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Nifedipine solid dispersion in microparticles of ammonio methacrylate copolymer and ethylcellulose binary blend for controlled drug delivery Effect of drug loading on release kinetics

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Abstract

In order to elucidate the controlled-release mechanism of a poorly water-soluble drug from microparticles of ammonio methacrylate copolymer and ethylcellulose binary blend prepared by a phase-separation method, nifedipine-loaded microparticles with different levels of drug loading were evaluated by micromeritic properties, drug physical state, matrix internal structure, drug dissolution, and release modeling. Drug release study indicated that nifedipine release from the microparticles followed the Fickian diffusion mechanism, which supported the study hypothesis that as a result of formation of a nifedipine molecular dispersion, nifedipine dissolution inside the matrix was no longer the rate-limiting step for drug release, and the drug diffusion in matrix became the slowest step instead. Moreover, study results indicated that even though drug loading did not significantly affect the microparticle size distribution and morphology, nifedipine release rate from those microparticles was more or less influenced by the level of drug loading, depending on matrix formulation. At lower levels of drug loading, nifedipine release was well described by the Baker and Lonsdale's matrix diffusion model for microspheres containing dissolved drug and nifedipine had a plasticizing effect on the polymers that caused an increase in drug effective diffusion coefficient with increasing drug loading. However, at higher levels of drug loading, probably due to formation of solid nifedipine domains in microparticles, a change in the release kinetics was observed.

Keywords: Nifedipine; Solid dispersion; Controlled release; Microparticles; Ethylcellulose; Eudragit RL

1. Introduction

According to Nixon (Nixon, 1983), three steps lead to drug release from microparticles into the aqueous medium: (1) imbibition of the release medium into the microparticles, (2) dissolution of the drug substance inside the microparticles, and (3) drug release by a diffusion process into the aqueous medium. In addition, in some cases the drug diffusion in the stagnant aqueous layer at the surface of microparticles may complicate the analysis of this process (Jalsenjak, 1992). Generally, the slowest step described above would be the rate-limiting step for the drug release from the microparticles into the aqueous medium.

For a poorly water-soluble drug, nifedipine (5.6 μg/mL at pH 7) (Ali, 1989), the drug dissolution from its stable crystalline form was reported being the slowest step during drug absorption process and causing low drug bioavailability (Benita et al., 1990). In addition, due to its fast clearance rate (Pfizer, 2003; Bayer, 2004), traditional immediate-release nifedipine oral solid dosage forms have to be administered three times a day, which results in a significant fluctuation in the plasma drug concentration and drug toxic side effects. Therefore, development of nifedipine controlled-release dosage forms is desirable for side effect reduction and for patient compliance. Usually, there were two formulation steps for a controlled-release dosage form of a poorly water-soluble drug. First, different technologies, such as reduction of particle size (Kornblum and Hirschorn, 1970) or solid dispersion of drug with polymers (Sekiguchi and Obi, 1961; Chiou and Riegelman, 1969), were utilized to improve the drug dissolution rate. Thereafter, a controlled-release technology was applied to achieve sustained release of drug (Pfizer,

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2003; Bayer, 2004). As a multi-particulate system, matrix-type microparticles or nanoparticles, which contain uniformly dispersed or dissolved drug, are another good example of controlled release dosage forms for an improvement of drug bioavailability and for a better controlling of drug delivery (Benita et al., 1990; Barkai et al., 1990; Bodmeier et al., 1991; Kim et al., 1997; Guyot and Fawaz, 1998). It has advantages in avoiding dose "dumping", reducing local irritation, minimizing erratic drug absorption, and achieving a more reproducible drug release rate. An improved bioavailability and a prolonged constant drug plasma concentration were reported after administration of this multi-particulate disperse system to rats (Kim et al., 1997). Moreover, these matrix-type microparticles may offer double benefits in dissolution rate improvement and controlled delivery of poorly water-soluble drugs in one formulation step. First, molecular dispersion of drug in polymer(s) would dramatically improve the drug dissolution rate locally within the microparticles by reducing particle size to a minimum level (Leuner and Dressman, 2000). Second, the presence of insoluble polymer(s) in the micro-matrix would modify the drug release rate into the dissolution medium by changing the matrix permeability.

To test these "double" concepts, nifedipine-loaded microparticles of a hydrophobic (ethylcellulose) and hydrophilic (Eudragit RL®) polymer binary blend were previously developed and explored for use in controlled release of nifedipine (Huang et al., 2006). A phase-separation methodology that is more flexible in choosing a solvent to avoid the use of toxic methylene chloride was used applied to prepare those microparticles. Enhancement or retardation of nifedipine release rate from the microparticles as compared to the micronized crystalline drug was achieved by adjusting the ratio of these two polymers. As a continuation of the previous study, investigations were further carried out to characterize the microparticles by the micromeritics properties, nifedipine physical state, microparticle internal structure, and drug in vitro release for the purpose to evaluate the effects of drug loading on the nifedipine release mechanism/kinetics from microparticles.

2. Materials and methods

2.1. Materials

Micronized crystalline nifedipine (NIF) was purchased from Sigma (St. Louis, MO). Ethylcellulose (EC) of N7 viscosity grade was kindly provided by Hercules (Wilmington, DE). Ammonio methacrylate copolymer, Eudragit RL100® (RL) granules were donated by Rohm America (Piscataway, NJ). Acetone and methanol were purchased from Sigma–Aldrich (St. Louis, MO). All other materials were at least of analytical grade. Because of the photosensitivity of nifedipine, drug samples were stored and handled under yellow light.

2.2. Methods

2.2.1. Microparticle preparation

Matrix-type microparticles, containing solid dispersion of nifedipine with polymers, were prepared by a phase-separation (co-precipitation) method. Different amounts of nifedipine were dissolved in a 50 mL acetone solution containing 7.5 g of ethylcellulose and Eudragit RL polymer blend at different EC/RL weight ratios. Under a constant stirring at 600 rpm, a 100 mL of non-solvent, purified water was added drop-wise (1 mL/min) to the drug and polymer solution. In the course of the water addition, the drug and polymer were co-precipitated out to form microparticles. At the end of the compounding, the resulted microparticle suspension was vacuum filtered with a Whatman # 5 filter disk, and then vacuum dried at room temperature for 72 h. The dried microparticles were stored in a desiccator at room temperature and protected from light until use.

2.2.2. Microscopic characterization

Microparticles were dispersed in mineral oil on a glass slide and covered with a cover glass. The microparticles were observed under an Olympus polarized light microscope equipped with a digital camera and image analysis software (Image-Pro® Plus 4.5 software for Windows™, San Diego, CA). A field containing approximately 100–300 microparticles was randomly selected for size analysis. The equivalent spherical diameter of a microparticle (d_s) was calculated from the projection area of the microparticle by Eq. (1). The geometric mean (median) diameter, the 50% size, was used to express the median particle size (Fonner et al., 1981). The size measurement was repeated with 3–7 replicates:

$$\text{diameter} = 2\sqrt{\frac{\text{area}}{\pi}}. \quad (1)$$

2.2.3. Analysis of nifedipine concentration

For nifedipine loading determination, an appropriate amount of microparticles was dissolved in methanol to obtain a theoretical nifedipine concentration of 20 mg/L. The drug concentration was then analyzed using a UV-visual spectrophotometer at 236 nm with a standard curve prepared using bracketed concentrations of nifedipine methanol solution. To determine nifedipine concentration in 0.5% (w/v) sodium dodecyl sulphate (SDS) aqueous solution for the dissolution study, the solution was measured using the same wavelength of 236 nm, and drug concentration was calculated with a standard curve prepared using bracketed concentrations of nifedipine aqueous solution with 0.5% SDS. No interference from the polymers or SDS on nifedipine assay was found at 236 nm.

