ELSEVIER

Thermochimica Acta 316 (1998) 29–36

thermochimica acta

Quantitative assessments of powder crystallinity: Estimates of heat and mass transfer to interpret isothermal microcalorimetry data

Patricia Darcy, Graham Buckton*

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

Received 8 December 1997; accepted 19 December 1997

Abstract

Isothermal microcalorimetry is used to study small quantities of amorphous materials in crystalline powders. The aim of this work is to better understand the isothermal microcalorimetry measurement with regard to the quantification of amorphous contents of materials. Amorphous lactose was crystallized in a sealed ampoule in an isothermal microcalorimeter at a range of temperatures (25–60°C) and humidities. Identical heat changes for crystallization were observed at all humidities at 25°C; however, the measured heat varied with humidity at higher temperatures. The heat measured by isothermal microcalorimetry was approximately the difference between the heat of crystallization and the heat of vaporization of the desorbed water. The isothermal microcalorimetry output for this process is now better understood and it can be seen that, in order to obtain quantitative data for crystallinity, it is necessary to have a slow supply of vapor. As the measured heat change is related to the extent of water desorption, care must be taken when using microcalorimetry to quantify the amorphous content of powders, especially when comparing data generated at different environmental conditions. © 1998 Elsevier Science B.V.

Keywords: Amorphous; Crystallinity; Glass transition; Isothermal microcalorimetry; Lactose; Water sorption

1. Introduction

There is a major interest in studies on amorphous content of pharmaceuticals, due to the fact that it can affect both the stability of the system and the performance of the material during product manufacture and use. Recent reviews highlight the major issues relating to the amorphous state in pharmaceuticals [1,2]. It is clear that small quantities of amorphous material in crystalline samples can be of importance, consequently techniques are being developed to assist in the study of amorphous contents amounting to <1% of the bulk weight of the material.

The most accurate detection of small quantities of amorphous content is through vapour-sorption techniques. One vapour-sorption approach is to monitor thermal changes in a sample when it is exposed to a defined vapour (often humidity) in an isothermal microcalorimeter [3–10]. Microcalorimetry is now widely available and used by many pharmaceutical research and development groups to study partially amorphous materials [2].

Much work has been published on isothermal microcalorimetry studies of both changes in crystallinity [3–5] and the crystal form [11] of lactose. Lactose is interesting, both as a model material and as a component of many pharmaceutical formulations. The simplest method is to load the isothermal micro-

*Corresponding author.

