The Effect of Fabrication Methods on the Mechanical and Thermal Properties of Poly(lactide-*co*-glycolide) Scaffolds

Rachel G. Forcino, Sriramakamal Jonnalagadda

University of the Sciences in Philadelphia, 600 South 43rd Street, Philadelphia, Pennsylvania 19104

Received 15 June 2006; accepted 6 November 2006
DOI 10.1002/app.25753
Published online in Wiley InterScience (www.interscience.wiley.com).

ABSTRACT: The purpose of this research was to evaluate the effects of the fabrication method, poly(ethylene glycol) (PEG) molecular weight, and PEG concentration on the mechanical and thermal properties of blended poly (lactide-*co*-glycolide) (PLGA)/PEG scaffolds. The manufacturing process was the dominant factor. The tested fabrication processes were compression, heat molding, and solvent casting/vacuum drying. The scaffolds produced by compression were strong and brittle with mechanical properties [compressive modulus (E) $\sim$ 400 N/mm^2] comparable to those of trabecular bone. The heat-molded scaffolds were weaker and more ductile ($E \sim 45$ N/mm^2) than the compressed scaffolds, so they were more applicable to nonload-bearing applications. The vacuum-dried scaffolds completely lacked compressive strength ($E \sim 5$ N/mm^2) and were considered unsuitable for scaffolding applications. The miscibilities of the blends were also affected by the processing method and were evaluated on the basis of the melting-point depression of crystalline PEG. The miscibility of PLGA in PEG was greatest with vacuum drying (6–13%), followed by heat molding (0.4–1.5%) and then compression (0.2–0.8%). The application of heat and solvent to the blend successfully altered the miscibility of the two polymers. Overall, this study demonstrates the ability to fabricate scaffolds with distinct thermal and mechanical characteristics by the manipulation of the fabrication method. © 2007 Wiley Periodicals, Inc. J Appl Polym Sci 104: 944–949, 2007

Key words: biodegradable; blends; differential scanning calorimetry (DSC); mechanical properties; polyesters

INTRODUCTION

Biodegradable poly(lactide-*co*-glycolide)s (PLGAs) have been widely investigated for controlled release[1] and tissue engineering applications.[2] These polymers are known to be biocompatible, and their availability in a wide range of molecular weights and copolymer ratios permits the fabrication of devices with a wide range of mechanical, degradation, and drug-release properties.

Despite extensive research, the clinical use of PLGAs has been limited primarily to controlled-release drug delivery. Scaffolds for tissue engineering must provide a three-dimensional substrate for cells that can serve as a template for regeneration.[3] Although the ability of PLGA copolymers to provide such networks has been established, there are several factors that limit their clinical use.[2] Biodegradable scaffolds for regeneration should have sufficient porosity and interconnectivity to accommodate the attachment, proliferation, and distribution of target cells.[3] Current scaffold fabrication techniques, such as solvent casting, particulate leaching, gas formation, emulsion freeze drying, and rapid prototyping, result in scaffolds with limited pore sizes and shapes, low porosity, or nonporous surfaces.[3,4] Other concerns of PLGA-based devices include (1) acidic degradation products that can result in high local acidity,[5] (2) heterogeneous catalytic degradation due to selective accumulation of acidic degradation products in the interior of devices,[6] and (3) foreign-body responses resulting in the formation of fibrous capsules that may prevent cell recruitment and proliferation within the scaffolds.[7] In addition, relatively hydrophobic PLGAs can also inhibit cell penetration into scaffolds, thereby limiting their regenerative potential.[8]

Numerous approaches continue to be investigated to overcome the drawbacks of PLGA devices for tissue engineering. A primary approach has included the use of block copolymers of polylactide (PLA) and poly(ethylene glycol) (PEG) to overcome the acidic interior environment,[8] irregular release,[9] and hydrophobicity.[8,10,11] A major limitation of PLA–PEG block copolymers is the low molecular weight of the synthesized polymers and the absence of mechanical strength in devices manufactured with these block copolymers.[12]

We have previously reported the use of physical blends of PEG with PLA or PLGA to overcome the drawbacks of existing PLGA devices while maintaining mechanical strength. This research examines the

Correspondence to: S. Jonnalagadda (s.jonnal@usip.edu).

effects of three processing methods—(1) direct compression, (2) heat molding, and (3) solvent casting/vacuum drying—on the physical properties of scaffolds fabricated from blends of PLGA (copolymer ratio = 75 : 25) and PEG. The molecular weights (0.4, 10, and 20 kDa) and concentrations of PEG (10 and 20 wt %) in the scaffolds were also varied.

