Journal of Thermal Analysis and Calorimetry, Vol. 68 (2002) 631–646

BINARY SYSTEMS OF NIFEDIPINE AND VARIOUS CYCLODEXTRINS IN THE SOLID STATE
Thermal, FTIR, XRD studies

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Abstract

Nifedipine complexes with β-cyclodextrin (β-CD), γ-cyclodextrin (γ-CD), 2-hydroxypropyl-β-cyclodextrin (2HP-β-CD), randomly methylated-β-cyclodextrin (RM-β-CD) and heptakis(2,6-*O*-dimethyl)-β-cyclodextrin (DM-β-CD) have been prepared by both kneading and heating methods and their behaviour studied by differential scanning calorimetry (DSC), diffuse reflectance mid-infrared spectroscopy (FTIR) and X-ray diffractometry (XRD). DSC revealed the nifedipine melting endotherm with onset at approximately 171°C for the kneaded mixtures with β-CD, γ-CD and 2HP-β-CD, thus confirming the presence of nifedipine in the crystalline state, while some decrease in crystallinity was observed in the DM-β-CD kneaded mixture. With RM-β-CD, however, broadening and shifting of the nifedipine endotherm and reduction in its intensity suggested that the kneading could have produced an amorphous inclusion complex. These differing extents of interaction of nifedipine with the cyclodextrins were confirmed by FTIR and XRD studies.

Keywords: cyclodextrins, DSC, FTIR, inclusion compounds, nifedipine, XRD

Introduction

In recent years, extensive pharmaceutical research has been conducted into cyclodextrin inclusion complexes, that are used to improve the solubility and/or stability and bioavailability of different pharmaceutically active ingredients [1, 2] However the poor aqueous solubility of the natural β-cyclodextrin and its tendency to crystallize in aqueous solution have limited its usefulness within the pharmaceutical field [3]. Modification of the cyclodextrin structure has resulted in the alkylated [4] and hydroxyalkylated derivatives with improved physico-chemical properties and toxicity profiles [5, 6].

Nifedipine (Fig. 1) is a highly photolabile, practically water-insoluble drug used therapeutically as a calcium channel antagonist for the treatment of various cardiovascular disorders [7]. Poor and erratic bioavailability is observed following oral administration of crystalline nifedipine, mostly due to its low aqueous solubility and slow dissolution rate within the gastrointestinal tract [8–10].

1418–2874/2002/ $ 5.00
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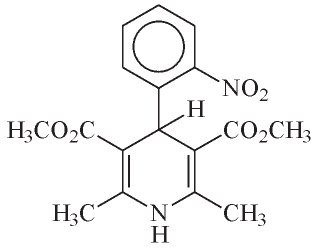

Fig. 1 Chemical structure of nifedipine

Cyclodextrins have been shown to be effective solubilisers of numerous hydrophobic drugs, permitting enhanced dissolution, absorption and bioavailability [2, 11–13]. The improved dissolution is most often attributed to a decrease in drug crystallinity and particle size, and increased solubility and wettability in the solid dispersed systems [2, 14, 15]. The surface free energy of powders may be a useful parameter for predicting their solubility and rate of dissolution. Powdered nifedipine has been shown to have low surface free energy, typical of a poorly wettable material with low aqueous solubility and dissolution rate [16]. Numerous methods are available for preparing solid-state cyclodextrin complexes. Kneading [17–20], spray drying and lyophilization are potentially suitable for the industrial-scale production of these inclusion compounds [21].

In this work, solid-state binary mixtures of nifedipine with β-CD, γ-CD, 2HP-β-CD, RM-β-CD or DM-β-CD in 1:1 molar ratios, and γ-CD and RM-β-CD in 2:1 and 1:2 molar ratios, have been prepared using a kneading method, while nifedipine-RM-β-CD mixtures in molar ratios of 4:1, 2:1, 4:3 and 1:1 have been prepared using a heating method. The resulting properties of these mixtures have been studied by DSC, FTIR and XRD techniques.

Experimental

Materials

Nifedipine (Sigma Chemical Co., St Louis, MO, USA) was kindly donated by South African Druggists Limited (Sandton, South Africa) and was used without further purification. The cyclodextrins used are listed below, together with their mass percentage of water determined by Karl Fischer titration; degree of substitution (DS) where relevant, determined by electrospray-ionization mass spectrometry, and molar mass:

β-CD, Amaizo, USA (14.9%, 1135.0 g mol^{-1});
γ-CD, Cyclolab, Hungary (11.2%, 1297.0 g mol^{-1});
2HP-β-CD, Cyclolab, Hungary (9.0%, DS 4.81, 1414.0 g mol^{-1});
DM-β-CD, Cyclolab, Hungary (2.5%, DS 14.00, 1331.0 g mol^{-1}), and
RM-β-CD, Cyclolab, Hungary (8.5%, DS 12.36, 1308.0 g mol^{-1}).

Preparation of the nifedipine-cyclodextrin binary systems

Binary mixtures of nifedipine with β-CD, γ-CD, 2HP-β-CD, RM-β-CD or DM-β-CD in 1:1 molar ratios, and γ-CD and RM-β-CD in 2:1 and 1:2 molar ratios, were prepared using the kneading method with an ethanol:water (50:50 mass/mass%) mixture as solvent. Solvent was added slowly to form a paste. The kneaded mixtures were then dried under vacuum over phosphorous pentoxide (Merck, Darmstadt, Germany) at 30±1.0°C for 24 h and screened through a 315 μm sieve.

