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A study on the differentiation between amorphous piroxicam:β-cyclodextrin complex and a mixture of the two amorphous components

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Abstract

Amorphous piroxicam was prepared by the melt spinning method to prove that freeze-dried amorphous piroxicam:β-cyclodextrin is a true inclusion compound and not a dispersed mixture of the two amorphous components. Differential scanning calorimetry (DSC) and near-infrared Fourier transform Raman spectroscopy (NIR FT-Raman) established the success of the encapsulation. Thermal analysis can also be used to evaluate the inclusion complex purity with regard to crystalline and/or amorphous free piroxicam content.

Keywords: Piroxicam; β-cyclodextrin; Inclusion complexes; Differential scanning calorimetry; NIR FT Raman; X-ray diffractometry; Glassy state; Melt spinning

Inclusion complexes between drugs and cyclodextrins (CD) are a topic of current interest to the pharmaceutical industry as they may improve the solubility, stability and bioavailability of the guest molecules (Frömming and Szejtli, 1994). Crystalline compounds can be identified by various physical methods since the guest inclusion gives rise to a new crystal lattice. So, different analytical techniques for inclusion confirmation have been reported such as single-crystal or powder X-ray diffraction (XRD), neutron diffraction, solid state ^{13}C NMR, IR and reflectance spectroscopies, thermal analysis and electrical conduction measurements (Hirayama and Uekama, 1987). In particular, single-crystal X-ray and neutron diffraction structure analyses allowed establish that CD form a channel- or cage-like clathrate surrounding the guest molecules which, in turn,

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are located in the intramolecular and intermolecular cavities or in some intermediate situation. The solvent molecules are a fundamental part of the crystal construction (Le Bas and Rysanek, 1987; Tsoucaris et al., 1990).

Unfortunately, the most applied manufacturing processes of inclusion compounds for industrial scale such as freeze-drying (Kurozumi et al., 1975) or spray-drying (Tokomura et al., 1985), usually yield amorphous products. In these cases, most of the spectroscopic methods and powder XRD are not able to discriminate whether the product obtained is a true inclusion complex or a homogeneous dispersed mixture of the two amorphous components as they give either very broad signals and a diffused diffraction pattern.

The present study aims at differentiating between the freeze-dried amorphous piroxicam:β-cyclodextrin (P:βCD) inclusion compound (Acerbi et al., 1990; Fronza et al., 1992; Selva et al., 1993). and a homogeneous physical mixture of the two amorphous components by differential scanning calorimetry (DSC) and near-infrared Fourier transform Raman spectroscopy (NIR FT-Raman).

Powder XRD patterns were recorded on a Philips PW 1050 diffractometer using Cu-Kα radiation over the interval 5–60°/2θ (speed: 1°/min). The DSC analyses were run out on a Pekin-Elmer DSC 7 Thermal Analysis System under a nitrogen atmosphere. Each sample was heated between 50° and 220°C with a scanning rate of 10°C/min. The thermal cycles were carried out by heating the samples until 125 or 150°C at 10°C/min, then cooling to 50°C at 2°C/min and reathing at 10°C/min. Raman spectra were recorded on a Perkin-Elmer 2000 FT-spectrometer equipped with an InGaAs detector and a diode pumped Nd:YAG laser operating at 1.064 μm.

The application of conventional amorphization techniques, such as liophylization, to piroxicam did not yield the expected results: only a partially amorphous product was obtained in all cases, indicating the high tendency of piroxicam to crystallize.

A complete amorphization was successfully obtained only by rapidly cooling after melting (Ford and Timmins, 1989; Fukuoka et al., 1986). Crystalline piroxicam was put in a fan-coil oven at 205–210°C. After complete melting it was held at this temperature for 5 min then cooled by means of a melt spinning apparatus (Capelletti et al., 1994) which allows an ultra-fast cooling rate (3–4 million degrees/min). The resulting product, in the form of brittle glassy flakes, exhibits the diffused diffraction pattern typical of an amorphous state (Fig. 1a). Nevertheless, the crystallization kinetics are very fast and almost completed at room temperature within few hours from the amorphous preparation (Fig. 1b). The crystallization process