EXPERIMENTAL

Materials

Poly(D,L-lactide-*co*-glycolide) (PLGA 75/25 DL 2A) was obtained from Alkermes (Cambridge, MA). PEGs with molecular weights of 400, 10,000, and 20,000 Da were purchased from Sigma–Aldrich (St. Louis, MO). Reagent-grade chloroform was obtained from Fisher Scientific (Hampton, NH).

Methods

Scaffold fabrication

Table I shows the three fabrication methods used to make the PLGA/PEG scaffolds. The methods were selected to enable the fabrication of scaffolds with a wide range of thermal and mechanical characteristics.

Initially, PLGA 75/25 granules were triturated with PEG in a glass mortar and pestle to prepare a powdered blend that was distributed into fractions for scaffold fabrication with direct compression and heat molding. All scaffolds were fabricated in triplicate and stored in a desiccator at 5°C until use.

Direct compression. The compressed scaffolds (100 mg each) were prepared by the compression of a PLGA/PEG mixture with 17.8 N of force for 5 min. A Carver press (Wabash, IN) and a 7/32″ F-press stainless steel tooling set (Natoli Engineering Company, Inc., St. Charles, MO) were used for the compression.

Heat molding. To fabricate scaffolds by heat molding, 100 mg of a PLGA/PEG mixture was placed in a 9/32″ F-press stainless steel tooling set. The assembly was placed on a hot plate adjusted to 60–70°C. This temperature was selected because it was above the glass-transition temperature (T_g) of PLGA 75/25 and the melting temperature (T_m) of the respective PEG. After the temperature was maintained for 5 min, the scaffolds were cooled in an ice bath for 10 min and removed from the mold.

Solvent casting/vacuum drying. Initially, PLGA was dissolved in chloroform at a concentration of 25% (w/v). Then, PEG was added at a specified concentration, as shown in Table I. The PLGA/PEG solution was poured into cylindrical, metallic molds in a Petri dish and cooled to −75°C for 24 h. The samples were then vacuum-dried at −40°C for an additional 72 h before they were vacuum-dried at room temperature for 48 h.

TABLE I
Formulations of the Blended PLGA/PEG Scaffolds

Fabrication method	Polymer	Excipient (PEG) Molecular weight (kDa)	Excipient (PEG) wt %
Direct compression	PLGA 75/25	0.4	10
			20
		10	10
			20
		20	10
			20
Heat-molded	PLGA 75/25	0.4	10
			20
		10	10
			20
		20	10
			20
Solvent-cast/ vacuum-dried[a]	PLGA 75/25	10	20
		20	10
			20

[a] The vacuum-drying method with 0.4-kDa PEG and 10% 10-kDa PEG did not form scaffolds.

Compression testing of the scaffolds

Compression testing was performed on a Shimadzu Autograph AGS-J (Columbia, MD) equipped with a 20-kN load cell and a 15-mm-diameter circular jig. The crosshead speed was set to 1 mm/min. The scaffolds were tested in triplicate at room temperature, and the data were analyzed by Trapezium2 version 2.22c (Shimadzu Corporation, Columbia, MD). The resulting stress–strain curves were used to determine the compressive modulus (E) and the maximum stress (σ_{max}) withheld by the scaffold before failure. The compressive modulus was determined from the slope of the curve in the elastic deformation region of the stress–strain curves.

Differential scanning calorimetry (DSC)

A Mettler–Toledo DSC822[e] (Columbus, OH) differential scanning calorimeter was used for the thermal analysis studies. Briefly, 5–15 mg of material was placed in a crimped aluminum pan with a pierced lid. The reference consisted of an empty, crimped, and pierced aluminum pan. Under a nitrogen purge, the samples were subjected to two heating cycles with an intermittent cooling cycle, all performed in the range of −50 to 150°C at a rate of 5°C/min. The resultant thermograms were analyzed with STAR[e] version 8.00 software (Mettler-Toledo Inc., Columbus, OH). The thermograms were evaluated for the melting transitions and glass transitions. For the melting transitions, the peak temperature (T_m) and enthalpy of fusion (ΔH_m) were determined, whereas for the glass transitions, the midpoint of the transition (T_g) was obtained.

