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Crystallisation of partially amorphous griseofulvin in water vapour: determination of kinetic parameters using isothermal heat conduction microcalorimetry

Humera Ahmed ^a, Graham Buckton ^{a,*}, David A. Rawlins ^b

^a Centre for Materials Science, School of Pharmacy, University of London, 29–39 Brunswick Square, London, WC1N 1AX, UK

^b MSD Laboratories, Hertford Road, Hoddesdon, Hertfordshire, UK

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Abstract

Griseofulvin was made partially amorphous by storing at its melting point and then rapidly cooling. The partially amorphous sample was seen to have a different differential scanning calorimetry profile to the starting crystalline material. The powder X-ray diffraction pattern of the starting and quenched materials were very similar, however, some evidence of an amorphous halo was seen for the quenched sample. Exposure to 65, 75 or 85% relative humidity (RH) in an isothermal microcalorimeter yielded an exotherm which was shown to be due to crystallisation of the amorphous content of the drug. The exotherm was modelled as a solid state reaction process to allow calculation of the rate constant, we believe this is the first time that such an experiment has been modelled to allow calculation of a rate constant. © 1998 Elsevier Science B.V. All rights reserved.

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1. Introduction

The physico-chemical properties of materials may change if the physical form is altered. The impact of changes in physical form may be altered dissolution rates, apparent solubility (Buckton and Beezer, 1992), stability (Pikal et al., 1978) and

bioavailability. Changes in physical form may be polymorphic transitions, changes in solvates or variation in the degree of crystallinity. It has been shown that changes in crystallinity can have a significant influence on product performance, even if the change is too small to be measured by techniques such as powder X-ray diffraction (XRPD). Saleki-Gerhardt et al. (1994) have shown that materials can contain as much as 10%

* Corresponding author.

amorphous content and still appear to be totally crystalline when studied by XRPD or differential scanning calorimetry (DSC). Our own data (Ahmed et al., 1996) have also shown this to be true.

It is known (Briggner et al., 1994; Ahmed et al., 1996) that processing of powders may produce amorphous material in crystalline samples, and that that amorphous material can recover when stored in suitable environmental conditions. For example, lactose will recrystallise when stored above ca. 48% RH. The presence of differences in the degree of crystallinity may cause batch-to-batch variability for 'at-risk' materials, such as those with low bioavailability (due to poor solubility), or those which have critical demands placed upon their interfacial behaviour (such as micronised drug in inhalation products).

In this study, griseofulvin has been treated to make it partially amorphous and has then been recrystallised by exposure to different RH environments. Griseofulvin is used as a model drug of relatively low water solubility (defined as very slightly soluble in The Pharmaceutical Codex, 1994) which has been known to exhibit changes in bioavailability due to dissolution rate changes.

2. Methods

Griseofulvin (Sigma, Batch 48FO787) was characterised by DSC and XRPD. The DSC was undertaken on a Perkin Elmer DSC7 using approximately 4 mg of sample, and scanning at 10°C/min from 40–250°C in a non-hermetically sealed pan. The XRPD was undertaken using a Philips PW3710 X-ray diffractometer.

The griseofulvin sample was stressed by raising to 225°C (just above the melting point) in an oven for 1 h and then flash cooled by submerging in liquid nitrogen. The stressed sample was examined by DSC, XRPD and isothermal microcalorimetry. The samples which had been exposed to differing vapours in the isothermal microcalorimeter (see method below) were then also assessed by DSC and XRPD.

The samples were investigated by isothermal microcalorimetry (Thermal Activity Monitor,

Thermometric, at 25°C) by loading approximately 100 mg into a glass calorimeter cell with a tube of saturated salt solution (such that the powder was physically separated from the liquid, but was exposed to the vapour produced). The glass cell was sealed with an air-tight stopper and lowered, after 30 min temperature equilibration, into the measuring position of the calorimeter. The responses detected by the calorimeter would be the net effect of all processes which occur, thus there is comparatively little by way of a wetting response as the enthalpy change for vaporisation is approximately equal and opposite to that for condensation of the vapour on the solid. Any change in crystal form will consequently be measured with relative ease in this system as baseline disturbance will be minimal in the absence of a large wetting response.

3. Results and discussion

The DSC of the starting material showed a melt at 222°C with a ΔH_{fus} of 100–112 J/g. When the starting material was exposed to humid air in the isothermal microcalorimeter the only response was the small initial thermal event, due to environmental equilibration and sample wetting within the closed cell.

