Mechanism Analysis of Poly(lactic acid) Particles Formation by Mesoscale Simulation

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ABSTRACT: In this work, the mechanism analysis of Poly(DL-lactic acid) (PLA) particles formation is investigated by dissipative particle dynamics (DPD) simulations. The polymer PLA is usually as a carrier at the drug deliver system (DDS), first, the mechanism analysis of PLA particles formation was preceded on the basis of the blank PLA particles. At the same time, the formation of PLA particles for drug delivery systems has been investigated. The results of DPD simulations indicate that the formation of PLA particles with drug or not, consists of three steps: (1) aggregation-individual PLA chains got aggregated with each other in solution; (2) formation and disruption of PLA particles; (3) solidification of PLA particles. At the same time, the effects of PLA, drug content on the aggregation morphology are investigated, which indicates the PLA particles with drug or not aggregated and formed is spherical particles, drug molecules are amorphously and homogeneously distributed inside the PLA matrix, the size of PLA particles increases with increasing the initial PLA content and drug content in the solution. The content of DMF is gotten when PLA chains and drug molecules begin to aggregate and form the particles. These could be used to guide the experimental preparation of PLA blank particles and DDS with desired properties. © 2010 Wiley Periodicals, Inc. J Appl Polym Sci 119: 3273–3281, 2011

Key words: drug delivery systems; simulations; self-assembly

INTRODUCTION

In the past decades, extensive efforts by numerous groups have been made in the development of Poly(DL-lactic acid) (PLA) particles for drug delivery. The sizes of biodegradable polymer particles have several critical implications for controlled-release drug delivery. For example, particle size influences allowable routes of administration[1] and the final disposition[2] of the spheres in the body. Importantly, particle size is a determining factor of drug release rates and controlled manipulation of the size may provide a means to tailor release rate profiles.[3] For all of these reasons, the part of authors have prepared PLA particles and with controlled size to provide an efficient route to clinical implementation by dialysis method.[4] The polymer PLA is dissolved in a solution of in a suitable water-miscible organic solvent; the solution is then introduced into a dialysis tube which is put into water. As the dialysis proceeds, the water diffuses into the organic solvent and the organic solvent also diffuses into the water, the polymer PLA precipitates into spherical particles, meanwhile, the active ingredient can be also encapsulated into the particles.

The mechanisms of the formation of blank PLA particles without drug are also suggested by investigating the dynamic process of particle formation during dialysis[4]; however, it is rather difficult for researchers to find the direct identification from experiment to investigate the formation mechanism of meso-structures. Thus, it is very important to study particles formation with the help of computer simulations. Simulations in couple with experiments might provide deeper insights for the preparation of controlled particles. Dissipative particle dynamics (DPD), introduced by Hoogerbrugge and Koelman[5] in 1992, was used to simulate soft spherical beads interacting through a simple-wise potential, and thermally equilibrated through hydrodynamics.[6] Español and Warren[7] pointed out that the total force acting on a particle is the sum of a conservative force, a dissipative force, and an additive random

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force. Groot and Warren[8,9] made an important contribution on DPD method by establishing a relationship between a simple functional form of the conservative repulsion and the Flory-Huggins parameter theory, which lead DPD method widely applied in the study of mesostructures of complex systems.[10–17] Groot[10–12] studied polymer-surfactant aggregation, electrostatic interactions, and morphology change by DPD method. Yuan et al.[13] drew the ternary phase diagram of AOT/water/iso-octane system using the results from DPD simulations. DPD is also used to calculate the solubility parameter, surface tension,[14] and the mean square end-to-end distance of polymer chain.[15] In addition, DPD simulation is also an efficient method to investigate the complex system consisting of polymer, surfactant, and water.[16,17] Qian et al.[18,19] investigated the effect of composition on the aggregate morphologies of polymer microspheres for the field of DDS, and indicated that DPD is plausible way for simulating a system containing PLA and water. However, DPD has seldom been applied to the mechanism analysis of DDS.

In this work, DPD simulation technique is employed to investigate mechanism analysis of poly(lactic acid) particles formation based on the previous work reported by parts of the authors.[4] The PLA is selected as a carrier, whereas the Epirubicin (EPI) as model drug. The process of PLA particles formed from the PLA solution by dialysis is simulated by different levels of water and DMF contents of the PLA solution to investigate the formation process of the PLA particles with drug or not. At the same time, the effect of the PLA content and drug content on aggregate morphologies of PLA particles and DDS was also investigated.

DPD SIMULATIONS

DPD theory

An important concept of DPD simulation is the coarse-grained model, which means that a DPD bead represents a group of atoms or a volume of fluid that is large on the atomistic scale but still macroscopically small.[20] In DPD simulations, all beads comply with Newton's equations of motion[8,21]:

$$\frac{dr_i}{dt} = v_i, \quad m_i \frac{dv_i}{dt} = f_i \tag{1}$$

where r_i, v_i, m_i, and f_i are the position, velocity, mass, and force of bead i, respectively. Dimensionless units are used in DPD simulations usually; the mass of each bead is set to 1 DPD mass unit, resulting in an equation between the force acting on a bead and its acceleration.[8] The force acting on a bead is a sum of a conservative force (F_{ij}^C), a dissipative force (F_{ij}^D), and a random force (F_{ij}^R)[12]:

$$f_i = \sum_{j \neq 1} \left(F_{ij}^C + F_{ij}^D + F_{ij}^R \right) \tag{2}$$

where the sum runs over all other particles within a certain cutoff radius r_c, which is set to be 1 in simulations. The conservation force is soft repulsion acting along the line of centers and is given by:

$$F_{ij}^C = \begin{cases} a_{ij} 1 - r_{ij}\hat{r}_{ij} & r_{ij} < 1 \\ 0 & r_{ij} \geq 1 \end{cases} \tag{3}$$

where a_{ij} is a maximum repulsion between bead i and bead j; $r_{ij} = r_i - r_j$, $r_{ij} = |r_{ij}|$, $\hat{r}_{ij} = \frac{r_{ij}}{|r_{ij}|}$. The other two forces are dissipative and random forces, which are given by:

$$F_{ij}^D = \frac{\sigma^2 \left(\omega(r_{ij})\right)^2}{2kT} \left(v_{ij} \cdot \hat{r}_{ij}\right) \hat{r}_{ij} \tag{4}$$

$$F_{ij}^R = \frac{\sigma \omega(r_{ij}) \hat{r}_{ij} \zeta}{\sqrt{\delta_t}} \tag{5}$$

where $v_{ij} = v_i - v_j$, ζ is a random variable with zero mean and variance 1, δ_t is the time step used, the r-dependent weight function.

