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Characterization and *in vitro* release of praziquantel from poly(ε-caprolactone) implants

Liang Cheng [a], Shengrong Guo [a,*], Weiping Wu [b]

[a] School of Pharmacy, Shanghai Jiao Tong University, 800 Dongchuan Road, Shanghai 200240, China
[b] National Institute for Parasitic Diseases, Chinese Center for Control and Prevention, 207 Ruijin 2nd Road, Shanghai 200025, China

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ABSTRACT

Poly(ε-caprolactone) (PCL) implants containing praziquantel (PZQ), a broad-spectrum antiparasite drug, are fabricated by injection molding and characterized in terms of content uniformity, morphology, drug physical state and stability. *In vitro* drug release from the implants is also studied. It is found that drug is dispersed uniformly in all implants and keeps stable over 365 days at 4 °C/60% RH. X-ray diffraction analysis reveals that PZQ exists primarily in its crystalline state in implants with high drug contents (50% and 25%). All implants exhibit similar release behaviors and about 70% of the drug is released after 365 days. The cross-sections of all implants present two distinct zones (i.e. peripheral white zone and inner pink zone) and the boundary between the two zones changes as time progresses. Drug content in the white zone is very low (less than 1%), but drug content in the pink zone is almost the same as the predefined value. Porous structures in the white zone but dense structures in the pink zone are observed by SEM. Obvious PCL degradation occurs till up to 365 days. These results show that the release process of PZQ is a gradual diffusion from the exterior to the interior of the implants.

1. Introduction

Hydatid disease caused by the larval stage of tapeworm is a worldwide parasitic disease especially in agricultural and pastoral regions (Anadol et al., 2001; Del Brutto, 2005). Dogs are the definitive hosts that harbor the adult stage of the parasite, thus, prevention of tapeworm infection to dogs is the decisive step avoiding transmission of this disease to humans and livestock (Jiao et al., 2005). Praziquantel (PZQ) is a broad-spectrum anthelmintic drug which is one of the first-line drugs to treat this disease (Liu and Weller, 1996; Urrea-Paris et al., 1999, 2001). Many programs have been implemented to remove the parasite in different countries/areas, such as administrating PZQ-contained tablets or baits to dogs once or twice a week (Gemmell et al., 1986). Although a decreasing morbidity is achieved, frequent administration is not only time-wasted, but also costly.

To overcome these shortcomings, Jiao et al. (2005) have developed a long-term sustained release implantable PZQ-containing bar. The preparation processes of this bar are detailed as follows: firstly, blend the drug and release modifier kaolin at a weight ratio of 9:1; then, fill the mixture into a silicone tube and seal the tube with silicone wafers. Even that the PZQ-loaded bars can exert an effective prevention of tapeworm infection to dogs, there are some deficiencies associated with this drug delivery system (DDS). The PZQ-loaded bar is a reservoir-type DDS which could run a risk of drug dumping after *in vivo* implantation. In addition, the preparation of this bar is tedious so that a large-scale manufacture is difficult to be carried out.

Poly(ε-caprolactone) (PCL), a polyester biomaterial, has been approved by FDA. Up to now, homo- and co-polymers derived from PCL are being extensively utilized to prepare various controlled or prolonged drug delivery systems for drugs and proteins (Zhang et al., 2009; Dhanaraju et al., 2003; Hombreiro Pérez et al., 2000; Wang et al., 2008; Wang and Guo, 2008; Wei et al., 2009; Jia et al., 2008). Since it exhibits slow biodegradation rate *via* bulk hydrolization of the ester bonds (Dordunoo et al., 1997; Fialho et al., 2008) and high permeability to low molecular species at body temperature (Edlund and Albertsson, 2002; Sinha et al., 2004), PCL is considered ideal for implantable long-term drug delivery. Capronor®, a 1-year contraceptive represents such a system. Moreover, due to the excellent thermal properties of PCL, such as a low melting temperature of 60 °C and a low glass transition temperature of −60 °C irrespective of the molecular weight (Barbato et al., 2001; Fialho et al., 2008), PCL-based long-term drug delivery devices can be prepared by means of some simple processing techniques, including melt molding (Carcaboso et al., 2008; Rosenberg et al., 2008) or extrusion (Lemmouchi et al., 1998a,b), without risking the integrity of the polymer and the

* Corresponding author. Tel.: +86 21 3420 4793; fax: +86 21 3420 4793.
E-mail address: srguo@sjtu.edu.cn (S. Guo).

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incorporated drug. In recent years, injection molding (IM) is introduced to pharmaceutical industry for its unique advantage over above-mentioned techniques, i.e., ease to large-scale manufacture. Several research groups have adopted this technique to prepare some implantable devices using PLA or polyanhydrides successfully (Soriano et al., 2006; Deng et al., 2001; Rothen-Weinhold et al., 1999).

Thus, one aim of this study is to prepare a PZQ-loaded sustained-release PCL implant by IM and to characterize the resultant implants in terms of drug thermal stability during the preparation process, content uniformity, physical state of drug and implant stability under storage condition (4 °C/60% RH).

