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Fluorescent microspheres of poly(ethylene glycol)-poly(lactic acid)-fluorescein copolymers synthesized by Ugi four-component condensation

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ABSTRACT: Microparticles formed by poly(lactic acid) (PLA) and poly(ethylene glycol) (PEG) diblock copolymers containing fluorescein grafted to the polymer chain were synthesized by a Ugi four-component condensation (UFCC) reaction. To synthesize these copolymers, lactide was first polymerized by a ring-opening polymerization with alcohol initiators containing functional groups to give carboxyl- and aldehyde-end-functionalized PLA. Two different fluorescent block copolymers (FCPs) of PEG–PLA conjugated to fluorescein (FCP 1 and FCP 2) were then synthesized by UFCC; they gave yields in the range 65–75%. These copolymers were characterized well according their chemical structures and thermal properties, and we prepared fluorescent microspheres (FMSs) from them with the single emulsion–solvent evaporation method (FMS 1 and FMS 2). A new application of UFCC in the preparation of biomasked drug-delivery systems is proposed. © 2015 Wiley Periodicals, Inc. J. Appl. Polym. Sci. **2016**, *133*, 42994.

KEYWORDS: biodegradable; biomedical applications; copolymers; drug-delivery systems

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INTRODUCTION

Ugi four-component condensation (UFCC) is a multicomponent reaction in which an aldehyde or a ketone, an amine, an isocyanide, and a carboxylic acid can form a bisamide in a one-pot reaction. Steps of this kind of condensation involve a first condensation of the amine with the carbonyl group of the aldehyde or ketone to yield an imine, which on protonation, reacts with both the carboxylic acid and the isocyanide to give an alkyl(acyl amine)amide.[1]

To take advantage of the UFCC features, the preparation of macromolecular conjugates has being reported.[1–5] Some of these reports have demonstrated the versatility of the UFCC method, showing even the possibility of the conjugation of water-soluble polymers with different active proteins.[1,3–5] The method allows, for example, the attachment of a poly(ethylene glycol) (PEG) chain, an active molecule, and a target group at a functional group of a polymer system selected from an amine, a carboxylic acid, or a synthetically introduced aldehyde or ketone. It also provides the possibility of preparing different conjugates in a combinatorial fashion if desired.[1]

On the other hand, PEGylation is one of the most common methods for increasing the stability of molecules during circulation in the bloodstream; it prevents protein adsorption and uptake by the reticuloendothelial system.[6–8] Furthermore, the conjugation of PEG to biodegradable polymers, such as poly (lactic acid-*co*-glycolic acid), have been used extensively for the design and synthesis of polymer conjugates because of their low immunogenicity, good biocompatibility, and excellent mechanical properties.[7,9–11]

Poly(lactic acid) (PLA) can be obtained from lactic acid by polycondensation. However, this is an equilibrium reaction, and difficulties in the complete removal of water can limit the maximum molecular weight attained. A solution to this problem is the use of the cyclic dimer lactide, which easily undergoes a ring-opening polymerization (ROP) in the presence of tin(II) 2-ethylhexanoate [$Sn(Oct)_2$] as a catalyst to give PLA. These method have been used to copolymerize lactide with PEG; this generates diblock copolymers that are able to form polymeric particles.[6,10]

Because of the advantages provided by these reactions, in this article, we report for the first time the use of UFCC for the synthesis of fluorescent PEGylated PLA block copolymers, in a one-pot reaction. These copolymers were able to form fluorescent microspheres (FMSs), which can be useful as a drug-delivery system. Therefore, they could be used for the encapsulation of any hydrophobic drug in the PLA phase by releasing the drug by diffusion or erosion of the polymer matrix.

EXPERIMENTAL

Materials

L-Lactide was supplied by Purac. $Sn(Oct)_2$, salicylaldehyde, salicylic acid, *tert*-butyl isocyanide, PEG–bisamine (molecular weight = 6000 Da), acid fluorescein, and acid 6-aldehyde fluorescein were supplied by Sigma Aldrich. Poly(vinyl alcohol) (PVA) was supplied by VETEC. All of the reactants were used as received, except lactide, which was purified by crystallization from toluene.

Polymerization

Functionalized poly(L-lactide) (end-activated PLA) was obtained from L-lactide polymerization in a 0.250-L glass flask with magnetic stirring, which was maintained at the initial part of the polymerization. The monomer (1 g) was fed into the flask with appropriate amounts of coinitiator (0.697 mmol) and $Sn(Oct)_2$ (0.0697 mmol). The choice of the coinitiator for each polymerization was dependent on the requested chain-end functional group. Salicylaldehyde and salicylic acid were used as coinitiators. The monomer/catalyst molar ratio was 100:1, and the coinitiator/catalyst molar ratio was 10:1. The reactions were carried out at 180°C for 1 h under a nitrogen atmosphere. After this time, the reaction flask was rapidly cooled to room temperature, and the solid product was dissolved in chloroform (10 mL). The solution was kept under stirring for 2 h before precipitation in cold ethanol (100 mL). The yield was calculated on the basis of the weight of the monomer at the feed and the precipitated polymer.

