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A novel fabrication of poly(ε-caprolactone) microspheres from blends of poly(ε-caprolactone) and poly(ethylene glycol)s

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Abstract

A novel method for the preparation of solvent-free poly(ε-caprolactone) (PCL) microspheres from PCL/poly(ethylene glycol) (PEG) blends was developed and demonstrated. The particle size of the PCL spheres are in the range of 1 to 20 μm (mostly in the range 5–10 μm). The influences of the molecular weight of PCL, the molecular weight of PEG, the type of emulsifier, the concentration of plasticizer, the homogenization time and temperature on the characteristics of the PCL microparticles were also investigated. No changes in PCL molecular weight distribution and peak shape were observed, which indicates that the PCL matrix is stable during the fabrication process. However, the microspheres made from low-molecular-weight PCL coalesced into a continuous mass on drying and lost their properties as individual spheres. The surface of the PCL microspheres prepared from PEG 300 as the external phase at 70°C is much smoother than that of the other formulations. The major advantage of this polymer-blend melt technique compared with traditional methods for microsphere preparation is the elimination of methylene chloride or other organic solvents for polymer solubilization. Therefore, the toxicity associated with organic solvent residues resulting from conventional solvent preparation of microspheres is not present with this polymer-blend melt technique.

Keywords: Poly(ε-caprolactone); Poly(ethylene glycol); Microspheres

1. Introduction

Biodegradable polyesters that are most commonly used for medical applications are copolymers of D,L-lactide/glycolide and poly(ε-caprolactone) (PCL). The advantages of these biodegradable polymers include no requirement for follow-up surgical removal once the drug supply is depleted, biocompatibility, predictability of degradation kinetics and their ease of fabrication [1]. Three general types of drug-delivery system based on these polymers are microparticles, implants and fibres. Microspheres and microcapsules of biodegradable polyesters have received the most attention in recent years for controlling the release of many conventional pharmaceutical agents and macromolecules (i.e., vaccines, peptides and proteins).

Several techniques may usually be used to produce polymeric microparticles from polyesters, including solvent evaporation, phase separation (coacervation) and spray drying [2]. Wagenaar et al. [3] claimed that spray drying allows the formation of particles in the micrometre size range, is rapid, easy to scale-up, and less dependent on the solubility characteristics of the drug and polymer than other techniques. Nevertheless, methylene chloride and other organic solvents (methanol, acetone, ethyl acetate) are still needed for polymer solubilization in all of these fabrication methods [4–10] The toxicity of the organic solvent residues presented in the final microspheres is a major problem incurred in the conventional microencapsulation process.

In our previous study, we observed a special phenomenon between PCL and poly(ethylene glycol) (PEG) in their blends, where the PEG was dispersed as spherical drops of size 2–5 μm throughout the PCL phase [11]. Since the melting point of PCL is quite low, around 60°C, both low melting temperature and this special phenomenon between PCL and PEG were applied further to prepare PCL microspheres. In this fabrication process, no methylene chloride was used for polymer solubilization. The influences of the molecular weight of PCL, the molecular weight of PEG, the type of emulsifier, the concentration of plasticizer, the

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homogenization time and temperature on the characteristics of the PCL microparticles were investigated.

2. Experimental

2.1. Materials

Poly(ε-caprolactone) flakes [molecular weight (*MW*) of 21 300] were from Scientific Polymer Products, Inc. (Ontario, NY). Poly(ε-caprolactone) diol ($MW = 5000$) and nine polystyrene standards ($MW = 600$ to 104 000) were from Polysciences, Inc. (Warrington, PA). Poly(ethylene glycol) 600 (PEG 600), poly(ethylene glycol) 300 (PEG 300), poly(oxyethylene sorbitan monooleate) (Tween 80), poly(oxyethylene 10) tricetyl ether (P-10) and Triacetin were from Sigma Chemical Company (St. Louis, MO). HPLC-grade chloroform was from Fisher Chemical Company (Fair Lawn, NJ).

2.2. Fabrication method

About 1.4 g (70%) of PEG 600 or PEG 300 containing 10% of emulsifier, Tween 80 or P-10, were melted at a specific temperature. High-molecular-weight or low-molecular-weight PCL (600 mg, 30%) was added and the whole melted mixture was then mixed for 1 to 3 min at 70% output power with a Sonic Dismembrator® (Fisher Scientific). The melted blends were cooled rapidly in a freezer at −20°C until solidified. Twelve formulations were prepared according to the fabrication conditions listed in Table 1.

2.3. Characteristics of microspheres by scanning electron microscopy (SEM)

The prepared blends were dispersed in HPLC-grade water. The suspension was taken and washed with distilled water. The washed solution was centrifuged and the supernatant was discarded. The above procedure was repeated, and the resulting PCL particles were then placed on a SEM stage for drying in vacuo in a desiccator prior to SEM analysis. All samples were coated under an argon atmosphere with gold–palladium and observed with a Hitachi model S-4000 scanning electron microscope.