On the basis of the strong hydrophobic property and a low degree of swelling in aqueous solutions (Yoon et al., 2000), the release mechanism of either hydrophilic or hydrophobic drug from PCL-based delivery devices has been demonstrated to be predominated by slow diffusion through PCL matrix or the voids left by the depleted drug (Rosenberg et al., 2007; Lao et al., 2008). Thereby, in most cases drug release rate from a PCL-based device should be somewhat dependent on the payload, i.e., the higher the payload, the faster the drug release. Interestingly, the fabricated PZQ-loaded PCL implants in the present study represent similar *in vitro* release profiles over 1 year independent of the drug payloads (6.25–50%). Upon that, the other aim of this study is to investigate the dynamic drug release in combination with a series of changes happened on the implants, including drug distribution in the implants, microstructures (surface and interior) of implants and degradation of PCL matrix.

2. Materials and methods

2.1. Materials

Poly(ε-caprolactone) was purchased from Daicel Polymer Ltd. (Minatoku, Tokyo, Japan). Praziquantel (PZQ) was obtained from Shanghai Jiachen Chemical Industry Co. Ltd. (Shanghai, China). All other chemicals were of analytical grade and used without further purification.

2.2. Fabrication of implants containing PZQ and PCL

The implants were prepared by fully blending PZQ particles with melting PCL at different ratios (1:15, 1:7, 1:3, 1:1, w/w) and then molding the blends into cylindrical implants by a lab-scale injection molder. In brief, PCL was fed into the hopper of HAAKE Rheocord System (Rheocord 90, HAAKE Mess-Technic GmbH, Germany) and heated until it was completely melted. Afterward PZQ was added slowly into the melting PCL and mixed at 70 °C for 20 min at a screw speed of 50 rpm. The resultant blends were collected and further molded into implants using a lab-scale injection molder at 70 °C (HAAKE MiniJet System, Germany). After cooling, the implants in the shape of pointed cylinder were obtained. The implant weighed 900 mg.

2.3. Scanning electron microscopy (SEM)

The implants were imaged using a JSM-7401F scanning electron microscopy (SEM) (JEOL, Tokyo, Japan). To image the interior structure of the implants, the cross-section of implant was obtained by first freezing it in liquid nitrogen and then fracturing by means of a scalpel. Samples were placed on metal sample holders and sputter coated (Emitech K-575 Sputter Coater) with a gold–palladium target for 30 s at 20 mA prior to imaging. Images were obtained at 1 kV accelerating voltage and 20 mA current.

2.4. Molecular weight determination

The molecular weight of PCL was determined by gel permeation chromatography (GPC) using a PerkinElmer 200 equipment (PerkinElmer, Inc., USA). Dimethyl formamide (DMF) was used as the mobile phase; the injected sample volume was 10 μl; the temperature of column was 25 °C; the flow rate was 1.0 ml/min. Polystyrene standards of known molecular weight were used as reference materials.

2.5. X-ray diffraction analysis (XRD)

X-ray diffraction patterns were obtained using an X-ray diffractometer (D/max 2200, Rigaku, Japan) equipped with Cu Kα radiation source (40 kV, 20 mA). Before analysis, all samples were pressed into thin films on a Compression Molding Machine (XLB-D, Shanghai No. 1 Rubber Machine Factory) at ambient temperature. Then the films were placed in a steel holder and scanned over a 2θ range of 5–45° at the rate of 5°/min.

2.6. Hot stage microscopy (HSM)

A hot stage plate (FP 52 Mettler, Greifensee, Switzerland), connected to a temperature controller, was used. A little amount of sample was placed on a glass slide and heated at 10 °C/min within the temperature range of 30–150 °C. The dynamic changes were monitored *via* an optical microscope.

2.7. High performance liquid chromatography analysis (HPLC)

PZQ was quantitatively analyzed by a Shimadzu HPLC apparatus using the method described in the Chinese pharmacopeia 2005 (CP2005). The HPLC system was equipped with a variable wavelength detector (SPD-10ADvp, Shimadzu, Japan). A pump (model LC-10AD, Shimadzu, Japan) was used at a constant flow rate of 1.0 ml/min. A C-18 reversed-phase column (4.6 mm × 250 mm, 5 μm, Dikma Technologies, Beijing, China) was used for analysis of the drug. The mobile phase was a mixture of methanol and ultrafiltrated water (100:40). The injection volume for the standard and the sample preparations was 20 μl, and the column effluent was monitored at the UV absorption wavelength of 263 nm. The linear dynamic range for the quantification of PZQ was calculated to be 0.1–1000 μg/ml.

2.8. In vitro release assay

The dissolution test was performed using the rotating basket method (CP2005) on a dissolution tester. Sink conditions were maintained throughout the assay period. Implants were weighed and placed in 900 ml release medium. The release medium used in the present study was 2 mg/ml sodium lauryl sulphate (SLS) aqueous solution. The rotating speed of the basket was set at 50 rpm, while the temperature of the release medium was maintained at 37 °C. At predetermined time intervals, aliquots of 5 ml were withdrawn, filtered and stored at 4 °C until HPLC analysis, whilst the whole release medium was replaced with fresh SLS-aqueous solution. The measurement was performed in triplicate for each batch.

2.9. Content uniformity and stability of PZQ in implant

The determination of content uniformity of PZQ in the PCL implants was performed according to the method stated in the general chapter of the CP2005. Ten implants were selected and weighed. Each implant was dissolved in 20 ml acetonitrile, subsequently 80 ml methanol was added to precipitate the PCL. The

resulting suspension was centrifuged at 10,000 × *g* for 10 min and the obtained supernatant was filtered and then analyzed by HPLC.

