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Effect of Solvent on Drug Release and a Spray-Coated Matrix of a Sirolimus-Eluting Stent Coated with Poly(lactic-*co*-glycolic acid)

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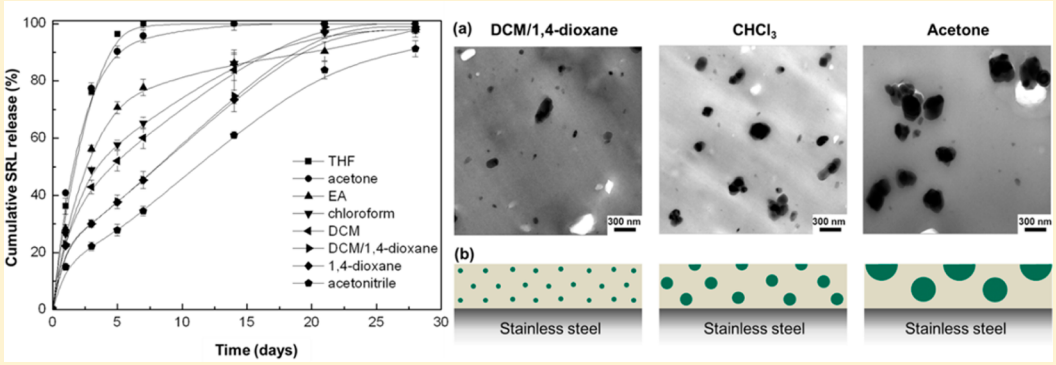
Supporting Information



ABSTRACT: Sirolimus (SRL) release from the biodegradable poly(L-lactic-*co*-glycolic acid) (PLGA) matrix was investigated for the application of drug-eluting stents (DES). In particular, this study focused on whether various organic solvents affect the interaction between SRL and PLGA and the formation of microstructures during ultrasonic coating. The SRL-loaded PLGA coated by tetrahydrofuran or acetone showed a significant initial burst, whereas that from acetonitrile was constantly released during a period of 21 days. On the basis of these results, the interactions at the molecular level of SRL with the polymer matrix were estimated according to various organic solvents. Although the topographies of the coated surface were obviously different, the correlation between surface roughness and SRL release was very poor. Irrespective of organic solvents, FT-IR data showed significantly weak SRL-PLGA interactions. From the result of wide-angle X-ray diffraction, it was confirmed that SRL was dispersed in an amorphous state in the polymer matrix after ultrasonic coating. The glass-transition temperature was also influenced by organic solvents, resulting in a plasticizing effect. The particle size of SRL appeared to determine the release profile from the PLGA matrix, which was the combination of diffusion and polymer degradation at an SRL size of more than 800 nm and the Fickian release at that of less than 300 nm. Therefore, organic solvents can lead to a heterogeneous microstructure in the SRL-loaded PLGA matrix, which is at or near the surface, consisting of aggregated drug- and polymer-rich regions. It is expected that the drug release can be controlled by physicochemical properties of organic solvents, and this study can be used effectively for localized drug release in biomedical devices such as drug-eluting stents.

1. INTRODUCTION

After bare metal stents (BMS) had emerged for the treatment of atherosclerotic coronary artery disease, in-stent restenosis due to neointimal hyperplasia has become a critical concern. Generally, it is believed that the mechanism of in-stent restenosis comprises two main processes: neointimal hyperplasia and vessel remodeling.[1] The implantation of a cardiovascular stent induces a mechanical injury of the blood vessel and provokes cellular and biochemical reactions, associated with thrombus formation and stent-induced inflammation.[2] These responses consequently trigger the migration and proliferation of vascular smooth muscle cells (VSMCs), resulting in the formation of neointimal hyperplasia that is the major determinant.[3] Meanwhile, the molecular mechanisms of artery remodeling are less well understood. The first-generation drug-eluting stents (DES) including Cypher (Cordis, USA) and Taxus (Boston Scientific, USA) were developed for the prevention of in-stent restenosis through the release of antiproliferative drugs such as sirolimus (SRL) and paclitaxel (PTX) from a nonbiodegradable polymer over a

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period of several months.[4−6] However, a synthetic nonbiodegradable polymer as a drug carrier may cause an exaggerated inflammatory response, hypersensitivity reactions, and late thrombotic stent occlusion.[7,8] It is necessary, therefore, to develop a DES with a biodegradable coating capable of improving biocompatibility and controlling drug release.

Poly(lactide-*co*-glycolide) (PLGA) has a lot of characteristic properties such as being an FDA-approved polymer, self-catalytic hydrolysis capability, good mechanical stability, and sufficient blood compatibility.[9] Recently, Peng et al. reported that a PLGA-coated stent did not induce more severe pathological responses than a BMS in porcine coronary arteries.[10] In addition, PLGA polymers have been commonly used for the controlled release of drugs[11] as well as for scaffolds in tissue engineering.[12]

SRL is a carboxylic lactone-lactam macrolide produced by an actinomycete (Streptomyces hygroscopicus) and is well known to have potent immunosuppressive activity and has poor water solubility.[13] After the binding of SRL to immunophilin FK506-binding protein 12 (FKBP12), the FKBP12/SRL complex binds to the mammalian target of rapamycin (mTOR).[14] Although the effects of inhibition of mTOR by SRL were not fully understood, SRL has been shown to restrict the proliferation of VMSCs by arresting cell-cycle progression at the G1/S transition stage in many mammalian cell lines such as SMCs and endothelial cells.[15]

Many different approaches for controlled drug release from polymeric materials have been developed for a wide range of applications such as micro/nanoparticles,[16−19] multilayers,[20,21] and injectable hydrogels.[22−24] In particular, drug−polymer composites have been used with conventional medical devices through various coating systems. As the dispersion of drug into the polymer matrix depends on the physicochemical properties of the materials, the drug−polymer−solvent interactions that are of fundamental importance may play a critical role in developing a controlled drug release.