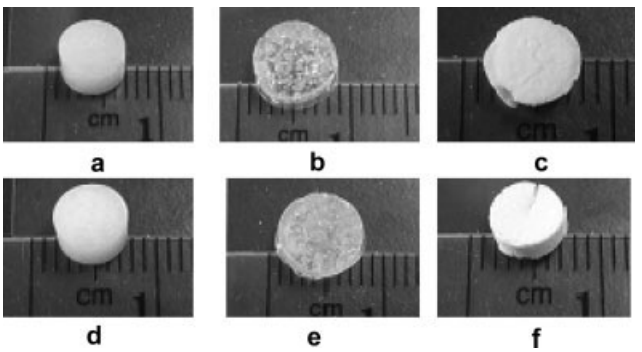

Figure 1 Digital images of PLGA 75/25–20-kDa PEG scaffolds containing 10% PEG [(a) compressed, (b) heat-molded, or (c) vacuum-dried] or 20% PEG [(d) compressed, (e) heat-molded, or (f) vacuum-dried].

RESULTS

Figure 1 shows representative images of scaffolds fabricated by all three procedures. The compressed and vacuum-dried scaffolds were opaque in comparison with the heat-molded scaffolds. No visual differences could be observed between scaffolds containing 10 or 20 wt % PEG.

Compression testing

Scaffolds fabricated by all three methods showed significant variations in mechanical properties (Fig. 2). Only 10-kDa PEG and 20-kDa PEG scaffolds could be characterized for mechanical properties. The mechanical properties of the scaffolds fabricated with 0.4-kDa PEG were below the detection limits of the instrument.

The compressed scaffolds containing 10- or 20-kDa PEG demonstrated the highest E values with clear break points. The E values of these scaffolds were around 400 N/mm^2, comparable to published moduli of trabecular bone (50–450 N/mm^2).[13–15] The maximum stress prior to failure of the compressed scaffolds, in the range of 25–38 N/mm^2, was greater than that of trabecular bone. The maximum stress of trabecular bone has been reported to be in the range of 1–10 N/mm^2.[13]

The heated scaffolds with 10- or 20-kDa PEG showed E values that were about 10 orders of magnitude lower than those of compressed scaffolds (shown in Table III). No distinguishable break points could be observed in these scaffolds. Similarly, the vacuum-dried scaffolds did not show break points and demonstrated the lowest mechanical strength, as characterized by E values in the range of 1.65–10 N/mm^2.

Tables II and III also suggest that the scaffolds fabricated with a lower concentration of PEG were stiffer when compressed or heated. However, this effect was considered to be statistically insignificant within the PEG concentration ranges used in this experiment.

DSC

Preliminary thermal scans were obtained for pure polymer and excipient samples, which were designated as controls. Control PLGA samples displayed a T_g in the range of 37–39°C and lacked a T_m because PLGA is known to be a completely amorphous polymer. The thermograms of the control PEGs did not show a T_g but could be characterized by melting endotherms with peaks (T_m) at 7, 64, and 67°C for 0.4-, 10-, and 20-kDa PEGs, respectively.

All scaffolds containing 10- or 20-kDa PEG demonstrated a T_g characteristic of amorphous regions (PLGA) and a T_m attributable to crystalline regions (PEG). The melting temperature of PEG in the scaffolds decreased compared with that of PEG controls (Fig. 3). Although T_g of PLGA was relatively constant at 37–43°C, the PEG melting endotherm was lowered considerably. The extent of lowering was highest in the vacuum-dried scaffolds (16–21%), which were followed by the heated scaffolds (4–15%), and was least in the compressed scaffolds (2–4%; Table IV). These trends were similar for both concentrations of 10-kDa PEG and 20-kDa PEG.

The melting-point depression could be used to calculate PLGA miscibility with PEG with the van't Hoff law for dilute solutions:[16]

$$m_{\mathrm{PLGA}} = \frac{\Delta T \cdot \Delta H_{\mathrm{PEG}}}{\mathrm{MW}_{\mathrm{PEG}} \cdot R \cdot T_o^2}$$

where m_{PLGA} is the molality of PLGA acting as an impurity in PEG, ΔT is the melting-point depression of PEG, ΔH_{PEG} is the molar enthalpy of melting, $\mathrm{MW}_{\mathrm{PEG}}$ denotes the molecular weight of PEG, R is the universal gas constant, and T_o represents the melting temperature of pure PEG. The obtained miscibility values are shown in Table V. These results suggest that the misci-

