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Effects of Drug and Polymer Molecular Weight on Drug Release from PLGA-mPEG Microspheres

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ABSTRACT: This study investigated the effects of drug and polymer molecular weight on release kinetics from poly (G-*co*-glycolic acid)-methoxypoly(ethyleneglycol) (PLGA-mPEG) microspheres. Bovine serum albumin (BSA, 66 kDa), lysozyme (LZ, 13.4 kDa), and vancomycin (VM, 1.45 kDa) were employed as the model drugs, and encapsulated in PLGA-mPEG microspheres of different molecular weight. Release of macromolecular BSA was mainly dependent on diffusion of drug at/ near the surface of the matrix initially and dependent on degradation of matrix at later stages, while, the small drug of vancomycin seemed to depend totally on diffusion for the duration of the release study. The release behavior of lysozyme was similar to bovine serum albumin, except a shorter lag period. PLGA-mPEG molecular weight also affected the release behavior of bovine serum albumin and lysozyme, but not obviously. PLGA-mPEG microspheres in smaller molecular weight seemed to degrade more quickly to obtain a mass lose and matrix erosion, and thus, an accelerated release rate of bovine serum albumin and lysozyme. Vancomycin released much faster than bovine serum albumin and lysozyme, and exhibited no lag period, as it is thought to be diffusion-controlled. Besides, vancomycin showed no difference in release behavior as PLGA-mPEG molecular weight change. © 2014 Wiley Periodicals, Inc. J. Appl. Polym. Sci. **2015**, *132*, 41431.

KEYWORDS: biodegradable; biomaterials; drug delivery systems

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INTRODUCTION

Polymeric microspheres have been widely utilized as a favorable tool in delivering various cytokines and proteins.[1–3] Biodegradable polyesters such as poly (lactide-*co*-glycolide) (PLGA), poly (lactic acid) (PLA) and poly (ε–caprolactone) (PCL) have been widely used as carriers in controlled-release delivery systems due to their biocompatibility and biodegradability.[3,4] Their degradation time can be varied from days to years by altering the type of polymer, the polymer molecular weight, or the structure of the microspheres.[5] Nevertheless, PLGA and PLA microspheres showed a low encapsulation efficiency of hydrophilic peptides and proteins due to their hydrophobic nature. Furthermore, during the initial drug release, the hydrophobic PLGA or PLA prevents the penetration of water into the center of microspheres, forming an acidic environment due to the accumulated acidic breakdown products, which can cause a degeneration of peptides or proteins.[6] Methoxypoly (ethylene glycol) (mPEG) segment is a hydrophilic part that can change the physicochemical properties of hydrophobic PLGA block segment and has been widely used to improve the biocompatibility of the blood contacting materials.[7] The mPEG chains of PLGA-mPEG acting as a surface modifier of hydrophobic PLGA network, could enhance the permeation of water into the center of microspheres thus increase the polymer degradation rate, reduce the acidic microenvironment because of the diffusion of acidic breakdown products, and accelerate the diffusion of drug in the matrix.[8] Diblock PLA-mPEG and PLGA-mPEG form more hydrophilic matrices than PLGA and are considered more suitable for the controlled delivery of proteins[9,10] and hydrophobic small drugs.[11] However, few studies had focused on the relationship between release behavior of water-soluble drug and PLGA-mPEG microspheres degradation, which may partly different from PLGA microspheres.

Macromolecular drug, as proteins and peptides, were preferred as water-soluble model drug for various microspheres release analysis[2,12] and the two main release mechanisms associated with drug release from PLGA microspheres were thought to be diffusion and degradation/erosion. However, few literatures concentrated on the release kinetics of small water-soluble drugs had been reported,[13] which may different from macromolecular drugs. The purpose of this article was to investigate the effects of drug and polymer molecular weight (Mw) on release kinetics from PLGA-mPEG microspheres. Bovine serum albumin (66 kDa), lysozyme (13.4 kDa), and vancomycin (1.45 kDa) are

employed as model drugs and encapsulated in PLGA-mPEG (14,573 Da, 24,914 Da, 36,647 Da) microspheres by the double emulsion–evaporation method. The drugs release rate are analyzed by BCA assay kit and UV-vis spectrophotometry, and the corresponding physicochemical changes of polymer molecular weight, mass loss and surface morphology of the microspheres are also investigated to analyze PLGA-mPEG degradation behavior. The effects of drug molecular weight and polymer degradation on drug release behavior are adequately discussed.

MATERIALS AND METHODS

Materials

Glycolide (GA), DL-Lactide (LA) and monomethoxypoly (ethylene glycol) (mPEG, $M_n = 5000$) were purchased from Aldrich. Bovine serum albumin, lysozyme, and Vancomycin were obtained from Sigma. Poly (vinyl alcohol) (PVA) from China National Medicines Corporation was used as a stabilizer in the emulsion. Methylene chloride and diethyl ether, as solvents, were of analytical grade and purchased from China National Medicines Corporation, and used without purification.