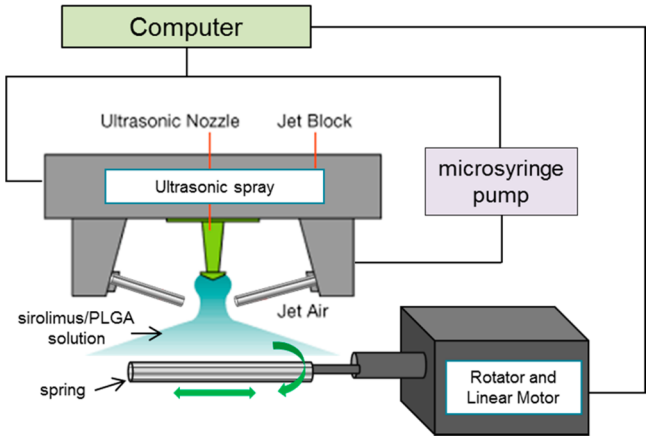
A number of studies have been reported in which drug release is influenced by multiple factors, such as particle size,[25] loading amount of drug,[26] polymer−drug miscibility,[27] drug−polymer interaction,[28] glass-transition temperature,[29] and molecular weight of the polymer matrix.[30] In particular, the properties of the organic solvent such as polarity and vapor pressure can control the chain mobility, and interfacial interaction of polymer,[31] and the drug solubility.[32] In general, the physical properties of organic solvents can influence the kinetic environment of polymer-chain mobility, and the interactions of both drug−drug and polymer−drug. Saylor et al. suggested that the evaporation rate of organic solvent during coating is associated with microstructure formation resulting in controlling drug release.[33] The aims of this study are to investigate the relationship among polymer, various organic solvents, and the drug and to control the release pattern from various microstructures produced by the organic solvents. As it was hypothesized that polymer−drug−organic solvent interactions may affect the microstructure formation corresponding to drug distribution due to organic solvents, polymer chains can be extended or become entangled with each other, resulting in differences in surface roughness, and the drug structure can change the crystalline structure to an amorphous structure as well. Sahana et al. reported that organic solvents play a significant role in nanoparticle formation and their physical properties influence the release behavior.[34] Crystallizing solvents having various polarities are preferred since the molecules in such solutions tend to form different types of hydrogen-bonded aggregates.[35] In this study, biodegradable PLGA was coated by an ultrasonic coating method as a reservoir for SRL. An ultrasonic atomizing spray method has been widely used to coat biomedical devices, such as stents and orthopedic implants, due to a thin and uniform coating. In addition, controlled SRL release from the PLGA surface was evaluated for the various effects by different solvents used.

2. MATERIALS AND METHODS

2.1. Materials. Poly(D,L-lactide-*co*-glycolide (PLGA 50:50, RG503H, M_w = 40 000 g/mol) was purchased from Boehringer Ingelheim (Germany). SRL was obtained from LC Laboratories (Woburn, MA, USA). Stainless-steel plates (SS, SUS316L, 10 × 10 mm^2) and springs (SUS316L, 1.8 mm (OD) × 18 mm (L)) were fabricated by Microspring (Korea), respectively. All organic solvents of analytical grade were used. Tetrahydrofuran (THF), acetone, ethyl acetate (EA), chloroform, dichloromethane (DCM), 1,4-dioxane, and acetonitrile were purchased from Sigma-Aldrich (USA) and were used without further purification. Table S1 lists the solvents used in this study and their physicochemical properties (Supporting Information).

2.2. Preparation of SRL-Loaded PLGA on Metal Substrates. SS plates and springs were cleaned ultrasonically in acetone and distilled water for 30 min and subsequently dried under vacuum at room temperature for 24 h. All PLGA solutions (0.3 wt %) were prepared by using SRL (20 wt %) with each different type of solvent (THF, acetone, EA, chloroform, DCM, DCM/1,4-dioxane, 1,4-dioxane, and acetonitrile). Coatings were deposited using an ultrasonic atomization operator (Sono-Tek, USA) as shown in Scheme 1.

Scheme 1. Process for Coating of Sirolimus/PLGA Solution with an Ultrasonic Atomizing Spray



Ultrasonic coating was carried out at a fixed nozzle-to-substrate distance of 9 mm, a flow rate of 0.05 mL/min, and an ultrasonic power of 4 W. After deposition, all samples were dried under vacuum at room temperature for 4 days.

2.3. In Vitro SRL Release. For the in vitro release of SRL-coated springs, samples were immersed in 1 mL of phosphate-buffered saline (PBS, pH 7.4) solution in glass vials at 37 °C with a rotational speed of 100 rpm. At predetermined time intervals, the buffer was replaced with 1 mL of fresh medium. SRL was extracted with DCM from 1 mL of the supernatant, subsequently dried under vacuum for 5 days, dissolved in eluent solvent, and measured by high-performance liquid chromatography (HPLC; LC1100, Agilent, Germany).[36] HPLC measurement conditions are as follows: mobile phase, acetonitrile (45 vol %)/methanol (40 vol %)/water (15 vol %); flow rate, 1.2 mL/min; detection, wavelength of 278 nm; SRL retention time, about 3 min.