Synthesis of the Fluorescent Block Copolymers (FCPs) by UFCC

Synthesis of FCP 1. PEG–bisamine (0.255 g) dissolved in 7.5 mL of a chloroform/methanol mixture (1:2 v/v) was reacted with an appropriate amount of aldehyde–fluorescein (0.063 mmol) overnight at room temperature under a nitrogen atmosphere with stirring (PEG–bisamine/aldehyde–fluorescein molar ratio = 1:5). The protonated imine that formed was reacted with carboxyl-end-terminated poly(lactic acid) (PLACOOH; 0.063 mmol) and *tert*-butyl isocyanide (0.012 mmol) at room temperature for 72 h under a nitrogen atmosphere with stirring (PEG–bisamine/aldehyde fluorescein/PLACOOH/*tert*-butyl isocyanide molar ratio = 5:5:1:1). After this time, the reaction product was precipitated in cold ethanol (75 mL). Finally, the product was dried. The yield was calculated on the basis of the weight of the feed end-activated PLA.

Synthesis of FCP 2. Aldehyde-end-terminated poly(lactic acid) (PLACOH; 0.3 g) dissolved in 7.5 mL of chloroform/methanol (1:2 v/v mixture) was reacted with an appropriate amount of PEG–bisamine (0.075 mmol) overnight at room temperature under a nitrogen atmosphere with stirring (PLACOH/PEG–bisamine molar ratio = 1:5). The protonated imine formed was reacted with acid fluorescein (0.075 mmol) and *tert*-butyl isocyanide (0.015 mmol) at room temperature for 72 h under a nitrogen atmosphere with stirring (PLACOH/PEG–bisamine/acid fluorescein/*tert*-butyl isocyanide molar ratio = 1:5:5:1). After this time, the reaction product was precipitated in cold ethanol (75 mL), and finally, the product was dried. The yield was calculated on the basis of the weight of the feed end-activated PLA.

Preparation of the FMSs

The FMSs were prepared with the single emulsion–solvent evaporation methodology[12] with some modifications. In this methodology, 100 mg of FCP was dispersed in 1 mL of dichloromethane. The FCP dispersion was then added to a beaker containing 5 mL of a cold PVA–water solution (0.5 wt %). This system was kept under stirring at 20,000 rpm for 5 min; it produced an oil-in-water emulsion, which was poured into a flask containing 50 mL of PVA (0.1 wt %). The system was kept under mechanical stirring for 2 h at room temperature (25°C) to harden the microspheres by solvent evaporation. The FMSs were then collected by centrifugation at 2500 rpm and washed with distilled water three times. Finally, the collected FMSs were freeze-dried by lyophilization and stored at 12°C.

Material Characterization

^{1}H-NMR measurements were carried out in a Varian Mercury VX-300 NMR spectrometer operating at 300 MHz (^{1}H). The samples (15 mg) were dissolved in chloroform–d_1 (0.8 mL) in 5-mm NMR tubes at room temperature.

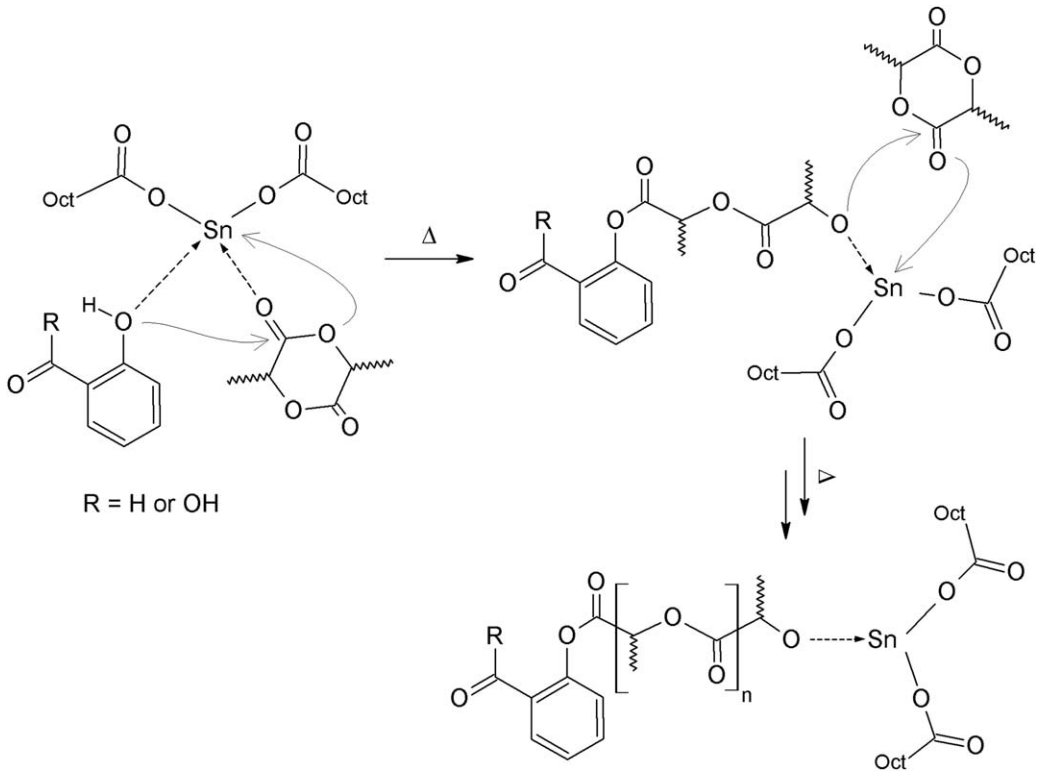
Gel permeation chromatography (GPC) was performed and the molecular weight (number-average molecular weight and weight-average molecular weight) and polydispersity index were determined with an Agilent 1200 series High Performance Liquid Chromatography (HPLC) instrument with a MIXED-C column. Chloroform was used as the solvent with a flow rate of 1.0 mL/min, and polystyrene was used as a standard.