2.4. Molecular weight determination by gel permeation chromatography (g.p.c.)

The PCL suspension (1 ml) was filtered through a 0.22 μm membrane (Millipore, GVWP) and washed with distilled water to remove PEG. The PCL particles were then dried in vacuo in a desiccator. About 5 mg of dried PCL was weighed and dissolved in 1 ml of chloroform. Each chloroform solution was filtered through a 0.45 μm filter (Millipore, sterile MillexHV) prior to g.p.c. analysis. An Ultrastyragel 7.8 mm × 30 cm linear column (Waters, Milford, MA) was used, with chloroform as the eluting solvent at a flow rate of 1 ml min^{-1} at 35°C and a refractive index detector (Shimadzu RID-6A, Columbia, MD). The g.p.c. procedure was calibrated by using polystyrene standards of different molecular weights. The molecular weight ($M_{g.p.c.}$) corresponding to the peak of the g.p.c. chromatogram was determined.

3. Results and discussion

The application of blends of PCL and PEG was explored to prepare PCL microspheres. The first batch of microspheres was prepared according to formulation 5, and the detailed fabrication conditions are shown in Table 2. The original molecular weight of PCL was 21 300, and the molecular weight of PEG, used as an external phase, was 600. The microspheres were prepared at 130°C for 2 min in the

Table 1
Fabrication conditions for preparation of blank PCL microspheres

Formula (no.)	*MW*		Emulsifier	Time (min)	Temp. (°C)	Plasticizer (Triacetin[a])
	PCL	PEG				
5	21 300	600	Tween 80	2	130	—
2	21 300	600	P-10[b]	2	130	—
3	5 000	600	Tween 80	2	130	—
4	5 000	600	P-10[b]	2	130	—
9	21 300	600	Tween 80	2	130	10%
10	21 300	600	Tween 80	2	130	5%
11	21 300	600	Tween 80	1	130	—
12	21 300	600	Tween 80	3	130	—
13	21 300	600	Tween 80	2	70	—
14	5 000	600	Tween 80	2	70	—
15	21 300	300	Tween 80	2	70	—
16	5 000	300	Tween 80	2	70	—

[a]1,2,3-Propanetriol triacetate.
[b]Poly(oxyethylene 10) tricetyl acetate.

Table 2
The fabrication condition for formulation 5

PCL phase	30% (w/w) PCL 21 300
PEG phase	70% (w/w) PEG 600
Emulsifier	10% (w/w) Tween 80 (based on PEG)
Homogenization time	2 min
Temperature	130°C

presence of an emulsifier (Tween 80) to stabilize the two phases. The morphology of the PCL microparticles thus prepared was examined by SEM, and the scanning electron micrograph is displayed in Fig. 1. The size of the PCL microparticles was in the range of 1 to 20 μm (mostly in the range 5–10 μm). This result suggested that PCL acts as an internal phase which is dispersed as spherical droplets in the PEG continuous phase during the fabrication process. The molecular weight of these freshly prepared PCL microparticles was examined by g.p.c. No change in the molecular weight distribution and peak shape was observed, which indicated that the PCL matrix was stable during the whole fabrication process.

An alternative method for the preparation of solvent-free PCL microspheres was successfully developed from PCL 21 300 and PEG 600 blends. The influences of the molecular weight of PCL, the molecular weight of PEG, the type of emulsifier, the concentration of plasticizer, the homogenization time and temperature on the characteristics of the PCL microparticles were investigated. A variety of formulations were designed based on formulation 5 in which the fabrication conditions were varied. The characteristics of microspheres obtained from the 12 formulations are listed in Table 3.

3.1. Effect of PCL molecular weight

Formulation 3 was similar to formulation 5 except that low-molecular-weight PCL (PCL 5000) was used in place of high-molecular-weight PCL (PCL 21 300). A scanning electron micrograph of formulation 3 is shown in Fig. 2. The microspheres made from formulation 3 were coalesced into a continuous mass on drying and lost their properties as individual spheres. A similar result has been reported by Coffin [12]. However, this observation is very different from what was observed for microspheres made from PCL with an initial molecular weight of 21 300. As in formulation 5, no change in the molecular weight of the PCL in freshly prepared microspheres was observed for formulation 3.