PII S0040-6031(98)00300-1

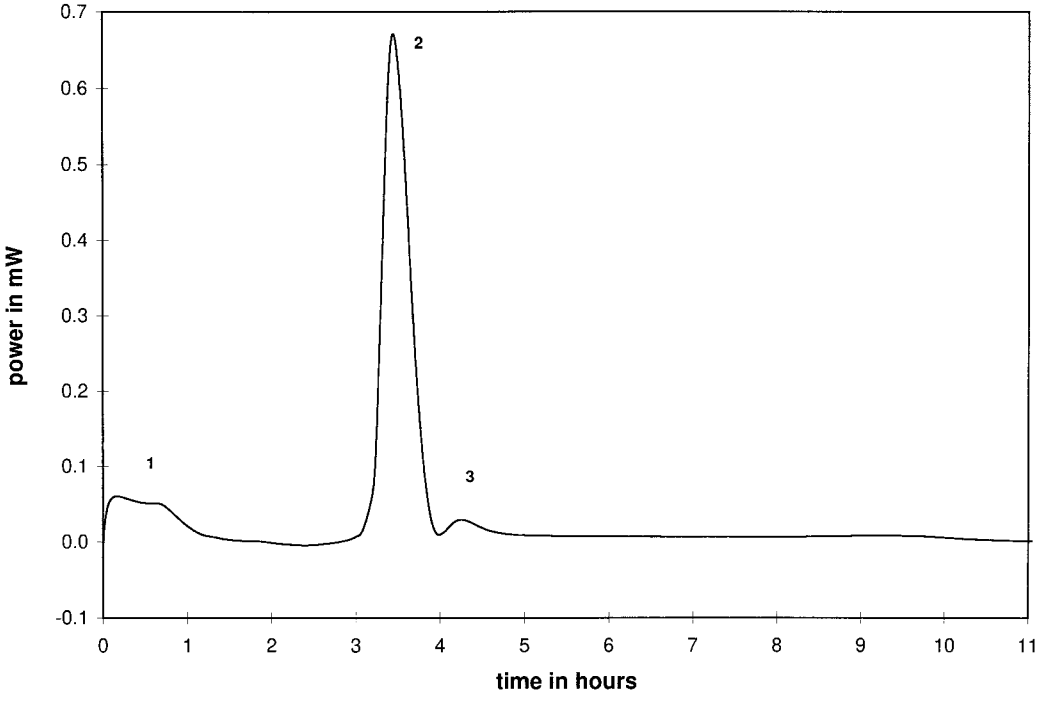

Fig. 1. Typical recrysallisation response for amorphous lactose exposed to 75% RH in a sealed cell of an isothermal microcalorimeter at 25°C, this being similar to data reported elsewhere [3,4,6].

calorimeter with a sealed ampoule containing the powder and a tube of saturated salt solution, to give a predefined relative humidity (RH) in the measuring cell. Calorimetry is non-specific, so every process which occurs in the cell is measured. The thermal response for amorphous lactose when exposed to 50% RH or greater at 25°C, is now well established and a typical example of such a response is shown in Fig. 1. Fig. 1 can be seen to have three distinct regions. Part 1 has been described as a wetting response [4] due to the absorption (exotherm) of water into the sample being slightly out of balance with the evaporation of the water vapour from the saturated salt solution reservoir (endotherm). More recently, it has been suggested that this initial response may, in part, be due to the amorphous material undergoing structural collapse following absorption of the water [12]; however, this needs further investigation. The second stage of the response (Fig. 1) is due to the crystallisation of the amorphous material. The third stage (Fig. 1) is suggested to be due to mutarotation of the lactose [4]; however, this remains unproven. The area under the curve for the sum of parts 2 and 3 has proved to be reproducible for spray-dried amorphous lactose samples produced in different laboratories [3], amounting to a mean net heat change of 48 J/g when measured at 25°C (ca. 32 J/g if only Section 2 of the response is measured [4]). At the present time, there is no consistency of approach in the literature, regarding whether the amorphous content is best assessed by use of the area under just part 2 or parts 2 and 3 of the power–time curve. It is necessary to develop an improved understanding of the power–time curve if a logical decision is to be made as to how such data can be used to provide a quantitative assessment of the amorphous content of lactose (and, subsequently, other powders). It has been noted that, at 25°C, the area under the power–time curve (including parts 2 and 3) is unaltered with the choice of RH (saturated salt solution) [3,4], the only difference being that the lag time between peaks 1 and 2 decreases as the RH is increased.

The aim of the current work is to explore the basis of the net heat change which is observed during

crystallisation in a humidity controlled cell of an isothermal microcalorimeter, to give an improved general understanding of what is being measured and to ensure the data is subsequently used in an appropriate manner when applied to material characterisation.

2. Materials and methods

Amorphous spray-dried lactose was prepared from a 15% w/v solution as used previously [3,6,12] and the material was stored at 20°C over silica gel until use.

The lactose (ca. 20 mg accurately weighed) was placed in a glass ampoule along with a tube containing a suitable saturated salt solution, to obtain a desired RH at the temperature of the experiment. Ampoules were sealed with an air-tight closure and lowered into the equilibration position, and then into the measuring position of the isothermal microcalorimeter (TAM, Thermometric). The calorimeter was set at 25, 35, 45, 50 or 60°C. The salt solution, powder and ampoule had all previously been temperature-equilibrated by storing in an oven set to 1°C above the calorimeter temperature, in order to keep the temperature equilibration within the calorimeter to an absolute minimum. The heat flow was measured as a function of time as the sample crystallised.