Ultraviolet–visible (UV–vis) spectroscopy was performed in a Varian model Cary 100 spectrophotometer. The samples were dissolved in chloroform (concentration = 0.1% w/v) and measured from 200 to 400 nm with the determination of the maximum absorbance.

Differential scanning calorimetry (DSC) of the polymers and copolymers were determined with a TA Q1000 calorimeter with a heating and cooling rate of 10°C/min from −10 to 200°C under a nitrogen flow. Values of the glass-transition temperature (T_g) and crystallization temperature on heating were obtained from a second heating after a quenching run. The melting temperature (T_m) and crystallization temperature on cooling (T_{cc}) were taken from a second heating run and subsequent cooling run, respectively.

Thermogravimetric analysis (TGA) was performed with a TA TGA Q500 thermoanalyzer. Measurements were carried out under a nitrogen flow at a heating rate of 20°C/min up to 700°C with a gas flow rate of 20 mL/min.

Fluorescence spectroscopy of the polymeric particles and fluorescein was carried out with a spectrophotometer (Shimadzu model RF-5301PC). The spectra were obtained from suspensions of



Scheme 1. Proposed mechanism for acid and aldehyde end groups in PLA obtained by ROP of lactide (LA) coinitiated by salicylaldehyde and salicylic acid.

particles and fluorescein in distilled water. Excitation was performed at 468 nm (the maximum absorbance of fluorescein determined previously by UV–vis spectroscopy), and the emission spectra were recorded over the range 400–600 nm. Both the excitation and emission slit widths were 5 nm. In addition, the quantum yield of fluorescence of the polymeric particles was determined by the following equation:

$$\Phi_S = \Phi_F \left(\frac{F_S}{F_F}\right)\left(\frac{1\text{-}10^{-\mathrm{Abs}(F)}}{1\text{-}10^{-\mathrm{Abs}(S)}}\right)\left(\frac{\eta_S^2}{\eta_F^2}\right)$$

where Φ_S and Φ_F are the photoluminescence quantum yield of the sample and that of the standard (fluorescein), Abs (F) and Abs (S) are the absorbance of the sample and stardard respectively; F_S and F_F are the integrated intensities (areas) of the sample and standard spectra, respectively; and η_S and η_F are the refractive indices of the sample and reference solution, respectively.[13]

Scanning electron microscopy (SEM) was performed with a JEOL JSM-5610 LV microscope with an acceleration voltage of 15 kV. The samples were coated with gold, and we sampled the materials by taking several images of various magnifications to ensure that the analysis was based on a representative region of the sample. The average particle diameter of the samples was determined with 100 polymeric microparticles with a JEOL JSM-5610 LV microscope.

RESULTS AND DISCUSSION

$Sn(Oct)_2$ has been reported as one of the most efficient initiators for the ROP of lactones as lactides.[14–16] Because of the mechanism of ROP, a hydroxyl functional group present in a coinitiator molecule coordinates to $Sn(Oct)_2$; this forms the initiating tin alkoxide linkage necessary to propagate monomer addition. After termination, a fragment of the coinitiator remains at the end of the chain.[15,17,18] In this study, salicylaldehyde and salicylic acid were used as coinitiators. As these molecules contain two different functional groups in addition to the hydroxyl one, we expected that aldehyde and carboxyl acid chain ends would be present in the PLA as a functional group for UFCC (Scheme 1).

Samples of PLACOOH and PLACOH were obtained with these aromatic coinitiators with moderate yields (75–80%) for 1 h of reaction at 180°C (lactide/$Sn(Oct)_2$ molar ratio = 100 and lactide/coinitiator molar ratio = 10). These results were in close agreement with those found in the literature, in which the yields were over 60% for end-functionalized PLA from an ROP with $Sn(Oct)_2$ as the initiator and nine different primary alcohols as coinitiators.[19] Even with water as a coinitiator in the presence of $Sn(Oct)_2$ as an initiator, yields of over 80% were reported.[20]

To verify the success of the end functionalization, the samples were characterized by ^{1}H-NMR, UV–vis spectroscopy, DSC, and TGA.the ^{1}H-NMR spectra of the end-activated PLA synthesized with the two coinitiators and their characteristic peaks are shown in Figure 1.

^{1}H-NMR spectra of the end-activated PLA showed peaks at δ values of 1.59 and 5.17 ppm due to the CH_3 and CH protons of PLA, respectively. On this basis, the salicylaldehyde or salicylic acid initiated the growth of lactide chains for one end and generated in each case the activated group at the end of the