$$\omega(r) = \begin{cases} r - 1 & r < 1 \\ 0 & r > 1 \end{cases} \tag{6}$$

Spring force

In the DPD model, atoms of each molecule are not directly represented, but they are grouped together into beads. Molecules are represented by several beads by a spring force to reproduce the typical nature of them. Groot and Warren[8] advocated the simple harmonic spring force on bead i:

$$F_i^S = \sum_j C r_{ij} \tag{7}$$

where C is the spring constant, and the mean distance between two consecutive chain beads is governed by the spring force and the repulsive interaction. The spring constant C is chosen so that, if all forces are considered, the average bead–bead distance along the chain has a reasonable value, which corresponds to the first peak in the bead–bead correlation function. In this study, the default value $C = 4$ has been used, resulting in a slightly smaller distance for bonded beads than for non-bonded ones.[9]

Models and simulation parameters used in DPD

Coarse-grained models of components are used in this study, as shown in Figure 1.

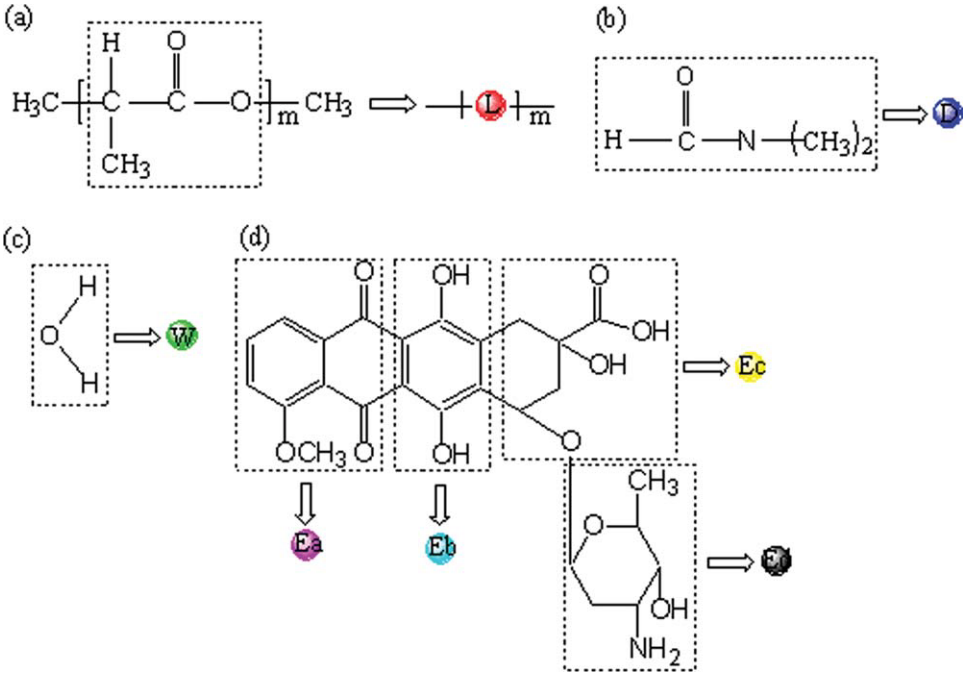

Figure 1 Coarse-grained models of (a) PLA, (b) DMF, (c) water, and (d) EPI. (red bead, PLA; blue bead, DMF; green bead, water; magenta, cyan, yellow and black beads, EPI. Legends of the beads are all same in the following figures as in this figures). [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

Each bead is represented by several atoms, which are closed by dashed lines. Each monomer of PLA is considered as one bead named L. One molecule of DMF is represented as a bead D. One molecule of water is represented as a bead W. The EPI molecule is divided into four types of beads, named Ea, Eb, Ec, Ed, respectively. To calculate the conservation force, repulsion parameters a_{ij} between any two beads should be calculated. The repulsion parameters between beads of the same type, a_{ii}, are chosen according to[9]:

$$a_{ii}\rho = 75k_BT \tag{8}$$

where k_B is the Boltzmann constant and T is the system temperature. The bead density $\rho = 3$ has been used in a previous work,[9] and $k_BT = 1$ has been used [10]. Repulsion parameters between different types of beads are linearly related with the Flory-Huggins parameters (χ_{ij}) according to

$$a_{ij} = a_{ii} + 3.27\chi_{ij} \tag{9}$$

χ_{ij} can be calculated from solubility parameters[9]

$$\chi_{ij} = \frac{V_{\text{bead}}}{kT}(\delta_i - \delta_i)^2 \tag{10}$$

where V_{bead} is the arithmetic average of molar volumes of two beads; δ_i and δ_j are the solubility parameters of bead i and j, which can be obtained from experiments or molecular dynamics simulations. In this study, solubility parameters are calculated according to Group Contribution Method or gained from handbook.[22]

DPD repulsion parameters are calculated by using eqs. (8)–(10), which are listed in Table I.

A $30 \times 30 \times 30\ r_c^3$ (r_c is the DPD length unit or the cutoff radius) box was adopted. In DPD simulations, each mesoscale molecule of PLA, water, DMF, and drug is coarse-grained into beads with the bead number of 10, 1, 1, and 4, respectively. The total number of beads, 81,000, was used in all simulations. Each DPD simulations run 40,000 steps with a time step of 0.05 to get a steady phase. The temperature was set constant at 298 K. All DPD simulations were performed by the Materials Studio software (Accelrys).

