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Improving furosemide polymorphs properties through supramolecular complexes of β-cyclodextrin

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ABSTRACT

In this work, complexes of β-cyclodextrin and the two solid forms of furosemide were prepared and characterized for their potential pharmaceutical applications, with the interactions between the two compounds being studied in the solution and solid states. The solubility studies revealed different behaviors of the polymorphs. In particular, it was observed that the binary complex significantly increased the solubility of furosemide form I in the gastric simulated fluid, which resulted in a rise in the bioavailability of this formulation after oral administration. In addition, results using ssNMR, FT-IR, DSC, TGA, SEM and XRPD provided evidence of the formation of complexes after utilizing kneading and freeze-drying methods. A comparison with previous developed complexes that used maltodextrin as the ligand was performed. Our results suggest that these novel supramolecular complexes showed promise to be used in drug delivery systems with an application in pharmaceutical formulations.

1. Introduction

It is well known that pharmaceutical solids can exist in various solid-state forms which have different physicochemical properties that affect their performance. In particular, polymorphic changes in the active pharmaceutical ingredient (API), may lead to significant effects on the bioavailability of the final product after oral administration [1].

Pharmaceutical complexes in solid state are usually developed in order to improve the profile of a single organic molecule, in terms of solubility, stability, bioavailability and organoleptic properties [2–4]. For example, supramolecular complexation is a commonly used technique to increase the solubility of poorly water-soluble drugs. Among the macromolecules utilized to solubilize drugs, the cyclodextrins (CDs) are the most widely used as they are an effective alternative. CDs are able to form inclusion complexes with many different types of appropriately sized and preferential non-polar molecules, both in solution and solid state [5–7].

Furosemide (FUR, Fig. 1) is widely applied as a strong loop diuretic in the treatment of edematous states associated with cardiac, renal, and hepatic failure, and also in the treatment of hypertension [8]. It is known to exist in seven polymorphic forms: four true polymorphs (I, II, III, IV), two solvates (IV – DMS and V – dioxane) and one amorphous form [9–11]. Since FUR has a low water solubility and low permeability, it belongs to Class IV in the Biopharmaceutics Classification System [12]. The relatively poor and variable oral absorption of FUR (60–70% [8]), which occurs site-specifically in the stomach and upper small intestine [13], has been ascribed to the poor dissolution of FUR at low pH as well as to the involvement of intestinal efflux proteins [14].

In previous reports, several approaches, including CDs, have been developed to increase the solubility and/or the dissolution rate of FUR [15–17]. However, to date, the effect of excipients on the performance of different solid-state forms has not been widely studied. A recent investigation of ours focused on supramolecular complexes of different polymorphs of FUR and maltodextrin [18], which certainly showed better solubility properties than their precursors.

Based on these above considerations, the aim of the present investigation was to prepare and characterize new supramolecular systems of FUR polymorphs I and II with β-cyclodextrin (βCD, Fig. 1). The objective of producing these complexes was to enhance the low solubility of FUR, which represents a limiting factor that is responsible for its poor and highly variable human bioavailability, and also to compare these complexes with the supramolecular ones with maltodextrin. Complexation was studied using solubility analysis, solid-state Nuclear Magnetic

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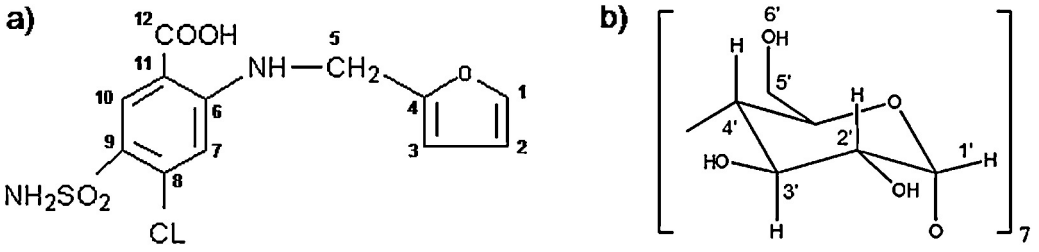

Fig. 1. Chemical structure of (a) FUR and (b) βCD, showing the carbon and proton numbering used in the NMR spectra.

Resonance (ssNMR), Fourier-transform Infrared Spectroscopy (FT-IR), Differential Scanning Calorimetry (DSC), Thermogravimetric Analysis (TGA), Scanning Electron Microscopy (SEM), and X-ray Powder Diffraction (XRPD). The ^{1}H spin lattice relaxation time measurements were also carried out in solid state.

