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Biodegradable nanoparticles for drug delivery and targeting

M.L. Hans, A.M. Lowman*

Biomaterials and Drug Delivery Laboratory, Department of Chemical Engineering, Drexel University, Philadelphia, PA 19104, USA

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Abstract

Throughout the world today, numerous researchers are exploring the potential use of polymeric nanoparticles as carriers for a wide range of drugs for therapeutic applications. Because of their versatility and wide range of properties, biodegradable polymeric nanoparticles are being used as novel drug delivery systems. In particular, this class of carrier holds tremendous promise in the areas of cancer therapy and controlled delivery of vaccines.

Keywords: Nanoparticles; PLGA; Drug delivery; Surface modifications

1. Introduction

Polymer nanoparticles are particles of less than 1 μm diameter that are prepared from natural or synthetic polymers. Nanoparticles have become an important area of research in the field of drug delivery because they have the ability to deliver a wide range of drugs to varying areas of the body for sustained periods of time. Natural polymers (i.e. proteins or polysaccharides) have not been widely used for this purpose since they vary in purity, and often require crosslinking that could denature the embedded drug. Consequently, synthetic polymers have received significantly more attention in this area. The most widely used polymers for nanoparticles have been poly(lactic acid) (PLA), poly(glycolic acid) (PGA), and their co-polymers, poly(lactide-co-glycolide) (PLGA). These polymers are known for both their biocompatibility and resorbability through natural pathways. Additionally, the degradation rate and accordingly the drug release rate can be manipulated by varying the ratio of PLA, increased hydrophobicity, to PGA, increased hydrophilicity.

During the 1980s and 1990s several drug delivery systems were developed to improve the efficiency of drugs and minimize toxic side effects [1]. The early nanoparticles and microparticles were mainly formulated from poly-(alkylcyanoacrylate). Initial promise for microparticles was dampened by the fact that there was a size limit for the particles to cross the intestinal lumen into the lymphatic system following oral delivery. Likewise, the therapeutic effect of drug-loaded nanoparticles was relatively poor due rapid clearance of the particles by phagocytosis post intravenous administration. In recent years this problem has been solved by the addition of surface modifications to nanoparticles.

Another promising class of nano-sized vehicles that have been considered in drug delivery applications is liposomes. These vesicles prepared from lipids have been used as potential drug carriers because of the protection they can offer drugs contained in their core. However, liposomes have shown a low encapsulation efficiency, poor storage stability, and rapid leakage of water-soluble drugs in the blood [2]. As such, their ability to control the release of many drugs may not be good. Solid, biodegradable nanoparticles have shown their advantage over liposomes by their increased stability and the unique ability to create a controlled release.

In recent years, significant research has been done using nanoparticles as oral drug delivery vehicles. In this application, the major interest is in lymphatic uptake of the nanoparticles by the Peyer's patches in the GALT (gut associated lymphoid tissue). Peyer's patches are characterized by M cells that overlie the lymphoid tissue and are specialized for endocytosis and transport into intraepithelial spaces and adjacent lymphoid tissue. Nanoparticles bind the apical membrane of the M cells, followed by a rapid internalization and a 'shuttling' to the lymphocytes [3,4]. The size and surface charge of the nanoparticles are crucial

*Corresponding author. Tel.: +1-215-895-2228.
E-mail address: alowman@cbis.ece.drexel.edu (A.M. Lowman).

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for their uptake. There have been many reports as to the optimum size for Peyer's Patch uptake ranging from less than 1 μm to less than 5 μm [5,6]. It has been shown that microparticles remain in the Peyer's patches while nanoparticles are disseminated systemically [7]. This application of nanoparticles in oral delivery holds tremendous promise for the development of oral vaccines and in cancer therapy.

Nanoparticles have a further advantage over larger microparticles, because they are better suited for intravenous (i.v.) delivery. The smallest capillaries in the body are 5–6 μm in diameter. The size of particles being distributed into the bloodstream must be significantly smaller than 5 μm, without forming aggregates, to ensure that the particles do not form an embolism.