2.4. Surface Roughness. For surface topography, PLGA was coated on SS plates by the ultrasonic coating method. Surface

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roughness was measured by a profilometer (Taylor-Hobson Form Taylsurf-120L, Rank Taylor Hobson Limited, England). The roughness parameter, R_a (arithmetic mean of the roughness height), was calculated by the average value of the roughness using Form Tailsurf Series, version 6.21, software.

2.5. Attenuated Total Reflectance-Fourier Transform Infrared Spectroscopy. ATR-FTIR spectra of PLGA coated on an SS plate with or without SRL were recorded by a PerkinElmer FTIR spectrometer (Spectrum 100, PerkinElmer, USA). The scan range was between 400 and 4000 cm^{-1}, and the scan resolution was 4 cm^{-1}.

2.6. X-ray Diffraction (XRD). XRD measurement of SRL-loaded PLGA films was performed with a D/MAX-2500 (Rigaku, Japan). Measurement conditions were the following: X-ray source, Cu Kα; voltage, 40 kV; and current, 98 mA. Samples were evaluated with a scan step of 0.02° 2θ between 3 and 100° 2θ at ambient temperature.

2.7. Differential Scanning Calorimetry (DSC). DSC analysis was performed with a DSC-Q20 (TA Instruments, USA). The SRL-loaded films (5 mg), prepared by solvent casting, were sealed in aluminum pans and heated at a rate of 10 °C/min up to 150 °C under a nitrogen atmosphere. The first heating cycle was performed to remove all thermal history. The data were recorded during the second run.

2.8. Particle Size Analysis in Suspension. For the particle size of SRL in different organic solvents, the sonication method was chosen.[32,37] Briefly, a suspension of 3.71 mL of water and 0.353 mL ofsurfactant (Tween 80) was put into a 20 mL vial. In another vial, 5.94 mg of SRL was dissolved in 0.742 mL of organic solvent, and then 5.19 mL of deionized water was added. And sonication was conducted on the solution in the 20 mL vial for 2 min at an amplitude of 30% with an ultrasonic machine of 20 kHz frequency. The solution in the 20 mL vial was released from the syringe pump to the 20 mL vial for 2 min. Then, sonication was conducted on the entire solution for 10 min. The completed suspension was dried under vacuum for 1 h to remove the residual solvent. The particle size of SRL in suspension was measured by a particle size analyzer (PSA; ELS-Z2, Otsuka Electronics Co., Japan) coupled with photon correlation spectroscopy (PCS). In the particle size analysis, the volume-weighted mean, surface-weighted mean, specific surface area, analysis of crystal population, and polydispersity index values were provided, and the value of the average diameter (cumulative result) was used in the experiment. All experiments were performed in triplicate at 25 °C.

2.9. In Vitro Degradation and Mass Loss. For polymer degradation and mass loss, PLGA films were prepared without SRL loading. Polymer solutions dissolved in various organic solvents were poured into a custom-designed vial. The solvents were evaporated for 1 day at room temperature and then completely under vacuum for 4 days. Each sample was then individually immersed in 20 mL of PBS in a glass vial and stored at 37 °C with a rotational speed of 100 rpm. At every defined time interval, samples were recovered, rinsed with pure water, and vacuum dried at 37 °C for 7 days. The degree of polymer degradation was determined using gel permeation chromatography (GPC; Water 410 differential refractometer, USA) with chloroform as a solvent. The percentage mass loss was calculated as follows with eq 1.

$$\text{mass loss}(\%) = \frac{\text{mass}_{\text{ini}} - \text{mass}_{\text{dry}}}{\text{mass}_{\text{ini}}} \times 100 \quad (1)$$

where mass_{ini} is the initial mass of the PLGA film and mass_{dry} is the mass of the dried PLGA film.

2.9. Morphology and Thickness of the Film. The surface morphology and thickness of the film were examined by field emission-scanning electron microscopy (FE-SEM; Hitachi S-2500C, 15 kV, Japan) after surface coating with platinum and palladium (Pt–Pb) ion sputtering (E–130, 0.1–0.05 Torr, 10 mA, and 80 s). For thickness, after PLGA containing sirolimus was used to coated the SUS substrate by the electrospray method, the sample was immersed in liquid N_2. It is easy to cut a cross section from a frozen sample; subsequently, the cross-sectioned sample was observed by SEM.

2.10. Transmission Electron Microscopy (TEM). To examine SRL particles aggregated in the PLGA layer, we embedded the samples in an epoxy resin. They were ultramicrotomed with a diamond knife under cryogenic conditions in cross section and approximately 85 nm in thickness. These foils were gathered onto Cu grids and were stained with a 2 wt % solution of uranyl acetate. The samples were observed with TEM (CM300, Philips) at 300 kV.

3. RESULTS AND DISCUSSION

Since the physical properties of organic solvents, such as polarity and evaporation rate, can be influenced in the microstructure formation of drug–polymers and drug release, the aim of this study was to investigate the effect of various organic solvents on the controlled SRL release. The following organic solvents were used: THF, acetone, EA, chloroform, 1,4-dioxane, acetonitrile, and DCM.