Stability test of PZQ in the four implant batches was performed at 4 °C/60% RH for 12 months. At pre-set time intervals, drug remaining in the implants was determined by HPLC and the relative drug content was calculated as the following expression.

Relative drug content (%) = drug content determined at different time points/initial drug content × 100.

3. Results and discussion

3.1. Characterization of PZQ-loaded PCL implants fabricated by IM

PZQ is a hydrophobic drug with a water solubility of 0.4 mg/ml and appears as white prismatic crystals (Fig. 1). Drug particles with the length smaller than 15 μm were used in this study. PCL, one white hydrophobic polyester material, functioned as binder during the fabrication process, due to its appropriate viscosity when heated up to 60 °C. It was interesting that all the fabricated implants with different drug loadings presented a color of slight pink. We speculated that the most possible reason for this discoloration was the decomposition of PZQ during the fabrication process, which involved heat, shear and pressure. To verify our hypothesis, PZQ was extracted from the implants and subjected to H^{-1} NMR and HPLC analysis. It exhibited that the extracted PZQ and the raw PZQ not only had identical NMR spectrums (not shown), but the same retention time (9.2 min). No additional peak was observed in the HPLC trace of the extracted PZQ. These results demonstrated that PZQ was stable during the fabrication process. The change in molecular weight of PCL was also determined by GPC right after the fabrication process. The weight average molecular weight (M_w) of the raw material was 90,000, while the M_w of PCL after IM was 91,140. There was no significant difference in M_w between the raw material and the processed material, revealing that PCL was also stable during the fabrication. The exact reason for the discoloration was not clear yet.

A possible drawback associated to IM is the uneven drug dispersion in resultant pharmaceutical products, due to a short mixing time. To avoid the occurrence of this case, a combination of twin-screw mixing and IM was adopted to fabricate PZQ-loaded PCL implants. As listed in Table 1, the determined PZQ content values in all implants were almost equal to the predefined values of PZQ in the implant formulations. Moreover, all calculated relative deviation values were quite low. These data indicated that PZQ particles were well dispersed in implants.

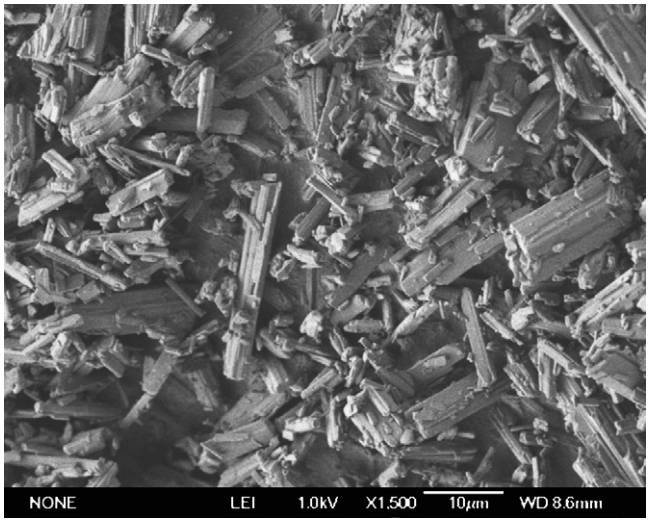

Fig. 1. SEM image of praziquantel (PZQ) particles.

Table 1
Drug contents in the fabricated PZQ-loaded PCL implants (A ± s, *n* = 10).

Implant	Feed ratio of PZQ and PCL (w/w)	Drug content (%)	
		Predefined value	Determined by HPLC
F1	50/50	50	49.9 ± 0.6
F2	25/75	25	24.5 ± 0.4
F3	12.5/87.5	12.5	12.3 ± 0.5
F4	6.25/93.75	6.25	6.1 ± 0.3

The morphologies (surface and cross-section) of implants were observed by SEM. The surfaces of all implants were smooth and devoid of any pores and cracks (shown in Fig. 2a), indicating a well coalescence of melting PCL after IM. However, there were obvious differences among the cross-sections of implants with different drug loadings. For F1 and F2, many prismatic particles, corresponding to PZQ crystals, were observed and they were distributed in PCL matrix separately (Fig. 2b). For F3 and F4, dense and homogeneous cross-sections with no drug particles were observed (Fig. 2c), which were similar to those of blank implant (Fig. 2d). The causes for the absence of drug particles in the cross-sections of F3 and F4 may be that in this situation, the majority of drug crystals were dissolved in the PCL matrix during the fabrication process. Many researches have reported the fact that partial crystalline drug changed into its amorphous or molecular state during a melting fabrication process (Quinten et al., 2009; Andrews et al., 2008).

