ELSEVIER

International Journal of Pharmaceutics 193 (2000) 247–259

international journal of pharmaceutics

www.elsevier.com/locate/ijpharm

Characterisation of the influence of micronisation on the crystallinity and physical stability of revatropate hydrobromide

Martyn D. Ticehurst [a,*], Patricia A. Basford [b], Christopher I. Dallman [b], Timothy M. Lukas [a], Peter V. Marshall [b], Gary Nichols [b], David Smith [b]

[a] *Formulation Design, Pharmaceutical R&D, Pfizer Central Research, Sandwich, Kent CT13 9NJ, UK*
[b] *Material Sciences, Pfizer Central Research, Sandwich, Kent CT13 9NJ, UK*

Received 25 June 1999; received in revised form 8 October 1999; accepted 8 October 1999

Abstract

Micronised particles of revatropate hydrobromide were observed to agglomerate when stored in uncontrolled conditions. Dynamic vapour sorption (DVS), isothermal microcalorimetry, microscopy and particle size measurement by laser diffraction have been used to study micronised revatropate hydrobromide. The rate and extent of agglomeration were dependent on the energy of the micronisation process, the sampling point for bulk within the mill and the humidity during storage. The agglomeration was attributed to the recrystallisation of disordered regions on the particles of revatropate hydrobromide generated during micronisation. This recrystallisation was assessed qualitatively and quantitatively, compared against spray-dried amorphous material, using DVS and isothermal microcalorimetry, respectively. A correlation was established between the energy of micronisation and the level of disorder within the micronised powder. A comparison of the DVS profiles of freshly prepared and aged micronised revatropate hydrobromide suggests an increased physical stability for the aged material. © 2000 Elsevier Science B.V. All rights reserved.

Keywords: Micronisation; Crystallinity; Particle size; Agglomeration; Dynamic vapour sorption; Microcalorimetry

1. Introduction

Revatropate hydrobromide is an anti-muscarinic drug in development for the treatment of asthma. It is a quaternary ammonium compound with an aqueous solubility of > 100 mg/ml at room temperature. Micronised particles of revatropate hydrobromide can agglomerate on storage after micronisation, as shown in Fig. 1a and b. This led to significant size enlargement and hence, an increase of material outside of the respirable range. This study was undertaken to determine the effect of the micronisation conditions, the point of collection from the mill and the duration

---

* Corresponding author. Tel.: + 44-1304-643915.
E-mail address: martyn_ticehurst@sandwich.pfizer.com (M. D. Ticehurst)

PII: S0378-5173(99)00347-6

and conditions of storage on the level of disorder in micronised revatropate hydrobromide.

Particles of a respirable size (typically < 5 μm) are required to formulate suspension metered dose inhalers or dry powder inhalers. Micronisation, using fluid energy or jet milling, is a commonly used process for the preparation of respirable sized particles. The micronisation process is extremely inefficient (Parrott, 1990) and, due to the high energy input, often leads to disruption within the crystal lattice of the micronised powder, creating disordered regions. These disordered regions are anticipated to be predominantly on the surface of the micronised particles and can be considered as amorphous.

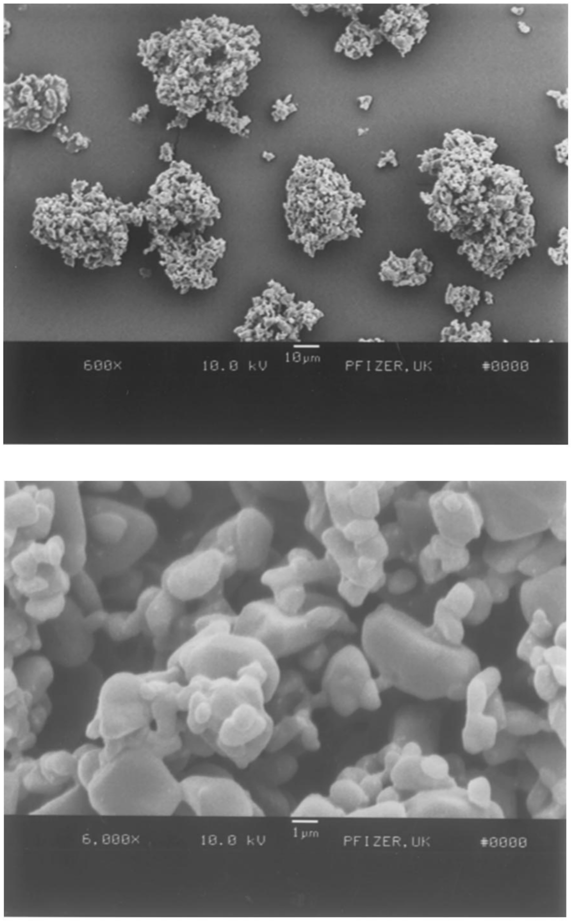

Fig. 1. Low magnification scanning electron micrograph illustrating revatropate hydrobromide agglomeration. (b) High magnification scanning electron micrograph illustrating revatropate hydrobromide agglomeration and bridging between particles.