2.2.4. X-ray powder diffraction

X-ray powder diffraction (XRPD2) was carried out with a Philips X'Pert powder diffractometer. A Cu Kα source operation (40 kV, 50 mA) was employed. The diffraction patterns were recorded over a 2θ angular range of 2–40° with a step size of 0.02°in 2θ and a 6 s counting per step at room temperature.

2.2.5. Fourier-transformed infrared

The Fourier-transformed Infrared (FTIR) spectra of samples were obtained, using an FTIR spectrophotometer (Nicollet Magna 560, Nicollet Instrument, WI). About 2 mg of the samples were mixed with dried potassium bromide of equal weight

and compressed to form a KBr disc. The samples were scanned 64 times from 400 to 4000 cm^{-1}.

2.2.6. DSC thermal analysis

Differential scanning calorimetry (DSC) thermal analysis was conducted using a TA instrument (Model: DSC 2910, TA Instruments Inc., DE). In an open aluminum pan under a 10 mL/min stream of nitrogen purge, samples of 2–5 mg were heated from room temperature to 200 °C at a heating rate of 10 °C/min. Universal Analysis (Version 2.5) software was used for analysis.

2.2.7. Dissolution study

United State Pharmacopoeia (USP) dissolution apparatus II (paddle method) was used for nifedipine microparticle release studies. A sample equivalent to 20 mg of nifedipine was added into a 1000 mL of de-ionized water containing 0.5% (w/v) SDS with a stirring speed of 50 rpm. The dissolution medium temperature was maintained at 37 ± 0.5 °C. Periodically, a 5 mL solution withdrawn from the dissolution medium was filtered with a 0.45 μm hydrophilic filter disk and measured by a UV-visual spectrophotometer at 236 nm. The filter used was pre-saturated with a nifedipine solution. A 5 mL blank dissolution medium, 0.5% SDS solution, was replaced back into the dissolution medium after each sampling in order to maintain a sink condition. The dissolution test was done with 2–3 replicates.

2.2.8. Regression analysis of drug release data

The SigmaPlot 2002 for Windows Version 8.0 (1986–2001, SPSS, Inc., Chicago, IL) was used for regression analysis of drug release data with model equations. The Marquardt–Levenberg algorithm is applied by this software to find the coefficients (constant parameters) of the independent variables. Both manual and automatic function of initial estimation of parameters were used, and the maximum number of regression iterations was set as 100 with a maximum stepsize of 100. The regression tolerance, which is a measurement of the difference between the norm. of the residuals from one iteration to the next, was set as 0.0001. Three criteria were used to statistically determine how well the release data fitted to model equations (Sigma Stat 2.03, 1992–1997; Costa et al., 2001). Taking into account of the varied number of independent variables, adjusted *R*-squared was used to measure how well the regression model describes the data. In addition, the standard errors (S.E.) that are estimates of the uncertainties in the estimates of the regression coefficients were also used to determine if the release data is best fitted to an equation. The *F*-statistic value that is equal to the residual mean square of analysis of variance (ANOVA) statistics for the regression was used to gauge the contribution of the independent variables in predicting the dependent variable.

3. Theoretical models for drug release

In the literature, drug release mechanisms from microparticles had been evaluated with first order (Wagner, 1969), zero order (Vudathala and Rogers, 1995), Hixson–Crowell cubic root (Hixson and Crowell, 1931), Weibull (Langenbucher, 1972), Korsmeyer–Peppas (Korsmeyer et al., 1983), Higuchi (Higuchi, 1963), Baker–Lonsdale models (Baker and Lonsdale, 1974), etc. However, for matrix-type polymeric microparticles containing molecularly dispersed drug in water-insoluble polymers, the drug diffusion process in the microparticle matrix usually is the rate-limiting step for a well disturbed dissolution system with a perfect sink condition. Therefore, Korsmeyer–Peppas and Baker–Lonsdale models that were derived from diffusion theories are chosen to describe nifedipine release kinetics from the polymeric microparticles of this study.

3.1. Contribution of Fickian diffusion and case-II transport to drug release

The Korsmeyer–Peppas model (Eq. (2)) (Korsmeyer et al., 1983), as a semi-empirical model correlating the drug release to time by a simple exponential equation for the fraction of released drug <0.6, has been used to evaluate drug release from controlled-release polymeric devices, especially when the drug release mechanism is unknown or when there are more than one release mechanism (Costa et al., 2001):

$$f_t = kt^n \tag{2}$$

where f_t is the fraction of released drug (M_t/M_∞); k the constant related to the drug and structural and geometric properties of the microparticles; n the release exponent for Fickian or non-Fickian diffusion; and t is the elapsed time, respectively.

Ritger and Peppas reported that the possible mechanisms involving drug release from a polymeric controlled-release device are Fickian diffusion, case-II transport (polymeric relaxation) or anomalous transport (combination of diffusion and polymeric relaxation), and each of them has different ranges of exponent (n) value. For polymeric spheres, where drug release follows the Fickian diffusion process, the n value could be in the range from 0.30 (±0.01) for spheres with broad size distribution to 0.432 (±0.007) for monodisperse spheres (Ritger and Peppas, 1987a,b). In order to account for burst release, the single term Korsmeyer–Peppas equation was modified with a term representing the surface free drug (Kim and Fassihi, 1997). Furthermore, since the drug release modeling indicated that drug release from matrix-type microspheres is a function of t/r^2 (t-time, r^2-square of microparticle radius) (Ritger and Peppas, 1987a,b), Eq. (2) was further normalized by the microparticle size (r^2) (Eq. (3)) in order to eliminate the variation in release rate due to a difference in particle size:

$$f_t = f_0 + a\left(\frac{t}{r^2}\right)^n \tag{3}$$

where f_t, t and n were defined previously; f_0 is the fraction of surface free drug that is available for burst release; a is equal to kr^{2n}; and r is the microparticle radius, respectively.

For a new matrix-type drug delivery system without prior knowledge, the heuristic equation developed by Peppas and

Sahlin (Peppas and Sahlin, 1989) was modified and used to quantify the contribution of Fickian diffusion and case-II transport on drug release for this study. Similarly, this equation was modified with a term representing the surface free drug fraction and normalized by the microparticle size (r^2) (Eq. (4)):

$$f_t = f_0 + a\left(\frac{t}{r^2}\right)^n + b\left(\frac{t}{r^2}\right)^{2n} \quad (4)$$

where f_t, f_0, r and t were defined previously; n and $2n$ are the release exponent for Fickian diffusion and case-II transport, respectively; a and b are a constant related to the drug and the structural and geometric properties of the microparticles. The ratio of b to a indicates the contribution of these two mechanisms to the drug release.

3.2. Determination of the matrix diffusion type for nifedipine release

The release equation for matrix of spherical shape containing dispersed drug (Eq. (5)) was developed by Baker and Lonsdale (Baker and Lonsdale, 1974) from Higuchi model (Higuchi, 1963). Under this model, it was assumed that drug was uniformly dispersed in the matrix and the drug concentration in the matrix was greater than the drug solid solubility in matrix:

$$\frac{3[1-(1-M_t/M_\infty)^{2/3}]}{2} - \frac{M_t}{M_\infty} = \frac{3DC_s}{C_o}\frac{t}{r^2} \quad (5)$$

where M_t and M_∞ are the amount of drug released at time t and the total drug loading; D, C_o and C_s are drug effective diffusion coefficient, drug concentration, and drug solid solubility in the matrix; r is the radius of the whole sphere, and t is the elapsed time for release, respectively.

For the microspheres containing dissolved drug, the original equations for drug release were derived by Baker and Lonsdale from diffusion theory (Baker and Lonsdale, 1974) (Eqs. (6) and (7)). Under this model, the drug loading is lower than drug solid solubility in the matrix.

For short time, valid for $M_t/M_\infty < 0.4$:

$$\frac{M_t}{M_\infty} = 6\left[\frac{Dt}{r^2\pi}\right]^{1/2} - \frac{3Dt}{r^2} \quad (6)$$

For long time, valid for $M_t/M_\infty > 0.6$:

$$\frac{M_t}{M_\infty} = 1 - \frac{6}{\pi^2}\exp\left[\frac{-\pi^2 Dt}{r^2}\right] \quad (7)$$

where M_t, M_∞, D, r, and t were defined previously.