Nifedipine and nifedipine–RM-β-CD mixtures (molar ratios of 4:1, 2:1, 4:3 and 1:1) were placed in glass Petri-dishes, covered with aluminium foil and heated to 200°C at a heating rate of 5–10°C min^{-1} in a Labcon Type FSOH forced circulation oven (Labcon Pty. Ltd., South Africa). The nifedipine and the nifedipine–RM-β-CD melts were immediately cooled in an ice water-bath for 5 min. The resulting solids were gently ground in a mortar and screened through a 315 μm sieve.

Physical mixtures were prepared in the same molar ratios as the kneaded and heat-treated mixtures, by simple blending of the individual components with a pestle in a mortar. For comparison of the results for physical mixtures with those of the heat-treated mixtures, samples of nifedipine and RM-β-CD were heated separately to 200°C and cooled in an ice water-bath for 5 min. The resulting individual solids were gently ground in a mortar and screened through a 315 μm sieve, prior to preparing physical mixtures. All mixtures were stored away from light in sealed containers in a dessicator at 2–4°C.

Analytical methods

DSC curves were recorded on a Perkin Elmer Series 7 thermal analysis system. Samples (6–9 mg) were accurately weighed (±μg), using a Sartorius MC5 electronic microbalance, into standard 25 μL aluminium pans, which were covered with aluminium lids but not crimped, and heated from 50 to 200°C at 10°C min^{-1} under flowing nitrogen. The DSC was calibrated for temperature and enthalpy measurements in the standard way, using the melting of pure indium metal.

Infrared spectra were acquired on a Perkin Elmer Spectrum 2000 Fourier transform infrared (FTIR) spectrophotometer with the use of a diffuse reflectance accessory. All samples were prepared using spectroscopic grade potassium bromide (Merck, Darmstadt, Germany) as diluent. Each spectrum represents 32 co-added scans obtained at a spectral resolution of 4 cm^{-1} over the 450–4000 cm^{-1} range. No grinding or compaction that could influence solid–solid interactions was involved.

The X-ray powder diffraction patterns were measured using a Rigaku Denki Max III diffractometer fitted with a horizontal goniometer, graphite monochromator and scintillation detector. Ni-filtered CuK_{α} radiation was generated at a voltage of 40 kV and a current of 20 mA. A fixed-time step-scanning method was employed. Step-scans were recorded for all samples from 2 to 32° 2θ with a step size of 0.02° 2θ at a fixed time of 1 s per step.

Results and discussion

Nifedipine exists in three monotropically related forms, namely the thermodynamically stable form I which melts at 169–175°C [22–25], the metastable form II which melts at 161–164, and the metastable form III which melts at 134–137°C [24, 25]. Hirayama *et al.* [22] prepared glassy nifedipine by heating nifedipine to 200°C and immediately cooling the melt to 0°C.

The DSC curve for the nifedipine sample used in this study (Fig. 2a) shows a single endotherm with onset at 170.9°C (maximum at 173.2°C), thus indicating that the sample is the thermodynamically stable form I [23, 26, 27]. The onset temperatures (T_{onset}), maximum temperatures (T_{max}) and enthalpies of melting (ΔH) of nifedipine, the physical mixtures and the kneaded mixtures are shown in Table 1.