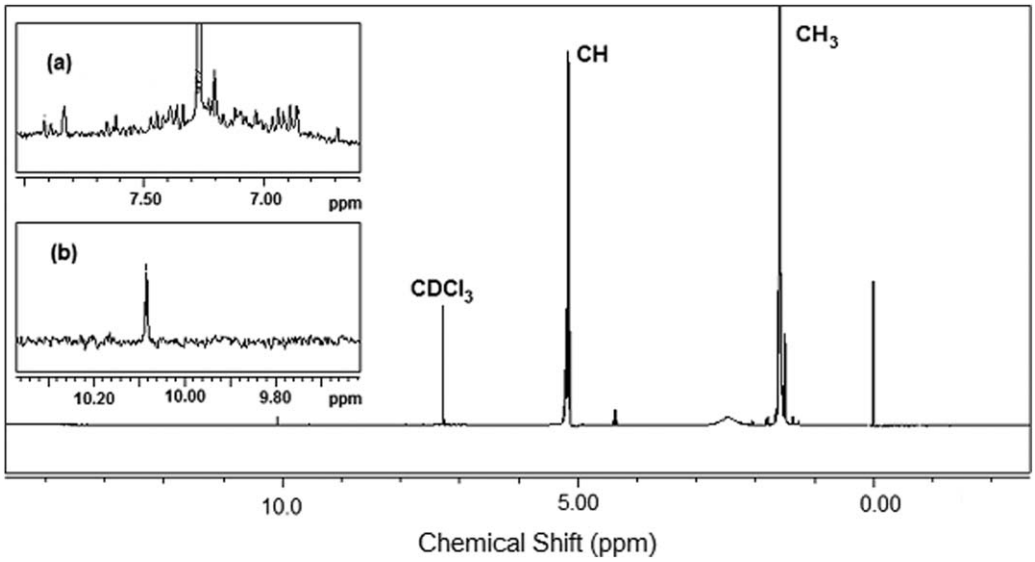


Figure 1. ^{1}H-NMR spectra of end-activated PLA detailing regions of characteristic peaks of (a) PLACOOH and (b) PLACOH.

chain. In turn, the peak at a δ value of 4.2 ppm probably corresponded to the CH_2 of $Sn(Oct)_2$ still linked at the end of the chain. The examination of the microstructure of PLACOH and PLACOOH by ^{1}H-NMR revealed resonance signals between δ values of 7.0 and 8.0 ppm; these were attributed to the aromatic protons of salicylaldehyde and salicylic acid, respectively [Figure 1(a)]. The spectrum of PLACOH showed the a peak at a δ of 10.0 ppm [Figure 1(b)]; this was attributed to the aldehyde proton of covalently linked salicylaldehyde. Because of the lower molar ratio of these end functional groups in the polymer, they made a poor contribution and resulted mainly in peaks of small intensity in the spectra.

The success of the end activation of PLA with aldehyde and carboxylic groups was also confirmed by UV–vis spectroscopy (Figure 2).

PLACOH had a strong absorbance at 260 nm [Figure 2(c)] because of the presence of aromatic aldehyde functional groups at the end of the chain, whereas PLA did not show this absorbance [Figure 2(a)]. In addition, the PLACOOH had a strong absorbance at 320 nm [Figure 2(b)], whereas PLA did not have either [Figure 2(a)]. The reaction with salicylic acid or salicylaldehyde caused this strong absorbance for these activated PLAs. In agreement with these results, the characterization of salicylaldehyde and salicylic acid by UV–vis spectroscopy showed the same maximum absorbance as reported in literature.[21,22] The absorbance with the maximum observed at 230 nm belonged to the solvent used in the sample preparation [Figure 2(a)].

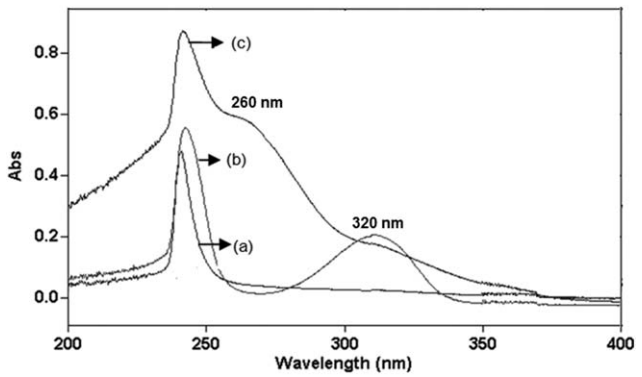


Figure 2. UV–vis spectra of (a) PLA, (b) PLACOOH, and (c) PLACOH. Abs, Absorbance.

The thermal properties of PLACOH and PLACOOH also suggested the end functionalization of PLA obtained with the aromatic initiators (Table I).