RESULTS AND DISCUSSION

The aggregate morphology of PLA particle

Several snapshots of configuration of the PLA particles investigated during one DPD simulation are

TABLE I
The Interaction Parameters a_{ij} Used in DPD Simulation

a_{ij}	L	W	D	Ea	Eb	Ec	Ed
L	25.0						
W	35.4	25.0					
D	25.1	26.0	25.0				
Ea	25.0	36.3	25.1	25.0			
Eb	25.3	32.3	25.0	25.4	25.0		
Ec	25.4	25.0	25.9	25.5	25.4	25.0	
Ed	25.0	25.5	25.1	25.0	25.3	27.0	25.0

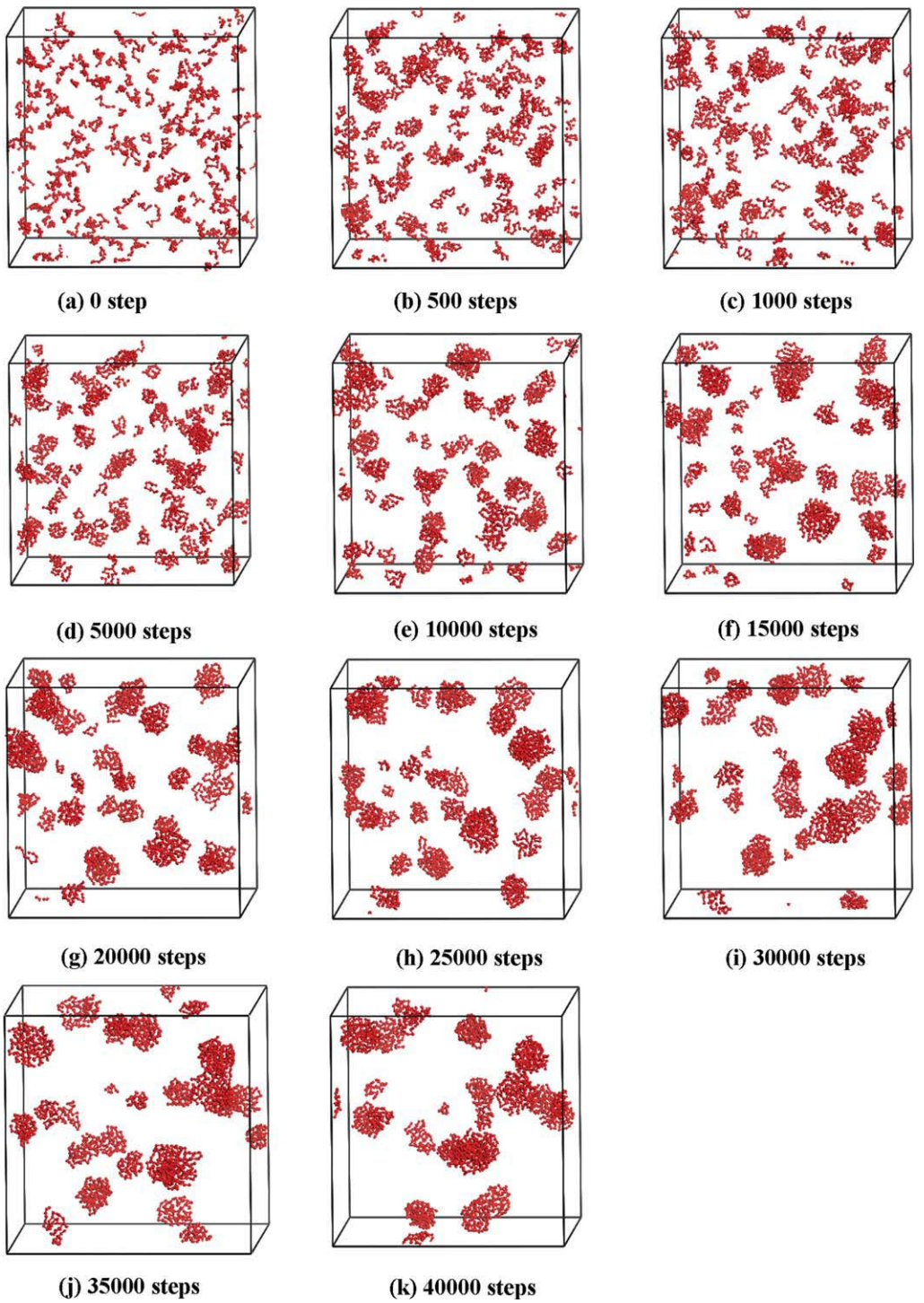

Figure 2 Change of aggregates with increasing simulation steps. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

shown in Figure 2. In the simulation system, the molar fraction of mesoscale molecule (PLA, water, DMF, and drug) is 3 : 97 : 0 : 0. To show the molecular aggregate morphologies clearly, the beads instead of water and DMF molecules have not been displayed (The same treatment was processed in all following figures).