To elucidate the different morphologies between the implants, all implant samples were subjected to XRD analysis. XRD patterns of PZQ, PCL, physical mixtures, and implants consisting of PCL and PZQ were presented in Fig. 3. PZQ is crystalline, as demonstrated by sharp and intense diffraction peaks at 6.22°, 7.92° and a series of peaks above 10°. Its XRD pattern corresponded to that of PZQ racemate crystal, as reported in literatures (Passerini et al., 2006; Liu et al., 2004). PCL displayed two characteristic peaks between 20° and 25°, confirming its semi-crystalline structure (Wang et al., 2008). For all the implants, there existed obvious crystalline peaks of PCL, indicating PCL was still in its crystalline state in implants. PZQ crystalline peaks for the implants with high drug loadings (F1 and F2) and all physical mixtures of PCL and PZQ were obvious, whereas those for implants with low drug loadings (F3 and F4) were hardly detectable. Moreover, slightly higher peak intensity was observed for physical mixtures with high drug loadings in comparison with their corresponding implants. This suggested that a certain fraction of PZQ crystals were dissolved or dispersed in molecular or amorphous state in the PCL matrix.

To further investigate the physical state of PZQ in implants, physical changes of all implant samples during heating were monitored by hot stage microscopy (HSM). Pure PZQ underwent melting in the range of 137–140 °C. PCL particles started to melt at about 56 °C, and then a transparent clear fusion formed at 63 °C. For F1 and F2, after the initial fusion of PCL during 56–65 °C, lots of prismatic crystals came into view and these crystals started to melt until heating to 110 °C. In contrast, the melting process of F3 and F4 was nearly the same as that of raw PCL. These results revealed that PZQ existed primarily in amorphous or molecular state in implant batch F3 and F4, on the contrary, in crystalline state in implant batch F1 and F2.

Generally, drug stability in preparations determined the shelf life, in the present study, PZQ content in the four implant batches was determined by HPLC at different time points. As shown in Fig. 4, values of drug content were nearly kept constant throughout the stability study. To verify whether the physical state of PZQ changed, all implants were simultaneously subjected to XRD analysis and identical XRD patterns were obtained. These results indicated that PZQ was stable in all implants under the storage condition over 365 days.

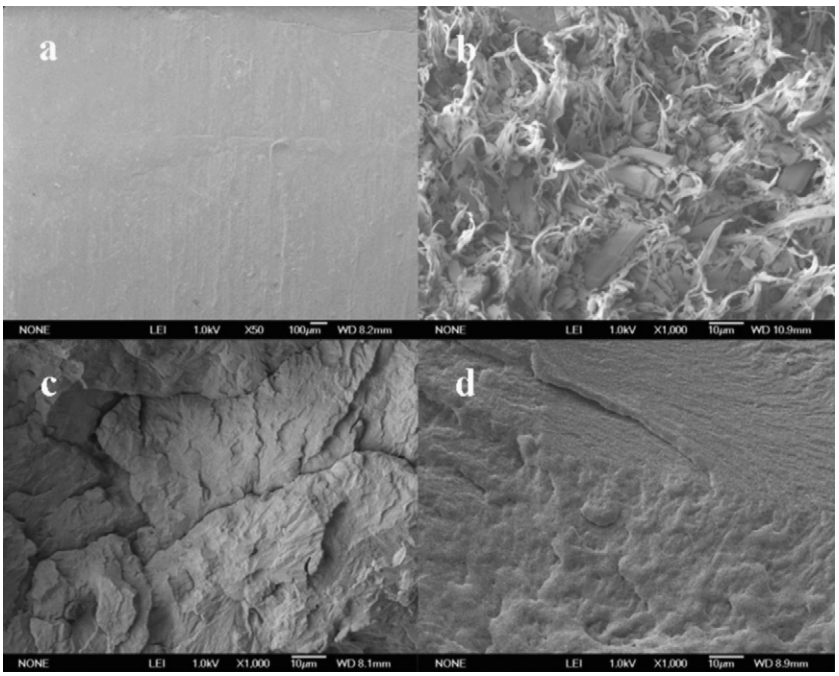

Fig. 2. Surface and cross-section SEM images of implants right after the fabrication process: (a) surface of F1 implant; (b) representative cross-section of F1 and F2; (c) representative cross-section of F3 and F4; (d) cross-section of blank implant.

3.2. *In vitro drug release test*

Fig. 5a shows the cumulative release curves of PZQ from PCL implants over a period of 365 days. The convex release curves could be divided into two phases—initial burst release phase and subsequent sustained release phase. The percentage of cumulative drug release during the first day was expressed as the initial burst release. There were $6.6 \pm 0.28\%$, $6.4 \pm 0.26\%$, $8.0 \pm 0.29\%$ and $8.4 \pm 0.41\%$ of PZQ released from the four implant batches within 1 day, respectively. The burst release could be ascribed to immediate dissolution of the drug located on or near the surface of implants following immersion in the release medium (Bourges et al., 2006). Following the burst release, PZQ release rate gradually slowed down and tended to be a constant regime (shown in Fig. 5b).