Synthesis and Characteristic of Copolymers PLGA-mPEG

Poly (DL-lactide-*co*-glycolic acid)-methoxypoly(ethyleneglycol) (PLGA-mPEG) copolymers of different composition (PLGA: mPEG molar ratio) were synthesized by bulk ring-opening polymerization method using mPEG as the macro-initiator and $Sn(Oct)_2$ as the catalyst.[14] Before the synthesis, LA and GA were recrystallized twice in ethyl acetate and sublimated. MPEG was lyophilized for 24 h in case of moisture. All glasses were heated in vacuum before use. The typical process for the polymerization is as follows: mPEG-5000 was stirred at 115°C in a three-necked flask under the protection of nitrogen for 30 min. LA and GA at a molar ratio of 3 : 1 were added and heated at 115°C to make them melted, then, 0.04 g of $Sn(Oct)_2$ were added and the reaction mixture was further heated at 125°C for 8 h under the protection of nitrogen. The following three copolymers were synthesized: (1). PLGA (10)-mPEG (5) with the composition LA : GA = 3 : 1, (2). PLGA (20)-mPEG (5) with the composition LA : GA = 3 : 1, (3). PLGA (35)-mPEG (5) with the composition LA : GA = 3 : 1. The synthesized polymer was purified by dissolving in dichloromethane followed by precipitation in diethyl ether. The precipitate was lyophilized under vacuum for 24 h.

The identity of the copolymers was examined by Gel permeation chromatography (GPC), infrared (IR) spectra, ^{1}H-nuclear magnetic resonance (^{1}H-NMR) and ^{13}C-nuclear magnetic resonance (^{13}C-NMR) spectroscopy. GPC in an Agilent 1100 apparatus with a differential refractometer as a detector was used to measure the molecular weight (M_W) of the polymers. The IR spectra were taken in a Bruker VERTEX 70 infrared spectrometer over the range 4000–500 cm^{-1}. KBr discs, containing 1% (w/w) of the copolymers being examined. ^{1}H-NMR (Bruker AV 400) spectrometer was used to characterize the chemical compositions of the copolymers with $CDCl_3$ as the solvent and TMS as standard.

Preparation of PLGA-mPEG Microspheres

Microspheres were prepared using the double emulsion-evaporation method.[15] First, 0.1 mL drug (BSA, LZ, and VM) solution (100 mg/mL) was added to 4 mL of dichloromethane containing 0.25 g PLGA-mPEG, the mixture was homogenized at 14,500 rpm for 10 s to obtain the primary emulsion. The primary emulsion was then, injected into 10 mL of 0.25% w/v PVA solution and emulsified at 8500 rpm for 10 s, creating the second emulsion. Next, the second emulsion was dispersed into 120 mL of 0.25% w/v PVA solution under magnetic stirring at 600 rpm for 3 h to evaporate the dichloromethane. The microspheres were collected by centrifugation at 3000 rpm and washed three times with distilled water. Subsequently, the microspheres were lyophilized and stored at 4°C. Besides, unloaded PLGA-mPEG microspheres in different M_w were also prepared under the same conditions for degradation analysis.

Characteristics of the Microspheres

The surface morphologies of microspheres in different M_w were observed using a scanning electron microscope (SEM, Quanta 200, Holland, FEI). Microspheres were mounted onto metal stubs using a double-sided adhesive tape. After vacuum-coated with a thin layer of gold, the microspheres were examined by SEM at 15 kV. For the mean size and size distribution analysis, 300 microspheres in each group were randomly chosen from the SEM micrographs and the software (Nano Measurer 1. 2) was applied to counting the size distribution.

The BSA and LZ encapsulation efficiencies of the microspheres were measured by the BCA assay,[16] and the VM encapsulation efficiency was measured by UV-vis spectrophotometry. Briefly, 10 mg of dried microspheres was dissolved in 1 mL of methylene chloride under stirring and 3 mL of PBS (pH 7.4, 0.01% sodium azide, 0.02% Tween 80) was then added. The mixture was vigorously agitated for 5 min to extract drug into PBS from the organic solution. After centrifuging, the aqueous phase was withdrawn and the amount of drug was analyzed using the BCA assay or UV-vis spectrophotometer. The drug encapsulation efficiency was expressed as follows:

Encapsulation efficiency (%) = (Actual drug: PLGA-mPEG ration)/(Initial drug: PLGA-mPEG ration) * 100

All the experiments were run in triplicate and data are shown as mean ± standard deviation.

In Vitro Microspheres Degradation Analysis

Unloaded microspheres in different Mw were applied in degradation study as it was found that unloaded microspheres exhibited a same degradation behavior with that drug-loaded.[13] The degradation behavior of the microspheres was evaluated by the molecular weight, microspheres mass reduction and microspheres morphology change with time upon their in vitro incubation in PBS (pH 7.4, 0.01% sodium azide, 0.02% Tween 80) at 37°C. Totally, 30 mg unloaded microspheres (eight samples for each molecular weight) fabricated in same conditions with drug-loaded were applied for degradation analysis and was incubated in 5 mL PBS under continuous shaking (50 rpm). As in the drug release experiments, mediums were removed at various time points for drug contents analysis. In the degradation study, all of the tubes were centrifuged at each time point and the same amount of supernatant was discarded to remove any acidic degradation products which would contribute to