Considering the possibility of the free drug present at the surface of microparticles, the original equations were modified with a new term (f_o) representing the fraction of surface free drug available for burst release.

For short time, valid for $6[Dt/r^2\pi]^{1/2} - 3Dt/r^2 < 0.4$ or $(M_t/M_\infty - f_0)/(1-f_0) < 0.4$:

$$\frac{M_t}{M_\infty} = f_0 + (1-f_0)\left\{6\left\lfloor\frac{Dt}{r^2\pi}\right\rfloor^{1/2} - \frac{3Dt}{r^2}\right\} \quad (8)$$

For long time, valid for $1-(6/\pi^2)\exp[-\pi^2 Dt/r^2] > 0.6$ or $(M_t/M_\infty - f_0)/(1-f_0) > 0.6$:

$$\frac{M_t}{M_\infty} = f_0 + (1-f_0)\left\{1 - \frac{6}{\pi^2}\exp\left[\frac{-\pi^2 Dt}{r^2}\right]\right\}$$
$$= 1 - (1-f_0)\left\{\frac{6}{\pi^2}\exp\left[\frac{-\pi^2 Dt}{r^2}\right]\right\} \quad (9)$$

where M_t/M_∞ is the fraction of released drug at time t, including the entire surface free drug released immediately at time zero and the drug released from inside of the microparticles for up to time t; D, r, and t were defined previously.

4. Results and discussion

4.1. Microparticle micromeritic properties

Study results indicated that the microparticle size distribution, median diameter, and shape were not significantly affected

Table 1
Micromeritic properties of microparticles with different drug loadings

Matrix composition	Formulation-weight ratio (NIF:RL:EC)	Drug loading (w/w) (%)	Medium size (S.D.) (μm)	Microparticle Shape	Size distribution
Formulation A (RL:EC = 2:1)	0.2:10:5	2	19.3 (0.4)	Mostly Spherical	Narrow
	0.8:10:5	7	15.0 (0.6)		
	2:10:5	11	13.3 (0.4)		
	3:10:5	21	20.9 (1.7)		
Formulation B (RL:EC = 1:1)	0.2:7.5:7.5	1	19.2 (0.5)	Mostly Spherical	Narrow[a]
	0.8:7.5:7.5	5	23.3 (1.4)		
	2:7.5:7.5	10	18.7 (1.0)		
	3:7.5:7.5	18	20.2 (3.1)		
Formulation C (RL:EC = 1:2)	0.2:5:10	1	29.7 (10.9)	Elongated	Broad
	0.8:5:10	4	31.7 (7.9)		
	2:5:10	9	38.4 (10.1)		
	3:5:10	16	32.6 (5.0)		

[a] Broad size distribution for 18% drug loading.

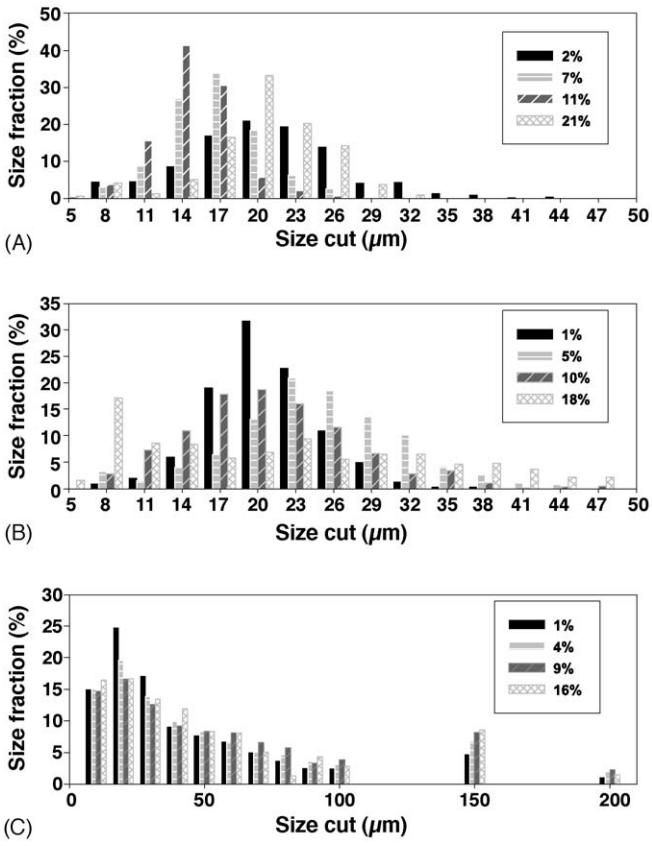

Fig. 1. Size distribution of nifedipine microparticles with different levels of drug loading. (A) Formulation A (RL/EC = 2:1), (B) formulation B (RL/EC = 1:1), and (C) formulation C (RL/EC = 1:2).

by changes of nifedipine loading (Table 1, Figs. 1–3). For formulations A and B, the physical characteristics, such as narrow size distribution and spherical shape of discrete microparticles were retained at different levels of drug loading except for formulation B with 18% of drug loading. No significant change in the microparticle median diameter with drug loading was observed for both formulations. For formulation C, no significant effects of drug loading on the microparticle median diameter and oblong shape were observed either. However, the size distribution plots (Fig. 1C) indicated that when drug loading was increased from 1% to 9%, there may be a reduction in the fine particle fraction (20–30 μm) and a corresponding slight increase in the fraction of coarse particles (150–200 μm).

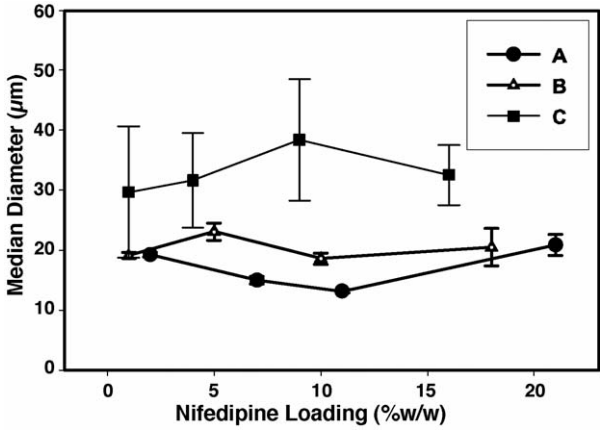

Fig. 2. Effect of nifedipine loading on the average microparticle median size. (A) Formulation A (RL/EC = 2:1), (B) formulation B (RL/EC = 1:1), and (C) formulation C (RL/EC = 1:2). Error bars represent the standard deviation of 3–7 measurements on the microparticles of the same batch.

4.2. Nifedipine physical state and microparticle internal structure

The physical states of nifedipine in microparticles of formulations A and C with different drug loadings were investigated by polarized light microscopy, X-ray, FT-infrared, and DSC. Since Microparticle formulation B (RL/EC = 1:1) is bracketed by formulation A (RL/EC = 2:1) and C (RL/EC = 1:2), no further solid-state characterization studies except light microscopy were performed on this formulation. The microscopic study (Fig. 3) indicated that microcrystalline or crystalline nifedipine was observed when drug loading was at 32% for formulation A, 18% for formulation B and 16% for formulation C, whereas no crystalline nifedipine was found for drug loadings at 21% for formulation A, 13% for formulation B and 9% for formulation C. This microscopic observation was confirmed by X-ray study (Fig. 4), which indicated that nifedipine crystalline started to appear between 21% and 32% of drug loading for formulation A and 9–16% for formulation C.