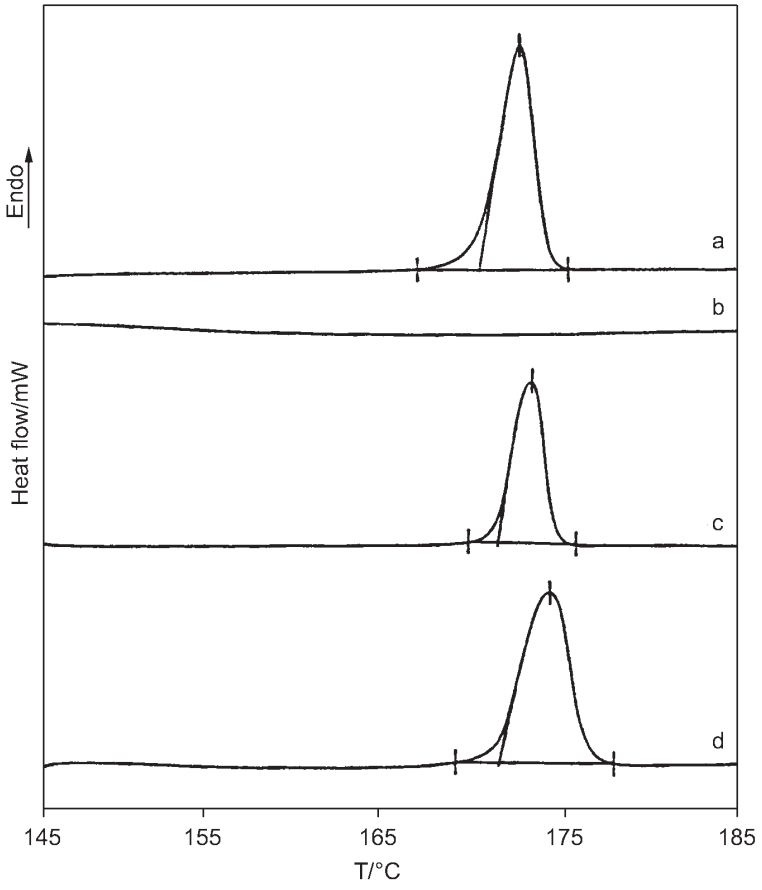

Fig. 2 DSC curves of a – nifedipine, b – β-CD, c – nifedipine–β-CD physical mixture (1:1 molar ratio) and d – nifedipine–β-CD kneaded mixture (1:1 molar ratio) (heated in nitrogen at 10°C min^{-1})

Table1 Onset temperatures, maximum temperatures and enthalpies of melting of nifedipine and nifedipine–cyclodextrin binary systems (given per g of nifedipine)

	Molar ratio	T_{onset}/°C	T_{max}/°C	ΔH/J g^{-1}
Nifedipine		170.9	173.2	107.1
Nifedipine-β-CD				
PM*	1:1	170.7	172.6	20.6
KM**	1:1	170.9	174.0	21.0
Nifedipine-γ-CD				
PM	2:1	171.5	173.9	32.7
	1:1	170.7	172.5	17.4
	1:2	171.2	172.8	10.4
KM	2:1	171.4	174.2	31.5
	1:1	170.7	173.1	18.0
	1:2	171.6	173.4	9.6
Nifedipine – 2HP-β-CD				
PM	1:1	170.7	173.1	18.0
KM	1:1	169.8	171.8	19.0
Nifedipine – RM-β-CD				
PM	2:1	157.9	164.1	31.0
	1:1	159.9	165.0	11.3
	1:2	150.7	164.8	8.0
KM	2:1	151.8	162.4	27.4
	1:1	146.6	157.9	12.2
	1:2	150.5	164.4	5.2
Nifedipine – DM-β-CD				
PM	1:1	169.6	171.7	20.9
KM	1:1	166.3	170.0	19.7

*PM: physical mixture; **KM: kneaded mixture

No thermal events were observed between 140 and 185°C for the cyclodextrins (curves b in Figs 2 to 4). They have no well-defined melting points and start to decompose at temperatures above 270°C [28, 29]. The amorphous nature of the sample of RM-β-CD used in this study explains the absence of thermal events in its DSC curve (Fig. 3b). The DSC curves of the nifedipine–β-CD physical mixture (Fig. 2c) and kneaded mixture (Fig. 2d) both show the nifedipine melting endotherm with onset at approximately 171°C. Similar curves were obtained for the nifedipine–γ-CD and nifedipine–2HP-β-CD binary systems (not illustrated), indicating that the original crystalline state of the nifedipine was not significantly altered by either of the mixing procedures.

Some broadening of the nifedipine melting endotherm is observed in the DSC curve for the nifedipine–DM-β-CD kneaded mixture (Fig. 3d) compared to the physical mixture (Fig. 3c), but the measured enthalpies of melting (Table 1) were not changed. The peak broadening could indicate that a slight decrease in nifedipine crystallinity had occurred through an interaction with the cyclodextrin.

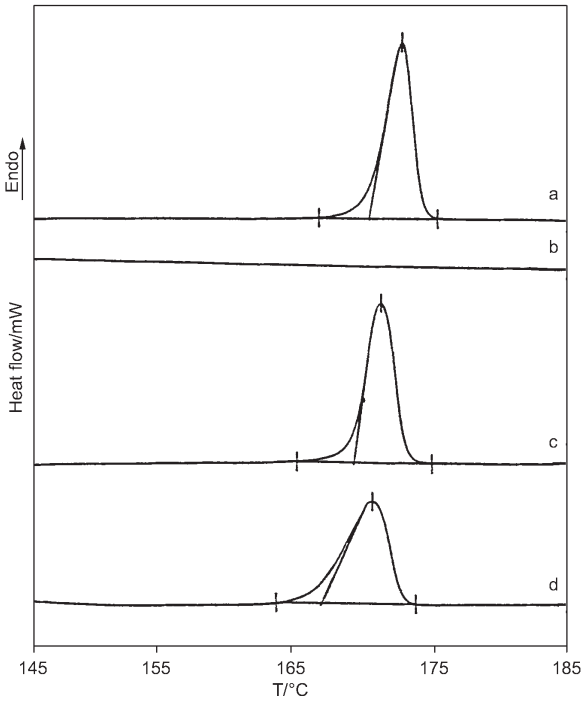

Fig. 3 DSC curves of a – nifedipine, b – DM-β-CD, c – nifedipine – DM-β-CD physical mixture (1:1 molar ratio) and d – nifedipine – DM-β-CD kneaded mixture (1:1 molar ratio) (heated in nitrogen at 10°C min^{-1})

The 1:1 nifedipine:RM-β-CD physical mixture exhibited a broad, weak endotherm which extended from 160 to 173°C with a maximum at 165°C (Fig. 4c). The corresponding kneaded mixture produced an equally broad but slightly weaker endotherm with onset at 147 and maximum at 158°C (Fig. 4d). Increasing the nifedipine content of the physical mixture (2:1 molar ratio) resulted in the appearance of a shoulder at approximately 168 on the main broad endotherm at 164°C (Fig. 5), while the kneaded mixture behaved in a similar manner to that of the 1:1 kneaded mixture. The 1:2 nifedipine:RM-β-CD physical mixture and kneaded mixture exhibited broad endotherms with onsets at approximately 151°C (Fig. 6). The lack of a distinct fusion endotherm at 171°C in the DSC curves for both the physical and the kneaded mixtures suggested that both types of mixing in this proportion had either resulted in drug inclusion in this CD or that the drug has been converted to an amorphous form by heating in the presence of RM-β-CD.