Following crystallisation in the isothermal microcalorimeter, each sample was removed and analysed by both differential scanning calorimetry (DSC) and thermogravimetric analysis (TGA). DSC was undertaken using a Perkin–Elmer DSC 7 with samples of ca. 5 mg enclosed in non-hermetically sealed pans. The scan rate was 10°C/min from 25°–250°C, under a nitrogen flush. The instrument was calibrated by use of the melting point of indium. TGA (TA Instruments) was undertaken over the same temperature range, at the same scan rate, in open pans using ca. 5 mg samples.

3. Results and discussion

The trace in Fig. 1, which has been discussed in Section 1, is typical of the response seen for crystallisation. The areas under the curve for parts 2 and 3 are shown in Table 1 for the various combinations of temperature and humidity which were used in this work. It can be seen that the heat changes are lower at lower temperatures. At higher temperatures, there is a general trend for the heat changes to increase with a decrease in the RH used to achieve crystallisation; however, this is not the case at 25°C. It is important to understand why different heat changes are measured under different environmental conditions.

3.1. Interpretation of the 25°C isothermal microcalorimetry data

The heat change measured for the crystallisation process using isothermal microcalorimetry (Table 1) was 48 J/g at each RH. However, the sum of the endotherms for a typical DSC trace (Fig. 2) was 209.2 J/g (consisting of 60.0 J/g for the loss of water of hydration, 5.5 J/g for the alpha lactose melt and

Table 1
Areas under the curve (J/g) for the isothermal microcalorimetry crystallisation response for amorphous lactose at various combinations of temperature and humidity (standard deviation of the mean)

Temperature (°C)	75% RH [a]	Sodium nitrite [b]	Magnesium nitrate [c]	Magnesium chloride [d]
25	48.9 (1.9)	47.9 (0.9)	48.2 (1.1)	no response
35	53.0 (0.6)	47.1 (3.3)	59.4 (0.3)	no response
45	62.1 (0.8)	63.2 (3.0)	66.5 (1.1)	variable response
50	—	58.3 (3.1)	70.9 (0.8)	variable response
60	—	—	72.6 (1.3)	74.9 (10)

[a] Saturated solution of Sodium chloride produces an RH of 75% across a wide temperature range.
[b] Saturated solution of sodium nitrite produces 65% RH at 25°C, 62% at 35°C, 59% RH at 50 and 60°C.
[c] Saturated solution of magnesium nitrate produces 53% RH at 25°C, 50% RH at 35°C, 46%RH at 50°C and 43% RH at 60°C.
[d] Saturated solution of magnesium chloride produces 33% RH at 25°C, 32% RH at 35°C, 31%RH at 50°C and 30% RH at 60°C. For this salt at 45° and 50°C the response was extremely variable, presumably because it was marginal as to whether the sample would crystallise under these conditions.