2. Materials and methods

2.1. Chemicals and reagents

Furosemide was provided by Parafarm (Argentina) and β-cyclodextrin (MW = 1135) was kindly supplied by the Ferromet agent of Roquette (France). All other chemicals were of analytical grade. A Millipore Milli Q Water Purification System (Millipore, Bedford, MA, USA) generated the water used in these studies.

2.2. Obtaining the polymorphic forms of furosemide

The two solid forms of FUR, I and II, were prepared as in our previous report [18]. Form I was recrystallised from a methanol solution and Form II was obtained from an acetone solution.

2.3. Solid sample preparations

The solid-state systems of FUR polymorphs I and II at an equimolar ratio with βCD were prepared as follows.

2.3.1. Kneading method (KN)

The FUR I:βCD (KN [kneading method] I) and FUR II:βCD (KN II) systems were prepared by accurately weighing appropriate amounts of βCD and then transferring these to a mortar. An ethanol–water (50:50, v/v) mixture was added to the βCD powder, and the resultant slurry was kneaded for about 10 min. For each system, the corresponding solid form of FUR was added at small amounts with the simultaneous addition of solvent in order to maintain a suitable consistency. This slurry was kneaded thoroughly for about 30 min, and the resultant paste was dried in a vacuum at 40 °C for 48 h and protected from light.

2.3.2. Freeze-drying method (FD)

To prepare the FUR I:βCD (FD [freeze-drying] I) and FUR II:βCD (FD II) systems, appropriate amounts of FUR (forms I or II) and βCD were suspended in water and sonicated at 25.0 ± 0.1 °C (constant water temperature) until the drug was completely dissolved. Solutions were then frozen at −40 °C for 24 h to ensure a complete solidification before the freeze-drying was started (Freeze Dry 4.5 Labconco Corp., Kansas City, MI).

2.3.3. Physical mixture (PM)

Physical binary mixtures of FUR I:βCD (PM I) and FUR II:βCD (PM II) were prepared by simply blending uniformly the corresponding components with a mortar and pestle.

2.4. Solubility studies

The effects of βCD on the solubility of solid forms of FUR were studied at 25.0 ± 0.1 °C in aqueous and buffered aqueous solutions of pH 2.0 and 6.5, with the solubility measurements being obtained according to the method of Higuchi and Connors [19]. An excess of the FUR forms I or II were added to solutions containing increasing concentrations of βCD ranging from 2.6 to 15 mM. FUR, in the absence of βCD, was used to determine the intrinsic solubility. The suspensions were sonicated for 15 min (ULTRASONIC LC 30 H Elma), before being placed for 72 h in a constant temperature water bath [Haake DC10 thermostat (Haake, Paramus, NJ, USA)]. These suspensions were then sonicated at several time intervals, and after the equilibrium was reached, the remaining solid FUR was removed by filtration through a 0.45 μm membrane filter (Millipore, USA). The clear solutions were suitably diluted and analyzed by UV–vis spectrophotometry (SHIMADZU UV-160A spectrophotometer) at λ = 277 nm.

2.5. Solid state NMR (ssNMR)

High resolution solid state ^{13}C spectra of FUR I and II, βCD and the PM I and II, KN I and II, FD I and II systems were recorded with the ramp cross polarization/magic angle spinning (CP-MAS) sequence, with proton decoupling during acquisition [20]. All ssNMR experiments were performed at room temperature in a Bruker Avance II spectrometer operating at 300.13 MHz for protons which was equipped with a 4 mm MAS probe. The operating frequency for carbons was 75.46 MHz. Glycine was used as an external reference for the ^{13}C spectra and for setting the Hartmann–Hahn matching condition in the cross-polarization experiments. Spectra were recorded with 2000 scans, with the contact time during CP being 2 ms and the recycling times being 5 s in all cases. The spinning rate for all the samples was 10 kHz.

^{1}H spin-lattice relaxation times in the laboratory frame (^{1}H T_1) were measured in static conditions with saturation recovery pulse sequence. In this experiment, the initial ^{1}H magnetization was saturated by a train of 40 pulses $\pi/2$ during a period of 160 μs and then allowed to recover along the z-axis during a time between 10 μs and 300 s. The recycling delay in these experiments was 5 s.

2.6. FT-IR spectroscopy

The FT-IR spectra were recorded on a Nicolet 5 SXC FT-IR Spectrophotometer (Madison, WI, USA), with the potassium bromide disks being prepared by compressing the powder.

2.7. Thermal analysis (DSC and TGA)

The DSC curves of the samples were obtained with a DSC TA 2920, and the TGA curves were recorded on a TG TA 2920. The samples were placed in aluminum hermetic pans, and the experiments were carried out under a nitrogen gas flow, at a heating rate of 10 °C/min, and over a temperature range of 25–350 °C.