Further study using FTIR spectroscopy indicated that nifedipine physical state did change with increasing drug loading. Since the frequency of the stretching vibration of a functional group is influenced by its local environment, any change in the interactions of a nifedipine functional group with its neighboring groups as a result of a physical state modification should be reflected by a shift in its infrared stretching vibration wavenumber. Literatures reported that the two carbonyl groups (–C=O) of crystalline nifedipine form hydrogen bonds with the neighboring nifedipine amine groups and have two strong stretching vibration bands at 1679 and 1689 cm^{-1}, respectively (Ali, 1989; Triggle et al., 1980). However, after nifedipine was incorporated into the microparticles, the stretching vibration of nifedipine carbonyl groups was observed shifting with drug loading (Table 2). At the lower levels of drug loadings, a new stretching vibration at 1705–1707 cm^{-1} was observed for the nifedipine carbonyl groups, completely replacing those of crystalline nifedipine carbonyl groups at 1679 and 1689 cm^{-1}. This new nifedipine carbonyl stretching vibration at 1705–1707 cm^{-1}, located between those of amorphous nifedipine (1701 cm^{-1}) (Burger and Koller, 1996) and "free", non-associate carbonyl group (1728 cm^{-1}) (Teraoka et al., 1999), indicated a formation of a new nifedipine physical state, most likely a molecular dispersion as a result of nifedipine–polymer interaction. Since no shift in the stretching vibration of nifedipine carbonyl group was observed on the physical mixture of crystalline nifedipine and an EC/RL polymer binary blend, this interaction between drug and polymers was proposed to be hydrogen-bond interaction. When drug loading was increased, the nifedipine carbonyl stretching vibrations at 1705 and 1701 cm^{-1} both appeared at 21% of drug loading for formulation A, suggesting coexistence of the nifedipine molec-

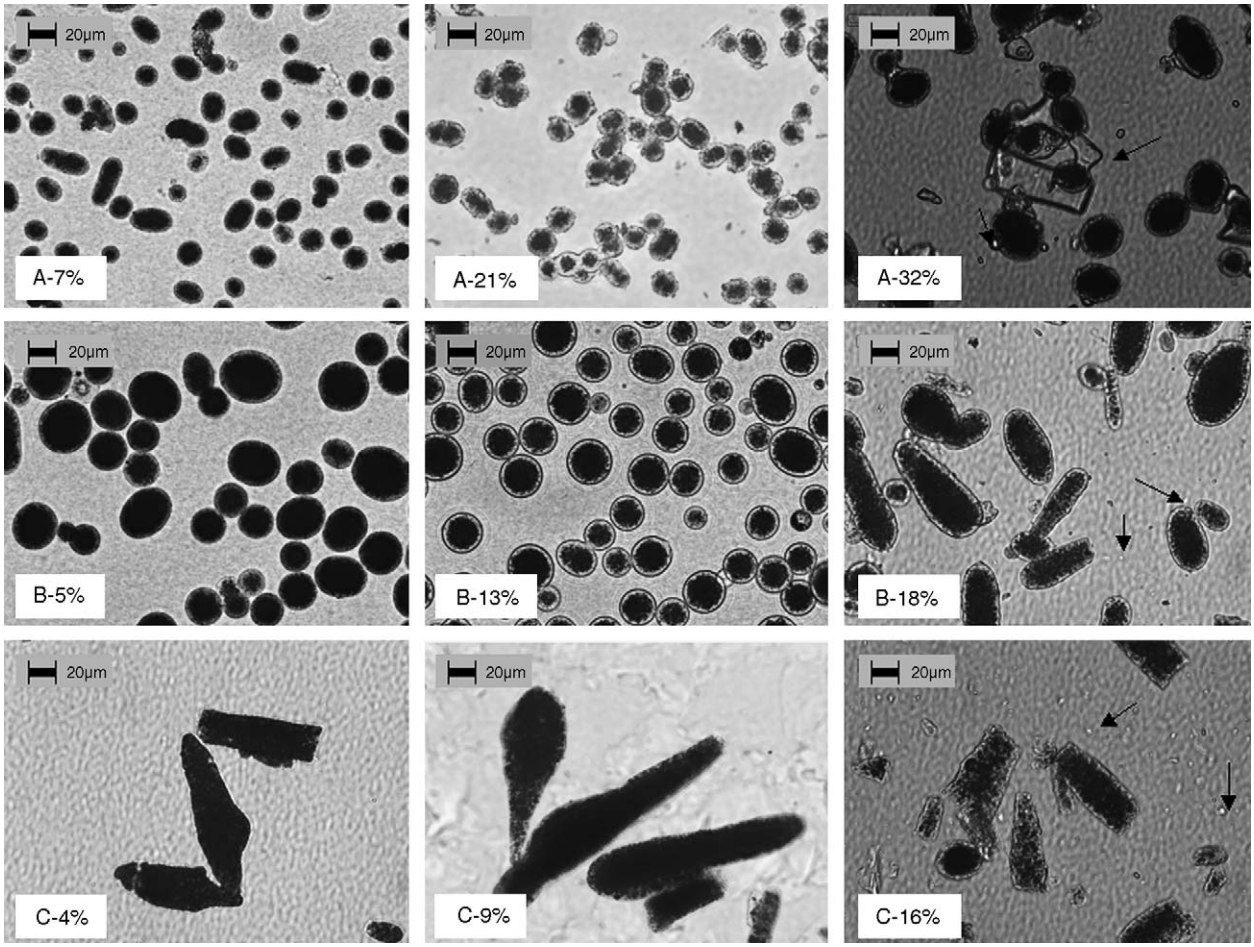

Fig. 3. Microscopic evaluation of microparticle morphology and nifedipine crystallinity. (A) Formulation A (RL/EC = 2:1), (B) formulation B (RL/EC = 1:1) and (C) formulation C (RL/EC = 1:2). Scale bar = 20 μm with a magnification of 40×; Arrows point to the appearance of surface microcrystalline or crystalline nifedipine.

Table 2
Shift of nifedipine carbonyl stretching vibration wavenumber at different physical states

Nifedipine (NIF) formulation	NIF loading (w/w) (%)	C=O (cm^{-1})	NIF physical state
Crystalline NIF[a]	–	1679, 1689	Crystalline NIF of stable form
PM[b] of crystalline NIF and EC/RL blend	–	1679, 1689	Crystalline NIF of stable form
Amorphous NIF[c]	–	1701	Amorphous NIF
Formulation A (RL:EC = 2:1)	7	1705	Molecular dispersion
	11	1705	Molecular dispersion
	21	1705	Molecular dispersion
		1701	+Amorphous NIF
Formulation C (RL:EC = 1:2)	4	1706	Molecular dispersion
	9	1707	Molecular dispersion
	16	1707	Molecular dispersion
		1701	+Amorphous NIF
		1686	+Crystalline NIF
Nifedipine nitro-derivative[d]	–	1728	Free, non-associated carbonyl group

[a] Ali (1989).
[b] PM: physical mixture.
[c] Burger and Koller (1996).
[d] Teraoka et al. (1999).

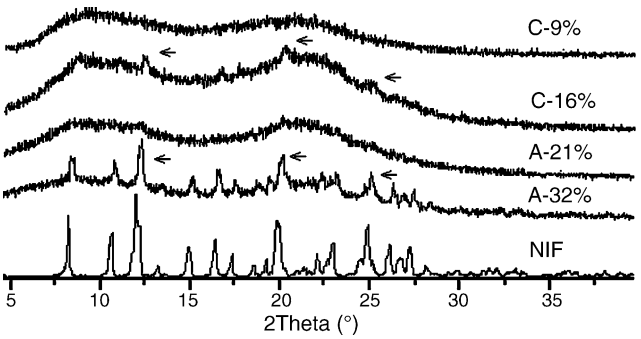

Fig. 4. Evaluation of nifedipine crystallinity by X-ray. (A) Formulation A (RL/EC = 2:1) and (C) formulation C (RL/EC = 1:2), NIF: crystalline nifedipine; arrows indicate the characteristic peaks of crystalline nifedipine.

ular dispersion and amorphous form. Whereas, the appearance of 1707, 1701 and 1686 cm^{-1} at 16% of drug loading for formulation C indicated coexistence of the nifedipine molecular dispersion, amorphous, and crystalline forms.