Figure 1 shows the release profile of SRL from the PLGA layer coated with different organic solvents by an ultrasonic spray method. All of the samples have nearly the same thickness of 5 μm. Although the total amounts of drug loaded on the spring surface were similar, the behavior of drug release was significantly different over 7 days (Supporting Information, Table S2). In acetone and DCM, the loading amounts of SRL were 70.5 and 74.0 μg, whereas the releases amounts were 63.5 (95.7%) and 38.5 (60.1%) μg, respectively. In chloroform and acetonitrile, while the loading amounts were 98.3 and 93.6 μg, the released amounts were 57.0 (65.3%) and 26.2 (34.5%) μg, respectively. In addition, the amounts of SRL released from THF, EA, DCM/1,4-dioxane, and 1,4-dioxane were 100, 77.7,

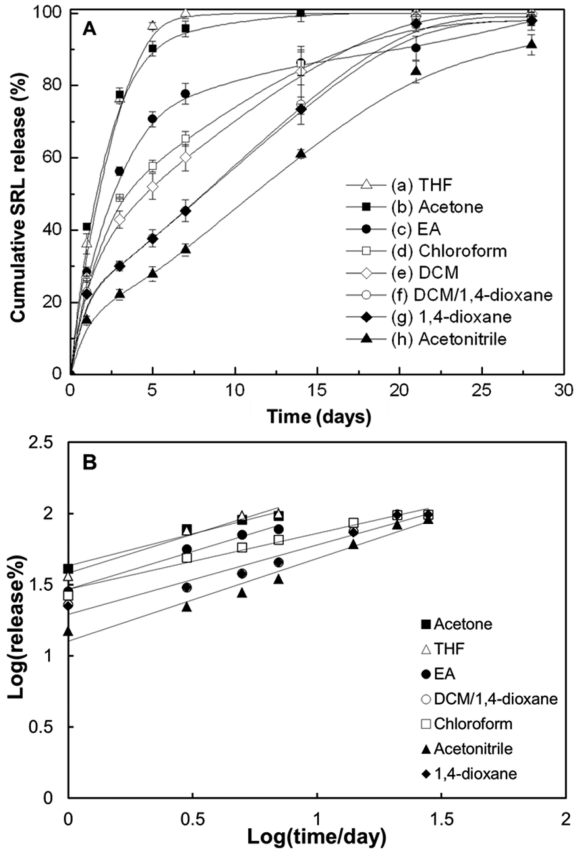

Figure 1. (A) Cumulative release profiles of SRL from the PLGA layer coated with ultrasonic spray and (B) the experimental data based on the power law model. Organic solvents used for coating were tetrahydrofuran (THF), acetone (AC), ethyl acetate (EA), chloroform, methylene chloride (DCM), 1,4-dioxane, and acetonitrile.

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Table 1. Drug Release Parameters Calculated by the Modified Peppa Equation

	acetonitrile	1,4-dioxane	DCM/1,4-dioxane	DCM	chloroform	EA	acetone	THF
n	0.581	0.490	0.488	0.430	0.392	0.530	0.451	0.545
k	1.103	1.285	1.292	1.423	1.468	1.468	1.631	1.581
r^2	0.969	0.965	0.958	0.999	0.976	0.984	0.962	0.965

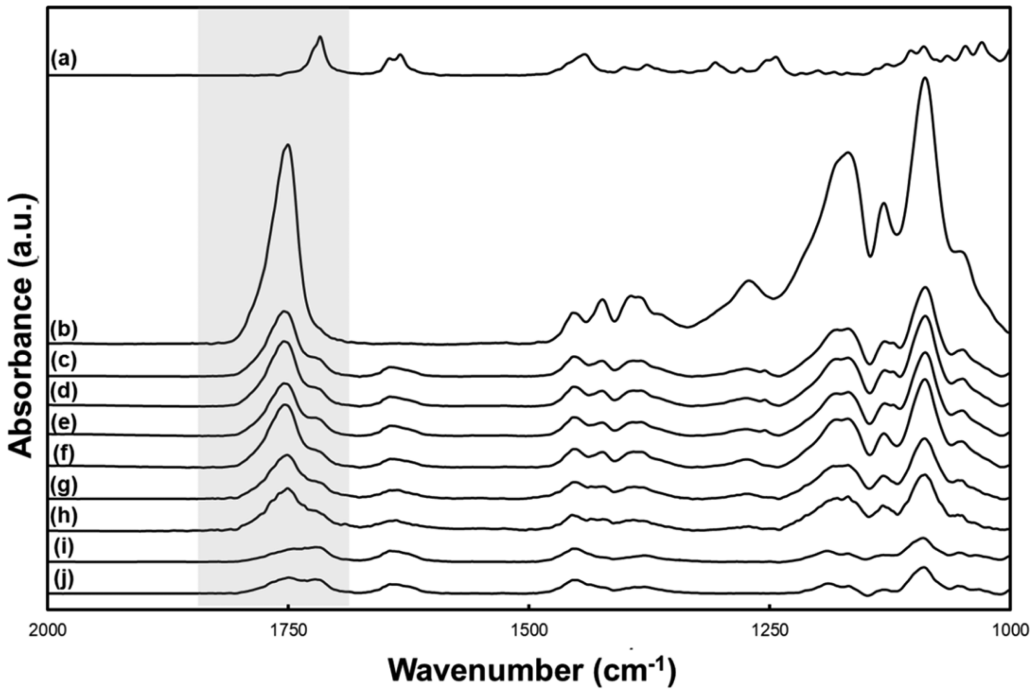

Figure 2. ATR-FTIR spectra of (a) SRL, (b) PLGA powder, and (c–j) PLGA films fabricated by the solution casting method using different organic solvents: (c) acetonitrile, (d) 1,4-dioxane, (e) DCM/1,4-dioxane, (f) DCM, (g) chloroform, (h) EA, (i) acetone, and (j) THF.