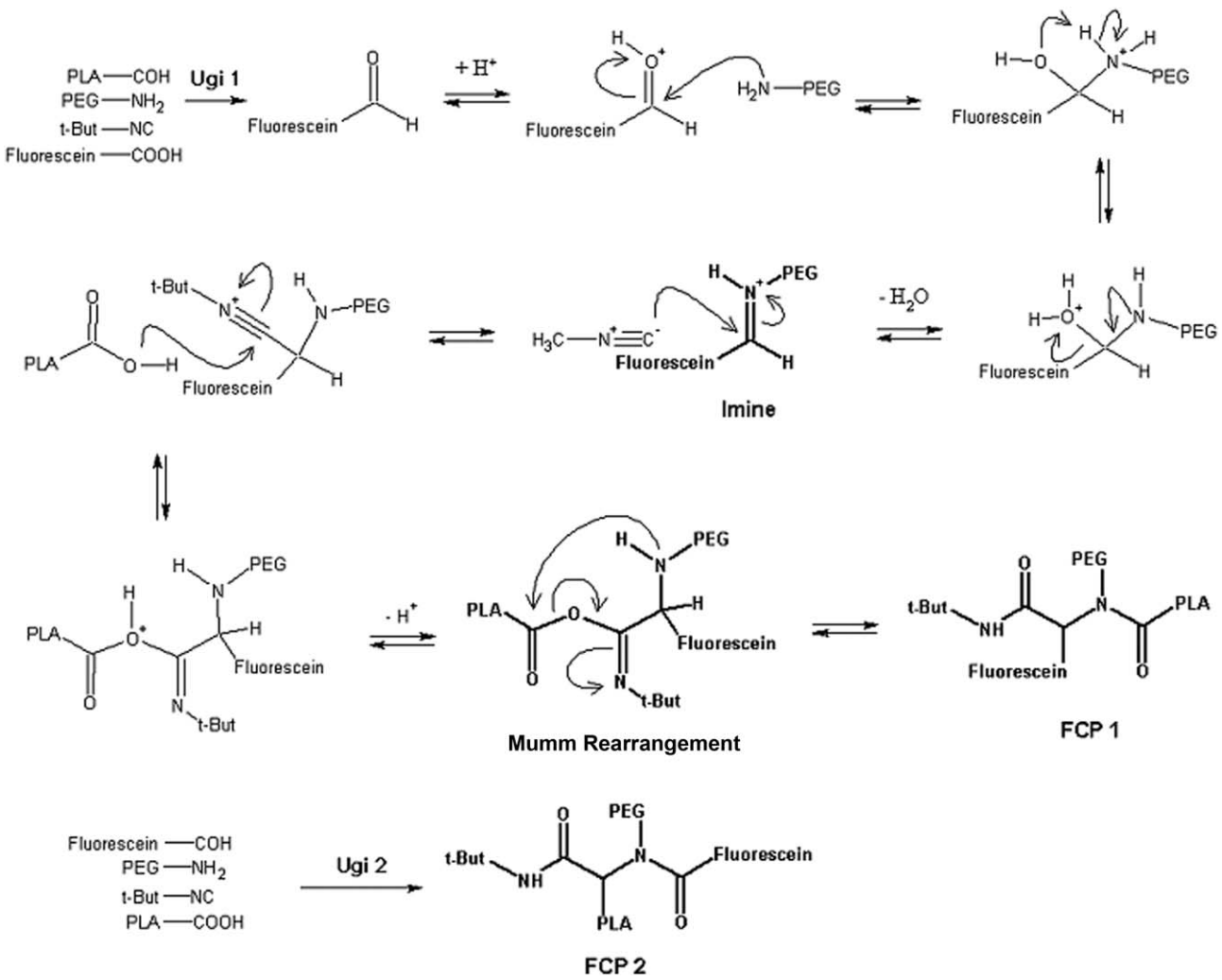
As shown in Table I, PLACOOH and PLACOH presented a higher degree of crystallinity and lower T_g and T_m values than PLA. These results were probably due to the presence of new interactions established by the end functional groups. These results agreed with those reported by the literature on the terminal functionalization of PLA with maleic anhydride.[24] On the other hand, the end-activated PLA showed lower values of the maximum degradation rate temperature obtained by TGA, probably because of the presence of these new functional groups and the lower molecular weight.

As explained before, in our study, the Ugi mechanism was used to PEGylate and introduce fluorescein into the end-activated

Table I. Thermal Properties of All of the Polymers involved in the Preparation of Fluorescent Particles

Sample	T_g (°C)	X_c (%)[a]	T_{cc} (°C)	T_m (°C)	T_d (°C)[b]
PLA	59.8	18.9	98.6	168.8	365.8
PLACOOH	55.6	31.6	100.9	162.0	266.1
PLACOH	53.1	26.5	107.8	149.0	260.1
PEG-NH_2	—	—	—	56.7	413.4
FCP 1	47.1	15.4	81.6	144.9 153.2	272.9 370.2
FCP 2	46.8	14.7	79.2	143.4 152.1	270.1 369.3

[a] Degree of crystallinity X_c determined on the basis of melting enthalpy (ΔH_m) (standard) = 106 J/g.[23]
[b] Degradation temperature determined with TGA.



Scheme 2. Mechanism for the synthesis of FCP 1 and FCP 2 by UFCC. Ugi 2 is the Ugi reaction starting from PLA-COOH. t-But; terc-butyl group.

PLA; this generated two different fluorescent copolymers (FCP 1 and FCP 2; Scheme 2).

Moreover, it was possible to use PLACOOH and PLACOH as components of the two different UFCCs; this made possible the reaction of each end-activated PLA with PEG–NH_2 and fluorescein–COH or fluorescein–COOH, respectively, in a one-pot reaction. Moderate yields (65–75%) were achieved after 72 h of reaction at room temperature with PLACOOH or PLACOH always as the limiting reactant. For the purification of the Ugi products, a nonsolvent of PLA in which all the residues were soluble was selected. On the other hand, to prevent crosslinking reactions between PEG–bisamine and the other molecules, 5 equiv of PEG was added to UFCC as an excess.

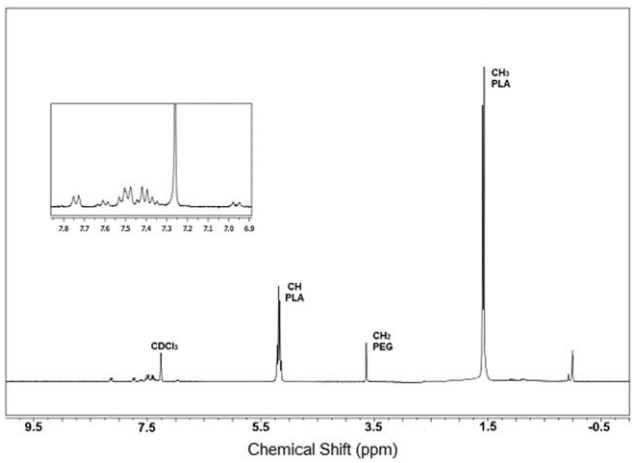


Figure 3. ^{1}H-NMR spectrum of a fluorescent Ugi copolymer (FCP 1).