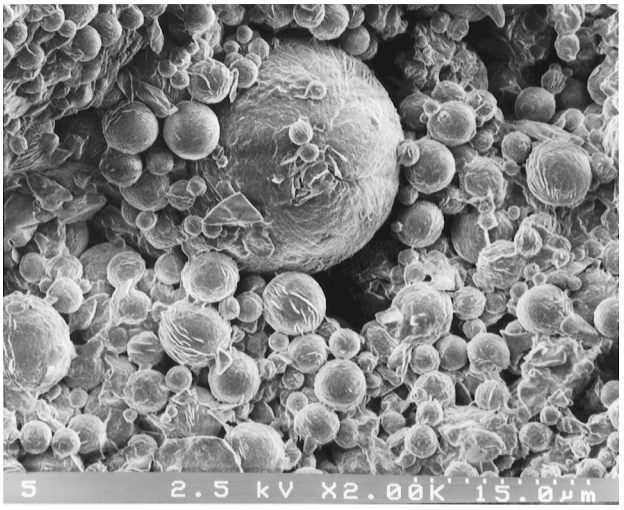

Fig. 1. Scanning electron micrograph of microspheres prepared from formulation 5 (PCL 21 300, PEG 600, Tween 80, 2 min, 130°C).

3.2. Effect of emulsifier

Formulation 2 was similar to formulation 5 except that another emulsifier, poly(oxyethylene 10) tricetyl acetate (P-10), was used instead of Tween 80. The morphology of the microspheres prepared, as observed by scanning electron micrography, was similar to that of formulation 5, and the molecular weights of PCL before and after fabrication were 21 300 and 22 200, respectively. The influence of emulsifier type on the characteristics of PCL microspheres was also investigated between formulations 3 and 4. Low-molecular-weight PCL (PCL 5000) was used in both formulations instead of high-molecular-weight PCL (PCL 21 300) for preparation of the microspheres. The molecular weight of PCL in freshly prepared microspheres was similar to the original one irrespective of the type of emulsifier. Again, coalescence of the PCL matrix was observed for formulations 3 and 4 where a low molecular weight of PCL (5000) was used.

3.3. Effect of plasticizer

The influence of plasticizer concentration on the fabrication of PCL microspheres was compared among three formulations (nos 5, 9 and 10). There was no plasticizer present in formulation 5; however, 10% and 5% of 1,2,3-propanetriol triacetate (Triacetin) were added in the external phase of formulations 9 and 10, respectively. Similar results for the morphology and molecular weight of PCL microspheres were obtained among the three formulations irrespective of the concentration of plasticizer. This result is not surprising, since the plasticizer would modify the glass transition temperature of the polymer in its glassy-rubbery state rather than in the melted state.

3.4. Effect of homogenization time

Three formulations were prepared to demonstrate the effect of mixing time on the characteristics and stability of PCL microspheres. The mixing time for formulations 11, 5 and 12 was 1, 2 and 3 min, respectively. Increasing the time period of mixing would decrease the particle size of the spheres slightly. The molecular weight of the PCL was

Table 3
Characteristics of PCL microspheres

Formula (no.)	$M_{g.p.c.}$		SEM
	Before	After	
5	21 300	21 300	sphere
2	21 300	22 200	sphere
3	5 000	5 000	coalesced
4	5 000	4 900	coalesced
9	21 300	20 500	sphere
10	21 300	20 100	sphere
11	21 300	20 600	sphere
12	21 300	20 100	sphere
13	21 300	20 100	sphere
14	5 000	5 000	coalesced
15	21 300	21 100	sphere, smooth
16	5 000	4 900	coalesced

changed by less than 6% even after mixing at 130°C for 3 min.

3.5. Effect of temperature

The melting point of PCL is quite low, around 60°C. Therefore 70°C, which is 10°C above the melting temperature of PCL, was selected as another fabrication temperature to demonstrate the effect of fabrication temperature on the morphology and stability of PCL microspheres. Two formulations, 5 and 13, were prepared and compared. No significant differences in both morphology and molecular weight of particles were observed when the lower temperature of 70°C was applied instead of 130°C during fabrication. This observation is very important, since the mild fabrication temperature used in formulation 13 would allow most pharmaceuticals, vaccines, peptides and proteins to be encapsulated with minimal decomposition and/or denaturation.

3.6. Effect of PEG molecular weight

It was demonstrated that a mild temperature (70°C) can be used to prepare PCL microspheres with the current polymer-melt technique. Hence, the influence of changes in viscosity of the external PEG phase on microsphere fabrication was investigated further between formulations 13 and 15. A scanning electron micrograph of formulation 15, which was prepared at 70°C with use of PEG 300 instead of PEG 600 as the external phase, is shown in Fig. 3. The surface of the microspheres obtained from formulation 15 was much smoother than that from formulation 13 and the other formulations. The stability of the PCL matrix remained, and no change in molecular weight was observed for PCL before and after fabrication. The drug-loading efficiency in PCL microspheres prepared from the polymer-blend melt technique was tested by using progesteron as a model drug. In this preliminary study, the loading efficiency of progesterone was shown to be high, above 90%, in the