Amorphous or disordered material is thermodynamically unstable and with time will recrystallise. Amorphous materials have a glass transition temperature above which the molecular mobility increases significantly and hence, crystallisation is more likely to occur. Water, from the atmosphere, can be absorbed into the amorphous regions and reduce the glass transition temperature (Zografi and Hancock, 1994; Elamin et al., 1995) thus decreasing the energy barrier to recrystallisation (Carstensen and Van Scooik, 1990; Buckton and Darcy, 1996). A small quantity of disordered material in a micronised powder may influence its physical and chemical stability, and hence, its performance in formulations (Buckton, 1997). Therefore the characterisation of crystalline disorder is a prerequisite in the formulation of micronised powders.

Often, changes in crystallinity after processing can not be detected by standard analytical techniques, such as differential scanning calorimetry and X-ray powder diffraction. This is because the disruption only effects a small quantity (in bulk terms) of the crystal lattice. The heat of crystallisation, measured using isothermal microcalorimetry has been used to quantify amorphous content down to a 1 or 2% level (Sebhatu et al., 1994). This sensitivity has prompted the use of microcalorimetry to quantify the amorphous content of processed materials (Briggner et al., 1994; Bystrom and Briggner, 1994). In addition, water absorbed into disordered regions of a powder can significantly influence its dynamic water vapour sorption isotherm, hence, DVS has been used to detect and, in some cases, quantify amorphous content (Saleki-Gerhardt et al., 1994; Buckton and Darcy, 1995; Ward and Schultz, 1995).

2. Materials and methods

2.1. Materials

Revatropate hydrobromide (batches 3601/185/2 and 4245/22/1, sourced from Pfizer) were 99 + % pure. $MgCl_2 \cdot 6H_2O$, $Mg(NO_3)_2 \cdot 6H_2O$ and NaCl were supplied by Sigma as Analar grade.

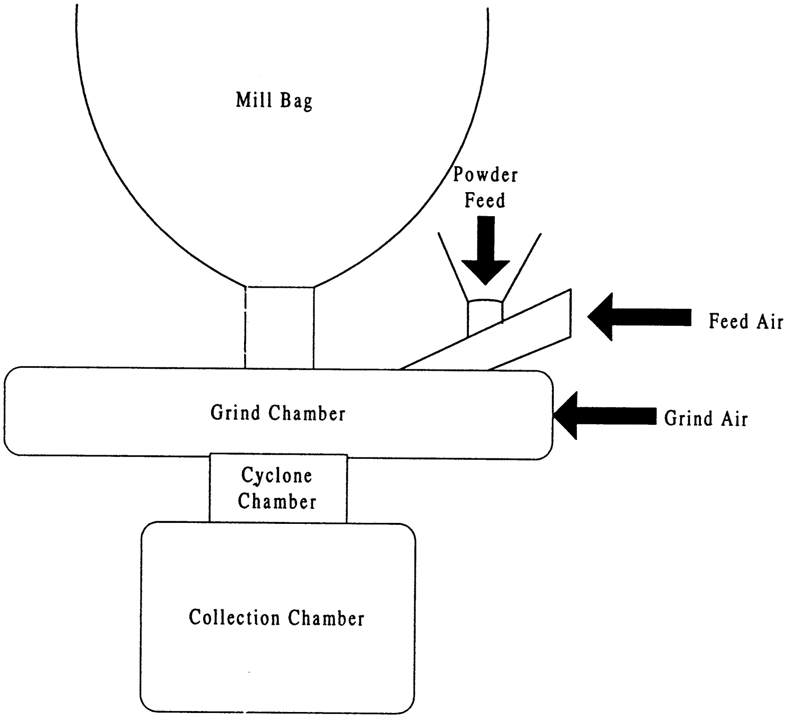

Fig. 2. Schematic diagram of an Apex pancake jet mill, highlighting the collection chamber and mill bag.