45.3, and 45.3%, respectively. Burst releases were observed by the samples made from THF and acetone, and SRL released 100% over 7 days. However, SRL from acetonitrile, 1,4-dioxane, and DCM/1,4-dioxane underwent sustained release, and 28 days was required to release up to 100% SRL.

In order to elucidate the mechanism of SRL release more precisely, the in vitro release behavior was analyzed by the semiempirical power law equation, which was introduced by Siepmann and Peppas[38] as given below (eq 2)

$$\frac{M_t}{M_\infty} = kt^n \tag{2}$$

where M_t/M_∞ is the fraction of drug released at time t, k is a characteristic constant of the system, and n is the kinetics and the release mechanism, which depend on the geometry of the system (Supporting Information)

If n is below 0.5, then the release is governed by Fickian diffusion. If n = 1, it corresponds to surface erosion, whereas both mechanisms play a role in the release if n has a value of between 0.5 and 1. Table 1 lists the calculated values of the diffusional exponent (n), release coefficients (k), and correlation efficiency (r^2). It is shown that the exponent n values for the release of SRL were within the limiting value of 0.5. This indicates Fickian release behavior. On the other hand, in the case of acetonitrile (0.581), EA (0.530), and THF (0.545), the exponent values were slightly higher than 0.5. Thus, the behavior of SRL release can be explained by the complex combination of diffusion and polymer degradation.

For the evaluation of the solvent effect that is capable of controlling the release rate, the surface roughness of the SRL-loaded PLGA layer was measured. The surfaces obtained via acetonitrile and acetone are relatively rougher than that of 1,4-dioxane. In addition, although the difference in the measured roughness between DCM (R_a = 0.42 μm) and EA (R_a = 0.40 μm) is only 0.02 μm, the release ratios of SRL at 5 days were 52 and 71%, respectively. Thus, it is suggested that the surface roughness is not the dominant factor in releasing SRL from the PLGA matrix.

ATR-FTIR spectra for SRL, PLGA, and SRL-loaded PLGA films prepared with various organic solvents are shown in Figure 2. It was used to estimate the extent of polymer–drug interaction on the molecular level. Figure 2a shows the spectrum for SRL. The peaks at 1717 and 1634 cm^{-1} can be assigned to the stretching vibrations of C═O and C═C, respectively. The peaks at 1443 and 1376 cm^{-1} are the asymmetric and symmetric C–H bending vibration peaks, the peak at 1090 cm^{-1} is characteristic of C–N, and the peak at 995 cm^{-1} is the out-of-plane vibration peak of C═C.[39] In Figure 2b, such unique properties of PLGA molecular bonding as the C═O group and C–O–C stretching peaks were measured at 1750 and 1088 cm^{-1}, respectively. Characteristic peaks for hydrogen bonding (– C═O···H–) were slightly shifted, and in particular there were significant shifts in the peaks that correspond to the carbonyl group. For acetonitrile (Figure 2c), the peak at 1750 cm^{-1} was shifted to 1752 cm^{-1}. Also, for acetone and THF, the peak was shifted to 1740 and 1749 cm^{-1}, respectively. This means that the interaction exists apparently

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between PLGA and SRL, and organic solvents used for PLGA-SRL films also affect the hydrogen bonding interactions even though residual solvents rarely remained in the films (data not shown). Moreover, it has been reported that the interaction between polymer and drug in solid dispersions is responsible both for the formation of amorphous or crystalline dispersions and possibly for the particle size distribution of the drug in the polymer matrix.[40] XRD measurements were carried out in order to confirm whether SRL particles exist in either the amorphous or crystalline state inside the PLGA matrix. It was also used to investigate whether organic solvents affect the PLGA-SRL interactions. As shown in Figure 3, SRL, PLGA,

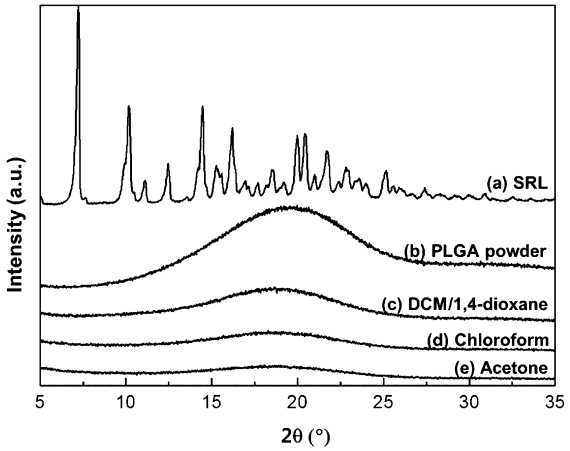

Figure 3. XRD patterns of (a) SRL, (b) PLGA powder, and (c–e) SRL-loaded PLGA films fabricated from different organic solvents: (c) DCM/1,4-dioxane, (d) chloroform, and (e) acetone.

and SRL-loaded PLGA films prepared by various organic solvents were measured. The pattern for SRL showed significantly sharp peaks at a 2θ diffraction angle: 7.2, 10.2, 12.2, 14.5, 15.8, 20.4, and 21.8 (Figure 3a). Meanwhile, as shown in Figure 3b–e, PLGA and SRL-loaded PLGA films fabricated with DCM/1,4-dioxane, chloroform, and acetone displayed only amorphous halo patterns. The peaks corresponding to SRL in the crystalline state disappeared in the PLGA film, which means that amorphous SRL particles were dispersed in the polymer matrix, irrespective of organic solvents.