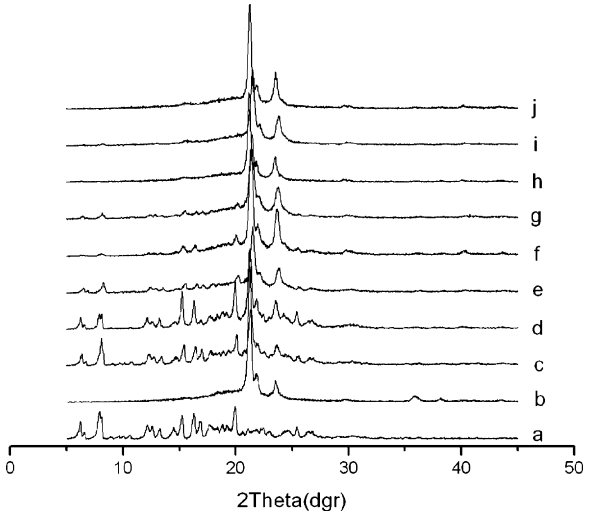

Fig. 3. X-ray diffraction patterns of pure PZQ (a), neat PCL (b), physical mixtures of PCL and PZQ with drug fractions as 50%, 25%, 12.5% and 6.25%, respectively (c, e, g and i); and implants with different drug contents as 50%, 25%, 12.5% and 6.25%, respectively (d, f, h and j).

Ritger–Peppas's equation (Ritger and Peppas, 1987) was utilized to illustrate the mechanism of drug release:

$$\frac{M_t}{M_\infty} = kt^n$$

where M_t/M_∞ is the fractional drug release, t is the release time, k is a constant, and n is the diffusional exponent characteristic of the release mechanism. It is shown that in cases of pure Fickian release the exponent n has the limiting values of 0.50, 0.45 and 0.43 for release from slabs, cylinders and spheres, respectively. The n values were calculated by the equation based on the four release profiles. All n values were less than 0.43 (Table 2), confirming that the release mechanism of PZQ from the four implant batches was a typical Fickian diffusion.

However, it was interesting that the four implant batches had similar cumulative release profiles over the whole *in vitro* release test. In order to explore the similarity of these release profiles, f_2 value, called as similarity factor, was introduced (Gao et al., 2007;

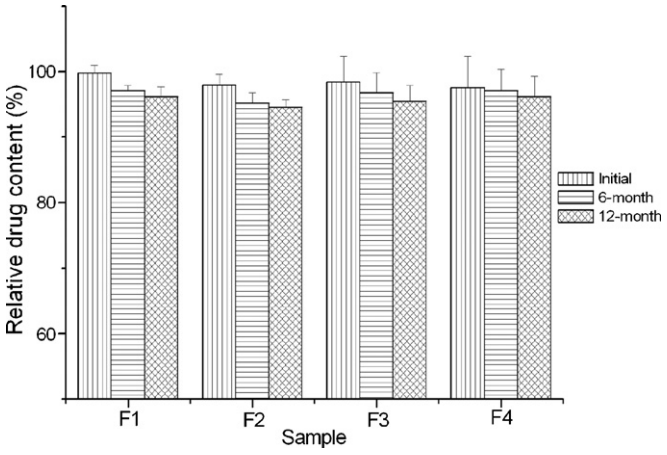

Fig. 4. Relative PZQ contents in implants at 4 °C for 12 months.

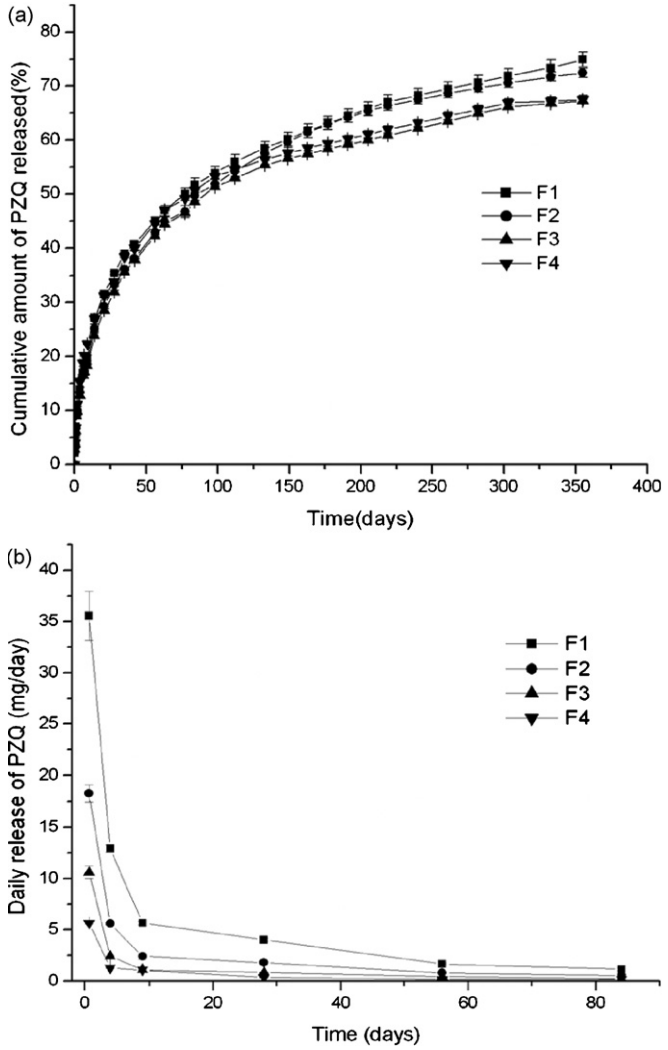

Fig. 5. Cumulative PZQ release from PCL implants containing different amount of PZQ for 365 days (a), daily release of PZQ (mg/day) as a function of time (b).