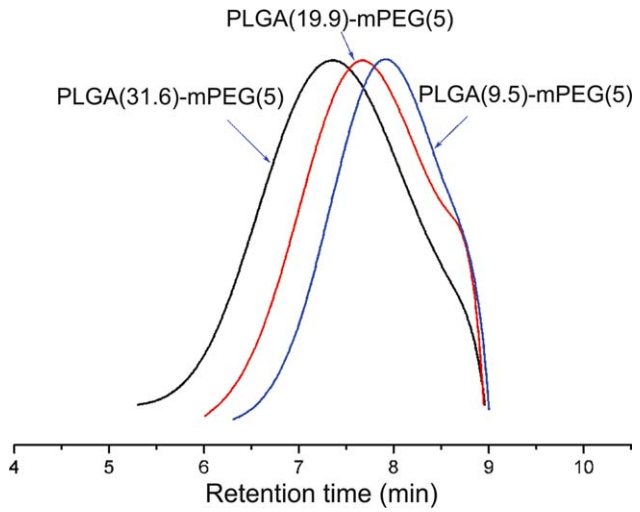

Figure 1. GPC traces of PLGA-mPEG with different molecular weight. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

auto-catalytic degradation of PLGA-mPEG. If the samples were to continue degradation, fresh medium was added back to the tube, if not, the remaining microspheres were lyophilized, and the final dry weight of the microspheres was compared to the original sample weight. Dried PLGA-mPEG microspheres for the mass lose study were also used to determine the molecular weight and molecular weight distribution of the degrading microspheres by GPC. The microspheres mean size changes during the degradation were also determined by the method described above.

In Vitro Drug Release Analysis

The in vitro drug release test was conducted by suspending microspheres in PBS (pH 7.4, 0.01% sodium azide, 0.02% Tween 80) and maintained at 37°C under continuous shaking (50 rpm). In triplicate, 30 mg of microspheres were suspended in 5 mL PBS and installed in centrifuge tubes. The tubes were then sealed and placed in a shaking water bath. At scheduled time intervals, the tubes were taken out and centrifuged, then, 3 mL of supernatant were withdrawn to determine the amount of drug released and were replenished by the same volume of fresh medium. The concentrations of BSA and LZ were measured by BCA assay kit and VM concentration was determined by UV-vis spectrophotometry.

RESULT AND DISCUSSION

Characteristic of Copolymers PLGA-mPEG

The PLGA-mPEG block copolymers were synthesized by the ring opening polymerization of DL-Lactide (LA) and glycolide (GA) using monomethoxy poly(ethylene glycol) (mPEG) (M_n =5000 g mol^{-1}) as a macroinitiator, in the presence of a catalytic amount of Sn(Oct)$_2$. The GPC analysis showed the synthesized PLGA-mPEG Mws were 14,573, 24915, and 36,647, respectively (Figure 1). The chemical structure of the PLGA-mPEG diblock copolymer was investigated via FTIR (Figure 2), ^{13}C-NMR and ^{1}H-NMR spectroscopy (Figure 3). The IR absorption peaks at 1748 and1082 cm^{-1} are assigned to a carbonyl (C═O) stretching mode of the PLGA and an ether

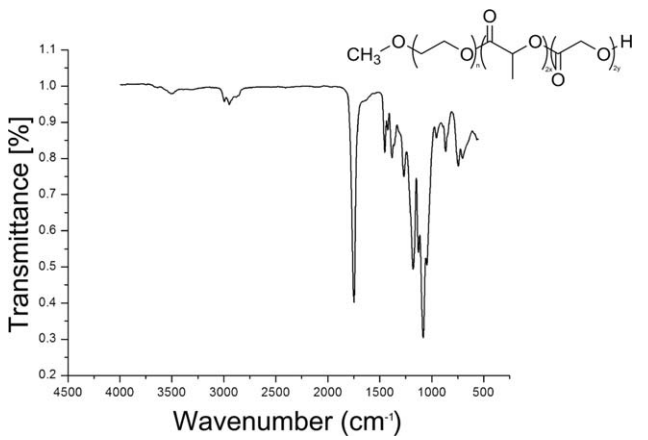

Figure 2. IR spectra of PLGA-mPEG, over the range 4000–500 cm^{-1}.

(C—O) bending mode of the mPEG and the PLGA ester, respectively. Also, the spectrum showed other peaks which mPEG-PLGA should had. The ^{1}H-NMR spectrum of the PLGA-mPEG block copolymer shows the terminal methoxy proton