Clearly, a wide variety of drugs can be delivered using nanoparticulate carriers via a number of routes. Nanoparticles can be used to deliver hydrophilic drugs, hydrophobic drugs, proteins, vaccines, biological macromolecules, etc. They can be formulated for targeted delivery to the lymphatic system, brain, arterial walls, lungs, liver, spleen, or made for long-term systemic circulation. Therefore, numerous protocols exist for synthesizing nanoparticles based on the type of drug used and the desired delivery route. Once a protocol is chosen, the parameters must be tailored to create the best possible characteristics for the nanoparticles. Four of the most important characteristics of nanoparticles are their size, encapsulation efficiency, zeta potential (surface charge), and release characteristics. In this review, we intend to summarize many of the techniques used for preparing polymeric nanoparticles, including the types of polymers and stabilizers used, and how these techniques affect the structure and properties of the nanoparticles. Additionally, we will discuss advances in surface modifications, drug encapsulation and targeted drug delivery applications.

2. Synthesis and characterization

As stated previously, there are several different methods for preparing nanoparticles. Additionally, numerous methods exist for incorporating drugs into the particles. For example, drugs can be entrapped in the polymer matrix, encapsulated in a nanoparticle core, surrounded by a shell-like polymer membrane, chemically conjugated to the polymer, or bound to the particle's surface by adsorption. A summary of these methods including the types of polymer, solvent, stabilizer and drugs used is given in Tables 1 and 2.

The most common method used for the preparation of solid, polymeric nanoparticles is the emulsification–solvent evaporation technique. This technique has been successful for encapsulating hydrophobic drugs, but has had poor results incorporating bioactive agents of a hydrophilic nature. Briefly, solvent evaporation is carried out by dissolving the polymer and the compound in an organic solvent. Frequently, dichloromethane or methylene chloride is used for PLGA copolymers. The emulsion is prepared by adding water and a surfactant to the polymer solution. In many cases, the nanosized polymer droplets are induced by sonication or homogenization. The organic solvent is then evaporated and the nanoparticles are usually collected by centrifugation and lyophilization [5,8–11].

A modification on this procedure has led to the protocol favored for encapsulating hydrophilic compounds and proteins, the double or multiple emulsion technique. First, a hydrophilic drug and a stabilizer are dissolved in water. The primary emulsion is prepared by dispersing the aqueous phase into an organic solvent containing a dissolved polymer. This is then reemulsified in an outer aqueous phase also containing stabilizer [6,7,9,12–14]. From here, the procedure for obtaining the nanoparticles is similar to the single emulsion technique for solvent removal. The main problem with trying to encapsulate a hydrophilic molecule like a protein or peptide-drug is the rapid diffusion of the molecule into the outer aqueous phase during the emulsification. This can result in poor encapsulation efficiency, i.e. drug loading. Therefore, it is critical to have an immediate deposit of a polymer membrane during the first water-in-oil emulsion. Song et al. [9] was able to accomplish this by dissolving a high concentration of high molecular weight PLGA into a the oil phase consisting of 80:20% weight dichloromethane/acetone solution. Additionally, the viscosity of the inner aqueous phase was increased by increasing the concentration of stabilizer, bovine serum albumin (BSA). The primary emulsion was then emulsified with Pluronic F68 resulting in drug-loaded particles of approximately 100 nm [9].

Another method that has been used to encapsulate insulin for oral delivery is phase inversion nanoencapsulation (PIN). Zn-insulin is dissolved in Tris–HCl and a portion of that is recrystallized by the addition of 10% $ZnSO_4$. The precipitate is added to a polymer solution of PLGA in methylene chloride. This mixture is emulsified and dispersed in 1 l of petroleum ether, which results in the spontaneous formation of nanoparticles [15].