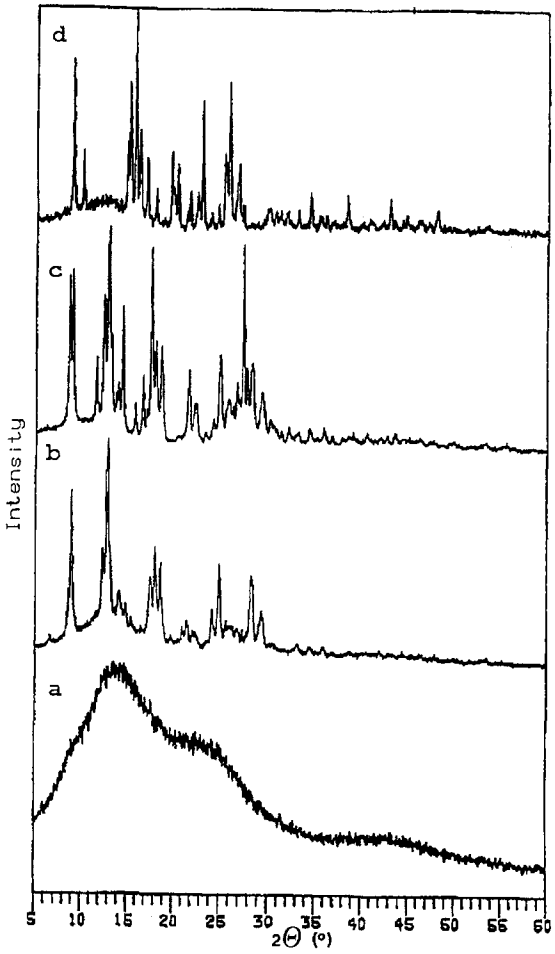

Fig. 1. XRD patterns of: (a) amorphous piroxicam (as obtained just after melt spinning); (b) melt-spinned piroxicam after 6 h at room temperature; (c) melt-spinned piroxicam after 6 days at room temperature and (d) crystalline piroxicam (starting material).

then evolves with time (Fig. 1c), giving rise to crystalline forms different from that of crystalline piroxicam used as the starting material (Fig. 1d). Therefore, it was necessary to prevent devitrification by keeping glassy piroxicam at a low temperature (− 80°C). The thermal trace of amorphous piroxicam shows a broad exotherm at 90–110°C corresponding to recrystallization followed by the endothermal melting peak at 190–200°C characteristic of crystalline piroxicam. Upon cooling and reheating, only the endothermal peak was observed, indicating that crystallization is an irreversible process. As far as amorphous βCD is concerned, it was obtained by simple freeze-drying. Like glassy piroxicam, the powder XRD shows only a diffused pattern. Its DSC curve evidences a broad endotherm between 50° and 150°C corresponding to the water loss. Melting with decomposition starts above 250°C according to the literature (Sztatisz et al., 1980). DSC is an useful technique for drugs that form inclusion complexes with CDs, being evidenced by the loss of the drug melting endotherm (Otagiri et al., 1983; Uekama et al., 1983; Chow and Karara, 1986; El-Gendy et al., 1986).

On the other hand, for freeze-dried products, the absence of the DSC signal, can also be expected because of their amorphous character (Funk et al., 1994). The thermogram of amorphous P:βCD in the scanning temperature range, evidences the endotherm due to βCD dehydration and the loss of the endothermic melting peak of crystalline piroxicam. On the contrary, in the thermogram of the mixture of the amorphous components, the peak is still present as free amorphous piroxicam recrystallizes before melting during the scanning time. This indicates that, in the freeze-dried complex, piroxicam is dispersed in the amorphous host clathrate and its crystallization is hampered. In the physical mixture, the crystallization of glassy piroxicam, occurring at 90–110°C, is hidden by the broad endotherm of βCD water, therefore the samples were also submitted to the cooling/reheating cycle. As expected, the drug melting peak was observed only in the DSC curve of the physical mixture of the amorphous components, confirming the previous findings. Typical DSC curves are displayed in Fig. 2a and Fig. 2b.