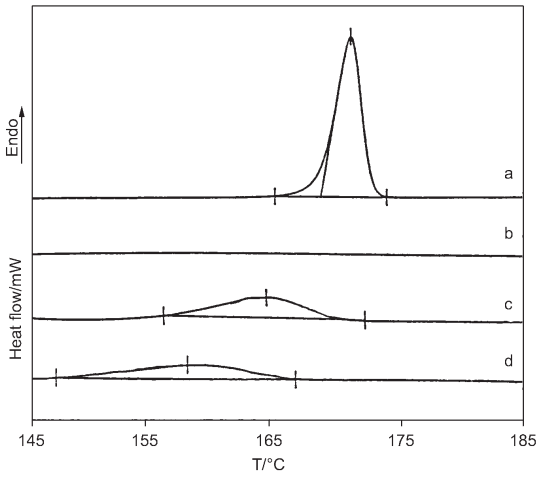

Fig. 4 DSC curves of a – nifedipine, b – RM-β-CD, c – nifedipine–RM-β-CD physical mixture (1:1 molar ratio) and d – nifedipine–RM-β-CD kneaded mixture (1:1 molar ratio) (heated in nitrogen at 10°C min^{-1})

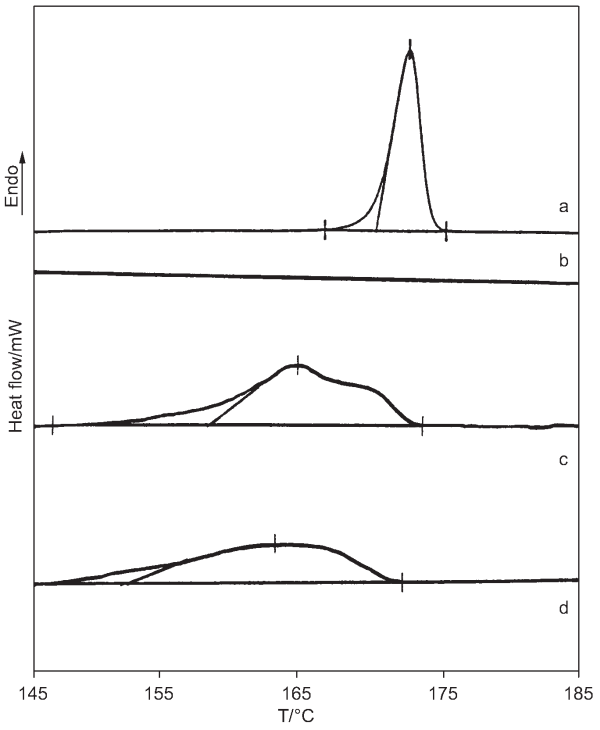

Fig. 5 DSC curves of a – nifedipine, b – RM-β-CD, c – nifedipine–RM-β-CD physical mixture (2:1 molar ratio) and d – nifedipine–RM-β-CD kneaded mixture (2:1 molar ratio) (heated in nitrogen at 10°C min^{-1})

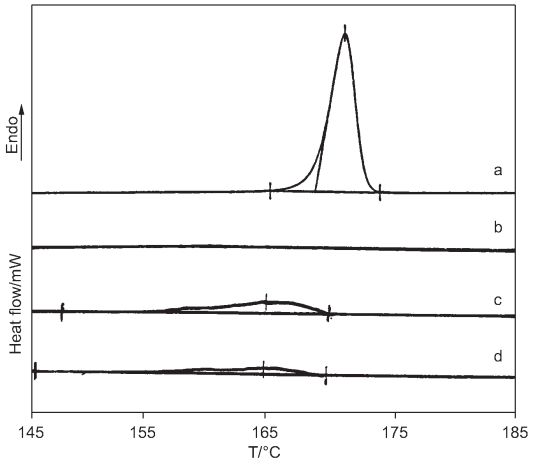

Fig. 6 DSC curves of a – nifedipine, b – RM-β-CD, c – nifedipine–RM-β-CD physical mixture (1:2 molar ratio) and d – nifedipine–RM-β-CD kneaded mixture (1:2 molar ratio) (heated in nitrogen at 10°C min^{-1})

Physical mixtures of benzoic acid with α-CD, β-CD and DM-β-CD, [30–32] clobazam with trimethyl-β-CD [33] and naproxen with RM-β-CD (DS 1.8) and hydroxyethyl-β-CD [33] produced similar DSC profiles to those in Fig. 4c and d and Bettinetti *et al*. [33] suggested that, because a physical mixture is an intimate blend of crystalline drug and cyclodextrin matrix, heating either disperses the drug molecules monomolecularly onto the surface of the cyclodextrin or results in their inclusion in the cyclodextrin cavity. Inclusion complexation has been cited as the main reason for the unusual thermal behaviour of heated physical mixtures [30, 32].