All of the previously mentioned techniques use toxic, chlorinated solvents that could degrade certain drugs and proteins if they come into contact during the process. Consequently, an effort has been made to develop other techniques in order to increase drug stability during the synthesis. One such technique is the emulsification–diffusion method. This method uses a partially water-soluble solvent like acetone or propylene carbonate. The polymer and bioactive compound is dissolved in the solvent and emulsified in the aqueous phase containing the stabilizer. The stabilizer prevents the aggregation of emulsion droplets by adsorbing of the surface of the droplets. Water is added to the emulsion, to allow for the diffusion of the solvent into the water. The solution is stirred leading to the

Table 1
Summary of methods used for preparation used for preparation of polymeric nanoparticles

Method	Polymer	Solvent	Stabilizer	Size (nm)	Reference
Solvent diffusion	PLGA	Acetone	Pluronic F-127	200	[33]
	PLGA	Acetone/DCM	PVA	200–300	[19]
	PLA-PEG	MC	PVA/PVP	~130	[45]
	PHDCA	THF	–	150	[38]
	PLGA	Acetone	Sodium cholate	161	[36]
	PLGA	Propylene carbonate	PVA or DMAB	~100	[16]
Solvent displacement	PLA	Acetone/MC	Pluronic F68	123±23	[41]
	SB-PVA-g-PLGA	Acetone/ethyl acetate	Poloxamer 188	~110	[32]
Nanoprecipitation	PLGA/PLA/PCL	Acetone	Pluronic F68	110–208	[20]
	PLGA	Acetonitrile	–	157.1±1.9	[18]
Solvent evaporation	PLA-PEG-PLA	DCM	–	193–335	[48]
	PLGA	DCM	PVA	800	[10]
	PEO-PLGA	MC	PVA	150±25	[8]
Multiple emulsion	PLGA	Ethyl acetate	–	>200	[37]
	PLGA	Ethyl acetate/MC	PVA/PVP	~280	[45]
	PLGA	Ethyl acetate/MC	PVA	335–743	[34]
	PLGA-mPEG	DCM	–	133.5±3.7–163.3±3.6	[49]
	PLGA	DCM	PVA	70–160	[42]
	PLGA	DCM	PVA	213.8±10.9	[14]
	PLGA	DCM/acetone	PVA	100	[9]
	PLGA	DCM	PVA	~250	[6]
	PLGA	Ethyl acetate	PVA	192±12	[12]
	PLGA	Ethyl acetate	PVA	~300	[7]
	PLGA	DCM	PVA	380±40–1720±110	[13]
Salting out	PLA	Acetone	PVA	300–700	[21]
Ionic gelation	Chitosan	TPP	–	278±03	[41]
Interfacial deposition	PLGA	Acetone	–	135	[40]
Phase inversion nanoencapsulation	PLGA	MC	–	>5 μm	[15]
Polymerization	CS-PAA	–	–	206±22	[25]
	PECA	–	Pluronic F68	320±12	[31,26]
	PE-2-CA	–	–	380±120	[30]

Size is in nm, unless otherwise indicated. DCM, dichloromethane; MC, methylene chloride; PVP, polyvinylpyrrolidone; PHDCA, poly(hexadecylcyanoacrylate); THF, tetrahydrofuran; SB-PVA-g-PLGA, sulfobutylated PVA, graft, PLGA; PCL, poly(epsilon-caprolactone); TPP, sodium tripolyphosphate; PAA, poly(acrylic acid); PECA, polyethylcyanoacrylate; PE-2-CA, polyethyl-2-cyanoacrylate.

nanoprecipitation of the particles. They can then be collected by centrifugation, or the solvent can be removed by dialysis [16,17].