Moore and Flanner, 1996):

$$f_2 = 50 \log \left\{ \left[1 + \frac{1}{n} \sum_{t=1}^{n} W_t (R_t - T_t)^2 \right]^{-0.5} \times 100 \right\}$$

where R_t is the reference drug release content at time point t, T_t is the test drug release content at time point t, n is the number of the sampling points, and W_t is an optional weight factors. Since all the release time points were treated equally, W_t was taken as 1. According to the guidance recommended by FDA, generally, two release profiles may be regarded as equivalence while the f_2 value is greater than 50. The values of f_2 were all more than the critical value 50 (shown in Table 3). This suggested that the change in drug loadings may not cause significant difference among the four release profiles. We speculated an appropriate explanation was that: for implant batches with low drug loading (F3 and F4), drug release was dominated solely by drug molecular diffusion through PCL matrix due to the formation of an amorphous solid solution, which was validated by XRD and HSM analysis; however, for implant batches with high drug loading (F1 and F2), drug release was dominated by two steps—the initial dissolution of drug crystals into release medium permeating into PCL matrix and subsequent drug molecular diffusion through PCL matrix or voids left by released drug particles. Generally, amorphous substances have higher solubility and dissolution rate than the corresponding thermodynamically stable crystalline forms, because their internal bonding forces are weak (Albers et al., 2008). Based on some reported experiments in attempts to improve the dissolution of PZQ (Passerini et al., 2006; Becket et al., 1999), the dissolution rate of PZQ in amorphous or molecular state can be at most 10-fold greater than that of drug crystals. Consequently, for implant batch F3 and F4 enhanced dissolution rate could counteract the retarding effect generated by the longer diffusion distance at some extent; while for implant batch F1 and F2 though drug diffusion distance was relatively short, additional time was needed to dissolve drug crystals.

Table 3
Values of similarity factor for the F1–F4 release profiles.

Similarity factor	Value
$f_{2\ (F1,F2)}$	86.7
$f_{2\ (F1,F3)}$	69.3
$f_{2\ (F1,F4)}$	73.7
$f_{2\ (F2,F3)}$	74.9
$f_{2\ (F2,F4)}$	76.0
$f_{2\ (F3,F4)}$	83.6

3.3. Study of PZQ release process from the PCL implants

3.3.1. Macroscopic study of drug release from PCL implants

At predefined time points, implants with different drug loadings were removed from release medium, washed by distilled water, then freeze-dried and imaged by digital camera. As shown in Fig. 6a, the implants were basically intact after a release period of 365 days, and there just existed some cracks on the surface of implants. Furthermore, an evident change in color of implant appearance was observed, pink implants turned into white ones. Cutting off the implants using a razor blade, the cross-sections of implants presented two distinct zones—peripheral white zone and inner pink zone. As release time extended, there existed a dynamic boundary between the above-mentioned two zones: peripheral white zone became increasingly thick, but inner pink zone became increasingly thin (Fig. 6b). Based on this phenomenon, we speculated that the changing two zones had direct relation to the drug release process. Thus, drug content in the two zones was determined by HPLC at different time points respectively.

Table 4 displayed a representative result of determined drug content in the white or pink zone for four implant batches at time point of 365 days. It was obvious that for all implant batches PZQ content in the white zone was very low (less than 1%) in compari-

Table 2
Data of the four *in vitro* drug release profiles fitted by Ritger–Peppas equation.

Implant batch	Equation[a]	n	r	p
F1	$W_t = 7.95t^{0.4239}$	0.4239	0.9934	<0.0001
F2	$W_t = 7.84t^{0.4147}$	0.4147	0.9972	<0.0001
F3	$W_t = 8.02t^{0.3969}$	0.3969	0.9943	<0.0001
F4	$W_t = 9.81t^{0.3633}$	0.3633	0.9935	<0.0001

[a] W_t is equal to M_t/M_∞, $W_t \leq 0.6$, except for M_t/M_∞ for the first day release.

Table 4
Drug contents in white zone and pink zone of the implants after 365-day release.

Implant (drug loading, %)	PZQ contents (w/w, %)	
	White zone	Pink zone
F1 (50)	0.87 ± 0.12	49.4 ± 2.83
F2 (25)	0.32 ± 0.04	23.6 ± 1.95
F3 (12.5)	0.11 ± 0.03	11.5 ± 0.92
F4 (6.25)	0.02 ± 0.00	6.6 ± 0.14