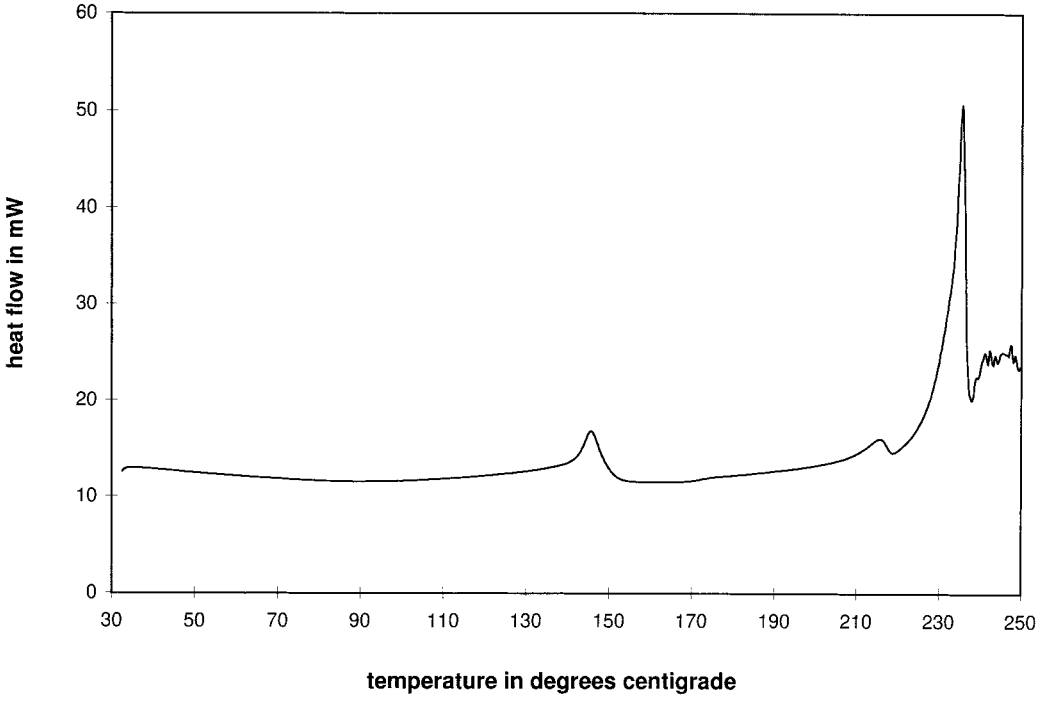

Fig. 2. Typical DSC trace for the crystallised product, following exposure to 55% RH in the isothermal microcalorimeter at 25°C. The first peak (130–150°C) is due to loss of water of hydration, the second peak (ca. 215°C) is the melt of the alpha-lactose, followed by the melt for beta-lactose, and subsequent decomposition.

143.7 J/g for the beta lactose melt). Even accepting that some decomposition will contribute to the DSC endotherm, there is a substantial difference between the apparent heat change associated with formation and that associated with the loss of the crystalline material which needs to be explored.

The proportions of alpha- and beta-lactose recorded on the DSC trace for samples which have been exposed to different humidities at 25°C were found to vary; however, the net recorded heat by isothermal microcalorimetry was always 48 J/g. Some mutarotation can occur during heating of a lactose sample, and as such the proportions of alpha- and beta-lactose which are present at their melting points are not necessarily the same that existed when the sample was loaded into the pan. However, evidence available to date makes it reasonable to conclude that the changes in measured heat at the higher temperatures are, simply, not due to the formation of different proportions of alpha and beta-lactose (Table 2).

Isothermal microcalorimetry measures all processes, as almost everything which occurs has an associated heat change. In order to understand the non-specific output, it is necessary to consider what happens during the process under consideration. The

Table 2
Typical areas under the curve (J/g) for DSC traces for lactose which has been crystallised at different temperatures in the isothermal microcalorimeter, showing a general increase in the measured alpha and decrease in beta content as the crystallisation temperatures were increased [a]

Temperature (°C)	Monohydrate loss	Alpha melt	Beta melt	Total area
25 (55% RH)	60.0	5.5	143.7	209.2
35 (55% RH)	40.8	3.1	135.3	179.2
45 (46%RH)	72.5	7.5	133.2	213.2
50 (46%RH)	59.5	17.1	97.3	173.8
60 (43%RH)	60.2	12.3	104.8	177.3

[a] NB: Typical areas are reported rather than mean data. These DSC data relate to specific samples removed at the end of an isothermal microcalorimetry run. To help with accuracy in the heat balance experiments, these specific DSC experiments have been related to the matching isothermal microcalorimetry experiment (rather than the mean isothermal microcalorimetry data, even though reproducibility between samples was good).

crystallisation of the amorphous lactose can be described by the following events:

