.

Muller, R. H., K. Mader, et al. (2000). "Solid lipid nanoparticles (SLN) for controlled drug delivery – a review of the state of the art." *Eur J Pharm Biopharm* **50**(1): 161–177.

Müller, R. H., Radtke, M. and Wissing, S. A. (2002). Solid lipid nanoparticles (SLN) and nanostructured lipid carriers (NLC) in cosmetic and dermatological preparations. *Adv Drug Del Rev*, **54**: S131–S155.

Nakayama, M., T. Okano, et al. (2006). "Molecular design of biodegradable polymeric micelles for temperature-responsive drug release." *J Control Release* **115**(1): 46–56.

Napoli, A., M. Valentini, et al. (2005). "Oxidation-responsive polymeric vesicles." *Nature Biomaterials* **3**(3): 183–189.

Niwa, T., H. Takeuchi, et al. (1993). "Preparation of biodegradable nanospheres of water-soluble and insoluble drugs with D,L-lactide/glycolide copolymer by a novel spontaneous emulsification solvent diffusion method, and the drug release behavior." *J Control Release* **25**: 89–93.

O'Reilly, R. K., C. J. Hawker, et al. (2006). "Cross-linked block copolymer micelles: functional nanostructures of great potential and versatility." *Chemical Society Reviews* **35**(11): 1068–1083.

Oh, K. T., T. K. Bronich, et al. (2006). "Block ionomer complexes as prospective nanocontainers for drug delivery." *J Control Release* **115**(1): 9–17.

Owens III, D. and N. Peppas (2006). "Integration of thermally responsive nanosphere hydrogels with gold nanoparticles for intelligent therapeutic applications." *Trans Soc Biomater* **31**: 150–151.

Panyam, J., W.-Z. Zhou, et al. (2002). "Rapid endo-lysosomal escape of poly (D,L-lactide-co-glycolide) nanoparticles: implications for drug and gene delivery." *FASEB* **16**(10): 1217–1226.

Pastorin, G., W. Wu, et al. (2006). "Double functionalization of carbon nanotubes for multimodal delivery." *Chem Commun* **11**: 1182–1184.

Patton, J. N. and A. F. Palmer (2005). "Engineering temperature-sensitive hydrogel nanoparticles entrapping hemoglobin as a novel type of oxygen carrier." *Biomacromolecules* **6**(4): 2204–2212.

Plank, C., F. Scherer, et al. (2003). "Magnetofection: enhancing and targeting gene delivery with superparamagnetic nanoparticles and magnetic fields." *J Liposome Res* **13**(1): 29–32.

Plank, C., U. Schillinger, et al. (2003). "The magnetofection method: using magnetic force to enhance gene delivery." *Biol Chem* **384**(5): 737–47.

Potineni, A., D. M. Lynn, et al. (2003). "Poly(ethylene oxide)-modified poly(beta-amino ester) nanoparticles as a pH-sensitive biodegradable system for paclitaxel delivery." *J Control Release* **86**(2–3): 223–234.

Radwan, M. A. (1995). "In-vitro evaluation of polyisobutylcyanoacrylate nanoparticles as a controlled drug carrier for theophylline." *Drug Dev Ind Pharm* **21**(20): 2371–2375.

Rehor, A., J. A. Hubbell, et al. (2005). "Oxidation-sensitive polymeric nanoparticles." *Langmuir* **21**(1): 411–417.

Rhim, J. W., S. I. Hong, et al. (2006). "Preparation and characterization of chitosan-based nanocomposite films with antimicrobial activity." *J Agric Food Chem* **54**(16): 5814–22.

Roskos, K. V., B. K. Fritzinger, et al. (1995). "Development of a drug-delivery system for the treatment of periodontal-disease based on bioerodible poly(ortho esters)." *Biomaterials* **16**(4): 313–317.

Sajeesh, S. and C. P. Sharma (2005). "Novel pH responsive polymethacrylic acid-chitosan-polyethylene glycol nanoparticles for oral peptide delivery." *J Biomat Mater Res B: Appl Biomat* **76B**(2): 298–305.

Sakurai, H., A. Katoh, et al. (2006). "Chemistry and biochemistry of insulin-mimetic vanadium and zinc complexes. Trial for treatment of diabetes mellitus." *Bull. Chem. Soc. Jpn.* **79**(11): 1645–1664

Sambhy, V., M. M. MacBride, et al. (2006). "Silver bromide nanoparticle/polymer composites: dual action tunable antimicrobial materials." *J Am Chem Soc* **128**(30): 9798–808.

Sawant, R. M., J. P. Hurley, et al. (2006). ""SMART" drug delivery systems: double-targeted pH-responsive pharmaceutical nanocarriers." Bioconjug Chem **17**(4): 943–949.

Schierholz, J. M., L. J. Lucas, et al. (1998). "Efficacy of silver-coated medical devices." *J Hosp Infect* **40**(4): 257–62.

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

Sershen, S. R., S. L. Westcott, et al. (2000). "Temperature-sensitive polymer-nanoshell composites for photothermally modulated drug delivery." *J Biomed Mater Res* **51**(3): 293–8.

Seymour, L. W., R. Duncan, et al. (1994). "Poly(ortho ester) matrices for controlled release of the antitumor agent 5-fluorouracil." *J Control Release* **31**: 201–206.

Shenoy, D., S. Little, et al. (2005). "Poly(ethylene oxide)-modified poly(beta-amino ester) nanoparticles as a pH-sensitive system for tumor-targeted delivery of hydrophobic drugs. Part 1 In vitro evaluations." *Mol Pharmacy* **2**(5): 357–366.