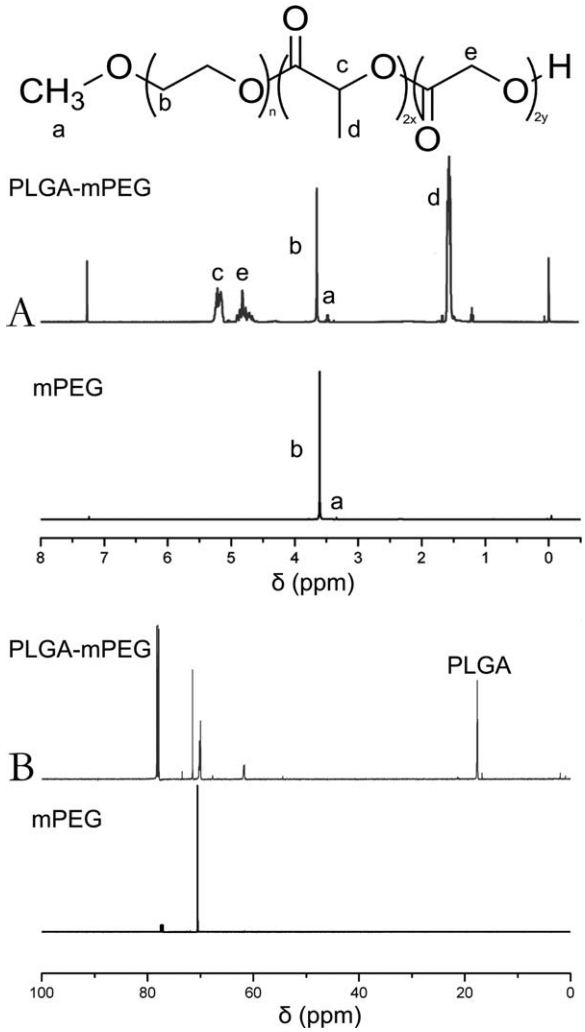

Figure 3. ^{1}H-NMR (A) and ^{13}C-NMR (B) spectrums of mPEG and copolymer in $CDCl_3$.

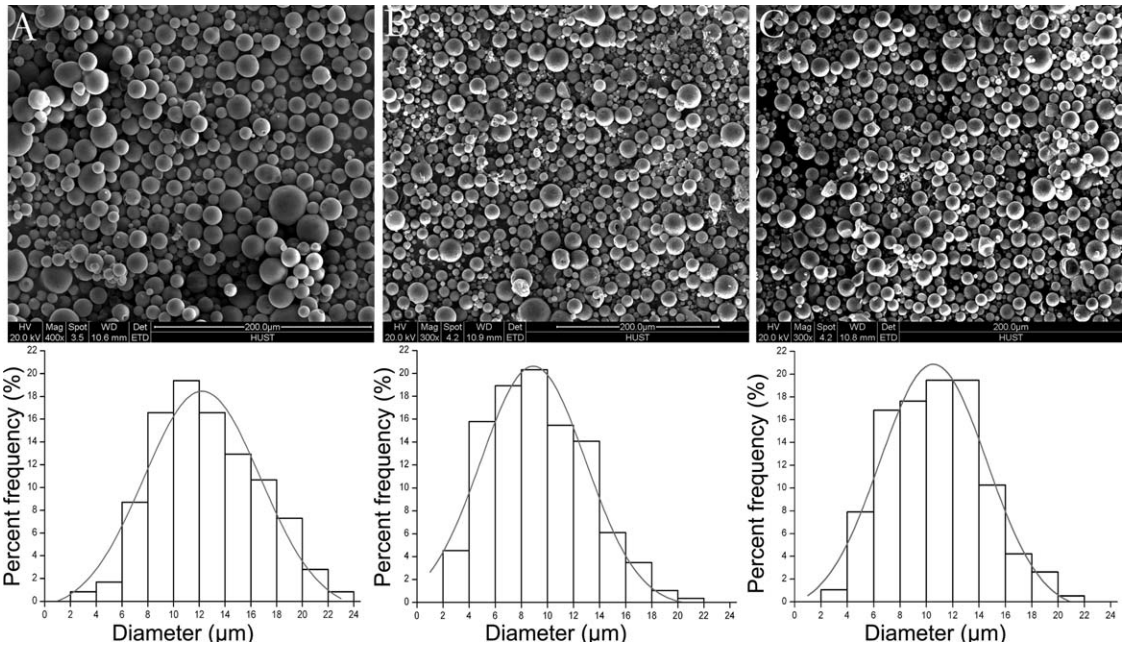

Figure 4. Scanning electron micrographs of BSA-loaded microspheres of different molecular weights and corresponding microspheres size distributions: (A) PLGA(9.5)-mPEG(5) microspheres; (B) PLGA(19.9)-mPEG(5) microspheres; (C) PLGA(31.6)-mPEG(5) microspheres.

signal of mPEG at $\delta = 3.36$ ppm and the methylene proton signal at $\delta = 3.65$ ppm as reported.[17,18] The methoxy proton signal of LA units in the PLGA segments was shown at $\delta = 1.55$ ppm and the methyne proton signal at $\delta = 5.18$ ppm. The methylene proton signal of GA units in the PLGA segments was shown at $\delta = 4.82$ pp. In comparison with the ^{1}H-NMR spectrum of mPEG, new proton signals which belonged to LA and GA appeared, indicating the successful synthesis of PLGA-mPEG block copolymer. ^{13}C-NMR spectrum also revealed that the copolymer get an additional signal at $\delta = 18.5$ ppm comparing with the ^{13}C-NMR spectrum of mPEG, which belonged to PLGA according to the available reports.[19]

Characteristic of PLGA-mPEG Microspheres

Figure 4 showed the morphology of BSA-loaded PLGA-mPEG microspheres in different Mw and corresponding microspheres size distribution. The SEM pictures revealed a similar surface morphology of microspheres in different M_w, with most microspheres appearing as spherical dense balls and low broken percentage. The size distribution analysis also showed a similar result between the three groups of microspheres, with an average size around 10 μm. LZ- and VM-loaded microspheres morphology and size distribution were similar to BSA microspheres (pictures not shown), as they were fabricated under the same conditions.