One problem with this technique is that water-soluble drugs tend to leak out of the polymer phase during the solvent diffusion step. To improve this process for water-soluble drugs, Takeuchi et al. [17] changed the dispersing medium from an aqueous solution to a medium chain triglyceride and added a surfactant, Span® 80, to the polymer phase. The nanoparticles are collected from the oily suspension by centrifugation. Several parameters can also be changed to benefit the encapsulation of hydrophilic molecules. Govender et al. [18] found that increasing the aqueous phase pH to 9.3 and incorporating pH-responsive excipients such as poly(methyl methacrylate-co-methacrylic acid) (PMMA-MAA), and lauric and caprylic acid increased hydrophilic drug encapsulation without effecting the particle size, morphology, or yield. Murakami et al. [19] effectively modified the solvent diffusion technique by using two water-miscible solvents, one with more affinity for PLGA and one with more affinity for the stabilizer, PVA, such as acetone and ethanol.

Nanoparticles can also be synthesized by the nanoprecipitation method. Briefly, the polymer and drug are dissolved in acetone and added to an aqueous solution containing Pluronic F68. The acetone is evaporated under reduced pressure and the nanoparticles remain in the suspension resulting in particles from 110 to 208 nm [20]. The salting-out process is another method that does not use chlorinated solvents. Using this technique, a water-in-oil emulsion is formed containing polymer, acetone, magnesium acetate tetrahydrate, stabilizer, and the active compound. Subsequently water is added until the volume is sufficient to allow for diffusion of the acetone into the

Table 2
Comparison of particle diameter for polymeric nanoparticles

Polymer	Drug	Size (nm)	Reference
PLGA	Doxorubicin	200	[33]
PLGA/PLA/PCL	Isradipine	110–208	[20]
PLGA	U-86983	144±37–88±41	[9]
PLGA	Rose Bengal	150	[40]
PLGA	Triptorelin	335–743	[34]
PLGA	Procaine hydrochloride	164±1.1–209.5±2.7	[18]
PLGA-mPEG	Cisplatin	133.5±3.7–163.3±3.6	[49]
PLGA	U-86983	70–160	[42]
PLGA	Insulin	>1 μm	[15]
PLGA	Hemagglutinin	~250	[6]
PLGA	Haloperidol	800	[10]
PLGA	Estrogen	~100	[16]
PEO-PLGA	Paclitaxel	150±25	[8]
PLA	Tetanus toxoid	>200	[37]
PLA	Savoxepine	~300–700	[21]
PLA	PDGFRβ tyrphostin inhibitor	123±23	[41]
PLA-PEG-PLA	Progesterone	193–335	[48]
PECA	Amoxicillin	320±12	[31]
Poly(butyl cyanoacrylate)	Dalargin	250	[29]
Chitosan	Cyclosporin A	283±24–281±05	[47]

Size is in nm, unless otherwise indicated.

water, which results in the formation of nanoparticles. This suspension is purified by cross-flow filtration and lyophilization [21]. However, one disadvantage to this procedure is that it uses salts that are incompatible with many bioactive compounds.

In most published techniques, nanoparticles are synthesized from the biocompatible polymers. However, it is possible to make biodegradable nanoparticles from monomers or macromonomers by polycondensation reactions [22,23]. These processes also result in sizes ranging from 200 to 300 nm. Nanoparticles can also be made from hydrophilic polysaccharides like chitosan (CS). CS-nanoparticles can be formed by the spontaneous ionic gelatin process [12,24]. CS-poly(acrylic acid) nanoparticles have also been made by polymerization of acrylic acid and the 'dropping method' [25]. The resulting nanoparticles have small sizes and positive surface potentials. This technique is promising as the particles can be prepared under mild conditions without using harmful organic solvents