As shown in Figure 2(a), all components are randomly mixed at the beginning of simulation. As simulation processes [Fig. 2(b–e)], some PLA molecules aggregate and form clusters. With the simulation time increasing, PLA chains gradually form the PLA particles [Fig. 2(f–k)]. The system reaches equilibrium after 30,000 steps, and the stable PLA particles

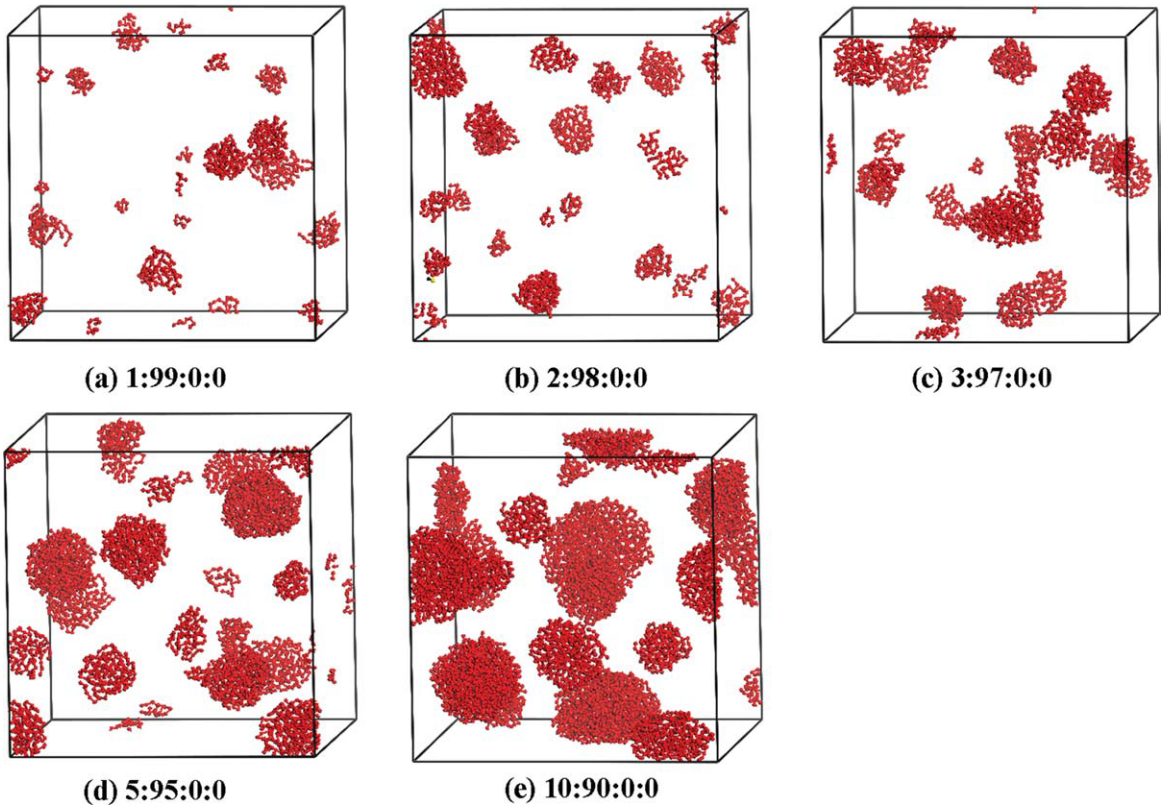

Figure 3 Aggregate morphologies of the PLA particles at the different PLA content. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

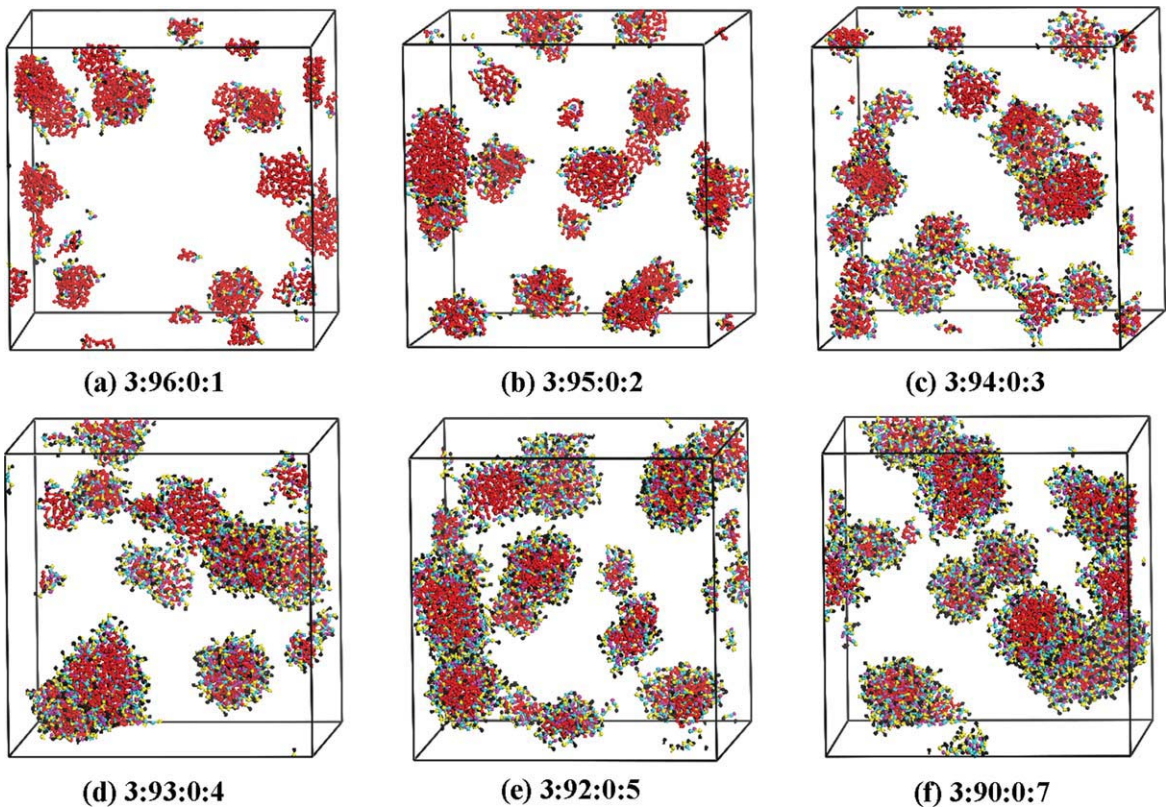

Figure 4 Aggregate morphologies of DDS at different content of drug. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