1. Moisture evaporates from the saturated salt solution, some salt will crystallise as this evaporation occurs. The moisture will absorb from the environment into the amorphous solid, causing evaporation to continue. These three processes will be a mixture of exotherms and endotherms, which collectively do not give a huge response (see peak 1 in Fig. 1).
2. The absorbed water lowers the glass-transition temperature (T_g) of the amorphous lactose, this will result in structural changes and then in structural collapse (believed to contribute to peak 1 in Fig. 1).
3. Following from structural collapse there will be a lag period during which there will be no significant changes in moisture content of the powder. This corresponds to the near baseline region between peaks 1 and 2 (Fig. 1).
4. Ultimately, the powder will start to crystallise. During crystallisation, some of the absorbed water will cause some of the sample to form into a monohydrate, the remainder will be desorbed. Some of the desorbed water is absorbed by the neighbouring amorphous material, where it will cause crystallisation to continue, and thus desorb again. This 'domino effect' gives rise to the sharp crystallisation response which is seen in the calorimeter. The crystallisation would be exothermic and the desorption endothermic.
5. The desorption of water into an equilibrium environment would leave an atmosphere with a higher RH than would be formed over a salt solution. This will result in condensation back into the solution (exotherm) with subsequent dissolution of some of the solid salt (exotherm or endotherm, depending upon the heat of solution of the salt). The condensation may be rather slower than the other processes due to the small surface area of the salt solution. If sufficient water vapour is desorbed, the atmosphere could exceed the dew point, and thus condensation onto the solid surfaces could occur, until such time as equilibrium RH conditions are re-established.

A surprising factor is that the isothermal microcalorimetry heat changes at 25°C are the same at each RH (Table 1). This must imply that the amount of water evaporating (i.e. that water which was absorbed in the amorphous structure and which is expelled during crystallisation) is the same at each RH when the sample is at 25°C. It is clear that amorphous lactose will reach a different equilibrium water content at each RH; for example, it is known that lactose peaks at ≈13% weight increase when exposed to 75% RH in a dynamic vapour sorption (DVS) apparatus at 25°C [12], whereas it would contain around 10% water when equilibrated to 50% RH. Given that the samples in the isothermal microcalorimeter must all desorb the same quantity of water (as the net heat change is the same in each case), it must be that the samples do not reach their full equilibrium moisture content. This would be possible if the water absorption were slow (due to the slow supply of water vapour in these ampoules), such that, at each RH, collapse and crystallisation occurred at the minimum water content required to lower T_g (and the collapse temperature) to below 25°C, rather than having the excess water content that would be achieved in the DVS (where the T_g would drop far below room temperature). It has been shown that the rate of onset of crystallisation of lactose in the ampoules in the microcalorimeter is related to the surface area of the salt solution, and the RH used [3]. Thus, at 25°C, it is probable that the actual water absorbed into the amorphous lactose in the sealed ampoules of the isothermal microcalorimeter would be lower than that achieved in the DVS (due to the slower rate of water supply). To get an estimate of the actual water content obtained in the isothermal microcalorimeter, samples were removed after the first peak had been measured and before crystallisation began (between peaks 1 and 2 in Fig. 1) for the 25°C samples. It was found that, at each RH, the water content (as measured by TGA) was ca. 7.8%. It is known that desorption from these collapsed structures will be slow [12] so this value is a true reflection of the water content. This measured value of ca. 8% water content is interesting as it is just above the quantity of water that is needed to lower T_g below 25°C [12], i.e. it is in keeping with the fact that the water content is the minimum amount needed to cause crystallisation to occur. Based on an absorbed water content of 8%, with 3% hydrate formation (which is the value detected in TGA experiments following crystallisation), the balance

of heats at 25°C would become 209.2 J/g from DSC, −112.8 J/g for desorption of water (based on the enthalpy of vaporisation of water of 40.6 kJ/mol, Merck Index), giving an expected measured value of 96.3 J/g. Compared with the true measured value of 48 J/g, there is a difference of ca. 48 J/g (i.e. 96.3–48 J/g) which would be the exotherm due to condensation (this being equivalent to ca. 1.2 mg of water condensing).

3.2. Understanding the data at higher temperatures

It can be seen (Table 1) that the net heats are higher and are also seen to vary with RH for the higher temperatures. The amount of water needed to plasticise the amorphous lactose is reduced as the temperature is raised, thus materials will crystallise with a lower water load and as such the water desorption contribution can be expected to be lower.