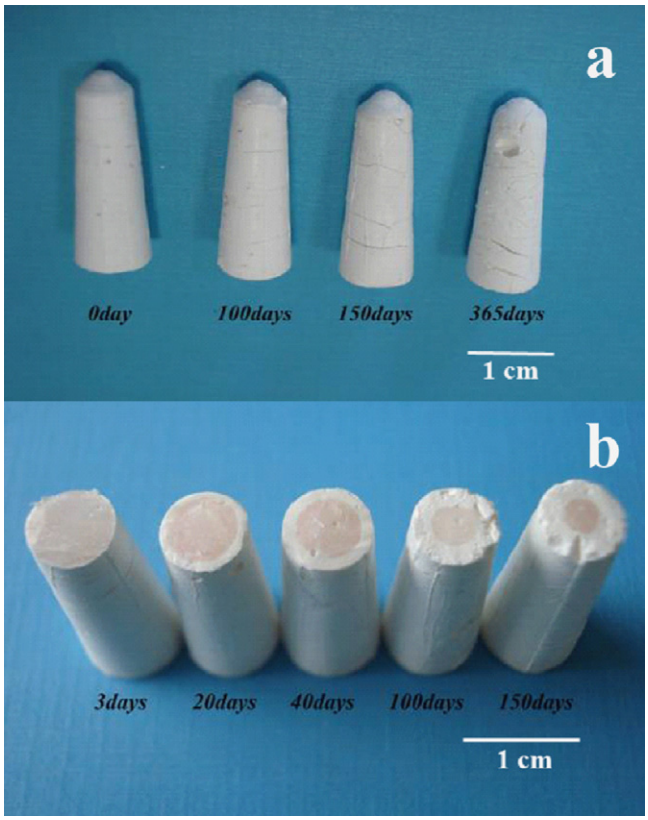

Fig. 6. Global appearance (a) of F1 after 0-, 100-, 150-, 365-day release and cross-section photographs (b) of F1 after 3-, 20-, 40-, 100- and 150-day release, taken by digital camera.

son with that in the pink zone, which was almost the same as the predefined drug loading. At other time points, similar results were also obtained (not shown). It revealed the white zone should be the remaining PCL matrix and functioned as a diffusion barrier after the original drug was depleted; whereas, the pink zone was the zone where drug was still unreleased. These results demonstrated that the *in vitro* release of PZQ from all implants was a process of gradual diffusion from the exterior to interior of implants.

3.3.2. Microstructure analysis by SEM

The surface and cross-section morphologies of implants after 365-day release were characterized by SEM. For F1 and F2, a coarse surface structure with lots of separated pores and cracks was observed (Fig. 7a and b), which could be attributed to the leakage of drug particles located on the surface and the partial erosion of PCL matrix. For F3 and F4, however, there only existed some cracks on the surface (Fig. 7c and d), which might be generated by PCL erosion. The structure of above-mentioned two zones (i.e. white zone and pink zone) was imaged respectively. Many cavities with the same shape as that of PZQ crystals were observed in the white zone of F1 and F2 (Fig. 7e and f), whereas a compact structure was observed in the pink zone at the same magnification (Fig. 7g). At a higher magnification (3000×), PZQ crystals were clearly observed (Fig. 7h). These results validated the foregoing conclusion related to drug release process. The structures of the two zones for F3 and F4 had no obvious difference and were similar to that of surfaces—only some cracks were observed (Fig. 7i and j), which may be also attributed to PCL erosion.

3.3.3. PCL matrix degradation

After *in vitro* release study, the changes in molecular weight (M_w and M_n) of PCL were analyzed by GPC. As shown in Table 5, compared with the initial molecular weight, molecular weights of PCL for both the white and pink zone of implants decreased whatever the sampling sites, indicating PCL degradation occurred. Moreover, an interesting phenomenon was observed that degradation extent of the white zone for F1 was higher than that for F4; but degradation extent of the pink zone for F1 was lower than that for F4. The difference in degradation extent could be attributed to that: in the former case, implants with higher drug loading generated more pores after drug release, hereby more release medium penetrated into the PCL matrix and subsequently facilitated the degradation process; in the later case, the existent weakly basic drug might play a role of an antacid of buffering the acidic degradation products and increasing the degradation half-life of PCL, and the buffering effect would be enhanced as the increasing drug loading. Some previous articles had demonstrated that an incorporation of antacid into polyesters matrix could result in a decreased degradation rate of polyesters (Varde and Pack, 2007; Gaozhong and Schwendeman, 2000; Jaganathan et al., 2005).

PCL mass loss was also studied during the *in vitro* release. As listed in Table 6, over a period of 365 days, F1 and F2 showed mass loss of 36.5% and 33.2% respectively; while F3 and F4 showed mass loss of 12.9% and 11.3% respectively. The more mass loss could be ascribed to the more severe degradation happened in the white zone of F1 and F2, confirmed by SEM and GPC analysis. There was no evident mass loss up to 150 days (shown in Table 7). This result indicated that PCL possessed a very low degradation rate over this period, which was also stated by other authors (Fialho et al., 2008). Although pronounced degradation of PCL occurred after the release study, close to 60% of PZQ was released from all implants, thus, the degradation of PCL just had limited influence on the whole release behavior for all implants.

Table 5
Molecular weights of PCL of F1 and F4 after 365-day release.

Parameters	Before release	After release			
		F1		F4	
		White zone	Pink zone	White zone	Pink zone
M_w	91,140	24,550	41,200	35,390	30,860
M_n	65,590	13,060	30,100	18,820	14,500
PDI	1.39	1.87	1.36	1.88	2.12

Table 6
Mass loss of PCL for four implant batches after 365-day release.

Implant batch	Drug released (%)[a]	Mass loss of PCL (%)[b]
F1	74.9	34.5
F2	72.4	33.2
F3	67.2	12.9
F4	67.5	11.3

[a] Weight of drug released/total weight of drug × 100%.
[b] (The initial weight of implant − the final weight of implant − the weight of drug released)/total weight of PCL × 100%.

Table 7
Mass loss of PCL for implant batch F1.