The production of nanoparticles has several independent variables. One key parameter is type of surfactant/stabilizer to use. A wide range of synthetic and natural molecules with varying properties has been proposed to prepare nanoparticles. Feng et al. [11] has investigated the use of phospholipids as a natural emulsifier. In their study, dipalmitoyl-phosphatidylcholine (DPPC) improved flow and phagocytal properties due to a denser packing of DPPC molecules on the surface of the nanoparticles leading to a smoother surface than particles made with the synthetic polymer, poly(vinyl alcohol) (PVA). DPPC also improved the encapsulation efficiency compared to PVA using the emulsification solvent evaporation method. In a different study conducted by Kwon et al. [16], PLGA nanoparticles prepared using didodecyl dimethyl ammonium bromide (DMAB) were smaller than particles prepared with PVA [16]. Lemoine et al. [6] found that the presence of PVA in the inner aqueous phase produced smaller particles than Span® 40 [6]. When Pluronic has been used a stabilizer, the grade used can have a distinct effect on the size of the nanoparticles. For example, particles prepared with Pluronic F68 were smaller than particles prepared with Pluronic F108 [26].

The amount of stabilizer used will also have an effect on the properties of the nanoparticles. Most importantly, if the concentration of the stabilizer is too low, aggregation of the polymer droplets will occur and little if any nanoparticles will be recovered. Alternatively, if too much of the stabilizer is used, the drug incorporation could be reduced due to interaction between the drug and stabilizer. However, when the stabilizer concentration is between the 'limits', adjusting the concentration can be a means of controlling nanoparticle size. For example, using the solvent evaporation technique, increasing the PVA concentration will decrease the particle size [6,9]. However, when using the emulsification diffusion method, Kwon et al. [16] found a that a PVA concentration from 2 to 4% was ideal for creating smaller nanoparticles, ~100 nm in diameter.

Another factor that can affect the nanoparticles properties is the final freeze-drying process. It has been reported that additives such as saccharides are necessary for cryoprotection of the nanoparticles in the freeze-drying process [27]. These saccharides may act as a spacing matrix to prevent particles aggregation. Because of the possibility of aggregation, freeze-drying procedure can

affect the 'effective' nanoparticle size and consequently their release behavior and accordingly the drug pharmacokinetics [28].

The polymer used to formulate the nanoparticles will also strongly affect the structure, properties and applications of the particles. As previously stated, PLGA has been the most common polymer used to make biodegradable nanoparticles, however, these are clearly not the optimal carrier for all drug delivery applications. For each application and drug, one must evaluate the properties of the system (drug and particle) and determine whether or not it is the optimal formulation for a given drug delivery application. For example, poly(butyl cyanoacrylate) nanoparticles have been successful in delivering drugs to the brain [29]. Other cyanoacrylate-based nanoparticles such as polyalkylcyanoacrylate (PACA) and polyethylcyanoacrylate (PECA), have also been prepared. They are considered to be promising drug delivery systems due to their mucoadhesive properties and ability to entrap a variety of biologically active compounds. These polymers are biodegradable, biocompatible, as well as compatible with a wide range of compatible drugs [26,30]. Furthermore, these polymers have a faster degradation rate than PLGA, which in some cases may be more desirable. PECA nanoparticles have been prepared by emulsion polymerization in the presence and absence of different molecular weight poly(ethylene glycol) (PEG), using Pluronic F68 as the stabilizer [31].

Other groups have successfully prepared nanoparticles from functionalized PLGA polymers. In one study, Jung et al. [32] synthesized nanoparticles made of a branched, biodegradable polymer, poly(2-sulfobutyl-vinyl alcohol)-g-LGA [32]. The purpose of using sulfobutyl groups attached to the hydrophilic backbone was to provide a higher affinity to proteins by electrostatic interactions that would favor adsorptive protein loading. Adjustments can be made to the characteristics nanoparticles by differing degrees of substitution of sulfobutyl groups. In another case, a carboxylic end group of PLGA was conjugated to a hydroxyl group of doxorubicin and formulated into nanoparticles. This modification produced a sustained release of the drug that was approximately six times longer than with unconjugated drug [33].

The molecular weight and concentration of the polymer used will also affect the nanoparticles. The molecular weight of the polymer has opposite effects on nanoparticle size and encapsulation efficiency. Smaller size nanoparticles, approximately 100 nm, can be prepared with lower molecular weight polymer, however, at the expense of reduced drug encapsulation efficiency. On the other hand, an increase in polymer concentration increases encapsulation efficiency and the size of the nanoparticles [7,9,16].