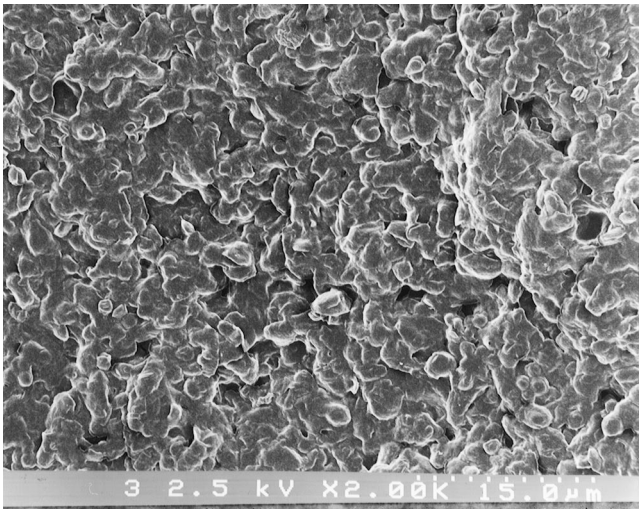

Fig. 2. Scanning electron micrograph of microspheres prepared from formulation 3 (PCL 5000, PEG 600, Tween 80, 2 min, 130°C).

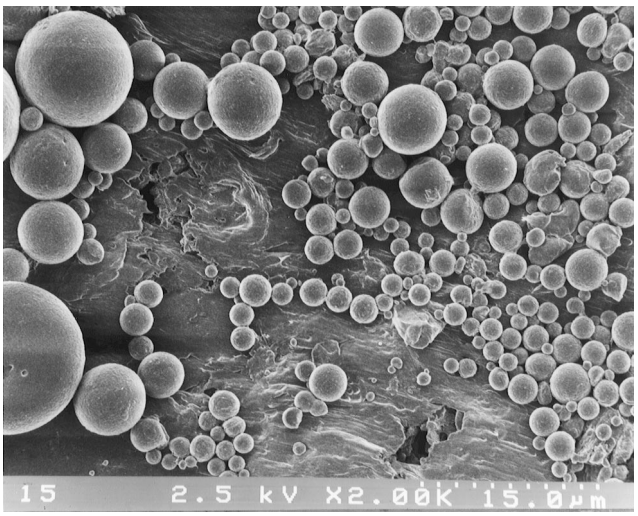

Fig. 3. Scanning electron micrograph of microspheres prepared from formulation 15 (PCL 21 300, PEG 300, Tween 80, 2 min, 70°C).

PCL microspheres prepared under the same conditions as formulation 15.

4. Conclusions

A novel method for the preparation of solvent-free PCL microspheres from PCL/PEG blends was developed and demonstrated. The major advantage of this polymer-blend melt technique compared with traditional methods for microsphere preparation is the elimination of methylene chloride and other organic solvents for polymer solubilization. Therefore, the toxicity associated with organic solvent residues resulting from the conventional solvent preparation of microspheres is not present with this polymer-blend melt technique. Moreover, the ease of fabrication, high drug loading and lack of solvent toxicity mean that the polymer-blend melt technique developed here has great potential to be applied in drug-delivery systems.

Acknowledgements

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References

[1] Lewis DH et al. In: Chasin M, Langer R editors. Biodegradable polymers as drug delivery systems. New York: Marcel Dekker 1990:1–41.
[2] Owusu-Ababio G, Rogers JA. Journal of Microencapsulation 1996;13:195.
[3] Wagenaar BW, Muller BW Biomaterials 1994;15:49.
[4] Guzman M, Molpeceres J, Garcia F, Aberturas MR. Journal of Microencapsulation 1996;13:25.
[5] Igartua M, Hernandez RM, Esquisabel A, Gascon AR, Calvo MB, Pedraz JL. Journal of Microencapsulation 1997;14:349.
[6] Conti B, Panico M, Ventura CA, Gianchedi P, Puglisi G. Journal of Microencapsulation 1997;14:303.
[7] Dash AK. Journal of Microencapsulation 1997;14:101.
[8] Shiga K, Muramatsu N, Kondo T. Journal of Pharmacy and Pharmacology 1996;48:891.
[9] Torres AI, Boisdron-Celle M, Benoit JP. Journal of Microencapsulation 1996;13:41.
[10] Uchida T, Yoshida K, Goto S. Journal of Microencapsulation 1996;13:219.
[11] Lin WJ. Degradation studies on poly(ε-caprolactone) and its blends, Ph.D. thesis, Iowa City, IA: University of Iowa, College of Pharmacy, 1995.
[12] Coffin MD. The development and physical–chemical properties of biodegradable pseudolatexes and their application to sustained release drug, Ph.D. thesis. Austin, TX: University of Texas at Austin, 1990.