As thermal analysis provides only negative evidence, spectroscopic methods were also applied for characterization. NIR FT-Raman (Hendra et al., 1991) has two important advantages over common IR spectroscopy: (i) bands are almost invariably sharp, very little overlap occurs and the spectra are far more detailed; (ii) no sample preparation is needed. The samples can simply be packed into a cup for the spectra to be quickly recorded. This is a remarkable feature because amorphous piroxicam rapidly converts to its crystalline form as reported above. The IR spectra of the two piroxicam forms, after preparing KBr pellets, are indeed superimposable, whereas their Raman spectra are significantly different. Furthermore, in the IR spectrum of P:βCD complex, the bands of the guest are almost completely obscured by the very intense and broad βCD bands, which are hardly influenced by complex formation.

Fig. 3 shows part of the Raman spectra (1650–1000 cm^{-1} range) of amorphous freeze-dried P:βCD inclusion compound and the homogeneous physical mixture of the two amorphous components. The hydroxyl groups of βCD give rise to much weaker bands, expecially in the diagnostic region of the amide stretching vibration. The absorption bands of piroxicam at 1614 and 1521 cm^{-1} experience a dramatic broadening in the spectrum of the inclusion compound; the former is also shifted toward lower frequencies. This change is probably related to the formation of intermolecular hydrogen bonding between the guest and the host (Uekama et al., 1983). Other differences were observed in the two spectra (in particular, a new peak at 1440 cm^{-1} appears in the inclusion compound spectrum), but no interpretation was attempted.

In conclusion, DSC and FT-Raman are convenient and diagnostic tools for proving that freeze-dried amorphous P:βCD is a true inclusion compound and not a homogeneous dispersed mixture of the two amorphous components. In particular, the thermogram of P:βCD evidences the loss of the endothermal melting peak of crystalline piroxicam which is still present in the thermogram of the physical mixture. For this reason, DSC also allows evaluation of the inclusion complex purity with regard to the free piroxicam content despite