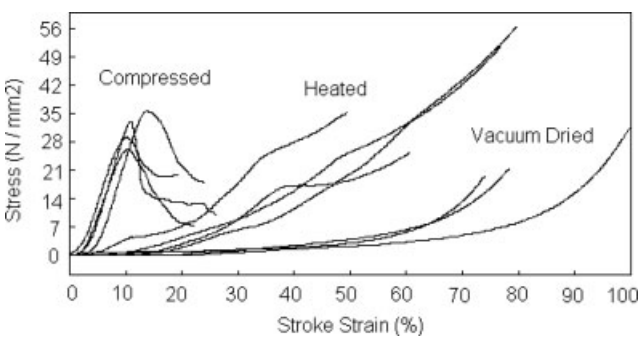

Figure 2 Representative stress–strain curves of PLGA/PEG scaffolds. Scaffolds containing 0.4-kDa PEG are represented by the curves along the x axis and do not follow the trends of 10-kDa PEG and 20-kDa PEG.

TABLE II
Mechanical Properties of the Compressed Scaffolds

Polymer	Excipient (PEG) Molecular weight (kDa)	wt %	E (N/mm^2)	σ_{max} (N/mm^2)
PLGA 75/25	10	10	439 ± 12	33.4 ± 0.2
		20	399 ± 12	25.3 ± 1.7
	20	10	419 ± 23	38.1 ± 2.0
		20	396 ± 41	28.3 ± 1.6

bility of PEG in PLGA was highest for the vacuum-dried scaffolds, followed first by the heated scaffolds and finally by the compressed scaffolds.

DISCUSSION

This study investigated the effects of three parameters on the thermal and mechanical properties of blended PLGA/PEG scaffolds. The examined parameters were the manufacturing method, PEG molecular weight, and PEG concentration. Although all three parameters affected the scaffold properties, the manufacturing process was found to be the most significant factor.

The mechanical strength of scaffolds is an important consideration in scaffold fabrication.[17] The desired application will define the required mechanical strength of the scaffold. For example, certain cases such as hip arthroplasty[18] and spinal fusion[19] may require load-bearing implants, whereas others such as breast reconstruction[20] and bone union[19] require soft, pliable biomaterials. Previous research involving PLGA- and PLA/PEG-based block copolymer scaffolds showed a significant lowering of the mechanical strength versus that of PLA.[8,13] Our results indicate that blended PLGA/PEG scaffolds fabricated with compression, heat, and high-molecular-weight PEGs (10 and 20 kDa) had sufficient mechanical strength for bone-scaffolding applications.

Among the three manufacturing methods examined in this study, compression produced scaffolds with the maximum mechanical strength, as shown by a high E value comparable to that of trabecular bone. However, a consideration in using these scaffolds is their susceptibility to fracture at stresses exceeding 25 N/mm^2. The mechanical strength of the heated scaffolds was lower than that of trabecular bone. Although these scaffolds may have limited value in load-bearing applications, they may be more suitable for soft-tissue scaffolding. The vacuum-dried scaffolds clearly lacked the mechanical properties required for bone and soft-tissue applications.

From Tables II and III, it is evident that identical blends fabricated into scaffolds showed highly variable mechanical strength dependent on the processing method. Therefore, the altered mechanical properties could be attributed to other factors such as the porosity of the individual scaffolds or miscibility of PLGA and PEG. Comparative values for the scaffold porosity were obtained with density measurements (data not shown). Although the vacuum-dried scaffolds showed high porosity, no significant differences in the porosity were observed between the compressed and heat-molded scaffolds.

TABLE III
***E* Values of the Heat-Molded and Vacuum-Dried Scaffolds**

Polymer	Excipient (PEG) Molecular weight (kDa)	wt %	E (N/mm^2) Heat-molded	E (N/mm^2) Vacuum-dried
PLGA 75/25	10	10	41 ± 5	—
		20	40 ± 13	9.91[a]
	20	10	53 ± 12	1.7 ± 1.3
		20	49 ± 12	2.1 ± 1.2

[a] Two scaffolds.