2.1.1. Sample preparation

Revatropate hydrobromide batches 3601/185/2M and 4245/22/1M were prepared by hammer milling crystallised batches 3601/185/2 and 4245/22/1, respectively, on an Apex MPX3 mill at high speed, hammers forward through a 1 mm mesh. An Apex ASS260 50 mm pancake jet mill was used to micronise batches 3601/185/2M and 4245/22/1M. Powder was fed manually into the feed inlet venturi at a rate of 0.3–0.4 kg h^{-1}. The feed pressure was set to 0.55 MPa and the grind pressure within the microniser chamber was varied, to prepare four micronised batches (0.24 MPa for batch 4245/22/1 M and 0.31, 0.41, and 0.52 MPa for batch 3601/185/2 M). This allowed the effect of energy input on the particle characteristics to be measured. The micronised particles were deposited either in the collection chamber or the mill bag, as illustrated in Fig. 2. Micronised powders were either stored over phosphorous pentoxide or activated clay, to protect them from moisture.

Spray-dried revatropate hydrobromide was prepared using a Buchi mini spray drier. A 10% w/v aqueous solution of revatropate hydrobromide was sprayed at 450 ml h^{-1} with inlet air at 170°C and outlet air at 96°C. The spray-dried material was stored over phosphorous pentoxide to protect it from moisture.

2.2. Methods

2.2.1. Powder X-ray diffraction

The powder X-ray diffraction (PXRD) patterns for revatropate hydrobromide were acquired at room temperature on a Bruker Axshtd D5000 diffractometer using Cu Kα radiation ($\lambda = 1.5406$ Å, tube operated at 40 kV, 40 mA), a θ-θ goniometer, automatic divergence and receiving slits, a graphite secondary monochromator, and a scintillation counter. The data were collected over an angular range of 2–55° 2 θ in continuous scan mode using a step size of 0.02° 2 θ and a step time of 5 s.

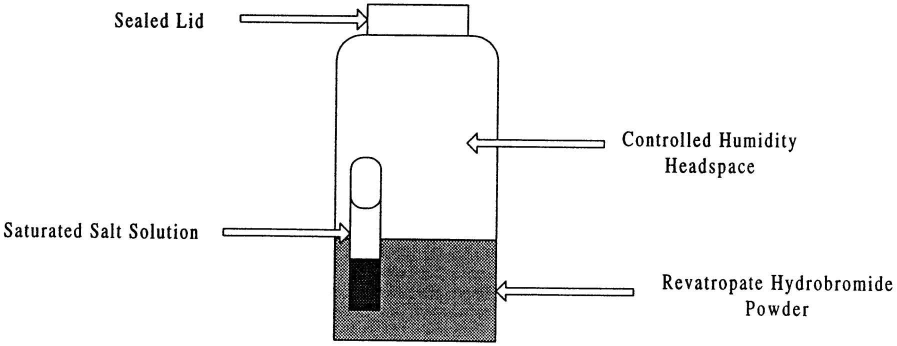

Fig. 3. Schematic diagram of sample set-up used for the isothermal microcalorimetry.

The powders were prepared as flat plate specimens using zero-background silicon wafers (The Gem Dugout, 1652 Princeton Drive, Pennsylvania State College, PA 16803). All specimens were rotated in their own plane during analysis.

2.2.2. Scanning electron microscopy

An Amray 1820-T scanning electron microscope (SEM) (Amray Inc, Bedford, MA) was used to examine the specimens. The powders were mounted onto 13 mm diameter aluminium stubs using double-sided adhesive tape and were sputter coated with gold. The scanning electron microscope was operated with a beam accelerating potential of 10.0 kV and images were collected in secondary electron mode.

2.2.3. Karl–Fischer analysis

Moisture content of samples were measured by Karl–Fischer titration on the Mitsubushi CA-06 Moisture Meter (Mitsubushi Chemical Corporation, Tokyo, Japan).

2.2.4. Isothermal microcalorimetry

A 2277 Thermal Activity Monitor (ThermoMetric, Jarfalla, Sweden) was used to monitor thermal events for 100 and 500 mg samples of revatropate hydrobromide. The powder was weighed into a glass vial. A micro test tube containing a saturated salt solution (33% RH ($MgCl_2 \cdot 6H_2O$), 53% RH ($Mg(NO_3)_2 \cdot 6H_2O$) and 75% RH (NaCl)) was added to the vial which was sealed, as shown in Fig. 3. The vial was lowered in the isothermal microcalorimeter and allowed to equilibrate at 25°C for 10 min before thermal monitoring commenced. Monitoring was stopped once the thermal event had occurred and the baseline returned to zero. The total integrated area under the thermal event profile (determined using Origin™ software) was used as the measure of the energy of the process.