As a result of nifedipine–polymer interaction, the changes in matrix internal structure resulting from the plasticizing effect of solute on polymer (Gillett et al., 1972; Dubernet et al., 1991; Brabander et al., 2002) were also observed on the nifedipine microparticles of this study (Fig. 5). DSC data showed that as nifedipine loading in the microparticles of formulation A was increased from 0% to 11%, the glass transition point (T_g) of the micro-matrix decreased from ~125 to ~115 °C, whereas the T_g of the micromatrices of formulation C shifted downward from ~143 to ~118 °C when the drug loading was increased from 0% to 9%. For microparticles with drug loadings higher than 11% w/w for formulation A and 9% for formulation C, no further reduction in the glass transition point of matrix was observed. The downward shift of the matrix glass transition point with increasing drug loading implied that nifedipine was incorporated into a solid solution with polymers, while nifedipine acted as a plasticizer for the polymer matrix. For microparticles with drug loading at 21% for formulation A and at 16% for formulation C, as suggested by an appearance of the glass transition of amorphous nifedipine, a phase separation between nifedipine and polymers may explain why no further reduction in the glass transition of polymeric matrix was observed.

The plasticizing effect of nifedipine on EC/RL polymer matrices was also indirectly reflected by a downward shift in the depolymerization temperature of EC polymer (Fig. 5). EC polymer was reported to undergo exothermal depolymerization degradation after its glass transition (Dubernet et al., 1991). For this study, the EC polymer degradation was observed in the DSC thermograms of untreated EC with an exothermic peak at ~200 °C. After incorporation of RL into microparticles with EC polymer, this peak shifted downward to ~175 °C. Moreover, loading those microparticles with nifedipine of various concentration levels caused an even greater decline in the EC degradation temperature down to ~160 °C. Since it was reported that the thermal degradation of the EC polymer depends on a formation of the rubbery state of EC polymer (Dubernet et al., 1991), the reduction in the EC degradation temperature with increasing drug loading may reflect a decrease in the EC polymer inter-chain interactions as a result of a higher portion of nifedipine incorporated into the microparticles.

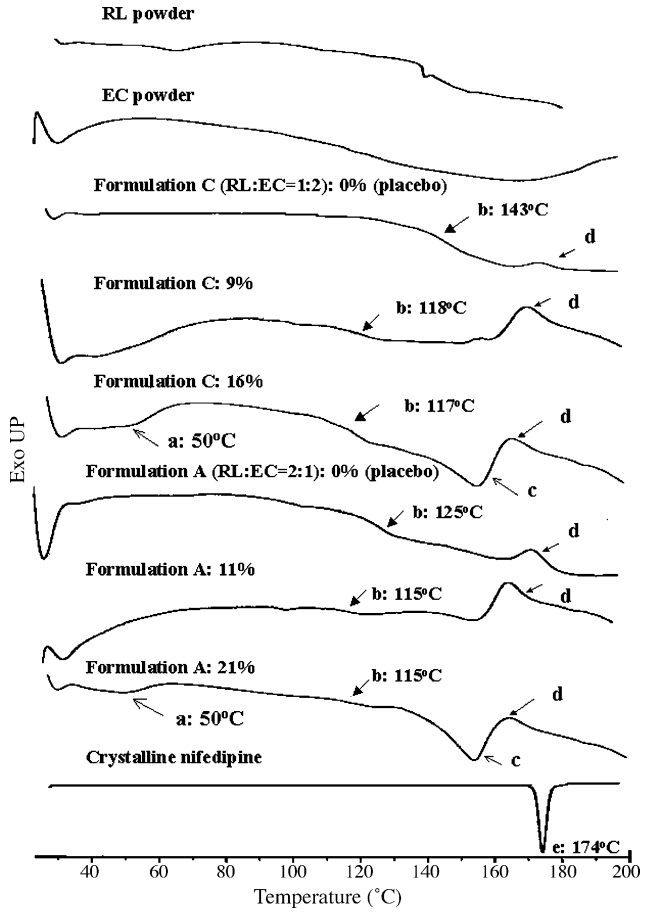

Fig. 5. Effect of nifedipine loading on the glass transition of polymeric matrices. (a) Glass transition of amorphous nifedipine, (b) glass transition of RL/EC micro-matrix; (c) melting of solid nifedipine in microparticles, (d) degradation of EC polymer, and (e) melting of crystalline nifedipine.

4.3. Nifedipine in vitro release from drug-loaded microparticles

The drug release profiles (Fig. 6) indicated that nifedipine release from the microparticles was more or less affected by the drug loading, depending on the microparticle matrix formulation. The percentage of drug released at 6 h for formulation A was observed increasing with drug loading from 2% to 11% w/w and then decreasing from 11% to 21% w/w. A burst release was observed when the drug loading was 32% w/w. For formulation B, the percentage of drug release at 6 h also increased with drug loading and a burst release was found at 18% w/w of drug loading. To the contrary, for formulation C, a slight reduction in the percentage of drug released at 6 h was observed with increasing nifedipine loading from 1% to 9% w/w; and a burst release of drug was observed at the nifedipine loading of 16% w/w. Based on the microscopic observations on those microparticles (Fig. 3), the observed burst release for microparticles with high levels of drug loadings was attributed to the surface nifedipine at the periphery of the microparticles.

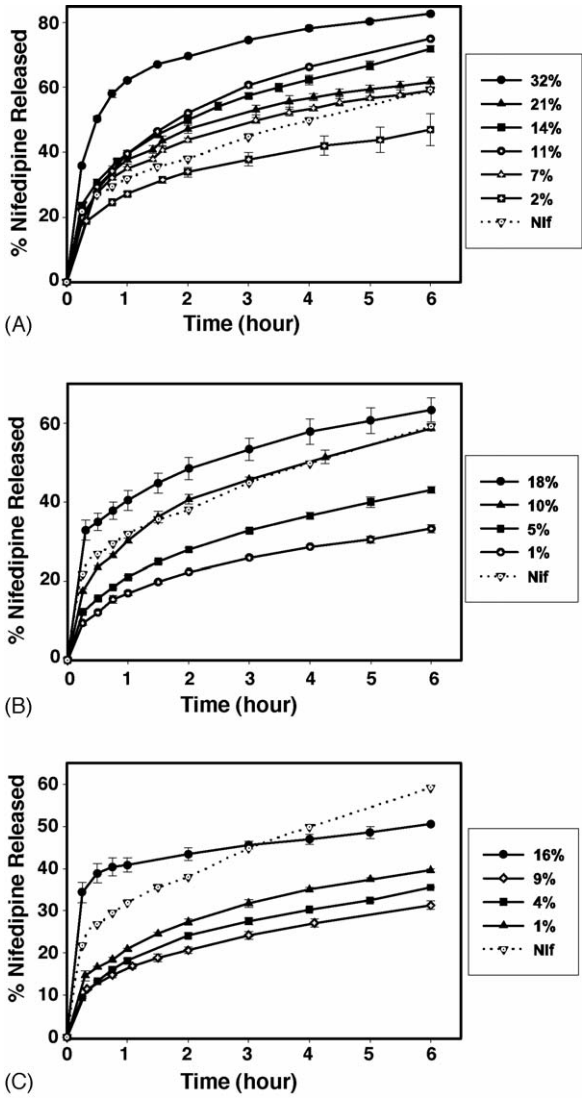

Fig. 6. Nifedipine release from microparticles with different levels of drug loading. (A) Formulation A (RL/EC = 2:1), (B) formulation B (RL/EC = 1:1), and (C) formulation C (RL/EC = 1:2). Dotted lines represent drug release from the micronized crystalline nifedipine (Nif). Error bars indicate the standard deviation of 2–3 replicates.