When considering a particular polymeric nanoparticle for a given drug delivery application, particle size and encapsulation efficiency are two of the most important characteristics of nanoparticles. One should determine what the goal of the nanoparticle delivery system is before determining the size desired. For example if the goal is rapid dissolution in the body or arterial uptake then the size of the nanoparticles should be approximately 100 nm or less. If prolonged dissolution is required, or targeting the mononuclear phagocytic system (MPS), larger particles around 800 nm would be preferable. A comparison of various drugs encapsulated and the resulting sizes of the particles are summarized in Table 2. From examination of these data, it appears that the encapsulation efficiency increases with the diameter of the nanoparticles. In one study, the encapsulation efficiency was maximized in the double emulsion solvent evaporation technique when the pH of the internal and the external aqueous phases were brought to the isoelectric point of the peptide being encapsulated, methylene chloride was used as a solvent, and the PLGA was rich in free carboxylic end groups [34].

Another characteristic of polymeric nanoparticles that is of extreme interest is zeta potential. The zeta potential is a measure of the charge of the particle, as such the larger the absolute value of the zeta potential the larger the amount of charge of the surface. In a sense, the zeta potential represents an index for particle stability. For the case of charged particles, as the zeta potential increases, the repulsive interactions will be larger leading to the formation of more stable particles with a more uniform size distribution. A physically stable nanosuspension solely stabilized by electrostatic repulsion will have a minimum zeta potential of ± 30 mV [35]. This stability is important in preventing aggregation. When a surface modification is added like PEG the negative zeta potential is lowered, increasing the nanoparticles stability [12].

3. Surface modification

Before deciding which of the techniques to use for synthesizing nanoparticles, one must consider what the nature of the drug used as well as the means and duration desired for the delivery. That will determine not only how the particles are synthesized but also what the nature of the particles should be. In particular, the body recognizes hydrophobic particles as foreign and thus they are rapidly taken up by the MPS. However, if sustained systemic circulation is required than the surface of the hydrophobic nanoparticles must be modified in order prevent phagocytosis.

Following intravenous administration, hydrophobic nanoparticles are rapidly cleared from the systemic circulation by the MPS, ending in the liver or the spleen. If the goal is to treat a condition in the liver, then the proper choice for the application would be a hydrophobic nanoparticle. While it would appear that the hydrophobic nature of most biodegradable particle would limit the applicability of these carriers in many drug delivery applications, one may overcome concerns of clearance by

the MPS through surface modification techniques. The goal of these modification techniques is to produce a particle that is not recognized by the MPS due to the hydrophilic nature of the surface.

Several types of surface modified nanoparticles that have been described in recent literature are summarized in Table 3. The most common moiety used for surface modification is poly(ethylene glycol) (PEG). PEG is a hydrophilic, nonionic polymer that has been shown to exhibit excellent biocompatibility. PEG molecules can be added to the particles via a number of different routes including covalent bonding, mixing in during nanoparticle preparation, or surface adsorption. The presence of a PEG-brush on the surface of nanoparticles can serve other functions besides increasing residence time in the systemic circulation. For one, PEG tethers on the particle surface can reduce protein and enzyme adsorption on the surface, which for PLGA based particles will retard degradation. The degree of protein adsorption can be minimized by altering the density and molecular weight of PEG on the surface [36]. The stability of PLA particles has been shown to increase in simulated gastric fluid (SGF) with the addition of PEG on the particle surface. After 4 h in SGF, 9% of the PLA nanoparticles converted to lactate versus 3% conversion for PEG-PLA particles [37]. PEG is also believed to facilitate transport through the Peyer's patches of the GALT [12].

