CONTROLLING POLYMORPHISM WITH CO_2 BASED PROCESSES: APPLICATION TO AN ANTI-HYPERTHENSIVE DRUG

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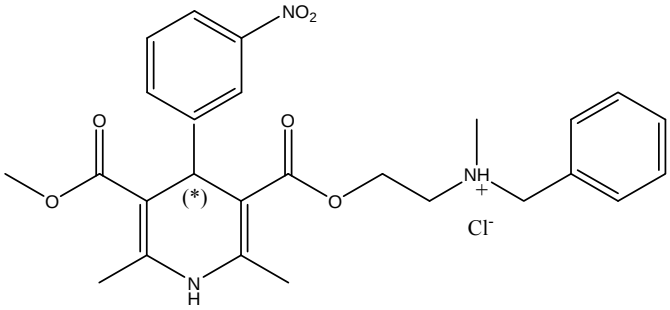
INTRODUCTION

Polymorphism, as defined by McCrone is "a solid crystalline phase of a given compound resulting from the possibility of at least two crystalline arrangements of that compound in the solid state". The phenomenon of polymorphism has been discussed in detail in the literature and an analysis of all the entries in the Cambridge Structural Database reveals that a large number of molecules are found to exhibit polymorphism. The occurrence of polymorphic modifications in molecular compounds is manifested not just as a consequence of minimum free energy of the crystalline phases but also by kinetics of crystal nucleation and growth. [1] Crystallization techniques using compressed fluids are pressure driven which is transmitted faster through space and time in solution than temperature and composition changes. For this reason, they're regarded as kinetically driven crystallization techniques. [2] These CO_2 based procedures have been shown to be suitable for pure polymorph production and polymorph selection just by changing pressure, temperature or compressed fluid flow rates. In literature are described several examples of polymorph selection using compressed fluids. However, almost all of them were carried out over organic molecules with medium molecular weights that show good crystallization behaviours. [3, 7] In this work, we have focused our study over a high molecular weight, flexible and highly conjugated molecule which belongs to the 1, 4-dihidropyridine's family: Nicardipine Hydrochloride (**1**) (M_W =516 g/mol)



(**1**)

This drug possesses multiple applications as for the treatment of cardiovascular and cerebrovascular disorders, and the treatment of oesophageal cancer in animals. However, it shows poor solubility in corporal fluids as well as it's degraded almost totally (80%) in the liver. [8] For this reason, production of new polymorphs, particle size reductions and new formulations are desirable for overcoming these inconvenients.

Although the existence of some polymorphic phases of this compound has been already reported in literature, its polymorphic behaviour has not been completely explored. Up to now, there are described only two crystalline phases of nicardipine hydrochloride: α (m.p =189 ºC), β (m.p =169ºC); which have been obtained using conventional cooling methods from organic solvents. [9]
In this work, we have used the DELOS crystallization technique as a tool for observing the effect of cooling rate over the crystallinity and polymorphic nature of solids obtained. [10]

MATERIALS AND METHODS

Nicardipine Hydrochloride (**1**) (purity 98%) was purchase from Sigma-Aldrich (Barcelona, Spain) as β fase (m.p. 169ºC), chloroform, methanol, ethanol, nitromethane and acetone from Romil Chemicals (Cambridge, UK) and used without further purification. Carbon Dioxide was kindly supplied by Carburos Metálicos-Air Products (Barcelona, Spain).
The crystallization of Nicardipine Hydrochloride through the DELOS process was performed using chloroform as conventional solvent and CO_2 as compressed fluid. The crystallization experiments have been performed using the same equipment described in previous works, [11] and the operational procedure and experimental conditions used were as follows. A volume of a solution of **1** in chloroform with a known value of supersaturation β_I, was charged into a high-pressure vessel, which was previously thermostatitzed at the working temperature, T_W =308 K. This temperature was kept constant during all the crystallization procedure. The initial solution was then pressurized up to a given pressure (P_W), by the addition of a given amount of CO_2, X_W. After leaving the system under the same conditions for 30-60 minutes, in order to achieve a complete homogenization and its thermal equilibration, the solution was depressurized over a non-return valve from P_W to atmospheric pressure. As the solution depressurization starts, the temperature monitored after the depressurization valve decreases suddenly until reaching a constant value. This temperature value is taken as T_F. The precipitate produced through a DELOS process, during the depressurization, was collected at atmospheric pressure on a filter placed after the depressurization valve. After the filtration, the cleaning of the precipitate was carried out with pure CO_2 at 3MPa and 293 K during 20 minutes.