The XRPD of the quenched material showed identical peaks to those seen for the starting material, however, there was some evidence of an amorphous halo. The DSC of the quenched material showed a difference from the starting sample, in that there was an exotherm at 121–124°C, followed by the melt at 222°C (Fig. 1(a and b)). The exotherm is attributed to a recrystallisation of amorphous material (such findings have been reported previously for DSC of quenched griseofulvin by Elamin et al., 1994). It follows that the quenched material was partially amorphous (DSC data), but substantially crystalline (XRPD data). Without an amorphous standard it is hard to quantify the extent of crystallinity of this sample. However, it has been shown (e.g. Saleki-Gerhardt et al., 1994) that (for other materials) the lower detection limit of DSC and XRPD is between 5–10% amorphous content in the sample, thus it

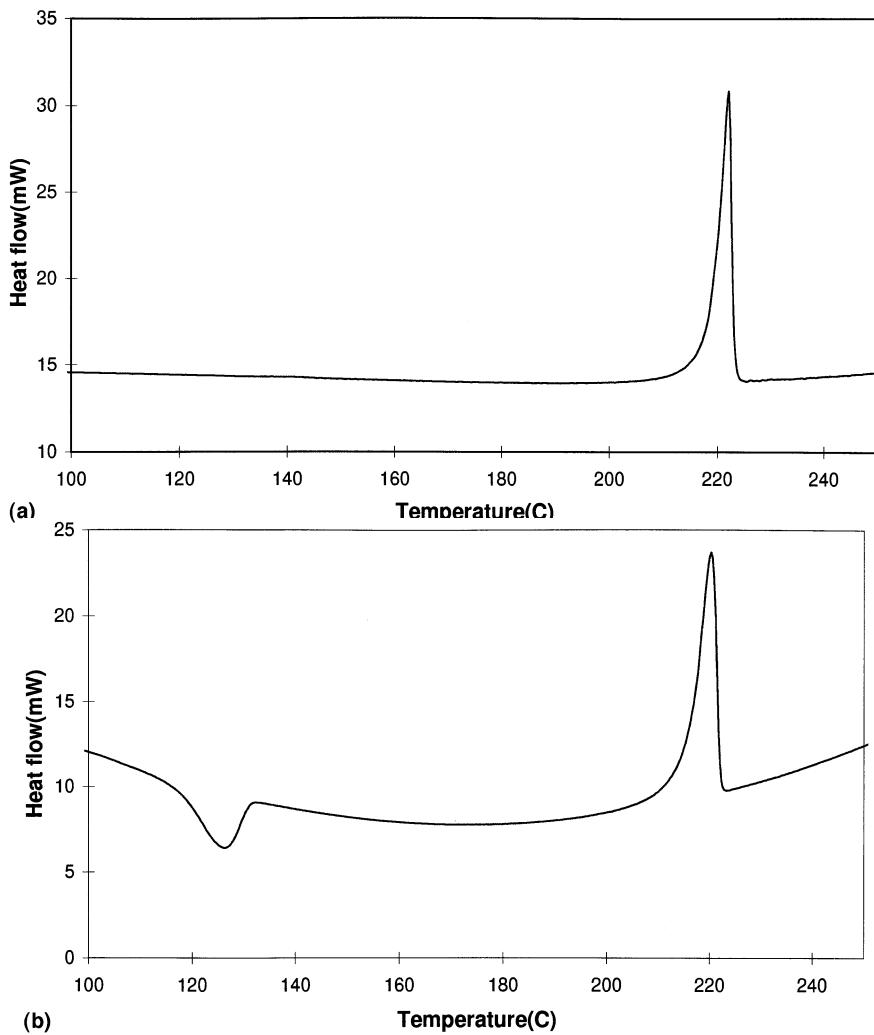


Fig. 1. DSC scan for (a) crystalline drug and (b) quenched drug with amorphous content.

is reasonable to assume that this sample may have an amorphous content which is greater than 5%, but not close to 100% (as the XRPD showed clear sharp peaks, typical of a highly crystalline sample).

When the quenched material was exposed to humid air in the isothermal microcalorimeter the sample produced a thermal event which was shown by DSC to be due to a crystallisation of the amorphous material (when removed from the isothermal microcalorimeter after the thermal event and run in the DSC, the crystallisation exotherm of the partially amorphous material was not observed). It was surprising that the am-

orphous material was able to convert to the crystalline form following exposure to a humid environment, given the low aqueous solubility of griseofulvin. It had been expected that this sample would behave like previous hydrophobic samples (Ahmed et al., 1996) and require an organic vapour to cause crystallisation to occur. It must be concluded, that the griseofulvin, whilst having low aqueous solubility, is able to absorb water into its amorphous structure which acts as a plasticiser, giving the increased mobility which allows the amorphous material to convert to the thermodynamically stable crystalline form.