Shenoy, D., S. Little, et al. (2005). "Poly(ethylene oxide)-modified poly(beta-amino ester) nanoparticles as a pH-sensitive system for tumor-targeted delivery of hydrophobic drugs: Part 2. In vivo distribution and tumor localization studies." *Pharm Res* **22**(12): 2107–2114.

Shikata, F., H. Tokumitsu, et al. (2002). "In vitro cellular accumulation of gadolinium incorporated into chitosan nanoparticles designed for neutron-capture therapy of cancer." *Eur. J. Pharm Biopharm*. **53**: 57–63.

Simonnet, J.-T. and Richart, P. (2002). Nanocapsules based on dendritic polymers, USA, L' Oreal, Patent 6,379,683.

Simonnet, J.-T., Richart, P. and Biatry, B. (2003). Nanocapsules based on poly (alkylene adipate), process for their preparation and cosmetic or dermatological compositions containing them, USA, L' Oreal, Patent 6,565,886.

Singh, R., D. Pantarotto, et al. (2006). "Tissue biodistribution and blood clearance rates of intravenously administered carbon nanotube radiotracers." *PNAS* **103**(9): 3357–3362.

Smaihi, M., E. Gavilan, et al. (2004). "Colloidal functionalized calcined zeolite nanocrystals " *J Mater Chem* **14**(8): 1347–1351.

Song, C. and Liu, S. (2005). "A new healthy sunscreen system for humans: Solid lipid nanoparticles as carrier for 3,4,5-trimethoxybenzoylchitin and the improvement by adding vitamin E." *Int J Bio Macromol*, **36**: 116–119.

Soppimath, K. S. and T. M. Aminabhavi (2002). "Ethyl acetate as a dispersing solvent in the production of poly(DL-lactide-co-glycolide) microspheres: effect of process parameters and polymer type." *J Microencapsulation* **19**(3): 281–292.

Soppimath, K. S., T. M. Aminabhavi, et al. (2001). "Biodegradable polymeric nanoparticles as drug delivery devices." *J Controlled Release* **70**(1–2): 1–20.

Soppimath, K. S., D. C.-W. Tan, et al. (2005). "pH-Triggered thermally responsive polymer core-shell nanoparticles for drug delivery." *Adv Mater* **17**(3): 318–323.

Steinbrink, J., H. Wabnitz, et al. (2001). "Determining changes in NIR absorption using a layered model of the human head." *Phys Med Biol* **46**(3): 879–896.

Taha, E. I., Samy, A. M., Kassem, A. A. and Khan, M. A. (2005). Response surface methodology for the development of self-nanoemulsified drug delivery system (SNEDDS) of all-trans-retinol acetate. *Pharm Dev Tech*, **10**: 363–370.

Tirelli, N. (2006). "(Bio)responsive nanoparticles." *Curr Opin Colloidal & Interface Sci* **11**(4): 210–216.

Tokumitsu, H., H. Ichikawa, et al. (1999). "Chitosan-gadopentetic acid complex nanoparticles for gadolinium neutron-capture therapy of cancer: Preparation by novel emulsion-droplet coalescence technique and characterization." *Pharm Res* **16**(12): 1830–1835.

Uhrich, K. E., S. M. Cannizzaro, et al. (1999). "Polymeric systems for controlled drug release." *Chem Rev* **99**(11): 3181–3198.

van Zutphen, S. and J. Reedijk (2005). "Targeting platinum anti-tumour drugs: Overview of strategies employed to reduce systemic toxicity." *Coordination Chem Rev* **249**(24): 2845–2853.

Vaupel, P., F. Kallinowski, et al. (1989). "Blood flow, oxygen and nutrient supply and metabolic microenvironment of human tumors: a review." *Cancer Res* **49**(23): 6449–6465.

Verma, A. K., K. Sachin, et al. (2005). "Release kinetics from bio-polymeric nanoparticles encapsulating protein synthesis inhibitor - Cycloheximide, for possible therapeutic applications." *Curr Pharm Biotechnol* **6**(2): 121–130.

Wei, H., X.-Z. Zhang, et al. (2006). "Self-assembled thermo- and pH-responsive micelles of poly(10-undecenoic acid-b-N-isopropylacrylamide) for drug delivery." *J Control Release* **116**(3): 266–274.

Wike-Hooley, J. A., J. Haveman, et al. (1984). "The relevance of tumor pH to the treatment of malignant disease." *Radiother Oncol* **2**(4): 343–366.

Wissing, S. A. and Müller, R. H. (2003). "Cosmetic applications for solid lipid nanoparticles (SLN)." *Int J Pharm*, 254, 65–68.

Wojtowicz, H., K. Kloc, et al. (2004). "Azaanalogues of ebselen as antimicrobial and antiviral agents: synthesis and properties." *Farmaco* **59**(11): 863–8.

Wu, W., S. Wieckowski, et al. (2005). "Targeted delivery of amphotericin B to cells by using functionalized carbon nanotubes." *Angew. Chem. Int. Ed.* **44**(39): 6358–6362.

Xu, P., E. A. Van Kirk, et al. (2006). "Anticancer Efficacies of Cisplatin-Releasing pH-Responsive Nanoparticles." *Biomacromolecules* **7**(3): 829–835.

Yoo, H. S., K. H. Lee, et al. (2000). "In vitro and in vivo anti-tumor activities of nanoparticles based on doxorubicin-PLGA conjugates." *J Control Release* **68**(3): 419–431.

Zhang, H., Y. Kim, et al. (2006). "Controlled release of paraquat from surface-modified zeolite Y." *Microporous and Mesoporous Materials* **88**(1–3): 312–318

Zwiorek, K., J. Kloeckner, et al. (2004). "Gelatin nanoparticles as a new and simple gene delivery system." *J Pharm Pharmac Sci* **7**(4): 22–28.