4.4. Release modeling

4.4.1. Contribution of Fickian diffusion and case-II transport to drug release

Hydrophilic RL polymer may swell as a result of macromolecular relaxation during release study. Therefore, it is possible that the drug release mechanism from the micromatrices of EC and RL binary mixture may be a coupling of diffusion and macromolecular relaxation kinetics. As no prior knowledge was found for nifedipine release from the EC/RL microparticles, the modified heuristic equation developed by Peppas and Sahlin (Peppas and Sahlin, 1989) was initially used to quantify the contribution of Fickian diffusion and case-II transport on nifedipine release (Eq. (4)).

The release exponent values (n) derived from Eq. (4) were in the range from 0.30 to 0.44 (Table 3), which matched the range (0.30–0.43) described by Ritger and Peppas (Ritger and Peppas, 1987a,b) for polydisperse spheres where the drug release is controlled by the Fickian diffusion mechanism. The derived b values that represent case-II transport were much less than a ($b/a \ll 0$), indicating that the contribution of case-II transport to the nifedipine release from the microparticles of this study was negligible. Further release modeling with the modified single-term Korsmeyer–Peppas equation (Eq. (3)) confirmed this observation. The n and a values derived from Eq. (3) were equal to those from Eq. (4) (Table 3). However, the adjusted regression correlation coefficients (adjusted R^2) (in the range from 0.9560 to 0.9997) and F-statistics (in the range from 89 to 12,402) of the non-linear regression using Eq. (3) were higher than those using Eq. (4), and the constant parameters derived from Eq. (3) had a much smaller standard error than Eq. (4), indicating that the release data is better described by Eq. (3) without using the term related to case-II transport.

Therefore, it was concluded that the nifedipine release from the microparticles was predominately controlled by the nifedipine Fickian diffusion kinetics rather than by the swelling of matrix (case-II transport), which supports the study hypothesis that due to formation of a nifedipine molecular dispersion, the nifedipine dissolution inside the microparticles is no longer the rate-limiting step and the drug diffusion in matrix becomes the slowest step for drug release.

4.4.2. Determination of the matrix diffusion type for nifedipine release

The release kinetics analysis with the semi-empirical Korsmeyer–Peppas model indicated that nifedipine release from the microparticles of EC and RL binary mixture was controlled by the Fickian diffusion mechanism. However, how nifedipine physical state and microparticle internal structure influenced the drug diffusivity in the polymeric matrix was still not clear. Since solid-state characterization studies suggested that depending on the drug loading, nifedipine was either dissolved or dispersed as a solid form in the microparticles, the matrix diffusion models for non-swelling microspheres containing dispersed or dissolved drug were compared in order to determine the fundament of drug release mechanism and to confirm nifedipine physical state in this dosage form.

Using SigmaPlot linear or non-linear regression program, the drug release data were analyzed using Baker and Lonsdale's model for microspheres containing dispersed drug (Eq. (5)) and Baker and Lonsdale's model for microspheres containing dissolved drug (Eqs. (8) and (9)), respectively. For non-linear regression of the release data using Eqs. (8) and (9), the surface free drug fraction, f_0, was first estimated by regression analysis using Eq. (8) for $M_t/M_\infty < 0.4$ and Eq. (9) (only if $M_t/M_\infty > 0.6$) for $M_t/M_\infty > 0.6$. Thereafter, based on the f_0 value determined from last regression, the regression analysis was repeated until a constant f_0 value was obtained from two consecutive regression analysis, using Eq. (8) for $(M_t/M_\infty - f_0)/(1 - f_0) < 0.4$ and Eq. (9) [only if $(M_t/M_\infty - f_0)/(1 - f_0) > 0.6$] for $(M_t/M_\infty - f_0)/(1 - f_0) > 0.6$. Even though the relative standard errors (standard error/constant parameter) of derived constant parameters were similar for both

Table 3
Results of non-linear regression with Korsmeyer-Peppas models

Matrix composition	Drug loading (w/w) (%)	Eq. (3)		Eq. (4)		
		n (S.E.)	*a* (S.E.)	*n* (S.E.)	*a* (S.E.)	*b* (S.E.)
Formulation A	2	0.30 (0.04)	1.07 (0.04)	0.30 (0.65)	1.1 (2.8)	1.9E−11 (3.9)
	7	0.31 (0.06)	1.21 (0.03)	0.31 (0.69)	1.2 (3.1)	1.2E−11 (4.1)
	11	0.41 (0.08)	1.81 (0.15)	0.4 (1.5)	1.8 (1.5)	5.3E−11 (14)
	14	0.35 (0.03)	1.81 (0.08)	0.35 (0.47)	1.8 (3.7)	2.5E−11 (5.4)
	21	0.32 (0.11)	1.52 (0.14)	0.3 (1.3)	1.5 (8.2)	8.2E−11 (11)
Formulation B	1	0.39 (0.05)	0.95 (0.07)	0.39 (0.51)	0.9 (2.0)	2.4E−11 (3.0)
	5	0.40 (0.02)	1.50 (0.04)	0.40 (0.15)	1.5 (1.0)	3.0E−11 (1.6)
	10	0.37 (0.05)	1.59 (0.11)	0.37 (0.54)	1.6 (3.5)	2.5E−11 (5.1)
	18	0.37 (0.04)	1.37 (0.01)	0.37 (0.47)	1.4 (2.7)	4.1E−10 (4.0)
Formulation C	1	0.37 (0.05)	1.30 (0.11)	0.37 (0.54)	1.3 (3.3)	3.4E−10 (5.1)
	4	0.39 (0.06)	1.50 (0.23)	0.39 (0.61)	1.5 (4.6)	3.1E−11 (8.1)
	9	0.43 (0.02)	1.57 (0.07)	0.43 (0.16)	1.6 (1.3)	2.3E−9 (2.7)
	16	0.44 (0.20)	0.97 (0.53)	0.44 (1.7)	1.0 (8.5)	1.0E−10 (17)

S.E.: standard error; *n*: release exponent; *a*: constant related to Fickian diffusion; *b*: constant related to case-II transport.

models, the regression analysis using Eqs. (8) and (9) (Table 4) had higher adjusted *R*-squared and *F*-statistic values than those of Eq. (5), indicating that the Baker and Lonsdale's model for microspheres containing dissolved drug described the release data better than the model for microspheres containing dispersed drug. Using the derived constant parameters, the normalized release profiles predicted by Eqs. (8) and (9) matched well with the actual normalized drug release curves (Fig. 7). Therefore, it was concluded that nifedipine release from microparticles was better described by the Baker and Lonsdale's model for microspheres containing dissolved drug, which in turn suggested that nifedipine was "dissolved" rather than "dispersed" in the microparticles of formulations A, B and C.