Figure 4 shows the DSC curves of SRL, PLGA, and PLGA films prepared by various organic solvents. While the glass-transition temperature (T_g) of the PLGA control was 46.5 °C, those of PLGA films prepared from chloroform, acetone, and DCM/1,4-dioxane were 35.5, 33.5, and 10.8 °C, respectively. The T_g value for DCM/1,4-dioxane was much lower than those for chloroform and acetone. Generally, it is known that T_g depends on chain flexibility and intermolecular interactions in polymers. Organic solvents can induce the molecular motions of amorphous chains, indicating that the solvents act as a plasticizer.[41,42] Besides, polymer chains that are solvated may entangle to a greater degree than aggregated bulky chains. Organic solvents can lead to specific interactions such as hydrogen bonding on specific parts of the polymer. Consequently, the T_g values can vary depending on the property of organic solvents. If the drug is in the noncrystalline state, then it will absorb moisture more readily than in the crystalline state at lower T_g.[43] The thermal properties of PLGA were also analyzed to show that PLGA starts to decompose

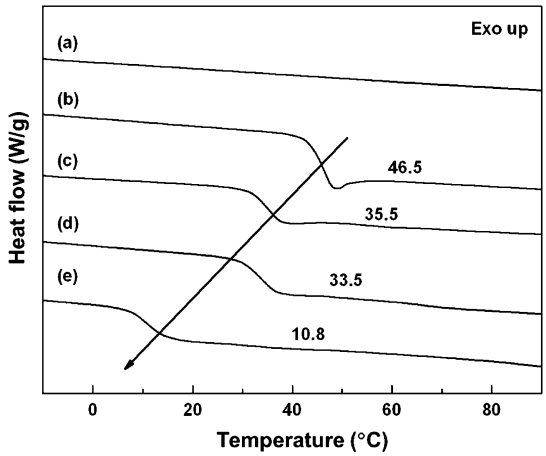

Figure 4. DSC curves of (a) SRL, (b) PLGA powder, and PLGA films fabricated using (c) chloroform, (d) acetone, and (e) DCM/1,4-dioxane.

thermally from 180 to 350 °C, and the melting point was changed to 175 °C, demonstrating the presence of SRL in the amorphous state in the PLGA matrix (Figure S1 and S2).

In order to analyze the relationship between the SRL particle size and drug-release behavior, the size of the SRL particles was determined by the suspension method in different organic solvents. From these results in Table 2, the size of SRL particles in suspension was found to be in the range of 10.6 nm to 2.3 μm for different organic solvents. As compared to other organic solvents, it can be observed that acetonitrile and 1,4-dioxane make significantly small particles of SRL, 10.6 and 11.4 nm, respectively. For acetone and THF, the SRL sizes were 1.9 and 2.3 μm. In particular, the particle sizes of SRL were 185 and 279 nm for DCM and chloroform. As suggested by Gandhi et al., the higher the solubility of SRL, the larger the aggregated particles and vice versa.[32] That is to say, the formation of SRL nuclei in suspension corresponded to the relative solubility against organic solvents. It can be associated with Ostwald ripening, which means that the nuclei formed in solution are redeposited over a period of time as long as the saturated solubility supports it.[32] Park et al. also reported that the shape and size of particles varied according to organic solvents.[44]

Although coatings were made under the same coating condition, SRL particles of different sizes were formed by different organic solvents, as shown in Figures 5 and 6A. Figure 5 displays SEM images of a cross-sectioned PLGA layer loading SRL with different organic solvents after 3 and 5 days of incubation in PBS solution at 37 °C. Depending on the degradation time, there were significant differences in the internal structures and surface morphologies (Figure S3). The initial smooth surfaces were found to be rough after 5 days. Interestingly, the PLGA layer fabricated by acetone was observed to have large pores, while the pore sizes of DCM/1,4-dioxane and chloroform were remarkably smaller than that of acetone. It is consistent with the results of particle size in Table 2.

In addition, TEM images were used to observe the particle size of SRL in the PLGA matrix as shown in Figure 6a. It seems that organic solvents influenced the size of SRL particles in the PLGA matrix. In DCM/1,4-dioxane, the particle sizes were detected in the range of 10–50 nm. In chloroform and acetone, the particle sizes were much larger, ranging within 50–150 and 80–330 nm, respectively. It was confirmed that SRL was

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Table 2. Effect of Various Organic Solvents on the Roughness of a PLGA-Coated Surface and the Particle Size of SRL

	acetonitrile	1,4-dioxane	DCM/1,4-dioxane	DCM	chloroform	EA	acetone	THF
R_a (μm)	1.74 ± 0.24	0.06 ± 0.01	0.07 ± 0.01	0.42 ± 0.03	0.20 ± 0.03	0.40 ± 0.07	1.24 ± 0.10	0.90 ± 0.09
particle size (nm)[a]	10.6 ± 0.1	11.4 ± 0.2	14.8 ± 0.6	185 ± 1	279 ± 5	860 ± 55	1973 ± 85	2295 ± 144

[a]Measured by DLS.