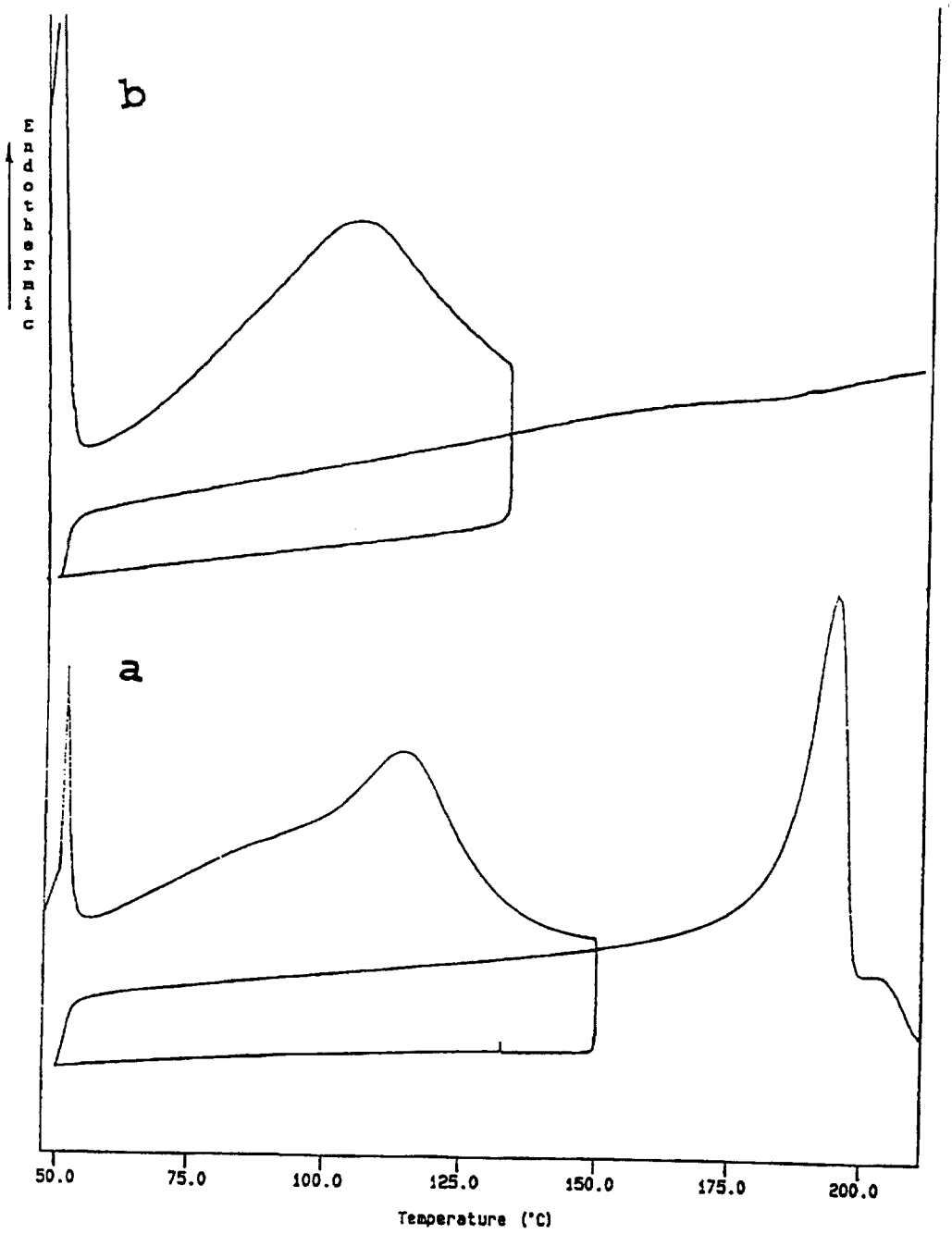

Fig. 2. DSC curve after cooling/reheating thermal cycle of: (a) physical mixture of the two amorphous components and (b) freeze-dried P:β-CD.

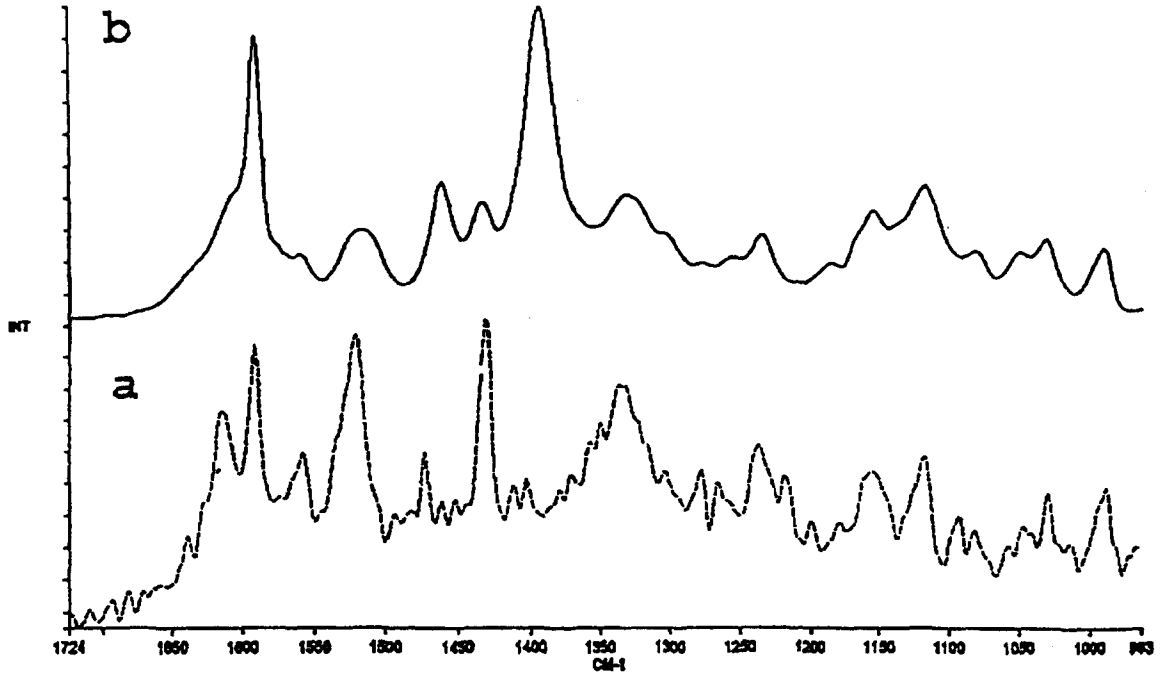

Fig. 3. Diagnostic region of NIR FT-Raman spectra of: (a) physical mixture of the two amorphous components and (b) freeze-dried P:β-CD (laser power: 30 mW/mm^2).

its amorphous or crystalline state (limit test $\leq 0.5\%$).

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