Modifying the procedure of Hirayama *et al.* [22] for preparing glassy nifedipine by heating nifedipine in the presence of 2HP-β-CD to 200°C and immediately cooling to 0°C, resulted in the appearance of an endotherm at 163°C, which was attributed to melting of the metastable form II. The extent of conversion of form I to form II increased with increasing amounts of 2HP-β-CD. Form I was almost entirely converted to the metastable form II at a molar ratio of 1:1 nifedipine:2HP-β-CD. The area of the endotherm associated with the form II remained constant at higher molar ratios [22].

Because the DSC curve for the nifedipine–RM-β-CD physical mixture (Fig. 4c) lacked the form I endotherm with onset at 171°C, the effect of heating nifedipine physical mixtures with RM-β-CD to 200°C, followed by rapid cooling, was investigated at 4:1, 2:1, 4:3 and 1:1 nifedipine:RM-β-CD molar ratios. The heated nifedipine produced a single sharp endotherm at 171.0°C, which was due to the melting of form I. The results for the heated 4:1 nifedipine:RM-β-CD mixture (Fig. 7) indicated that the original form I was absent, but a very small endotherm at 163°C appeared in the DSC curve as a result of melting of the metastable form II. At lower molar ratios of the drug, namely 2:1, 4:3 and 1:1 nifedipine:RM-β-CD, the DSC curves for the heated products were devoid of thermal events, suggesting that nifedipine existed either in an amorphous state or was included in the cyclodextrin.

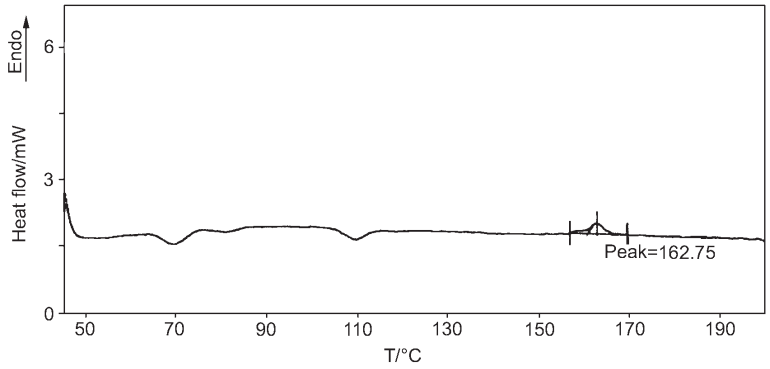

Fig. 7 DSC curve of a nifedipine–RM-β-CD kneaded mixture (4:1 molar ratio) (heated in nitrogen at 10°C min^{-1})

The principal IR absorption peaks of nifedipine at 3332 cm^{-1} (NH stretching), 1689 cm^{-1} (C=O ester), 1679 cm^{-1} (C=O ester), 1624 cm^{-1} (–C=C–aromatic), 1529 cm^{-1} (NO_2), 1380 cm^{-1} (–C–CH_3) and 1122 cm^{-1} (–C–O–ester) [8] were unchanged in the spectra of the physical mixtures and kneaded mixtures. The spectra were thus simply superimpositions of the spectra of the individual components (Fig. 8).

The spectra of the heated nifedipine–RM-β-CD mixtures (Fig. 9), unlike those for the kneaded mixtures, were found to differ slightly from the spectra of the physical mixtures. The individual components, nifedipine and RM-β-CD, were given the same heat-treatment prior to mixing as was given to the mixtures, in order to ensure that any spectral differences noted could be attributed to the presence of RM-β-CD and not simply to changes in crystallinity induced by the heat treatment. The IR spectra of crystalline nifedipine and heated nifedipine differed only in the region of the carbonyl stretching bands (Fig. 10). The carbonyl bands at 1689 and 1679 cm^{-1} were present in the heated nifedipine, but an additional small shoulder appeared at 1702 cm^{-1}. In the crystalline state, adjacent nifedipine molecules form intermolecular hydrogen bonds between the N1 hydrogen atoms of the dihydropyridine rings and the carbonyl oxygens of the carbomethoxy groups (Fig. 1) [34]. The carbonyl bands at 1689 and 1679 cm^{-1} observed in crystalline nifedipine can therefore be assigned to 'hydrogen bonded' carbonyl groups. The process of heating nifedipine to 200°C followed by rapid cooling may distort the crystal lattice to some extent, resulting in dissociation of hydrogen bonds. The absorption band attributed to the 'free' carbonyl groups was therefore shifted to a higher frequency at 1702 cm^{-1}.

The NH absorption band at 3332 cm^{-1}, due to stretching vibrations of the N–H bond in the dihydropyridine ring of nifedipine, protruded from the broad O–H stretching band (3100–3700 cm^{-1}) of RM-β-CD in the physical mixtures, but disappeared in the 4:1, 2:1, 4:3 and 1:1 molar ratio heat-treated nifedipine:RM-β-CD mixtures. Mielcarek and Sadaj [18] observed a similar change upon kneading/roll mixing nifedipine with β-CD in a 1:1 molar ratio and suggested that it was due to inclusion complexation.