Table I. Encapsulation Efficiencies of Drugs in PLGA-mPEG Microspheres in Different Molecular Weight

Drug	PLGA (9.5)-mPEG(5)	PLGA(19.9)-mPEG(5)	PLGA(31.6)-mPEG(5)
BSA	63.6 ± 2.13%	53.12 ± 0.73%	52.75 ± 0.92%
LZ	21.92 ± 1.63%	14.15 ± 0.66%	16.77 ± 1.07%
VM	53.48 ± 2.34%	32.00 ± 1.97%	29.13 ± 1.63%

Encapsulation efficiencies were found to be between 52.8 and 63.6% for BSA in PLGA-mPEG microspheres of various M_w, much higher than 16.8 and 21.9% for LZ (shown in Table I), which may due to the higher M_w of BSA, thus, a bigger space block effect that hindered its escape to outer water during solvent evaporation and polymer precipitation process. However, VM showed higher encapsulation efficiency than LZ though it had the smallest Mw, which may be ascribed to the LZ denaturation caused by the organic solvent during the fabrication process. PLGA (9.5)-mPEG(5) microspheres showed a little higher encapsulation efficiency of all the three types of drugs than PLGA (19.9)-mPEG(5) and PLGA (31.6)-mPEG(5), which may due to its higher proportion of mPEG in the polymer chains, thus a increased hydrophilicity, which can hindered the hydrophilic drug escape to outer water more efficiently.

In Vitro Microspheres Degradation Analysis

Maryellen Sandor reported that encapsulated drug does not affect degradation or erosion of microspheres.[13] Unloaded microsphere results were, therefore, taken to be representative of all types of microspheres examined in this study. The M_w of all types of PLGA-mPEG decreased drastically with degradation time, as shown in Figure 5. Within an incubation period of 1 week, significant M_w reduction of the microspheres occurred, 68.81% of original M_w remained for PLGA (9.5)-mPEG(5) and 58.79%, 47.69% for PLGA(19.9)-mPEG(5) and PLGA(31.6)-mPEG(5). Obviously, the M_w reduction rate was more rapid as PLGA-mPEG M_w increased according to the data, which could probably be explained by the more hydrolytic cleavable ester groups in the polymer chain of higher M_w, and thus, more rapid random hydrolytic cleavage. However, despite the rapid

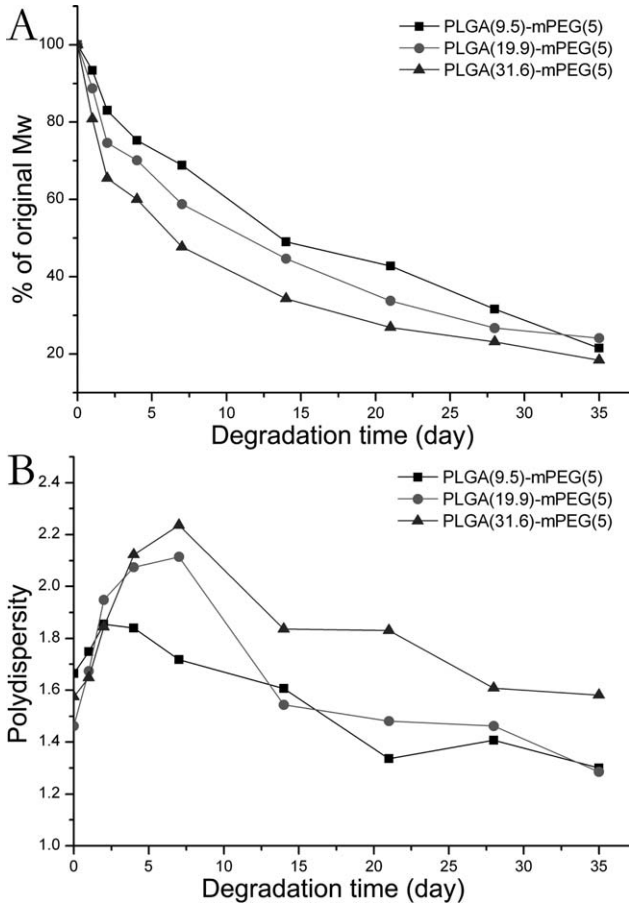

Figure 5. A: Percent of original polymer molecular weight of unloaded PLGA-mPEG microspheres during degradation in PBS. B: Polydispersity change of unloaded PLGA-mPEG microspheres during degradation in PBS.

M_w reduction rate during the first week, mass loss from the microspheres were not obvious, only 4.5% of original loss for PLGA (9.5)-mPEG(5), and 1.4%, 1% for PLGA(19.9)-mPEG(5) and PLGA(31.6)-mPEG(5), respectively. It could probably due to the few soluble degradation products generated in the early period of incubation. The results showed that the rate of mass reduction may also depend on the polymer composition, increasing with an increase in the proportion of mPEG in the polymer chains (Figure 6), which may be attributed to their increased hydrophilicity that apparently overrides their decreased content of cleavable ester bonds.[20] In the latter stages of the degradation, the M_w reduction rate slowed down and decreased to a low value, while, the mass loss rate speed up at this time. It was probably due to the abundant soluble degradation products generated at this stage, which could be confirmed by the obvious erosion of microspheres, shown in Figure 7. This was consistent with the common hydrolysis rule that the weight loss would not take place until the M_w of samples had decreased to a critical value able to dissolve in water.[21] Microspheres in smallest M_w showed a relative more severe erosion in surface after 1 week degradation [Figure 7(d)], and its mass reduction rate accelerated after 4 days degradation according to the data, earlier than PLGA(19.9)-mPEG(5) and PLGA(31.6)-mPEG(5), showed in Figure 6. It meant that PLGA (9.5)-mPEG(5) microspheres degraded easier to reach the M_w critical value to dissolve in water, though it showed the slowest M_w reduction rate. All the three types of PLGA-mPEG microspheres kept a rapid mass loss rate after 2-week incubation, accompanied with a continuous erosion of microspheres, and degradation eventually proceeded to the point of eradication of any existing microspheres structure after 5 weeks, showed in Figure 7. The microspheres mean size decreases were similar with mass lose, accelerated after 2-week incubation, and PLGA(9.5)-mPEG(5) microspheres also showed a faster size decrease rate, shown in Figure 8. All the microspheres loss the structure after 5 weeks degradation, so we just analyzed the size changes among the first 4 weeks.