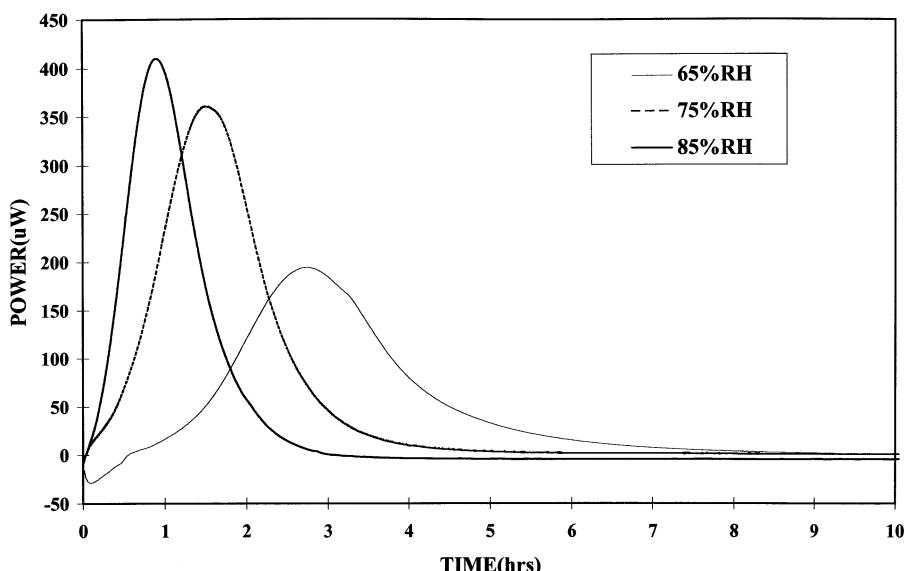


Fig. 2. Isothermal microcalorimetry responses showing the crystallisation of the amorphous content of the quenched drug. Saturated salt solution were used to achieve the humidities as indicated on the key.

The shape of the crystallisation responses which were seen in the isothermal microcalorimeter were different from the typical response seen for other materials upon recrystallisation of amorphous solids (e.g. Briggner et al., 1994; Buckton et al., 1995). The griseofulvin crystallisation responses (Fig. 2) occur over a longer time period than has been observed previously (lactose crystallisation occurring in 1 h or less (Briggner et al., 1994) and salbutamol sulphate in under 2 h (Buckton et al., 1995). The crystallisation events for griseofulvin (Fig. 2) have a reasonable degree of symmetry and last for up to 6 h. The responses here have a rapid onset which could be slowed only slightly by reducing the RH of the air (Fig. 2). This is different to previous studies, where reducing the RH resulted in substantial lag times when crystallising lactose (Briggner et al., 1994) or salbutamol sulphate (Buckton et al., 1995). The shape of the calorimetric output changes with RH (Fig. 2), with the higher RH resulting in responses which are initiated from a zero baseline (approximately) whilst the 65% RH responses are seen to start with an endotherm which is not separated from the crystallisation exotherm. The areas under the curve (best approximations given displacement of

response from baseline) are presented in Table 1. It can be seen that each RH gives reproducible data within its own set of results, however, the data for each RH are different from that obtained at each other RH. The differences in measured area under the curve are probably related to the fact that the crystallisation responses are not being measured in their entirety, due to overlapping events such as the wetting of the powder surface and changes in the salt solution (evaporation of water and crystallisation of the dissolved salt) being superimposed on the early part of the calorimetric response. Part of these early wetting/environment equilibration events will be lost during sealing and temperature equilibration of the sample in the microcalorimeter water bath. The

Table 1

Areas under the recrystallisation response following exposure of quenched griseofulvin to different RH environments in the isothermal microcalorimeter

	Areas (J/g)	Mean (J/g)
65% RH	17.9; 16.4; 18.0; 14.3	16.7
75% RH	19.3; 19.2; 20.3; 20.7	19.9
85% RH	15.8; 14.0; 12.9; 16.8	14.9

Table 2

Values obtained for the enthalpy of reaction, the rate constant and the number of moles reacting, by fitting the microcalorimetry data using the curve fitting programme

RH	Enthalpy (kJ/mol)	Rate constant ($s^{-1} \times 10^4$)	No. of moles ($\times 10^4$)
65%	4.6	2.6	
	5.0	3.1	3.5
	4.3	3.2	4.1
	4.8	4.4	3.7
65% mean	4.7	3.3	3.6
75%	4.4	6.5	4.3
	5.0	7.0	4.0
	4.6	6.3	4.5
	4.5	7.9	4.4
75% mean	4.6	6.9	4.3
85%	4.6	7.9	3.7
	4.0	8.4	4.0
	3.1	7.8	4.3
	3.8	8.9	3.4
85% mean	3.9	8.3	3.9

impact of the early events on the measured crystallisation responses is clearly marginal, but still detectable (Table 1). In order to remove the impact of these early wetting/environmental equilibration events, it is necessary to separate the bulk of the crystallisation event from the other thermal events which occur in the cell. Previously this has been achieved by changes in sample load and RH, but as described above, this did not result in a sufficient lag time prior to crystallisation in this case. Consequently, it was decided to model the data theoretically to attempt to get a better measure of the crystallisation process.