Thermal analysis has been used extensively to study the solid-state properties of polymeric blends.[21] Correlations between the elastic moduli and temperature with respect to T_g are well established in the literature.[22] The glass transition temperature of miscible blended polymers can be determined with the Fox equation:[23]

$$\frac{1}{T_g} = \frac{w_1}{T_{g1}} + \frac{w_2}{T_{g2}}$$

where T_g is the glass-transition temperature of the miscible polymer blend and T_{g1} and T_{g2} are the individual glass-transition temperatures of the blended polymers with amorphous weight fractions w_1 and w_2, respectively. With a T_g of about 40°C for PLGA and a T_g of about −55 to −70°C for PEG, miscible blends of PLGA and PEG (10–20 wt %) could be expected to have a T_g in the range of 7–25°C. How-

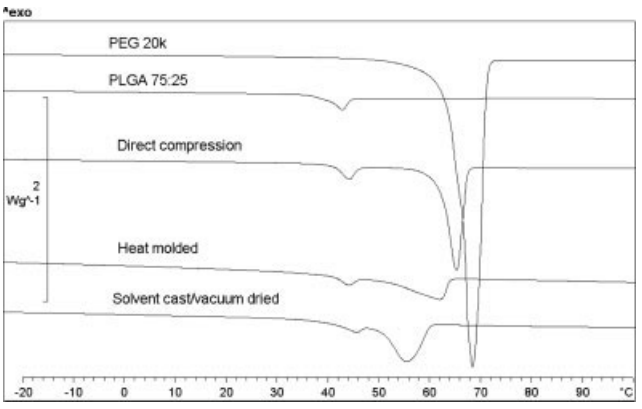

Figure 3 Representative DSC thermograms showing the effect of the fabrication method.

TABLE IV
Thermal Properties of the Controls and Scaffolds

Fabrication method	Polymer	Excipient (PEG) Molecular weight (kDa)	Excipient (PEG) wt %	T_m (°C)	ΔH_m (J/g)	T_g (°C)
Control	PLGA 75/25	NA	0	NT	NT	37.22
	PEG	0.4	100	6.51	104.48	NT
		10	100	65.22	178.68	NT
		20	100	67.81	171.02	NT
Direct compression	PLGA 75/25	0.4	10	NT	NT	39.37
			20	NT	NT	39.08
		10	10	63.54	25.90	39.95
			20	63.63	46.73	40.34
		20	10	65.26	32.01	40.07
			20	65.06	44.06	40.23
Heat-molded	PLGA 75/25	0.4	10	NT	NT	38.44
			20	NT	NT	5.14
		10	10	56.16	5.57	36.6
			20	57.04	19.99	40.01
		20	10	64.9	19.88	42.09
			20	63.1	48.84	42.54
Solvent-cast/vacuum-dried	PLGA 75/25	10	20	54.66	26.70	39.73
		20	10	54.13	10.30	37.87
			20	55.46	24.79	41.35

NA = not applicable; NT = no transition detected.

ever, Figure 3 and Table IV show that T_g of PLGA was relatively unaffected in most scaffolds. This is not surprising because of the highly crystalline nature of pure PEG.[24] The lowering of T_g was observed only in the heat-molded scaffold containing 0.4-kDa PEG at a concentration of 20 wt % (Table IV).

An interesting phenomenon observed in this study was a significant lowering of the PEG melting endotherm as a function of the processing method. The phenomenon may be explained by purity analysis, as described in the Results section, or by the formation of a new polymorphic state of PEG. Polymorphic states for PEGs have been previously reported in the literature.[25] For instance, PEG 6000 is known to exist as lamellae with chains fully extended, folded once, or folded twice according to the crystallization environment.[25] The existence of polymorphic states for PEG is further supported by the fact that ΔH_m was greater than the predicted values in all compressed and heated 20-kDa PEG scaffolds.

The theories described here are feasible explanations involving the thermal transitions of the polymer blends. The true description may even be a combination of both theories, but further analysis such as X-ray diffraction is required to elucidate the state of the crystalline regions within the polymeric blend.

CONCLUSIONS

This study demonstrates the capacity to fabricate scaffolds with distinct thermal and mechanical characteristics by the variation of the manufacturing

TABLE V
Miscibility Obtained from the Melting-Point-Depression Analysis

Fabrication method	Excipient (PEG) Molecular weight (kDa)	Excipient (PEG) wt %	Melting-point depression (°C)	Molality[a]	Miscibility (%)[b]
Direct compression	10	10	−1.68	0.00032	0.22
		20	−1.59	0.00030	0.48
	20	10	−2.55	0.00045	0.35
		20	−2.75	0.00049	0.78
Heat-molded	10	10	−9.06	0.00170	1.24
		20	−8.18	0.00154	2.44
	20	10	−2.91	0.00052	0.39
		20	−4.71	0.00083	1.43
Solvent-cast/vacuum-dried	10	20	−10.56	0.00198	8.59
	20	10	−13.68	0.00242	6.66
		20	−12.35	0.00219	12.96