The molecular weight distribution (polydispersity) turned wider in the first stage of degradation, due to the accumulation of degradation products with M_w decrease; however, it turned narrower at the latter stage as a result of the loss of degradation products, as present in Figure 5.

In Vitro Drug Release Analysis

Three drugs commonly used as molecular weight markers, bovine serum albumin, lysozyme, and vancomycin, were encapsulated in PLGA-mPEG of different M_w in order to determine how drug molecular weight and polymer degradation affect release from microspheres. Drugs were released by suspending microspheres in PBS and maintained at 37°C under continuous shaking, and the results were shown in Figure 9. A typical triphasic release profile was observed during BSA and LZ release from PLGA-mPEG microspheres in different M_w, while, for VM, the smallest drug, a bi-phasic release profile was observed.

The initial burst release during the first day may be attributed to the fraction of drug content which was adsorbed or close to the surface of the microspheres. Upon addition of the microspheres to the release medium, this part of drug diffused rapidly into the surrounding liquid, accounting for the rapid initial part of the release profile. The smallest drug, VM, revealed a most severe burst release in microspheres of various M_w, around 50% of total encapsulated VM, much higher than the 10–20%

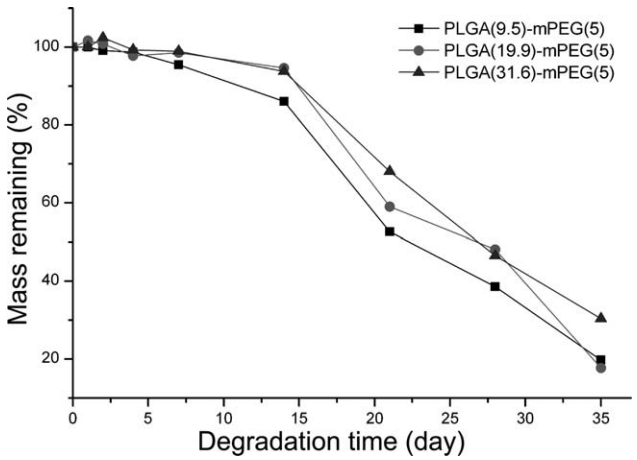

Figure 6. Percent of original total mass of unloaded PLGA-mPEG microspheres during degradation in PBS.