As stated previously though, the primary reason for interest in preparing PEG functionalized particles is to improve the long-term systemic circulation of the nanoparticles. The PEG functionalized particles are not seen as a foreign body and are therefore not taken up by the body, allowing them to circulate longer providing for a sustained systemic drug release. Because of their behavior these PEG functionalized nanoparticles are often called 'stealth nanoparticles' [38]. Furthermore, it has been determined that PEG MW is important with respect to MPS uptake. For example, Leroux et al. [21] showed that an increase in PEG molecular weight in PLGA nanoparticles was associated with less interaction with the MPS, and longer systemic circulation. Also, PEG-containing PLGA nanoparticles synthesized by Li et al. [14] were able to extend the half life of BSA in a rat model to 4.5 h from 13.6 min [14]. Another study compared the dosages of PLGA nanoparticles versus PEG-PLGA nanoparticles. The PLGA nanoparticles pharmacokinetics seemed to depend on MPS saturation. However, the pharmacokinetics of PEG-PLGA dosages did not exhibit the same dependence on dosage/MPS saturation due to their stealth nature [39].

Poloxamer and poloxamines have also been shown to reduce capture by macrophages and increase the time for systemic circulation. Similarly PLGA particles coated with poloxamer 407 and poloxamine 908 extended the half life of rose bengal, a hydrophilic model drug, with ~30% left in the bloodstream after 1 h post nanoparticle administration, as opposed to 8% present after 5 min post free drug administration [40].

Another polymer used for surface modification is chitosan. The addition of CS to the surface of PLGA nanoparticles, resulted in increased penetration of macromolecules in mucosal surfaces [24]. CS coated PLGA particles were able to increase the positive zeta potential of the particles and increase the efficiency of tetanus toxoid protein encapsulation. Radiolabelled tetanus toxoid was used to show the enhanced transport across nasal and intestinal epithelium using CS coated particles versus uncoated particles, with a higher percentage of ^{125}I present in the lymph nodes for CS coated particles [12].

4. Targeted drug delivery using nanoparticles

Another exciting application of surface modified particles is targeted drug delivery to tumors or organs. Kreuter et al. [29] were able to deliver several drugs successfully through the blood brain barrier using polysorbate 80 coated poly(butylcyanoacrylate) nanoparticles [29]. It is thought that after administration of the polysorbate 80-coated

Table 3
Comparison of nanoparticles modified with the addition of polymers to the surface

Polymer	Surface modification	Size (nm)	Reference
PLGA	Poloxamine 908	~160	[40]
PLGA	Poloxamer 407	~160	[40]
PLGA	Chitosan	500±29	[12]
PLGA-mPEG	mPEG	133.5±3.7–163.3±3.6	[49]
PLGA-mPEG	mPEG	113.5±14.3	[39]
PLGA-PEG	PEG	198.1±11.1	[14]
PLA	PEG	164–270	[36]
PLA	PEG 6000	295	[21]
PLA-PEG	PEG	>200	[37]
PLA-PEG	PEG	~130	[45]
PHDCA	PEG	~150	[38]
PECA	PEG	220±10–280±8	[31]
PBCA	Polysorbate 80	250	[29]

particles, apolipoprotein E (ApoE) adsorbs onto the surface coating. The ApoE protein mimics low density lipoprotein (LDL) causing the particles to be transported via the LDL receptors into the brain.

There are other specific areas where nanoparticle administration may have an advantage over microparticulate-based drug delivery systems. One area that has been of recent interest is in prevention of restenosis. Restenosis is a major postoperative concern following arterial surgery. In order to inhibit vascular smooth muscle cell proliferation, drugs must be delivered at a high concentration over a long period of time. Nanoparticles offer an advantage because the medication would not have to be delivered systemically as they are small enough for cellular internalization, and connective tissue permeation. Several types of drugs including antiproliferative agents have been used to test this method of delivery. PLA nanoparticles were loaded with platelet-derived growth factor receptor β tyrphostin inhibitor and delivered intraluminally to an injured rat carotid artery [41]. The drug had the desired effect of preventing restenosis, but of significance was the absence of drug in other areas of the arteries and systemic circulation. Song et al. [42] found that specific additive after nanoparticles formation, such as heparin, DMAB, or fibrinogen, could enhance arterial retention of the particles. Suh et al. [8] created poly(ethylene oxide)-PLGA nanoparticles which had an initial burst release of 40% of the antiproliferative drug in the first 3 days [8]. However, a total of 85% of the drug was released after 4 weeks. This shows that nanoparticles have a great potential for long term arterial drug delivery.