The distinct carbonyl stretching bands of heated nifedipine observed in the physical mixtures at 1689 cm and 1679 cm^{-1}, shifted to higher wavenumbers in the heat-treated mixtures and, irrespective of the molar ratio, disappeared in favour of the absorption band at 1702 cm^{-1}. These shifts were accompanied by an increase in intensity of the 'free' carbonyl absorption band at 1702 cm^{-1}. The shift of the carbonyl absorption band to a higher frequency can be attributed to dissociation of intermolecular hydrogen bonds in the nifedipine crystal lattice as it converts to an amorphous state. The increased intensity of the carbonyl absorption band suggests that inclusion complexation is not the predominant mechanism by which nifedipine is converted to an amorphous state, because the vibrational motions of an included drug moiety are

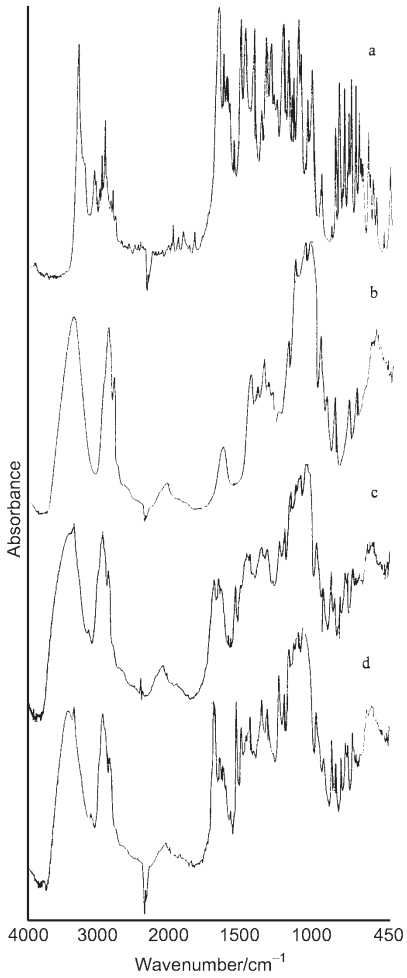

Fig. 8 Infrared spectra of a – nifedipine, b – RM-β-CD, c – nifedipine–RM-β-CD physical mixture (1:1 molar ratio) and d – nifedipine–RM-β-CD kneaded mixture (1:1 molar ratio)

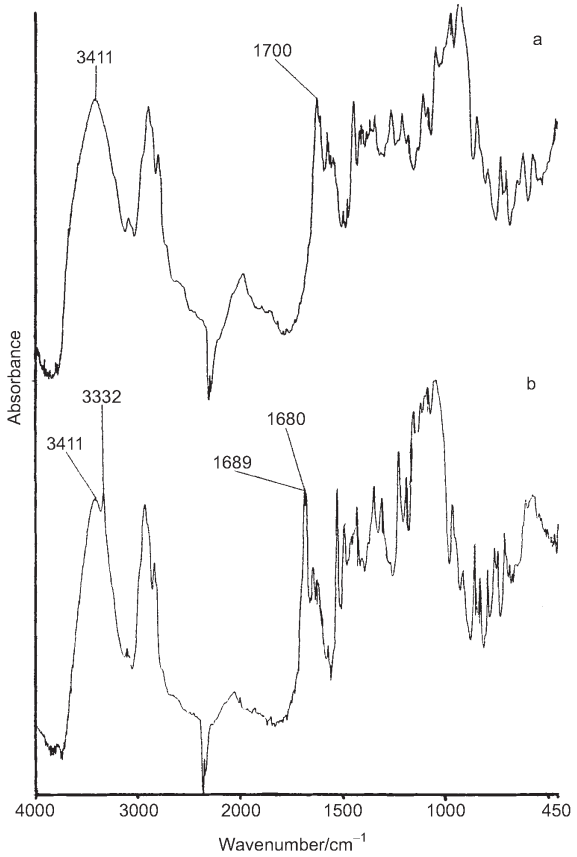

Fig. 9 Infrared spectra of a – heated nifedipine–RM-β-CD mixture (1:1 molar ratio) and b – nifedipine – RM-β-CD kneaded mixture (1:1 molar ratio)

usually restricted in the cyclodextrin cavity resulting in a decreased absorption band intensity [35]. A minor absorption band appeared at 1281 cm^{-1} in the spectra of the heated nifedipine–RM-β-CD mixtures with its intensity and frequency unaffected by changes in molar ratio.