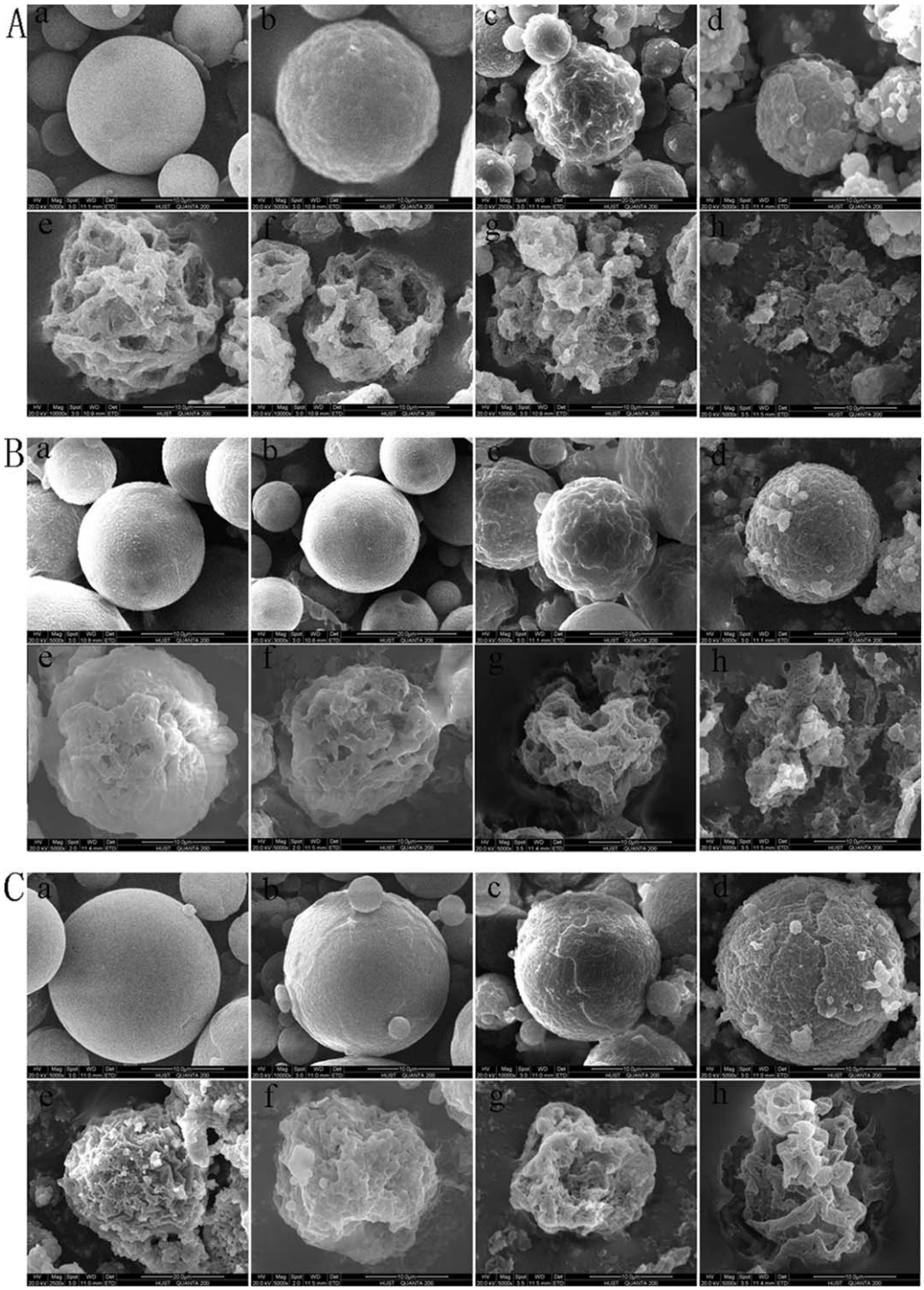

Figure 7. SEM images of PLGA-mPEG microspheres morphology change during degradation in PBS. A, PLGA (9.5)-mPEG(5) microspheres; B, PLGA (19.9)-mPEG(5) microspheres; C, PLGA (31.6)-mPEG(5) microspheres. a. original, b. 1 day, c. 4 days, d. 7 days, e. 14 days, f. 21 days, g. 28 days, h. 35 days.

burst release of other two drugs. It was assumed that VM could diffuse out faster and easier, and not only the external VM, but the internal part that may transport through the polymer phase and the small water-filled pore,[22] due to its small molecular volume. The initial burst release was assumed to be diffusion controlled[23] and in this study, we found that the amount of drug released during the burst release period had no obvious relationship with PLGA-mPEG M_w, shown in Figure 9(D–F). The second phase, called lag period, was only present for BSA and LZ. During this period, BSA and LZ diffused hardly either through the relatively dense polymer or through the few existing pore, which may due to their macromolecular volume. For

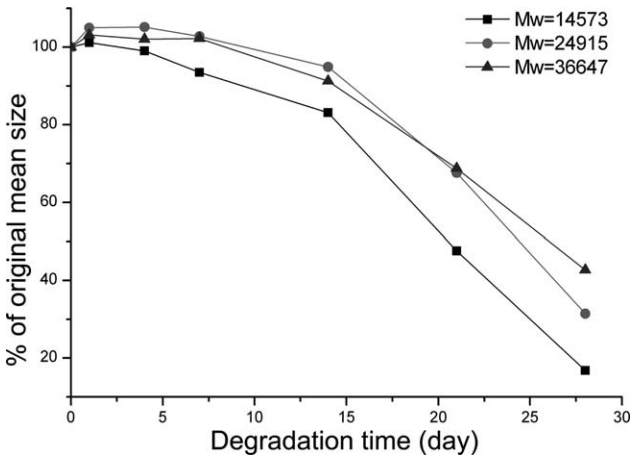

Figure 8. Percent of original mean size of unloaded PLGA-mPEG microspheres during degradation in PBS. Three hundred microspheres in each group were randomly chosen for the mean size analysis.

LZ, the lag period lasted ~1 day, while, BSA showed a prolonged lag period lasted ~6 days. It may be ascribed to their different Mw and LZ could partially diffused out after the massive swelling and the deformation of PLGA-mPEG microspheres,[24] due to its smaller M_w. VM-loaded microspheres didn't undergo this period, it kept a continuous release of the remaining drug after the burst release, which could probably be explained by its small molecular volume, thus, it can transport through the polymer phase and the small water-filled pores. Besides, VM exhibited a similar release pattern despite the change of PLGA-mPEG M_w [Figure 9(F)], further indicating that the VM release behavior was diffusion-controlled during the whole release period. The third phase, a quick additional spurt of the remaining drug following the lag period, occurred between 1 and 5 weeks for BSA. By the time between 1 and 2 weeks, accelerated mass reduction rate and severe erosion of microspheres were observed (Figures 6 and 7), creating and widening pores for drug diffusion, which should be the main reason accelerating the release, therefore, it was assumed that the BSA release was diffusion controlled initially but degradation controlled at latter stages. The results in Figure 9(d) showed that BSA release accelerated after 1 week for microspheres of different M_w based on the available data and since samples were not analyzed at times between 1 and 2 weeks. Maybe the third phase began later as the polymer M_w increased, indicated by the microspheres mass loss data and morphology changes during 1 week incubation, shown in Figures 6 and 7. LZ showed an advanced spurt release, which may due to the same reason for its shorter lag period, and it kept a fast release rate in the following 3 weeks until 90% of total drug were released, then, the release rate slow down for the remaining 10% drug. LZ also showed a faster release rate than BSA [Figures 9(A–C)], which should be also ascribed to its smaller molecular volume.