Wilson et al. (1995) have presented a method by which it is possible to fit data obtained from isothermal microcalorimetry experiments to suitable models in order to simultaneously determine thermodynamic and kinetic parameters for any detectable event. The transition from a partially amorphous to crystalline form of griseofulvin can be described as a solid state reaction and as such was fitted to the equation attributed to Ng by Wilson et al. (1995):

$$dx/dt = Ak(x/A)^m(1 - (x/A))^n \quad (1)$$

where dx/dt is the rate of reaction, x/A is the

fraction of reacted material, k is the rate constant and m and n are constants. This equation can be transformed to an equivalent which would describe the calorimetric experiment (Wilson et al., 1995):

$$dq/dt = Ak\Delta H(q/A\Delta H)^m(1 - (q/(A\Delta H))^n) \quad (2)$$

where dq/dt is the rate of change of heat (q) with time (t), and ΔH is the enthalpy change of the reaction, with A being the concentration term.

The data for the crystallisation event were modelled exactly as described by Wilson et al. (1995), using a computer program to solve Eq. (2) for the unknowns (A , k , ΔH and the constants) by a process of iteration. The output of the computer modelling is summarised in Table 2.

Although it is possible to calculate an enthalpy change and a value for the number of moles reacting (Table 2), it has been shown by Gaisford (1997) that the method of estimating these values can result in more than one pair of values for the enthalpy and concentration terms which would fit to the equation. Thus unless either the enthalpy or the amount reacting are known it is not possible to have confidence in the value which is predicted

for either of these terms. This becomes clear when the amount of material reacting is considered. The calculated value (Table 2) is essentially the same at each RH at ca 4×10^{-4} moles. The sample load in the calorimeter was 100 mg, which is 3×10^{-4} moles of griseofulvin, and it has been shown that only a small amount of this was amorphous, thus the moles of griseofulvin crystallising cannot amount to the calculated 4×10^{-4} moles which have been calculated to be involved in this process. It may be that the amount reacting related to the water content in the sample, however, we have measured the water uptake by the quenched sample using a Dynamic Vapour Sorption microbalance system (not shown) and have found the maximum uptake to be in the region of 1×10^{-4} moles of water into a 100 mg quenched griseofulvin sample, i.e. a quarter of the amount calculated for the material reacting. As the calculated number of moles reacting seems to be greater than the quantity of material available to react, it can be concluded that the enthalpy and concentration terms are indeed unreliable as noted by Gaisford (1997). In the system studied here, it is certain that more than one process will contribute to the measured enthalpy change (i.e. contributions will exist from at least the processes of crystallisation and water desorption, and potentially other events as well), as such it may become more appropriate to model this event by parallel or sequential reaction mechanisms (such a scheme has been proposed by Gaisford (1997) for sequential chemical reactions). The problem of identifying either a concentration or enthalpy term (for each reaction step) remains as the major problem to overcome if such an advance is to be made.

The calculated rate of the crystallisation (Table 2) is not subject to difficulties during the modelling process (Gaisford, 1997). The apparent rate constants for the measured response are seen to increase as the RH is increased, demonstrating the vital role that the RH plays in lowering the glass transition of the material and thus facilitating the crystallisation. It follows that it is possible to investigate the effect of humidity on the rate of

solid state transformations by modelling of calorimetric data. Such rate constant data are difficult to obtain by other means, especially for samples such as this which are only partially amorphous.

4. Conclusion

It has been shown that partially amorphous griseofulvin can convert to the stable crystalline form in the presence of water vapour. To the best of our knowledge the data presented here are the first attempt to determine the thermodynamic and kinetic parameters associated with such a transition, by use of isothermal microcalorimetry. Changes in RH have been shown to alter the rate of crystallisation, and the rate constant can be calculated. As such the RH is clearly a catalyst in the process. The amount reacting, however, is greater than the amount of griseofulvin present and this demonstrates that it remains impossible to calculate the enthalpy change and the amount reacting unless one of these terms is known. For complex solid state processes there is still no method to determine one of these two terms. This is especially true in this case where more than one process will contribute to the enthalpy term.

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