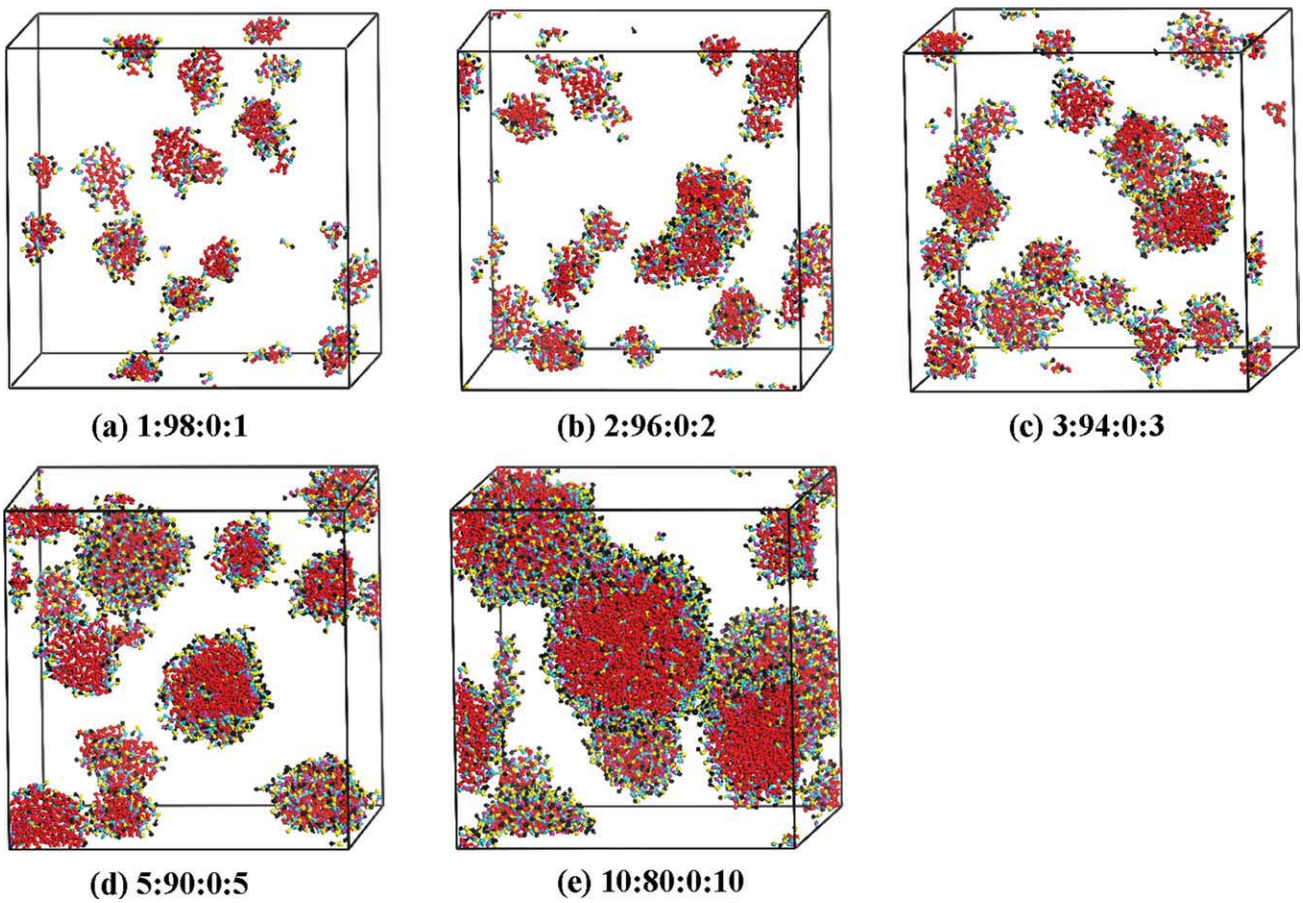

Figure 5 Aggregate morphologies of DDS at different content of PLA and drug. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

have been formed. The aggregate morphology does not change significantly with extra simulation steps. All the following simulations are set to 40,000 steps.

The effect of PLA content on aggregate morphology of PLA particles

The aggregate morphologies of the PLA particles at different levels of PLA content are shown in the Figure 3. The results showed the PLA particles observed all are spherical structures under different PLA content, at the same time, the size of PLA particles increases with the polymer PLA content increasing [from 1% to 10% corresponding to Fig. 3(a–e)], which is in agreement with a general observation found in experimental preparation of polymer particles.[4,23] These indicated that the polymer concentration was an important factor on the size of particles.

The effect of drug content on aggregate morphology of DDS

The aggregate morphologies of DDS at different levels of drug contents are investigated and shown in Figure 4. The molar fraction of mesoscale molecule of PLA is fixed at 3%. With the molar fraction of mesoscale molecule of drug (EPI) increasing, a stable spherical particle is observed, and the drug molecules were distributed inside the PLA matrix. However, the distribution of drug in the PLA matrix is not homogeneous, which is some different from the experimental results.[4] The reason may be the driving force of the DDS formation includes the electrostatic interactions and hydrophilicity/hydrophobic of PLA and drug's. Compared Figure 4 with Figure 3(c), the size of PLA particle increases slightly for the drug added, at the same time, the size of PLA particle increase obviously with the EPI content increasing. When molar fraction of the drug increases to 5% [Fig. 4(e)], the particles cannot encapsulate all drug molecules inside the PLA matrix. The excess drug molecules will be encapsulated outside the particles when the drug content exceeds the maximum. More and more drug molecules are encapsulated outside the particles with the increase of drug content [Fig. 4(f)].

The effect of PLA and drug content on aggregate morphology of DDS

The aggregate morphologies of the DDS at different levels of PLA content and drug are shown in Figure 5.