It was found that BSA and LZ released a little faster in PLGA (9.5)-mPEG(5) microspheres [Figures 9(D,E)], which should due to its earlier and faster mass reduction, while, there was no significant difference between PLGA(19.9)-mPEG(5) and PLGA(31.6)-mPEG(5) microspheres, which may due to the unobvious difference of microspheres erosion and mass lose rate between them (Figures 7 and 6), thought they showed different Mw decreasing rate. The two main release mechanisms associated with drug release from PLGA microspheres were diffusion and degradation/erosion. In this study, we found the release rate was diffusion-controlled initially for BSA and degradation/erosion-controlled during the latter stage of the release period, and as the drug M_w decreased, the degradation/erosion-

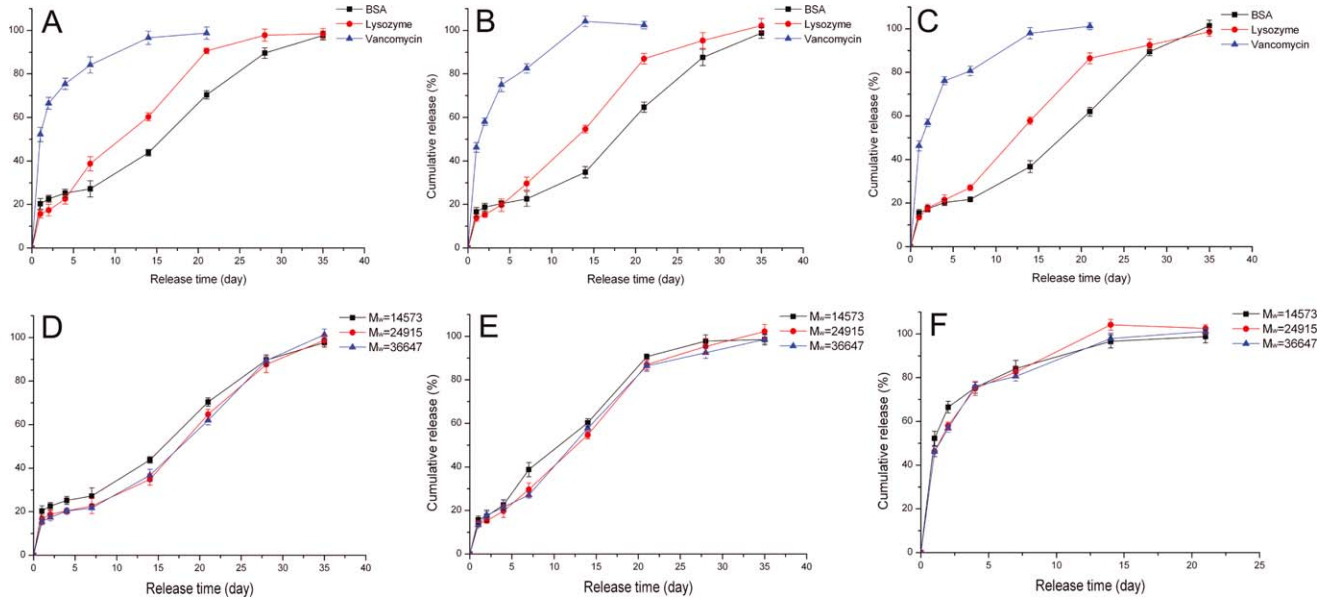

Figure 9. The release behavior of : A, Drugs in PLGA (9.5)-mPEG(5) microspheres; B, drugs in PLGA (19.9)-mPEG(5) microspheres; C, drugs in PLGA (31.6)-mPEG(5) microspheres; D, BSA in microspheres of different PLGA-mPEG M_w; E, LZ in microspheres of different PLGA-mPEG M_w; F, VM in microspheres of different PLGA-mPEG Mw. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

controlled period got shorter (for LZ), and even disappeared (for VM). The results indicated that the drug M_w had a significant effect on the release behavior.

CONCLUSIONS

In this research, PLGA-mPEG of different Mw were successfully synthesized and applied for preparing microspheres encapsulating drugs of various M_w, in order to investigate the effects of drug and polymer M_w on release kinetics from PLGA-mPEG microspheres. The macromolecular drug was found to be diffusion controlled initially but degradation/erosion controlled at latter stages. As drug M_w decreased, LZ showed a reduced lag period and faster release rate than BSA, which may due to its smaller molecular weight. To the smallest drug, VM revealed a fastest release rate and didn't undergo the lag period, and the VM release curves were similar in microspheres of different PLGA-mPEG Mw, indicating the release pattern was absolutely diffusion-controlled during the whole release period. The PLGA-mPEG Mw affected the release kinetics for BSA and LZ to some extent. PLGA (9.5)-mPEG(5) microspheres showed a higher release rate due to its faster degradation and structure erosion, while, the release difference between PLGA(19.9)-mPEG(5) and PLGA(31.6)-mPEG(5) microspheres were not obvious, as the degradation difference was insufficient to cause significant microspheres erosion difference.

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