[a] mol of PLGA/kg of PEG.
[b] Miscible PLGA/total PLGA (w/w).

method. The three fabrication methods used in this study—compression, heat molding, and solvent casting/vacuum drying—produced scaffolds with E values ranging from 1.65 to 440 N/mm^2 and break strengths ranging from 0 to 15 N/mm^2. The thermal properties of the scaffolds showed that PEG had a minimal effect on T_g of PLGA, suggesting that amorphous solid-state miscibility was not responsible for changes in the mechanical strength. This may be a major advantage for the use of blending rather than block copolymerization in scaffold design. The miscibility of PLGA in PEG, on the other hand, was dependent on the manufacturing method and ranged from 0.22 to 13%. Overall, these results demonstrate the ability to use blending and fabrication processes to design biodegradable scaffolds for a wide range of biomedical applications.

References

1. Jain, R. A. Biomaterials 2000, 21, 2475.
2. Wu, Y. C.; Shaw, S. Y.; Lin, H. R.; Lee, T. M.; Yang, C. Y. Biomaterials 2006, 27, 896.
3. Liu, X.; Ma, P. X. Ann Biomed Eng 2004, 32, 477.
4. Sachlos, E.; Czernuszka, J. T. Eur Cell Mater 2003, 5, 29.
5. Kim, J. H.; Taluja, A.; Knutson, K.; Han Bae, Y. J Controlled Release 2005, 109, 86.
6. Siepmann, J.; Elkharraz, K.; Siepmann, F.; Klose, D. Biomacromolecules 2005, 6, 2312.
7. Iwasaki, Y.; Sawada, S.; Ishihara, K.; Khang, G.; Lee, H. B. Biomaterials 2002, 23, 3897.
8. Wan, Y.; Chen, W.; Yang, J.; Bei, J.; Wang, S. Biomaterials 2003, 24, 2195.
9. Mallarde, D.; Boutignon, F.; Moine, F.; Barre, E.; David, S.; Touchet, H.; Ferruti, P.; Deghenghi, R. Int J Pharm 2003, 261, 69.
10. Saito, N.; Okada, T.; Toba, S.; Miyamoto, S.; Takaoka, K. J Biomed Mater Res 1999, 47, 104.
11. Otsuka, H.; Nagasaki, Y.; Kataoka, K. Biomacromolecules 2000, 1, 39.
12. Kim, K.; Yu, M.; Zong, X.; Chiu, J.; Fang, D.; Seo, Y. S.; Hsiao, B. S.; Chu, B.; Hadjiargyrou, M. Biomaterials 2003, 24, 4977.
13. Lin, A. S.; Barrows, T. H.; Cartmell, S. H.; Guldberg, R. E. Biomaterials 2003, 24, 481.
14. Karageorgiou, V.; Kaplan, D. Biomaterials 2005, 26, 5474.
15. Rohlmann, A.; Zilch, H.; Bergmann, G.; Kolbel, R. Arch Orthop Trauma Surg 1980, 97, 95.
16. Laidler, K. J.; Meiser, J. H. Physical Chemistry; Houghton Mifflin: Boston, 1999; p 216.
17. Rezwan, K.; Chen, Q. Z.; Blaker, J. J.; Boccaccini, A. R. Biomaterials 2006, 27, 3413.
18. Buma, P.; Schreurs, W.; Verdonschot, N. Biomaterials 2004, 25, 1487.
19. Wozney, J. M.; Seeherman, H. J. Curr Opin Biotechnol 2004, 15, 392.
20. Croll, T. I.; O'Connor, A. J.; Stevens, G. W.; Cooper-White, J. J. Biomacromolecules 2004, 5, 463.
21. Mano, J. F.; Koniarova, D.; Reis, R. L. J Mater Sci: Mater Med 2003, 14, 127.
22. Van der Voort Maarschalk, K.; Zuurman, K.; Van Steenbergen, M. J.; Hennink, W. E.; Vromans, H.; Bolhuis, G. K.; Lerk, C. F. Pharm Res 1997, 14, 415.
23. Ao, Z. M.; Jiang, Q. Langmuir 2006, 22, 1241.
24. Pielichowski, K.; Flejtuch, K. Polym Adv Technol 2003, 13, 690.
25. Buckley, C. P.; Kovacs, A. J. Colloid Polym Sci 1976, 254, 695.