Because the DSC determinations showed that the nifedipine crystallinity was largely unaltered in the nifedipine–cyclodextrin kneaded mixtures, further analysis of these binary systems by powder X-ray diffraction was not performed. The X-ray diffraction patterns of the 2:1 molar ratio heated nifedipine:RM-β-CD mixture and of the physical mixture were recorded and compared in order to determine any changes in the crystalline state of nifedipine that may have occurred in the presence of the cyclodextrin. The X-ray diffraction patterns of crystalline nifedipine and heated nifedipine are shown in Fig. 11. Nifedipine exhibited characteristic diffraction peaks at 2θ=8.12, 11.81, 16.25, 19.63 and 24.47°, attributable to the crystal planes of Miller indices 100, 002, 200, 211 and 300 or 221, respectively [29]. The diffraction pattern

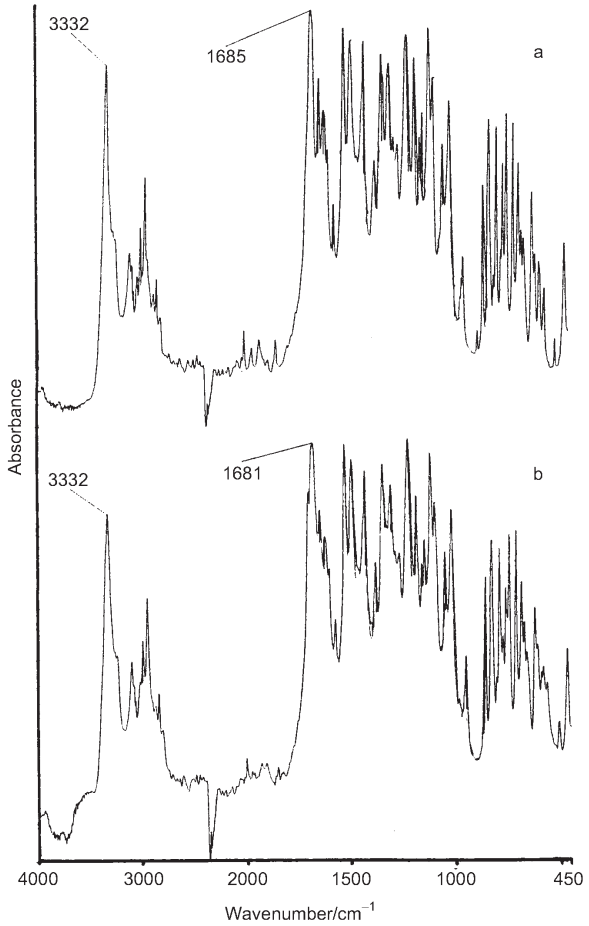

Fig. 10 Infrared spectra of a – nifedipine and b – heated nifedipine

of the heated nifedipine was different from crystalline nifedipine in both reflection angle (2θ) and peak intensity. The characteristic peaks at 2θ=8.12, 11.81, 16.25, and 24.47° disappeared or were greatly reduced in the diffractogram of the heated nifedipine, while minor peaks appeared at 2θ=7.45, 10.72, 12.36, 16.91 and 24.21°. The diffraction pattern of the metastable nifedipine polymorph, form B, is distinguishable from crystalline nifedipine by the presence of peaks at 2θ= ~7.1° and ~23.9° [29]. Hirayama *et al.* [22] suggested that the diffraction peak at 2θ= ~7.1° may be attributed to the reflection from the 100 crystal plane, indicating that the metastable form II has a slightly longer a-axis, thereby implying that the hydrogen-bonded nifedipine molecular layers in the bc plane are stacked along the a-axis in a loose and less-structured manner than in crystalline nifedipine. The appearance of the diffraction peak at 2θ= ~7.45° and the reduction in peak intensities observed in

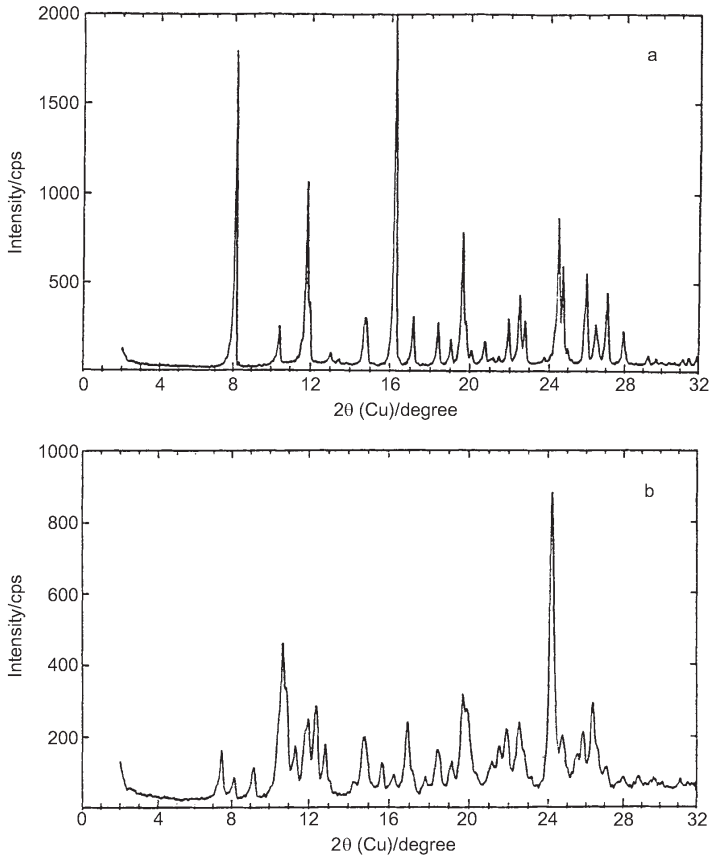

Fig. 11 X-ray powder diffractograms of a – crystalline nifedipine and b – heated nifedipine

the X-ray diffractogram of heated nifedipine, suggest that the heating and cooling of nifedipine produces a less-structured crystalline or partially amorphous state, from which the presence of the metastable form II polymorph cannot be entirely excluded. These findings are in agreement with the appearance of a 'free' carbonyl absorption band in the IR spectrum of heated nifedipine, which indicated that dissociation of intermolecular hydrogen bonds had occurred due to distortion or disruption of the crystalline lattice.