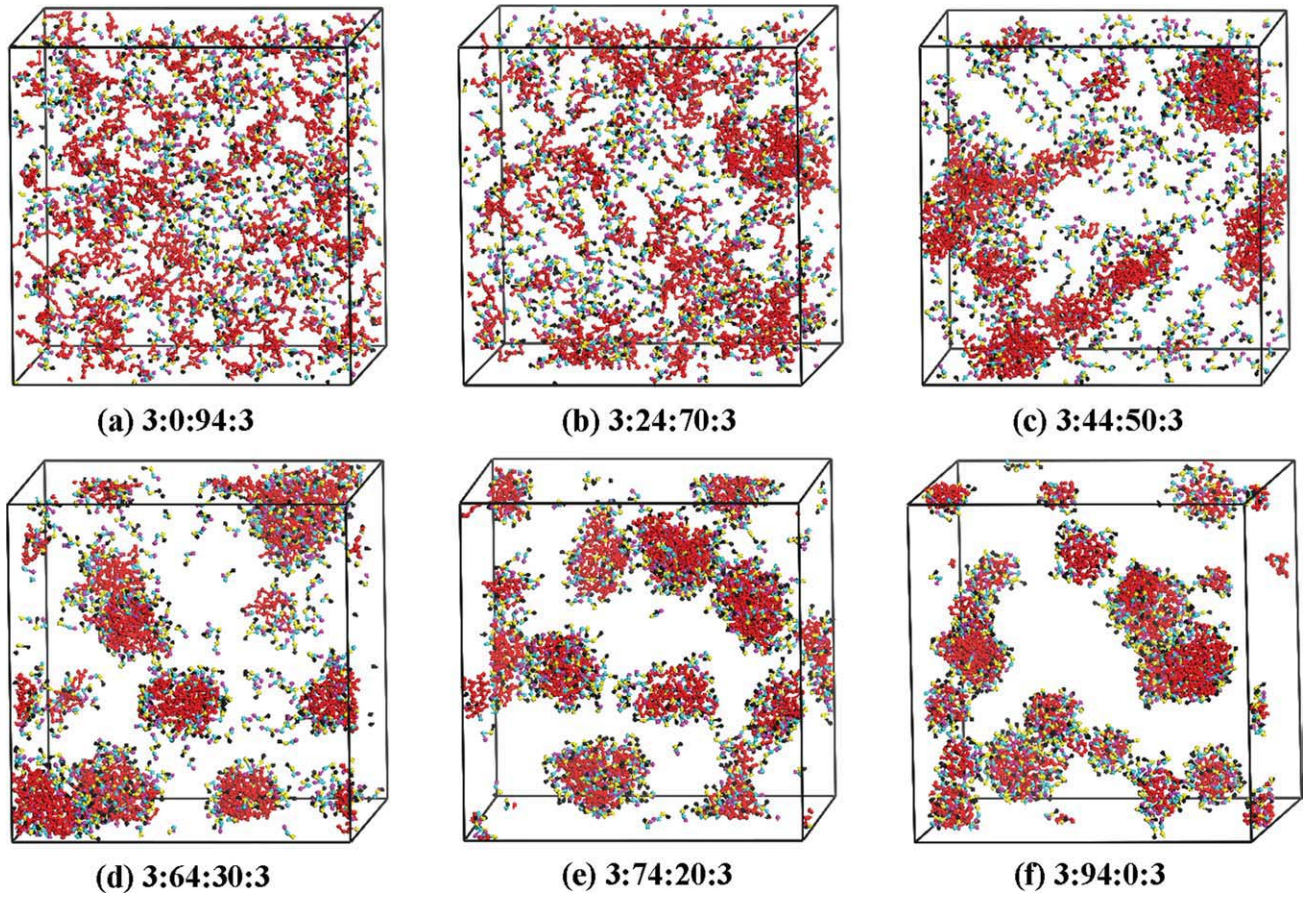

Figure 6 Aggregate morphologies of PLA particles with drug at the different DMF content. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

The molar ratio of mesoscale molecules of PLA and drug is fixed at 1 : 1. A stable spherical particle is observed with drug molecules distributed inside the PLA matrix. The size of PLA particles increases obviously with the PLA and EPI content increasing. When molar ratios of mesoscale molecules (PLA and drug) increased to 10%, the self aggregated PLA chains appeared and some drug molecule cannot be encapsulated inside the PLA matrix, but encapsulated outside the PLA particles. The excess drug molecules will be encapsulated outside the particles when the drug content exceeds the molar ratio (10%). More and more drug molecules are encapsulated outside the particles with the increase of drug content.

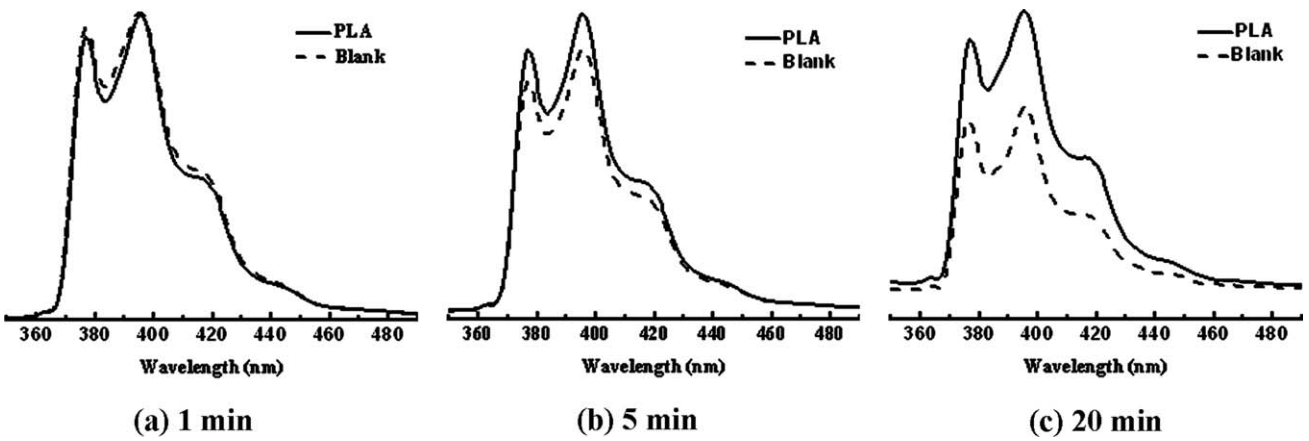

Figure 7 Fluorescence emission spectra of samples in the presence of pyrene at different dialysis time.