Much attention has also been given to lymphatic targeting using nanoparticles. The lymphatic absorption of a drug via the GALT has an advantage over a portal blood route since it avoids any liver pre-systemic metabolism, known as the first pass effect. This could be beneficial for anticancer treatment, mucosal immunity, as well as the potential for staining the lymph nodes prior to surgery [43]. Nanoparticles can also be used to carry antisense oligonucleotides and plasmid DNA that can be used to treat some forms of cancer and viral infections, as well as a new vaccination approach. Antisense oligonucleotides normally have poor stability and cannot easily penetrate cells, but are easily encapsulated in nanoparticles [44,45].

Nanoparticles have also had some success as a new delivery vehicle for vaccines. CS-nanoparticles have been successful as a nasal vaccine in some animal studies producing significant IgG serum responses and superior IgA secretory responses when using influenza, pertussis, and diphtheria vaccines [46].

Attention has been given to the absorption via the intestinal tract because of its availability to the lymphatic system. However, high doses of antibiotics and antiparasitics are given to treat gastrointestinal bacteria and parasites because only 10–15% of the drug administered is absorbed [35]. The increased mucoadhesivity of nanoparticles could be effective in treating these pathogens with lower doses of drugs.

The lungs are another target area for nanoparticles due to their large surface area, good mucosal permeation, well-developed vascular system, thin alveolar walls, and low activity of drug metabolizing enzymes. When CS coated nanoparticles were administered via the lungs, there were detectable blood levels of the drug 24 h after administration, as opposed to 8 h for the noncoated particles [17].

Nanoparticles have been used to target mucosal surfaces. Long-term extraocular (cornea and conjunctiva) drug delivery with nanoparticles provides an improvement in conventional drug delivery in this region. It was possible to deliver drug loaded CS-nanoparticles to the extraocular structures over the course of 48 h at higher levels than with free drug, without exposing the inner ocular structures (iris and aqueous humour) to the drug [47].

5. Release characteristics

The release characteristics of polymeric nanoparticles are one of the most important features of the drug/polymer formulations because of the proposed application in sustained drug delivery. There are several factors that affect the release rate of the entrapped drug. Larger particles have a smaller initial burst release and longer sustained release than smaller particles. In addition, the greater the drug loading the greater the burst and the faster the release rate. For example, PLA nanoparticles containing 16.7% savoxepine released 90% of their drug load in 24 h, as opposed to particles containing 7.1% savoxepine, which released their content over 3 weeks [21]. The initial burst release is thought to be caused by poorly entrapped drug, or drug adsorbed onto the outside of the particles. When using polymers, which interact with a drug, like PLGA with a free COOH group and proteins, the burst release is lower and in some cases absent, and drug release is prolonged [7,34].

The addition of other polymers to PLA based polymers can also be used to control drug release. For example, PEG has been polymerized into a PLA homopolymer creating a PLA-PEG-PLA copolymer [48]. The amount of the drug, in this case progesterone, released increased with the PEG content and the molecular weight of the copolymers. The drug release continued to increase as the total molecular weight of the copolymers decreased. The initial burst was decreased in the absence of lower molecular weight polymers. The content of PEG in the copolymer affected the size of the particles as well as the degradation of the polymers. Similar effects were seen with PLGA-mPEG nanoparticles loaded with cisplatin [49]. Consequently it would be possible to alter the release rate of the drug by changing the amount of PEG in the copolymer as well as the molecular weights of the polymers.

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