At 45°C and 45% RH, the sample equilibrated to an 8.7% weight increase before crystallising, with a hydrate content following crystallisation of ca. 3% weight. The mean measured heat change in the isothermal microcalorimeter was 66.5 J/g (Table 1). The isothermal microcalorimetry experiment which resulted in the sample reported in Table 2 as the typical DSC run had a measured heat change of 65.1 J/g. If the sample reaches the same equilibrium moisture content in the isothermal microcalorimeter as it did in the DVS experiment, then the desorbed water will be the difference between ca. 9% absorbed and the ca. 3% retained as a monohydrate (which equates to 135.3 J/g by use of the enthalpy of vaporisation of water). The difference between the DSC heat (213.2 J/g, Table 2) and the heat of vaporisation (135.3 J/g) is 77.9 J/g i.e. ca. 12 J/g different from the measured value. The difference between the measured and estimated heat changes is easily explained due to any of the following:

(a) small differences in the water uptake in the ampoule compared with the DVS experiment;

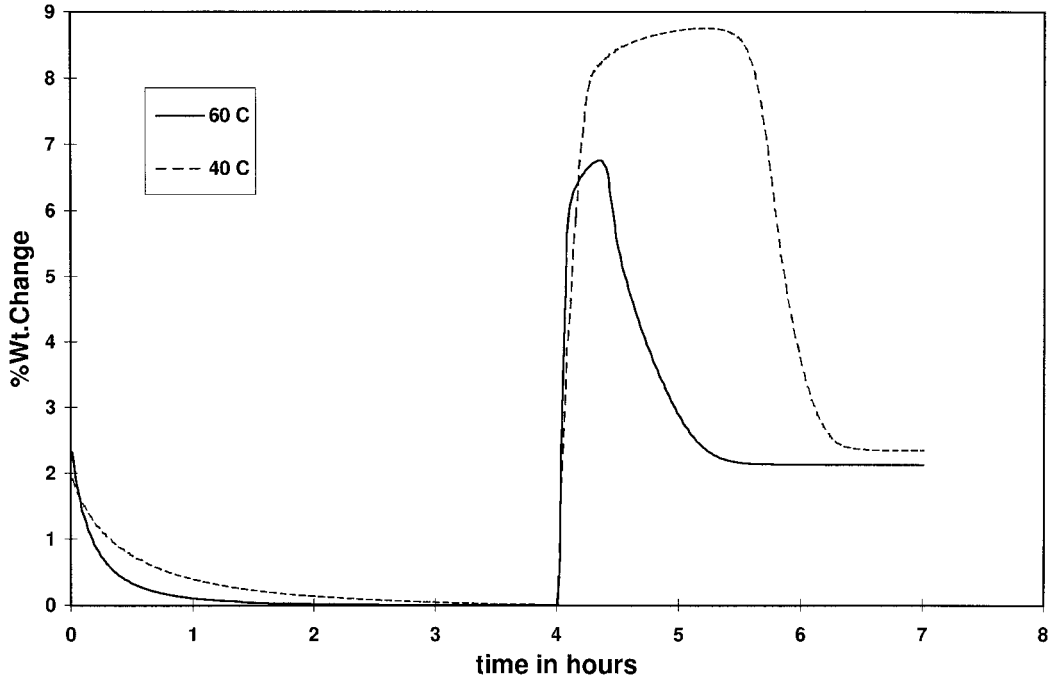

Fig. 3. Gravimetric sorption for amorphous lactose showing desorption over the first 4 h due to exposure to 0% RH, followed by exposure to 45% RH, 8.7% at 45°C and 6.76% at 60°C. The loss of weight following this peak uptake is due to the crystallisation resulting in desorption of the water. The retained water at the end of the experiment is due to the formation of a hydrate in part of the sample.

(b) possible differences in the heat of formation of alpha and beta lactose; and
(c) condensation effects after desorption.