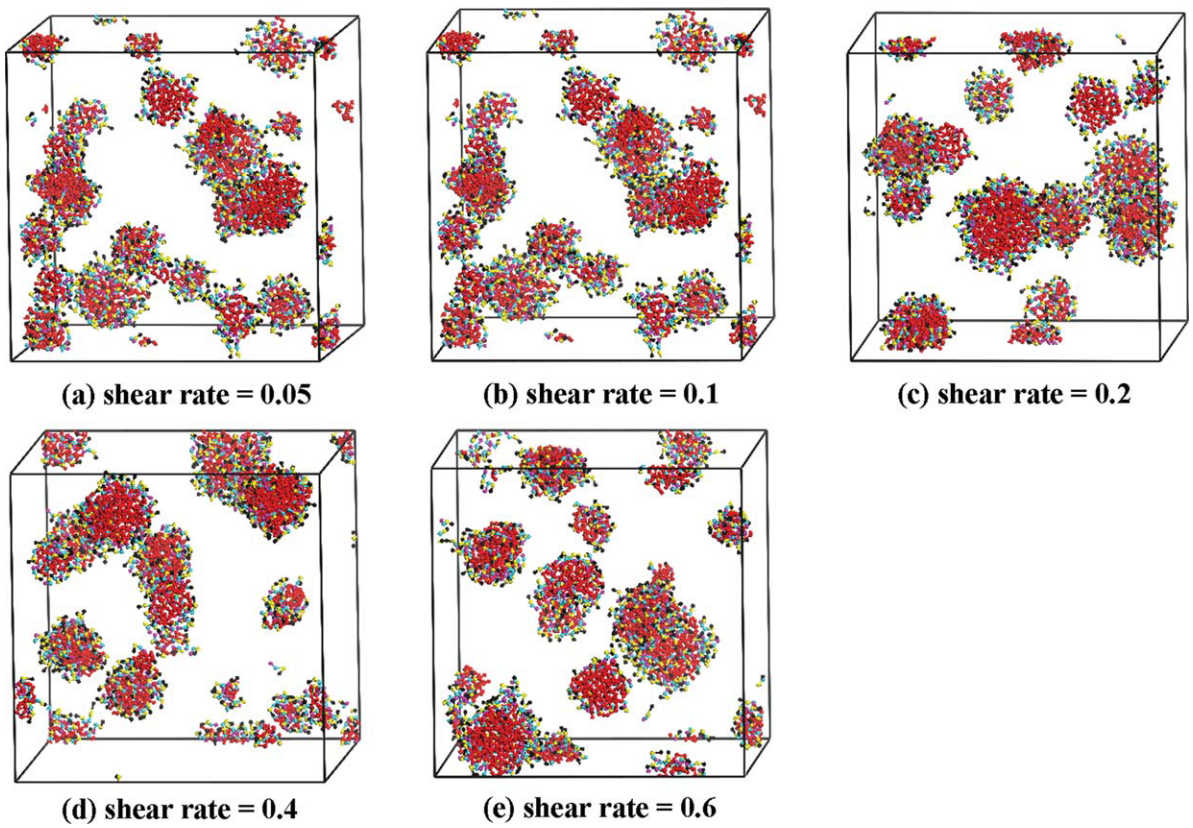

Figure 8 Aggregate morphologies of PLA particles with drug at the different shear rate. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

Effect of the DMF content on the aggregate morphology of DDS

Effect of the DMF content on the aggregate morphology of PLA particles with drug are investigated and shown in Figure 6. It is clear that the formation process of PLA particles with drug is the same as that of the PLA particles without drug in the dialysis process. When the PLA and EPI are dissolved in the DMF, they are dispersed well [Fig. 6(a)]. However, with the DMF content decreasing and water content increasing, the aggregation of PLA chains and the drug molecules, the PLA chains, and the drug molecules appeared gradually. Of course, the aggregation of the same molecules is more obvious than that of the different molecules at the beginning [Fig. 6(b–c)]. The PLA particles with drug begin to form and disrupt further [Fig. 6(d–e)] with the DMF content decreasing and water content increasing further. At last, the PLA particles with drug is transformed from swollen state to densely packed state [Fig. 6(f)] with DMF decreasing gradually to be instead of water completely.

The growth process of PLA particles prepared by dialysis were monitored by SEM and DLS[4]; at the same time, the method of Fluorescence emission spectra of pyrene was used to inspect the formation of PLA particle.[24] The pyrene is easy self-destroy in the polarity solution, so the intensity of Fluorescence is very low. However, when the micro-district was formed in the solution, the pyrene can produce the strong radiation, so the fluorescence is strong. The Fluorescence emission spectra of PLA spheres and blank samples in the presence of pyrene at different dialysis time were shown in Figure 7. The composite of solvent is varied with the dialysis process. However, the Fluorescence intensity of pyrene in the different solvent is very different. So, the Fluorescence intensity of the same content pyrene in the samples of different dialysis time is not comparable. The contradistinguish method is used to inspect the formation of PLA particles. It is to say, the samples which own the same composite solvent and pyrene with the sample of the dialysis were prepared.

The results showed that the Fluorescence intensity of pyrene of PLA and blank samples is the same when the dialysis proceed for 1 min [Fig. 7(a)], which confirms there is no micro-district formed in the solvent. When the dialysis time is 5 min or over, the Fluorescence intensity of pyrene in the PLA solution is higher than that of blank samples [Fig. 7(b,c)], which confirms there is micro-district formed in the PLA solution. These results agree very well with the results of SEM, DLS, and DPD simulation.