The X-ray diffractogram of RM-β-CD predictably showed no sharp peaks and produced a halo-pattern typical of an amorphous compound. The diffraction pattern did not change when RM-β-CD was heated to 200°C and cooled. The X-ray diffraction pattern of the 2:1 molar ratio nifedipine:RM-β-CD physical mixture was a superimposition of the separately heated individual components (Fig. 12a). The sharp diffraction peaks could be attributed to heated nifedipine. No sharp peaks due to nifedipine or heated nifedipine were observed in the X-ray diffractogram of the re-

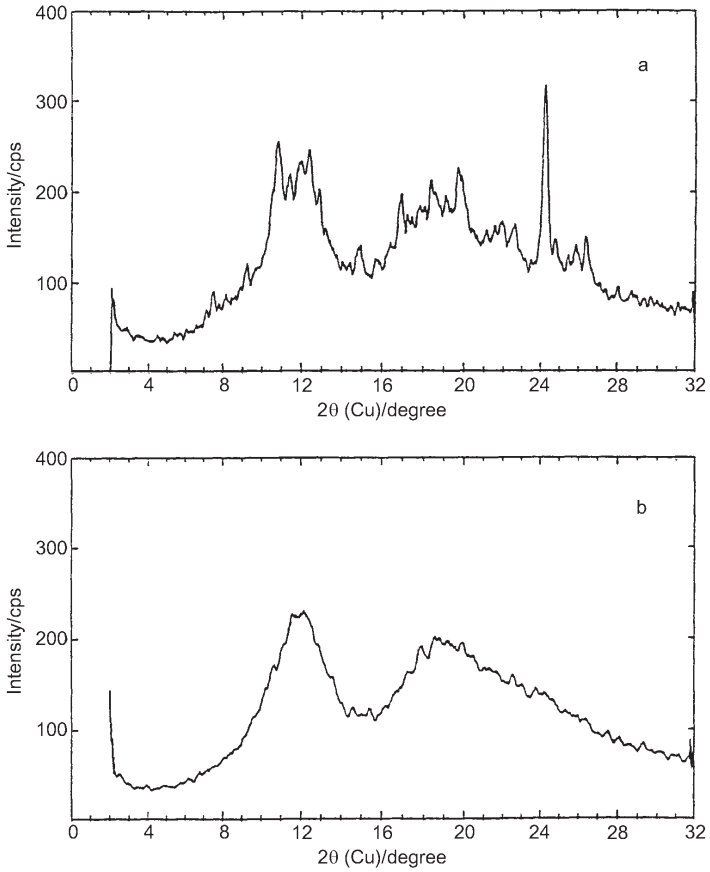

Fig. 12 X-ray powder diffractograms of nifedipine of the nifedipine–RM-β-CD a – physical mixture and b) heated mixture prepared in a 2:1 molar ratio

spective heated mixture (Fig. 12b), suggesting that nifedipine crystals were transformed into an amorphous state or were included within the cyclodextrin as a result of the heating and subsequent rapid cooling.

Conclusions

In this study, attempts were made to prepare solid-state nifedipine–cyclodextrin inclusion complexes utilizing processes that could be easily and cost-effectively implemented in an industrial environment.

The properties of kneaded binary mixtures of nifedipine with β-CD, γ-CD, 2HP-β-CD or DM-β-CD strongly resembled those of the physical mixtures. DSC and IR spectroscopy suggested that the kneading process was not successful in producing any significant amounts of inclusion complex for these mixtures.

The thermal behaviour of both the physical and kneaded mixtures of nifedipine–RM-β-CD was notably different from the behaviour of the mixtures with the other cyclodextrins. Absence of the nifedipine melting endotherm in the DSC curves of both the physical mixture and the kneaded mixture suggested that crystalline nifedipine was converted into an amorphous or included state by simply heating in the presence of RM-β-CD. Physical mixtures of nifedipine–RM-β-CD were prepared over a range of molar ratios and were then heated to 200°C followed by rapid cooling. DSC, IR spectroscopy and powder X-ray diffraction studies of these heat-treated mixtures showed that nifedipine was in an amorphous state or included within the cyclodextrin at a nifedipine–RM-β-CD molar ratio of 4:1.

The high drug ratio at which the changes in state of the nifedipine occurs (4:1 nifedipine:RM-β-CD) in the heated nifedipine–RM-β-CD mixture, suggests that inclusion complexation is not the dominant mechanism, but rather monomolecular dispersion of nifedipine within the cyclodextrin matrix. Some formation of an inclusion complex under these conditions is not ruled out.

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