To verify the success of the PEGylation by UFCC, the synthesized FCPs were characterized by ^{1}H-NMR, GPC, DSC, and TGA.

The ^{1}H-NMR spectra of both products of the Ugi reaction (FCPs) showed the characteristic peaks of these molecules (Figure 3).

The ^{1}H-NMR spectrum of FCP 1 (Figure 3) was similar to the spectrum of FCP 2 and presented peaks at δ values of 1.59 and 5.17 ppm of the CH_3 and CH protons of end-activated PLA. They showed also a new peak at a δ of 3.65 ppm; this corresponded to the CH_2 protons of PEG. These spectra suggest a covalent linkage between the end-activated PLA and PEG after UFCC. The peak shown in Figure 3 at a δ of 3.65 ppm also matched the peak of different PLA–PEG copolymers reported in literature.[25] The examination of the microstructure of the FCP

Table II. Molecular Weights and Polydispersities of the FCPs Synthesized by UFCC and Their Corresponding End-Activated PLas Used as Components of the Ugi Reaction

Ugi coinitiator	Sample	Number-average molecular weight (g/mol)	Weight-average molecular weight (g/mol)	Polydispersity index
Salicylic acid	PLACOOH	3956	6722	2.0
	FCP 1	8301	19,700	1.6
Salicyl aldehyde	PLACOH	4248	7934	1.8
	FCP 2	9510	19,481	2.3

1 by ^{1}H-NMR revealed resonance signals between δ values of 6.0 and 8.0 ppm; these were attributed to the aromatic protons of the fluorescein linked to the copolymers. On the other hand, because of the lower molar ratio of the fluorescein in the polymer, they made a poor contribution and resulted mainly in small-intensity peaks in the spectra. Similar results were reported previously for fluorescent macromolecules functionalized with fluorescein.[26,27]

As determined by GPC, unimodal molecular weight distributions and moderate polydispersities were observed in the GPC chromatograms of PLACOH, PLACOOH, FCP 1, and FCP 2 (Table II).

GPC analyses also revealed increases in the molecular weights of both FCPs in relation to the molecular weights of the corresponding end-activated PLA used in each UFCC. This result confirmed the success of the covalent linkage between PEG and PLA after 72 h of UFCC at room temperature.

Moreover, from the thermal properties of the synthesized FCPs (Table I), it was clear that they were more amorphous than PLA, PLACOOH, and PLACOH, probably because of the presence of PEG linked to the PLA; this decreased the FCP degree of crystallinity.[28] Otherwise, T_g and T_{cc} also decreased in the synthesized FCPs. The decrease in T_{cc} indicated a decrease in the PLA chain capability of crystallizing in the FCPs. These results were in agreement with those previously reported on the thermal properties of copolymers containing chemical linkages between PLA and PEG.[8] In relation to T_m, the FCPs showed bimodal melting peaks with maxima at 144.9 and 153.2°C; this is commonly observed in PLA. Although PEG was a crystallizable polymer, no PEG crystallization was observed in the FCPs.

On the other hand, TGAs of those copolymers showed two steps of mass loss; this suggested the presence of two different blocks of chemically bonded PEG and PLA.[8,29]

As mentioned previously, the FMSs were prepared by a single emulsion–solvent evaporation procedure. These FMSs were characterized by fluorescence spectroscopy and SEM.

Fluorescence spectroscopy confirmed that FMS 1 and FMS 2 contained the fluorescein framework covalently bonded to the PLA–PEG block copolymers (Figures 4–6).

As shown in Figure 4, we clearly observed the same maximum absorbance of the fluorescein at 468 nm for FMS 1 and FMS 2; this confirmed the presence of fluorescein bounded to the polymeric microspheres. The excitation of FMS 1 and FMS 2 at 468 nm in the fluorescence spectroscopy analyses (Figure 5) showed the first harmonic emission bands at 524 and 517 nm for FMS 1 and FMS 2, respectively. A similar value (515 nm) was registered for the free fluorescein exited at 468 nm (Figure 6). In addition, the calculated quantum yield of the fluorescence of the polymeric particles gave values of 0.031 and 0.032 for FMS 1 and FMS 2, respectively. This calculated quantum yield of fluorescence was 3% of the quantum yield of fluorescence reported for free fluorescein (0.98).[13,30]

This performances of FMS 1 and FMS 2 were still poor. However, some materials with quantum yields of fluorescence between 1 and 20% have been used for biological images.[31–34] On the other hand, further optimization of the Ugi reaction is

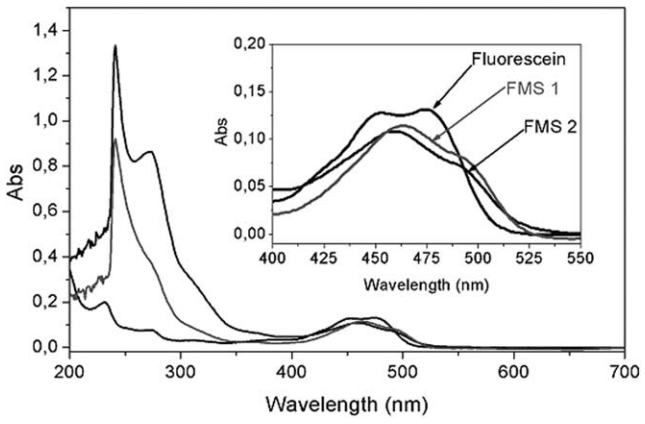


Figure 4. UV–vis spectra of fluorescein, FMS 1, and FMS 2. Abs, Absorbance.