4.5. Effect of drug loading on nifedipine release kinetics

The best-fit Baker and Lonsdale's model for microspheres containing dissolved drug (Eqs. (8) and (9)) indicates that drug release is affected only by drug effective diffusion coefficient (D) and t/r^2. Therefore, the nifedipine release profiles normalized with respect to the microparticle median size (r^2) (Fig. 7) were further evaluated for the purpose to compare nifedipine diffusivity in matrix at different drug loading levels. Changes in the normalized release curves with drug loading should be related to changes in the drug effective diffusion coefficient in the matrix. Interestingly, at lower levels of drug loading, different effects of drug loading on the normalized release profiles were observed on different formulations. For formulation A (RL/EC = 2:1) and formulation B (RL/EC = 1:1), the normalized release rate indicated by steepness of the normalized release curve appeared to increase with drug loading except for that of formulation A at 21% of drug loading. However, for formulation C (RL/EC = 1:2), only a slight, but statistically insignificant, difference in the normalized release rate was found at different levels of drug loading. To further investigate the effect of nifedipine loading on drug diffusivity in the matrix, the drug effective diffusion coefficient (D) (Table 4) was obtained from non-linear regression of the release data with the Baker and Lonsdale's model for dissolved drug (Eqs. (8) and (9)). A rise in the drug effective diffusion coefficient value with increasing drug loading was

Table 4
Results of non-linear regression with Baker and Lonsdale matrix diffusion model for microspheres containing dissolved drug (Eqs. (8) and (9))

Matrix composition	Drug loading (w/w) (%)	f_0 (S.E.)	D (S.E.) ($\times 10^{-8}$ cm^2/h)	Adjusted R^2	F-statistics
Formulation A	2	0.11 (0.0082)	0.29 (0.016)	0.9925	1058
	7	0.070 (0.0097)	0.50 (0.028)	0.9973	4792
	11	0.046 (0.0059)	0.61 (0.011)	0.9994	12949
	14	0.084 (0.0054)	0.99 (0.024)	0.9991	12260
	21	0.104 (0.026)	0.71 (0.12)	0.9757	524
Formulation B	1	0.035 (0.0054)	0.16 (0.0076)	0.9937	1417
	5	0.032 (0.0019)	0.43 (0.0054)	0.9996	20629
	10	0.079 (0.013)	0.51 (0.038)	0.9938	1445
	18	0.24 (0.0019)	0.41 (0.010)	0.9998	59382
Formulation C	1	0.058 (0.0038)	0.41 (0.013)	0.9980	4578
	4	0.033 (0.0071)	0.50 (0.032)	0.9932	1164
	9	0.048 (0.0014)	0.46 (0.0073)	0.9997	23620
	16	0.33 (0.0086)	0.32 (0.042)	0.9610	198

SE: standard error; f_0: surface drug fraction available for burst release; D: drug effective diffusion coefficient.

observed for formulations A and B at the lower levels of drug loading. However, at higher levels of drug loading, it appeared that there was a slight reduction in nifedipine effective diffusion coefficient. For formulation C, a similar trend in the changes of drug effective diffusion coefficient with increasing drug loading was observed. However, the normalized release profiles shown in Fig. 7C indicate that the slight difference in the drug effective diffusion coefficient values might not be statistically significant for formulation C.

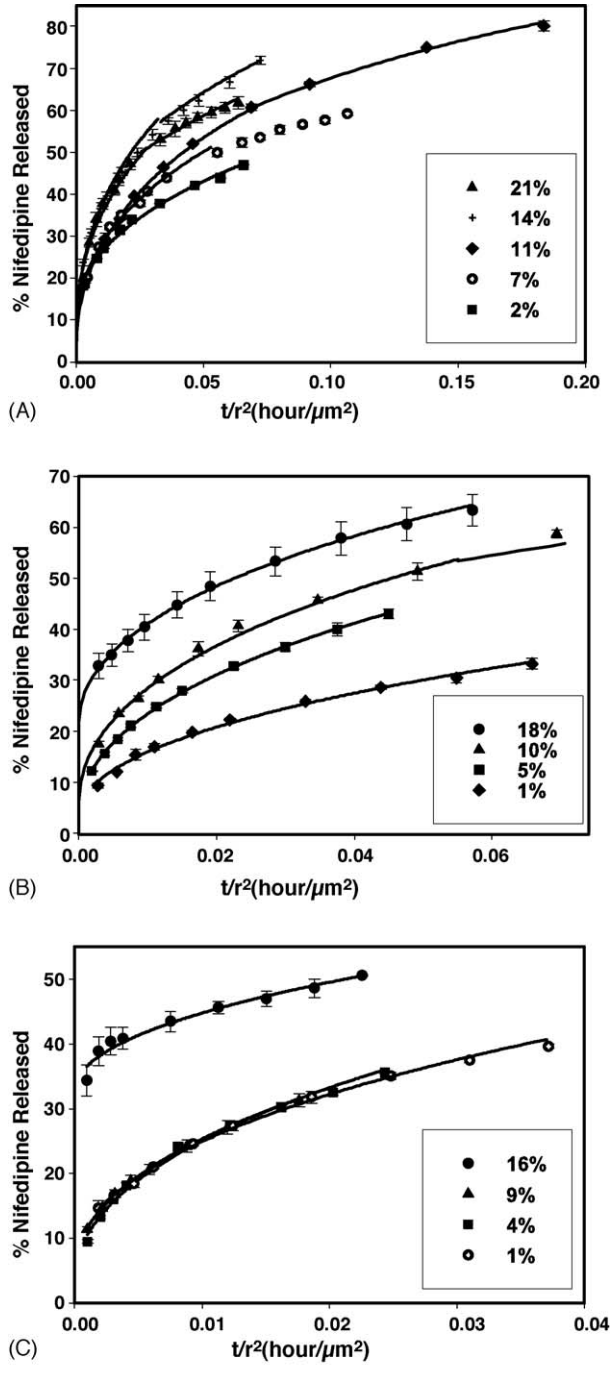

Fig. 7. Effect of nifedipine loading on normalized drug release with respect to microparticle size. (A) Formulation A (RL/EC = 2:1), (B) formulation B (RL/EC = 1:1), and (C) formulation C (RL/EC = 1:2). Solid lines represent the drug release predicted by Baker and Lonsdale model for microspheres containing dissolved drug (Eqs. (8) and (9)).

In the literature, drug–polymer interaction (Frisch, 1965) had been reported affecting the drug release rate. Two types of Fickian diffusion were described by Frisch (Frisch, 1965) for homogenous polymeric system in term of solute–polymer interaction. Type A Fickian diffusion was characterized as a concentration-independent diffusion coefficient for an ideal system where there is no solute–polymer interaction, whereas the diffusion coefficient of type B Fickian diffusion is concentration dependent due to strong polymer–solute interaction. Those observed different trends in the changes of drug effective diffusion coefficient with increasing nifedipine loading indicated that formulations A and B had a stronger drug–polymer interaction than formulation C probably as a result of different combinations of EC and RL polymers of different physical–chemical properties.

Furthermore, for the effects of drug loading on the changes in drug effective diffusion coefficient for each formulation, as indicated by a reduction in the matrix glass transition (T_g) and a corresponding increase in the drug effective diffusion coefficient (D), the increase in D was attributed to the plasticizing effect of nifedipine on matrices as a result of drug–polymer interactions, specifically the hydrogen-bond interactions between nifedipine and polymers. The interactions between plasticizer and polymer were known to reduce the polymer inter-chain interaction and to increase polymer chain mobility, which in turn increase drug diffusivity by an increase in the free volume for diffusion (Flynn et al., 1974). The reduction in the effective diffusion coefficient at the higher levels of drug loading might indicate formation of a solid nifedipine domain inside the microparticles as observed by the solid-state characterization. An increase in nifedipine loading would no longer have further plasticizing effects on the matrices due to the phase-separation between drug and polymers. To the contrary, the solid nifedipine domains may serve as reservoirs for drug, which would slow down the inward movement rate of solvent diffusion front by dissolving (Fan and Singh, 1989). As a result, the "apparent drug diffusion coefficient" derived from the Baker and Lonsdale's model for dissolved drug decreased accordingly.

5. Conclusions

Studies have indicated that nifedipine loading affected the nifedipine release kinetics/mechanism from microparticles of EC and RL binary mixture. No significant effects of drug loading on the microparticle size, morphology, and size distribution were observed. However, an increase in nifedipine loading may have cause a change in nifedipine physical state and microparticle internal structure. At the low levels of drug loading, nifedipine–polymer interaction generated a plasticizing effect on the matrix that caused an increase in drug effective diffusion coefficient with increasing drug loading. The best fit of release data with the Baker and Lonsdale's matrix diffusion model for microspheres containing dissolved drug supported that nifedipine was in a solid solution state at the low levels of drug loading. However, at the higher levels of drug loading, the

existence of solid nifedipine domain may cause a change in drug release kinetics by formation of reservoirs for nifedipine.