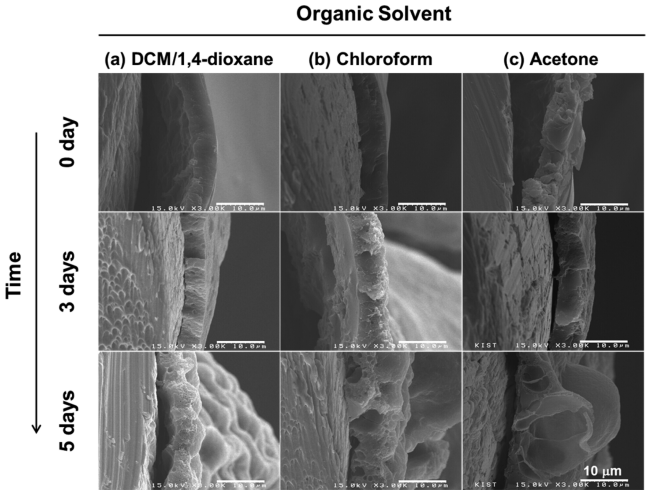

Figure 5. SEM images of cross-sectioned PLGA layers coated on SS plates by using (a) DCM/1,4-dioxane, (b) chloroform, and (c) acetone after 0, 3, and 5 days of incubation at 37 °C.

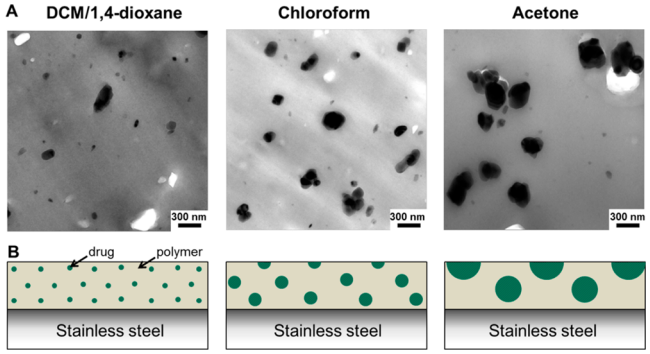

Figure 6. (A) TEM images of an SRL-loaded PLGA matrix prepared by using DCM/1,4-dioxane, chloroform, and acetone and (B) a schematic illustration of the polymer−drug microstructure by organic solvents.

dispersed in an amorphous state within PLGA (Figure 3). Although the result of particle size measured by DLS using a suspension method shows an indirect correlation with the size of drug particles in the polymer matrix, it implies that individual SRL particles tend to aggregate in organic solvents. As a result, it is obvious that SRL particles can form aggregated-drug or drug-poor regions at or near the surface and consequently the concentration gradient leads to a significant initial burst release.[33] Therefore, organic solvents can not only control the aggregate size of SRL particles but also contribute to their rate of release from the PLGA matrix.

With increasing particle size, drug release appeared to increase for 7 days. If the drug was relatively uniformly distributed inside the polymer matrix without a large crystalline region, then the initial release rate would be low and drug delivery would take place slowly. However, Kim et al. demonstrated that the large aggregated-drug regions at or near the surface coatings readily dissolve in the media and give rise to the initial burst.[45] Although the evaporation rate of organic solvent led to the development of heterogeneous structure during film formation, the effect might be weakened by the process of ultrasonic coating, unlike the situation for the solution casting. Consequently, it is assumed that the large aggregated-drug regions may be formed at or near the surface in THF and acetone. On the other hand, in the case of acetonitrile and 1,4-dioxane, drug may be homogeneously distributed over a small area in the PLGA matrix (Figure 6b).

Figure 7 shows the results of mass loss, degradation, and pH change in PLGA. It is observed that the molecular weight

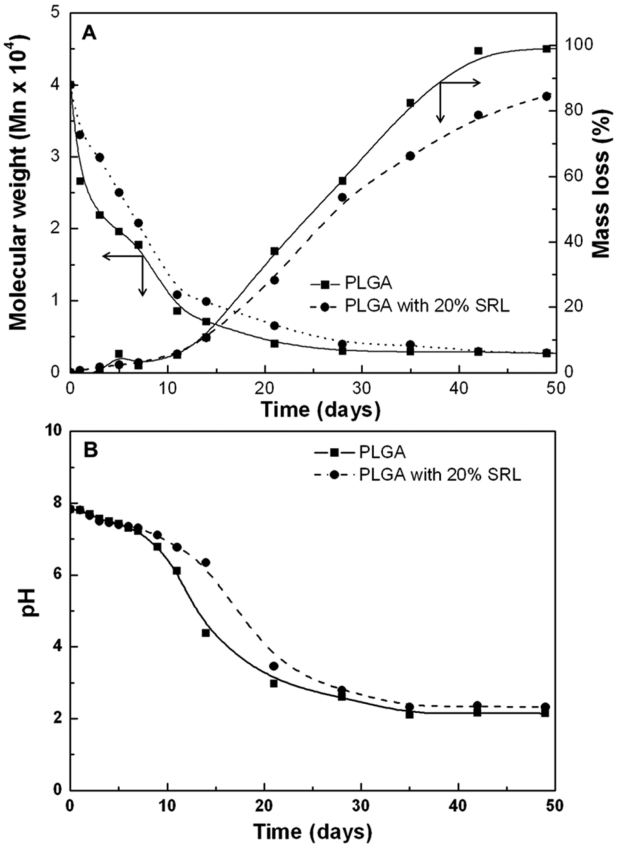

Figure 7. (A) Molecular weight loss and mass loss and (B) pH changes in PLGA coated on a stent.

decreased in two stages: an initial significant decrease for 9 days and a subsequently a steady decrease (Figure 7a). Moreover, mass loss occurred rapidly after 11 days. The patterns of polymer degradation and mass loss were similar, irrespective of SRL loading. The measured pH values also showed a decreasing tendency as the loss of molecular weight increases (Figure 7b).