At 60°C and 43% RH, the isothermal microcalorimetry heat for the crystallisation response was 71.7 J/g (Table 1) and the DSC heat was 177.2 J/g (Table 2). The TGA response for this sample when crystallised showed a 2.4% weight loss for the hydrate, and the DVS uptake (Fig. 3) was 6.8%. If 7% is taken as the absorbed water content and, thus, (7−2.4=4.6) 4.6% is the amount of water desorbed, then the heat balance is such that the vaporisation of water subtracted from the DSC heat approximates the measured heat in the calorimeter (73.7 J/g calculated, 71.7 J/g measured). In this instance, it must be noted that the rate of evaporation for the saturated salt solution is sufficiently fast (due to the increased temperature) and the water content required to achieve crystallisation is sufficiently low, that the sample in the isothermal microcalorimeter reaches the same water content as the equivalent sample in the DVS experiment.

3.3. *Considerations for quantitative amorphous content determination using isothermal microcalorimetry*

The consistency of the isothermal microcalorimetry data for different RH values at 25°C would indicate that this is the best condition at which to attempt quantitative analysis of the amorphous content of lactose. At higher RH, any variation in amorphous content between samples may result in different levels of absorbed water in the amorphous regions prior to collapse and crystallisation, thus giving different measured net heat changes, due to the more rapid supply of water vapour at elevated temperatures. At 25°C, for lactose it is reasonable to assume that the rate of water vapour supply is sufficiently slow that the collapse will occur with a consistent water content at any RH, i.e the rate of supply of water becomes the rate limiting event, rather than the kinetic events (for molecular movements, collapse, nucleation, crystallisation) within the sample.

4. Conclusion

The heat changes produced during crystallisation of amorphous lactose in an isothermal microcalorimeter are identical for all saturated salt solutions at 25°C, but become progressively larger with increases in the temperature of the experiment and with the solutions which yield lower RH at the higher temperatures. By considering a heat balance between DSC and the isothermal microcalorimeter responses, and a mass balance based upon an estimate of absorbed water content, hydrate water and, thus, desorbed water, it was possible to calculate a net heat change in keeping with that which was measured experimentally.

At higher temperatures, the higher measured net heat changes reflect the lower water sorption and, thus, lower water desorption contributions. At these higher temperatures, there is a difference in the isothermal microcalorimetry response for the saturated salt solutions used due to changes in water uptake prior to collapse of the structure.

The crystallisation response that is measured in the isothermal microcalorimeter has been explained to a greater extent, thus providing assistance for those who use such measurements to quantitatively assess amorphous content of powders. However, this study reveals that the complex nature of the processes which give the composite isothermal microcalorimetry response makes it necessary to be very careful in selecting appropriate conditions for the quantitative assessment of the amorphous contents of powders. It is particularly important to be careful when trying to compare data generated using different temperatures and/or humidities to induce crystallisation. Further work is still needed to obtain a complete understanding of how to design isothermal microcalorimetry experiments and how to use the data to assess the amorphous content of powders.

References

[1] E.Y. Shalev, G. Zografi, J. Pharm. Sci. 85 (1996) 1137.
[2] B.C. Hancock, G. Zografi, J. Pharm. Sci. 86 (1997) 1.
[3] L.-E. Briggner, G. Buckton, K. Bystrom, P. Darcy, Int. J. Pharm. 105 (1994) 125.
[4] T. Sebhatu, M. Angberg, C. Ahlneck, Int. J. Pharm. 104 (1994).

[5] G. Buckton, P. Darcy, D. Greenleaf, P. Holbrook, Int. J. Pharm. 116 (1995) 113.
[6] G. Buckton, P. Darcy, Int. J. Pharm. 121 (1995) 81.
[7] Y. Aso, S. Yoshioka, T. Otsuka, S. Kojima, Chem. Pharm. Bull. 43 (1995) 300.
[8] H. Ahmed, G. Buckton, D.A. Rawlins, Int. J. Pharm. 130 (1996) 195.
[9] T. Osterberg, A. Fatouros, Pharm. Res. 13 (1996) S-92.
[10] N.M. Vemuri, J.C. Croney, Z. Chrzan, R. Carlton, D. Toledo-Velasquez, Pharm. Res. 13 (1996) S346.
[11] M. Angberg, Thermochim. Acta 248 (1995) 161.
[12] G. Buckton, P. Darcy, Int. J. Pharm. 136 (1996) 141.