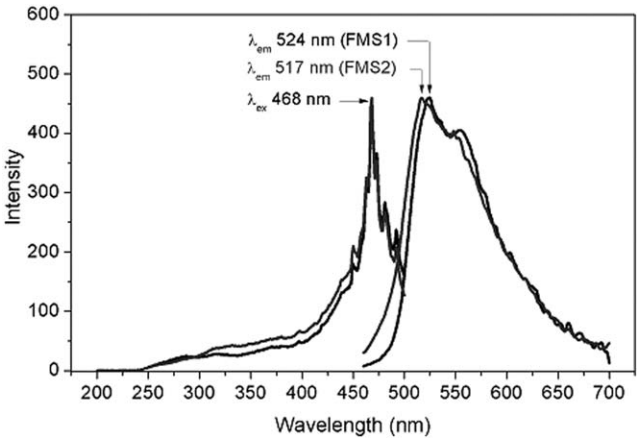


Figure 5. Fluorescence spectra of FMS 1 and FMS 2. λ_{em}, emission wavelength; λ_{ex}, excitation wavelength.

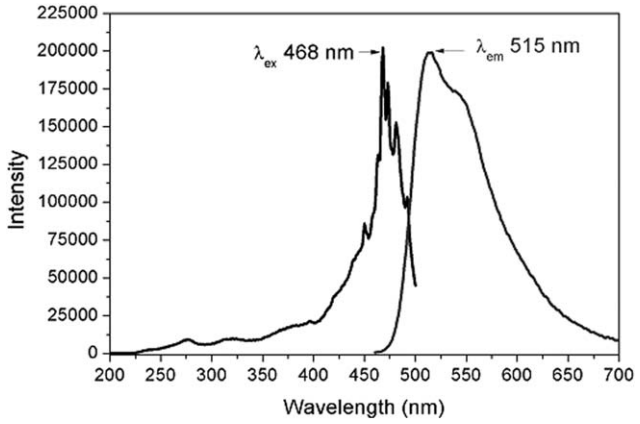


Figure 6. Fluorescence spectra of fluorescein. λ_{em}, emission wavelength; λ_{ex}, excitation wavelength.

under progress by our group to enhance the reactivity of fluorescein during the Ugi reaction. As expected, the curve profiles of the fluorescence of FMS 1 and FMS 2 showed a slight difference, probably because of the structural differences in both fluoresceins used in each UFCC reaction (Figure 7). Similar results were previously reported in fluorescence spectroscopy analyses of fluorescent macromolecules functionalized with fluorescein.[35–37]

Furthermore, both FCPs were able to form microspheres loaded with fluorescein; this could make them useful as target and diagnostic materials. The morphological characterization of FMS was also carried out by SEM analyses (Figure 8).

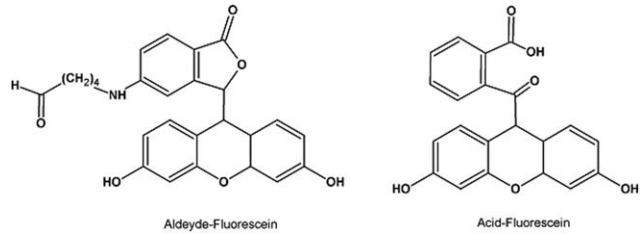


Figure 7. Chemical structures of aldehyde and acid fluorescein used in the syntheses of FCP 1 and FCP 2 by UFCC.

Figure 8 shows representative SEM images of the microspheres prepared from the PLA–PEG–fluorescein block copolymer. Both FCPs were able to form spherical microparticles with a slightly rough surface and no aggregation [Figure 8(a,b)]. To date, others studies of biodegradable microspheres prepared with PLA and PEG polymers have been reported, and such microspheres have been reported to have a spherical shape and a smooth or slightly rough surface.[38,39] However, as shown in Figure 8(b,c), the presence of some small pores randomly distributed on the surface of both microspheres was clear; these were probably due to the influence of the amount of PEG bonded to PLA. The ability of PEG to form small pores on the surface of microspheres was reported for microparticles based on blends of PLGA/PLA and PEG and applied also to microparticles formed by a diblock copolymer.[40,41]

Particle size analyses were done from the SEM images, and average diameters of 13.19 ± 5.42 and 12.73 ± 4.6 μm were calculated for FMS 1 and FMS 2, respectively. For both microspheres, nonhomogeneous size distributions were observed (Figure 9).