Time (day)	Drug released (%)	Mass loss of PCL (%)
3	11.7	–
20	31.3	3.3
40	40.5	4.6
100	54.0	6.3
150	57.6	8.7
365	74.9	34.5

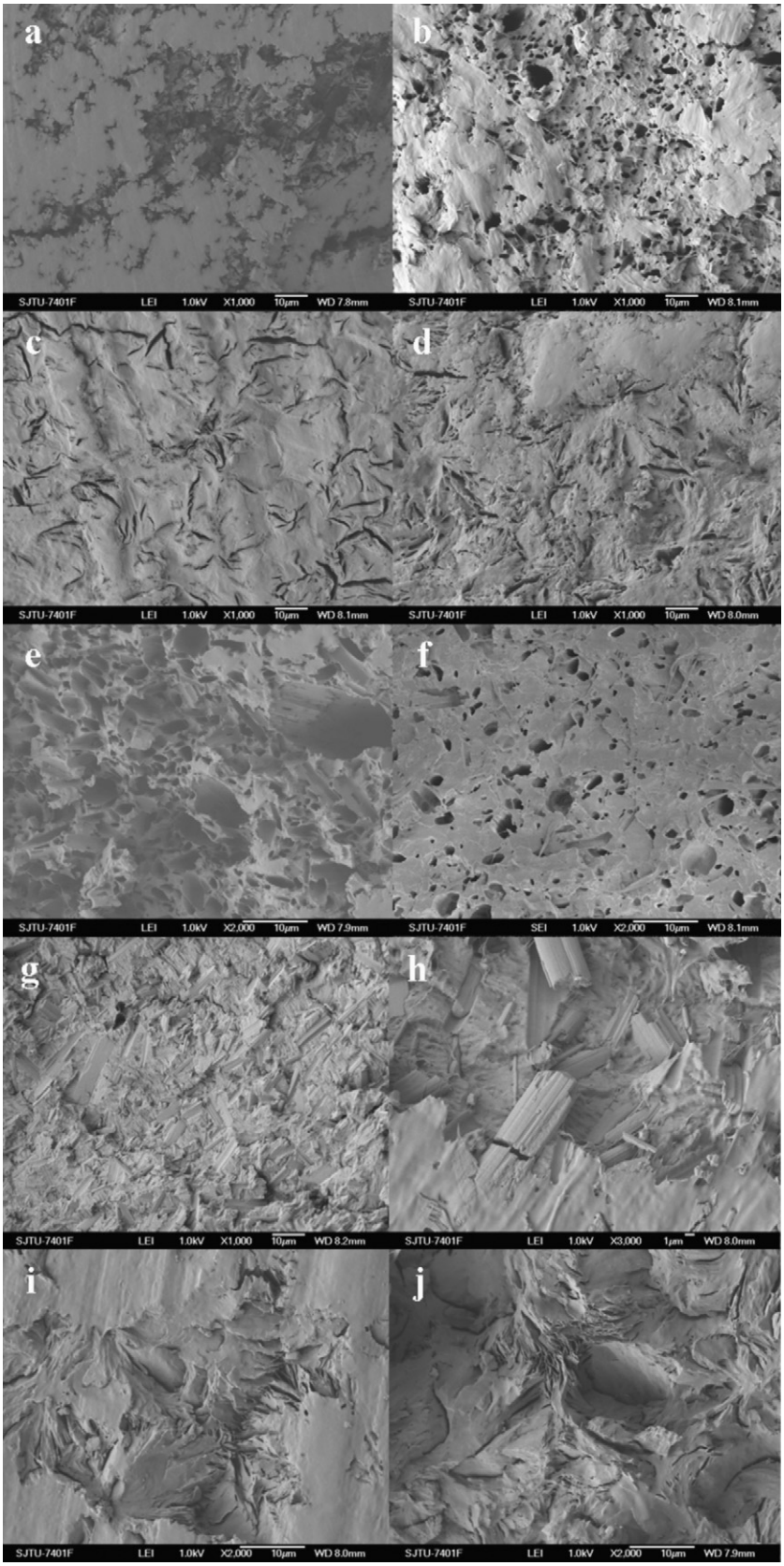

Fig. 7. Surface and cross-section SEM images of implants after 365-day release: (a–d) surface morphology of F1–F4 respectively; (e and f) white zone morphology of F1 and F2 respectively; (g and h) pink zone morphology of F1 and F2 at magnification 1000× and 3000× respectively; (i and j) cross-section morphology of F3 and F4 respectively.

4. Conclusions

This study validates that IM is an alternative to produce a sustained-release polymer-based drug delivery device. Combination of twin-screw mixing and IM can make the specific drug disperse uniformly in the PCL matrix whatever the drug loadings (6.25–50%). A mild processing condition (70 °C, a little higher than the melting point of PCL) is beneficial to the thermal stability of drugs and shows a promising prospect for the preparation of pharmaceutical products incorporating heat-labile drugs. *In vitro* study reveals that the dynamic release process of PZQ is gradual diffusion from the exterior to interior of the implants.

Over the whole *in vitro* release study, it seems that variation of drug loadings will not have significant influence on the release behaviors, the essential reason for this phenomenon is not clear yet. Many factors, including the composition of release medium and other parameters involved in the release test, could have a bearing on the rate and mechanism of drug dissolution. In our future study, these factors will be deeply investigated.

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