Taken altogether, SRL release as well as SRL-rich and SRL-poor regions can be controlled by the solvent effect. Some factors can be suggested as the reasons: (1) the particle size of SRL in an amorphous region within the PLGA matrix; (2) the

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distribution of SRL-rich regions at or near the surface, depending on the physical properties of organic solvents; (3) the formation of microstructures due to the interaction between SRL and PLGA; and (4) T_g of PLGA that is lowered by the molecular interaction between PLGA and organic solvents. Thus, PLGA with T_g lower than 37 °C exists in the rubbery state and increases the mobility of polymer chains, water absorption, and drug release from a polymer matrix.

The solvent effect on SRL release and coating properties may be elucidated on the basis of the some physicochemical properties of organic solvents (Table S1). Our finding shows that the release rate of SRL depends on the particle size of SRL, which was demonstrated by depicting a direct correlation between particle size and release rate (Figure S4). The correlation is that the larger the particle size, the faster the SRL release. Gandhi et al. have shown that the higher solubility of SRL facilitates the formation of larger particles and vice versa. That is to say, the formation of SRL nuclei in suspension corresponds to relative solubility against organic solvent. It can be associated with Ostwald ripening, which means that the nuclei formed in solutions are redeposited over a period of time as long as the saturated solubility supports it. Drug−polymer composites also show phase-separated morphologies depending on the physicochemical properties of materials and environmental conditions. According to Kim and Saylor et al., the drug loading and structural heterogeneities are associated with the solvent evaporation rate during film fabrication. Therefore, organic solvent can be a critical parameter in controlling drug release. However, in the case of ultrasonic spray coating, more parameters should be multiply considered, such as the drug solubility, boiling point, polarity, and vapor pressure of organic solvents.

Solvents can affect the distribution of SRL in a coating matrix of PLGA. Primarily, the formation of SRL particles with different sizes due to the different solubility in various organic solvents leads to a different distribution that is closely associated with the release behavior of SRL. Second, the evaporation rate of solvent that is affected by the inherent volatility of the solvent, temperature, and vapor pressure can influence the vertical distribution of SRL during the drying process. Because coatings were dried under the same conditions, the inherent volatility of solvent can mainly influence the vertical drug distribution. On the basis of the fact that the volatility of solvent is inversely related to the boiling point (Table S1), more volatile solvents such as dichloromethane and acetone might facilitate the vertical distribution of SRL, which was not analyzed in the current study.

The surface roughness of the PLGA coating matrix with SRL has no correlation to any properties of organic solvents including the relative polarity and boiling point and even the particle size of SRL. The surface roughness is more strongly correlated with solubility of PLGA rather than with the presented parameters because smoother surfaces were made for chlorinated solvents that are known good solvents for PLGA.

The solvent effect is also related to the bulk properties of the coating matrix containing drug, such as microstructure and crystallinity. Basically, all PLGAs are amorphous rather than crystalline and show a T_g in the range of 40−60 °C. Decreases in T_g in different solvents are probably attributed to differences in the solubility of PLGA and volatility during drying. The influence on T_g is roughly stronger at higher solubility for less-volatile solvents. However, the presence of drug in the matrix makes the elucidation more complex. Interaction between PLGA and SRL can elucidate the effects on the microstructure that are related to the release behavior of SRL. Although the relative polarity of solvents can be a clue to explaining the interaction, there was no correlation between the relative polarity of solvent and coating parameters in the current experimental results. This is probably because there are many other intra- and intermolecular interactions such as PLGA−solvent and SRL−solvent besides PLGA-SRL.

An in-depth discussion of solvent effects on release behavior and coating matrix properties enables us to give some criteria for the preparation of an ideally optimized drug-eluting stent (DES). However, incomplete correlations and other parameters should be sufficiently considered for the successful development of DES.

4. CONCLUSIONS

On the basis of these results, it was confirmed that the physicochemical properties of organic solvents, when coated on the metal surface through an ultrasonic spray method, affected the surface roughness of drug-containing polymer, the size of drug particles within a polymer matrix, the distribution at or near the surface of either aggregated drug or polymer-rich regions, and the loading amount of drug. The larger the particle size, the faster the drug release in vitro. It is considered that both the particle size and the distribution of the aggregated-drug region affected the release profile and the degradation of the polymer matrix as a result of water absorption and swelling. Therefore, a variety of drug-release patterns were obtained through the molecular interaction among polymer, organic solvent, and drug molecules, and this interesting finding may be helpful for future potential applications of controlling drug release in the field of biomedical devices, including DES.

■ ASSOCIATED CONTENT

Supporting Information

Characteristic properties of organic solvents used for ultrasonic coatings. Loading amounts of SRL in the PLGA matrix. TGA curve of a PLGA film under nitrogen. DSC thermograms of PLGA and the SRL/PLGA film. SEM images showing the surface morphology after incubation. Correlation between the SRL size and the SRL release. This material is available free of charge via the Internet at http://pubs.acs.org.

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Notes

The authors declare no competing financial interest.

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