References

Ali, S.L., 1989. Nifedipine. In: Florey, K. (Ed.), Analytical Profile of Drug Substances, vol. 18. Academic Press Inc., New York, pp. 221–288.

Baker, R.W., Lonsdale, H.K., 1974. Controlled release: mechanism and rates. In: Tanquary, A.C., Lacey, R.E. (Eds.), Controlled Release of Biological Active Agents. Plenum Press, New York, pp. 15–71.

Barkai, A., Pathak, Y.V., Benita, S., 1990. Polyacrylate (Eudragit Retard) microspheres for oral controlled release of nifedipine. I. Formulation design and process optimisation. Drug Dev. Ind. Pharm. 16, 2057–2075.

Bayer Pharmaceuticals Corporation, 2004. The Package Insert of Adalat CC® (Nifedipine) Extended Release Tablet. Bayer Pharmaceuticals Corporation.

Benita, S., Barkai, A., Pathak, Y.V., 1990. Effect of drug loading extent on the in vitro release kinetic behavior of nifedipine from polyacrylate microspheres. J. Control. Rel. 12, 213.

Burger, A., Koller, K.T., 1996. Polymorphism and pesudopolymorphism on nifedipine. Sci Pharm. 64, 293–301.

Bodmeier, R., Chen, H., Tyle, P., Jarosz, P., 1991. Spontaneous formation of drug-containing acrylic nanoparticles. J. Microencapsulation 8, 161–170.

Brabander, C.D., Mooter, G.V.D., Vervaet, C., Remon, J.P., 2002. Characterization of ibuprofen as a nontraditional plasticizer of ethyl cellulose. J. Pharm. Sci. 91, 1678–1685.

Chiou, W.L., Riegelman, S., 1969. Preparation and discussion characterization of several fast-release solid dispersion of griseofulvin. J. Pharm. Sci. 58, 1505–1510.

Costa, P., Manuel, J., Lobo, S., 2001. Modeling and comparison of dissolution profiles. Eur. J. Pharm. Sci. 13, 123–133.

Dubernet, C., Rouland, J.C., Benoit, J.P., 1991. Ibuprofen-loaded ethylcellulose microspheres: analysis of the structure by thermal analysis. J. Pharm Sci. 80, 1029–1033.

Fan, L.T., Singh, S.K., 1989. Controlled Release: A Quantitative Treatment. Springer-Verlag, Berlin/New York, pp. 34–40.

Flynn, G.L., Yalkowsky, S.H., Roseman, T.J., 1974. Mass transport phenomena and models: theoretical concept. J. Pharm. Sci. 63, 479–510.

Fonner, D.E., Anderson, N.R., Banker, G.S., 1981. Granulation and tablet characteristics. In: Lieberman, H.A., Lachman, L. (Eds.), Pharmaceutical Dosage Forms, Tablets, vol. 2. Marcel Dekker, Inc., New York/Basel, p. 193.

Frisch, H.L., 1965. Mechanism of Fickian diffusion of penetrants in polymers. J. Polym. Sci. Polym. Lett. 3, 13–16.

Gillett, J.W., Harr, J.R., Lindstrom, F.T., Mount, D.A., St. Clair, A.D., Weber, L.J., 1972. Evaluation of human health hazards on use of dichlorovos (DDVP), especially in resin strips. Residue Rev. 44, 115–159.

Guyot, M., Fawaz, F., 1998. Nifedipine loaded-polymeric microspheres: preparation and physical characterization. Int. J. Pharm. 175, 61–74.

Higuchi, T., 1963. Mechanism of sustained-action medication: theoretical analysis of rate of release of solid dispersed in solid matrices. J. Pharm. Sci. 52, 1145–1149.

Hixson, A.W., Crowell, J.H., 1931. Dependence of reaction velocity upon surface and agitation. Ind. Eng. Chem. 23, 923–931.

Huang, J., Wigent, R.J., Schwartz, J.B. 2006. Nifedipine molecular dispersion in microparticles of ammonio methacrylate copolymer and ethylcellulose binary blends for drug controlled delivery. Effects of matrix composition. Drug Dev. Ind. Pharm. (accepted).

Jalsenjak, I., 1992. In vitro release from microcapsules and microspheres. In: Donbrow, M. (Ed.), Microcapsules and Nanoparticles in Medical and Pharmacy. CRC Press, Boca Raton, FL, p. 194.

Kim, H., Fassihi, R., 1997. Application of a binary polymer system in drug release rate modulation. 1. Characterization of release mechanism. J. Pharm. Sci. 86, 316–322.

Kim, Y.I., Flukinger, L., Hoffman, M., Lartaud-Idjouadiene, I., Atkinson, J., Maincent, P., 1997. The antihypertensive affect of orally administrated nifedipine-loaded nanoparticles in spontaneously hypertensive rats. Br. J. Pharmacol. 120, 399–404.

Kornblum, S.S., Hirschorn, J.O., 1970. Dissolution of poorly water soluble drugs. I. Some physical parameter related to method of micronization and tablet manufacture of a quinazolinone compound. J. Pharm. Sci. 59, 606–609.

Korsmeyer, R.W., Gurny, R., Doelker, E.M., Buri, P., Peppas, N.A., 1983. Mechanism of solute release from porous hydrophilic polymers. Int. J. Pharm. 15, 25–35.

Langenbucher, F., 1972. Linearization of dissolution rate curves by the Weibull distribution. J. Pharm. Pharmacol. 24, 979–981.

Leuner, C., Dressman, J., 2000. Improving drug solubility for oral delivery using solid dispersions. Eur. J. Pharm. Biopharm. 50, 47–60.

Nixon, J.R., 1983. Release characterization of microcapsules. In: Lim, F. (Ed.), Biomedical Applications of Microcapsulation. CRC Press, Boca Raton, FL, p. 19.

Peppas, N.A., Sahlin, J.J., 1989. A simple equation for the description of solute release. III. Coupling of diffuision and relaxation. Int. J. Pharm. 57, 169–172.

Pfizer and Inc., 2003. The Package Insert of Procardia XL® (Nifedipine) Extended Release Tablet. Pfizer Inc.

Ritger, P.L., Peppas, N.A., 1987a. A simple equation for description of solute release. I. Fickian and non-Fickian release from non-swellable devices in the form of slabs, spheres, cylinders or discs. J. Control. Rel. 5, 23–36.

Ritger, P.L., Peppas, N.A., 1987b. A simple equation for description of solute release. II. Fickian and anomalous release from swellable devices. J. Control. Rel. 5, 37–42.

Sekiguchi, K., Obi, N., 1961. Studies on absorption of eutectic mixtures. I. A comparison of the behavior of eutectic mixture of sulfathiazole and that of ordinary sulfathiazole in man. Chem. Pharm. Bull. 9, 866–872.

Sigma Stat 2.03, 1992–1997. Electronic User Manual. SPSS, Inc., Chicago, IL.

Teraoka, R., Otsuka, M., Matsuda, Y., 1999. Evaluation of photostability of solid-state dimethyl 1,4-dihydro-2,6-dimethyl-4-(2-nitro-phenyl)-3,5-pyridinedicarboxylate by using Fourier-transformed reflection–absorption infrared spectroscopy. Intl. J. Pharm. 184, 35–43.

Triggle, A.M., Shefter, E., Triggle, D.J., 1980. Crystal structure of calcium channel antagonists: 2,6-dimethyl-3,5-dicarbomethoxy-4-{2-nitro-, 3-cyano-, 4-(dimethylamino)-, and 2,3,5,6-pentafluorophenyl}-1,4-dihydropyridine. J. Med. Chem. 23, 1442–1445.

Vudathala, G.K., Rogers, J.A., 1995. Dissolution of fludrocortisone from biphasic polymers hydrogels. J. Control. Rel. 34, 185–192.

Wagner, J.G., 1969. Intepretation of percentage dissolved-time plots derived from in vitro testing of conventional tablets and capsules. J. Pharm. Sci. 58, 1253–1257.