RESULTS AND DISCUSSION

Nicardipine hydrochloride has been precipitated from CO_2 expanded chloroform by the DELOS process with a crystallization yield of 35% when initial supersaturation was β_I =0.55. The solid collected as well as the raw solid powder of **1**, were characterized by means of Scanning Electron Microscopy (SEM), Differential Scanning Calorimetry (DSC) and X- Ray Powder Diffraction (XRPD). As can be ascertained from the SEM images of Figure 1, DELOS crystallized powders of nicardipine hydrochloride are constituted by irregular plates near one order of magnitude lower in size respect to raw powders of **1**. In addition, processed and raw solid powders of **1** show different DSC profiles (Figure 2) and different XRDP patterns (Figure 3), indicating that solid phase of **1** obtained by DELOS process has a different crystalline structure than the starting solids of **1**, which corresponds to the already known β phase. For further comparison, crystals of α phase were obtained as described in literature [9] and its DSC and XRPD pattern was also determined and included in Figure 3 and Figure 4. As can be ascertained, the DSC and XRPD patterns of α phase and the crystallized solids by DELOS process are also different; suggesting that the solid obtained from chloroform/CO_2 is another crystal phase. As far as we know, this is the first time that this phase has been obtained and described. The melting point of this new phase is 120ºC; near 50ºC lower than the β phase, and 70ºC than α phase. This melting temperature decrease indicates the presence of weaker molecular interactions in the new phase, named as γ, than in α and β polymorphic forms and, also that this phase is metastable respect to α and β, at the absolute zero. Work is currently done in order to determine the crystal structure of this new phase. In other hand, this phase has been tried to obtain by fast cooling of chloroform solutions at the same experimental conditions used in DELOS crystallization experiments, and no precipitation of **1** was observed. The same experiments were carried out using methanol,

ethanol, acetone and nitromethane obtaining the same result. However, if the cooling rate is lower, β phase can be obtained from solutions of acetone and methanol with $\beta_I = 0.55$ whereas no precipitation occurs when the solvent used is chloroform.

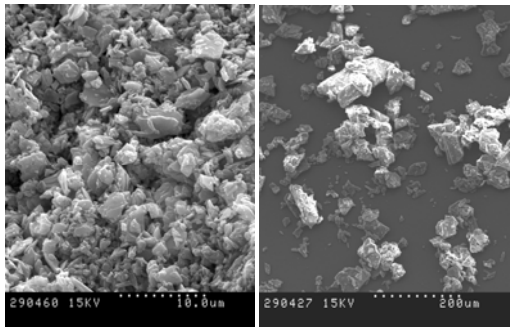


Figure 1: SEM images of Nicardipine hydrochloride: (right) unprocessed β phase; (left) crystallized by DELOS from "chloroform/CO_2" mixtures.

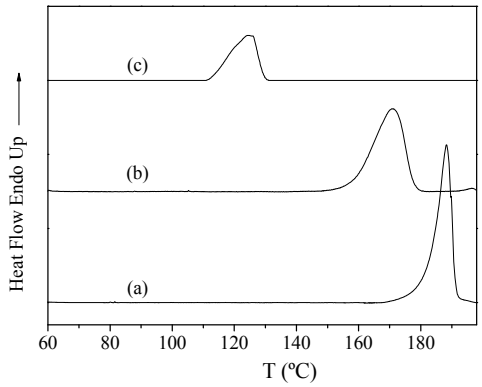


Figure 2: DSC profiles of Nicardipine: (a) α phase, **(**b) β phase (raw Nicardipine powder) and (c) crystallized by DELOS from "chloroform/CO_2".

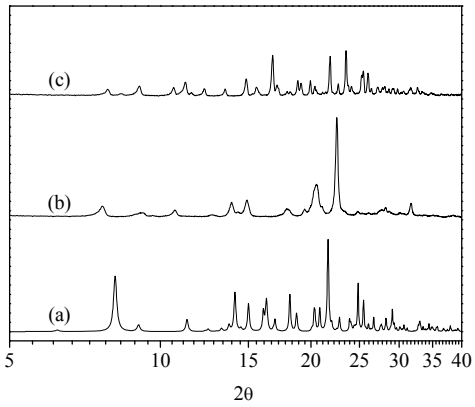


Figure 3: PXRD patterns of Nicardipine (a) α phase, (b) β phase, and (c) crystallized by DELOS from "chloroform/CO_2".

CONCLUSIONS

In the present work, we have obtained a new phase of nicardipine hydrochloride from chloroform/CO_2 applying the fast cooling rates intrinsic of the DELOS process. We also have checked that this phase is difficult to be obtained, from liquid chloroform, by conventional batch cooling precipitation, either when using high or low cooling rates.

ACKNOWLEDGMENT

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