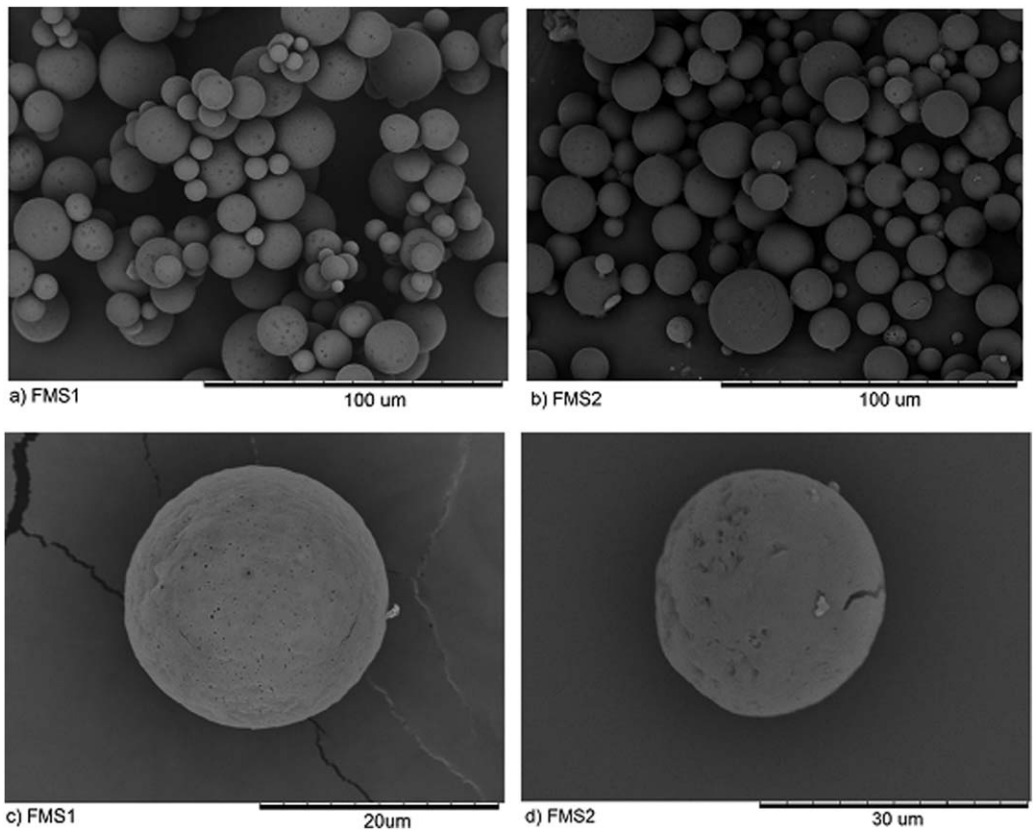


Figure 8. SEM images of (a,c) FMS 1 and (b,d) FMS 2.

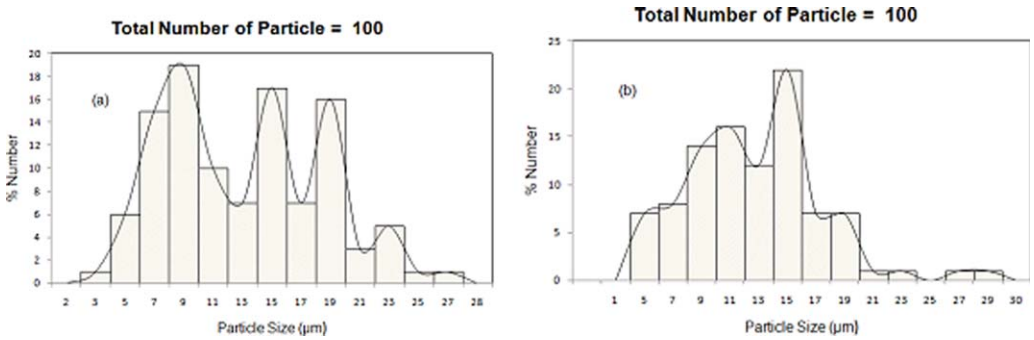


Figure 9. Size distributions of (a) FMS 1 and (b) FMS 2. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

In both cases, the sizes were within the range of few micrometers and below 30 μm. For parenteral administration, the size control of the microparticles is an important factor because very large microparticles (>100 μm) with a broad particle size distribution are acceptable for embolization. Microparticles in the size range 10–100 μm can be used for subcutaneous and intramuscular administration. Here, the particle size distribution is not a critical factor.[42]

The ability of this material to form small microspheres suggests that it could be used to encapsulate various therapeutic agents. For example, these drugs could be included by the double emulsion and solvent evaporation methods or spray drying. The inclusion of fluorescein could allow biodistribution studies of these particles, and it also could be replaced by a targeting moiety by the same synthetic rout of the Ugi reaction.

CONCLUSIONS

ROP was used for the synthesis of PLACOH and PLACOOH. UFCC made possible the PEGylation of these end-functionalized PLAs and their chemical linkage with fluorescein, which acted as a fluorescent cellular target. The structure and thermal properties of all of the fluorescent PEGylated PLA copolymers was elucidated by ^{1}H-NMR, GPC, DSC, TGA, and fluorescence and UV–vis spectroscopy. Both fluorescent copolymers were capable of forming microspheres with diameters in the range 3–25 μm with slightly rough surface morphologies and a few pores randomly distributed on the surface. In addition, these microspheres could be potentially useful for the site-specific delivery of any cytotoxic agent for cancer therapy or diagnostics. Therefore, from the results, it was possible to demonstrate the utility of the UFCC for the synthesis and design of versatile biodegradable materials that would be very useful in the preparation of a new drug-delivery system.

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