The results of the DPD simulation on the basis of particle growth process confirmed the formation

mechanism of PLA particles[4] suggested according to experimental phenomenon is reasonable. The formation of PLA particles consists of three steps: (1) aggregation-individual PLA chains got aggregated with each other in solution [Figs. 6(b) and 7(a)]; (2) formation and disruption of PLA particles, particles with irregular shape were formed due to the association of PLA chains, meanwhile, the particles tended to disrupt into small particles to obtain small surface energy [Figs. 6(c) and7(b)], at the end of this period, PLA spheres with slack chains were obtained [Fig. 6(d)]; (3) solidification—PLA chains turned from slack state to compact state which was corresponding to PLA spheres′ transition from swollen state to densely packed state [Figs. 6(e–f) and 7(c)].

The effect of shear rate on aggregate morphology of DDS

In the dialysis experiments, it happens on that there is some effect of perturbation on preparation of PLA particles. To investigate the effect of the perturbation on the formation of PLA particles in detail, the aggregate morphologies of DDS at different shear rate are investigated and shown in Figure 8. Where, the molar fraction of mesoscale molecule (PLA, water, DMF, and drug) is 3 : 94 : 0 : 3. The result showed the effect of shear rate on aggregate morphologies of DDS is not very obvious. However, the shear rate of 0.2 makes the distribution of drug in PLA matrix more uniform [Fig. 8(c)]. At the same time, there is no effect on aggregate morphologies of DDS at the lower shear rate [Fig. 8(b)]. With the shear rate increasing unceasingly [Fig. 8(d,e)], the size of PLA particles decrease slightly, the size distribution of PLA particles is not the same narrow as Figure 8(c). Because the shear rate effects the solution local concentration at the dialysis. When every point's concentration of solution was kept uniform and the solutes have equal chance to touch each other, shear rate is enough high. When the shear rate is very high some solutes may have no enough time to touch and form microspheres, resulting in decreasing PLA particles' size.

CONCLUSIONS

DPD approach has been successfully applied to the investigation of the formation of PLA particles. The simulation results indicate that the formation of PLA particles consists of aggregation of individual PLA chains and drug molecules, formation and disruption of PLA particles, solidification (from swollen state to densely packed state) three steps. The optimal molar fraction of mesoscale molecule of PLA or drug is fixed at 3%. Under different PLA and drug content, the particles observed are spherical structures. The size of PLA particles increases with the PLA increasing. At the same time, the size of PLA particles with drug also increases slightly with the drug content increasing. The solvent (DMF) content of PLA particle formation and disruption has been also proposed. DPD simulations could not only provide insight into the mechanism of mesoscopic structures of PLA particles, but also serve as a complement to experiments and more efficiently guide the experimental preparation of PLA particles and DDS with desired properties.

References

1. Smith, P. F. IDrugs 2003, 6, 1173.
2. Nomura, T.; Koreeda, N.; Yamashita, F.; Takakura, Y.; Hashida, M. Pharm Res 1998, 15, 128.
3. Waterman, K. C.; Sutton, S. C. J Controlled Release 2003, 86, 293.
4. Liu, M.; Zhou, Z. M.; Wang, X. F.; Xu, J.; Yang, K.; Cui, Q.; Chen, X.; Cao, M. Y.; Weng, J.; Zhang, Q. Q. Polymer 2007, 48, 5767.
5. Hoogerbrugge, P. J.; Koelman, J. Europhys Lett 1992, 19, 155.
6. Koelman, J.; Hoogerbrugge, P. J. Europhys Lett 1993, 21, 363.
7. Español, P.; Warren, P. Europhys Lett 1995, 30, 191.
8. Groot, R. D.; Warren, P. B. J Chem Phys 1997, 107, 4423.
9. Groot, R. D.; Madden, T. J. J Chem Phys 1998, 108, 8713.
10. Groot, R. D. Langmuir 2000, 16, 7493.
11. Groot, R. D. J Chem Phys 2003, 118, 11265.
12. Groot, R. D.; Rabone, K. L. Biophys J 2001, 81, 725.
13. Yuan, S. L.; Cai, Z. T.; Xu, G. Y.; Jiang, Y. S. Chem Phys Lett 2002, 365, 347.
14. Maiti, A.; McGrother, S. J Chem Phys 2004, 120, 1594.
15. Li, Y. M.; Xu, G. Y.; Luan, Y. X.; Yuan, S. L.; Zhang, Z. Q. Colloid Surf A 2005, 257–258, 385.
16. van Vliet, R. E.; Hoefsloot, H. C.; Iedema, P. D. Polymer 2003, 44, 1757.
17. Yuan, S. L.; Xu, G. Y.; Luan, Y. X.; Liu, C. B. Colloid Surf A 2005, 256, 43.
18. Guo, X. D.; Zhang, L. J.; Qian, Y.; Zhou, J. Chem Eng J 2007, 131, 195.
19. Long, C. X.; Zhang, L. J.; Qian, Y. Chem Eng J 2006, 119, 99.
20. Qian, H. J.; Lu, Z. Y.; Chen, L. J.; Li, Z. S.; Sun, C. C. J Chem Phys 2005, 122, 184907.
21. Yamamoto, S.; Hyodo, S. J Chem Phys 2005, 122, 204907.
22. Brandrup, J.; Immergut, E. H. Polymer Handbook, 3rd ed.; John Wiley & Sons Inc.: New York, 1989.
23. Hu, J. L.; Tang, Z. H.; Qiu, X. Y.; Pang, X.; Yang, Y. K.; Chen, X. S.; Jing, X. B. Biomacromolecules 2005, 6, 2843.
24. Glushiko, V.; Thaleer, M. S. R.; Karp, C. D. Arch Biochem Biophys 1981, 210, 33.