2.2.5. Dynamic vapour sorption

The change in weight of samples as humidity was varied was recorded using a Dynamic Vapour Sorption (DVS), series 1 (DVS-1) Automated Water Sorption Analyser (Surface Measurement Systems Ltd, London). Samples were typically of the order of 10 mg, with humidity varied between 0–90% RH at 30°C.

2.2.6. Particle sizing

The particle size distributions of revatropate hydrobromide batches were measured using a Malvern Mastersizer S (Malvern, UK). For suspension sizing, the powder was dispersed in a saturated solution of revatropate hydrobromide in toluene and SPAN 80. Sonication was required to completely disperse the powder in the suspending liquid. A sonication time of 5 min was identified to produce a size distribution which did not decrease significantly with further sonication. Care was taken to ensure that the suspension did not heat up during sonication. A uniform dispersion of revatropate hydrobromide was maintained using a magnetic stirrer. For dry sizing, the powder was introduced through the Malvern dry powder feeder.

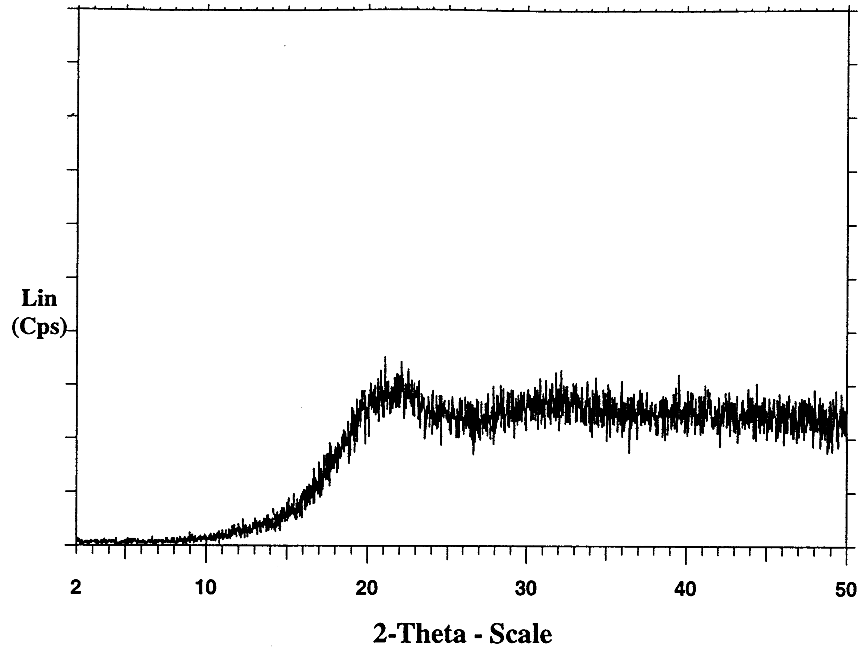

Fig. 4. Powder X-ray diffraction pattern of spray-dried revatropate hydrobromide.

Comparable particle size distributions were determined using the 'wet' and 'dry' deaggregation methods for material micronised at 0.31 MPa.

3. Results and discussion

3.1. Spray-dried material

The PXRD pattern of spray-dried revatropate hydrobromide is shown in Fig. 4. The characteristic halo was taken as an indication that the material was amorphous. This material had been stored with phosphorous pentoxide to protect it from moisture and hence prevent spontaneous recrystallisation.

The spray-dried material was characterised on the DVS using a stepwise increase in relative humidity between 0 and 50% RH. The sorption profile of the spray-dried material at 30°C with 2% step size is shown in Fig. 5. The profile shows a weight increase up to 29% RH followed by a dramatic weight loss. Following the event at 29% RH the revatropate hydrobromide was found to be crystalline by PXRD, with a pattern equivalent to standard crystalline material, as shown in Fig. 6. The event was thus attributed to the absorption of water which plasticises the amorphous material leading to recrystallisation and expulsion of absorbed water from the crystal lattice (revatropate hydrobromide has only one anhydrous polymorph and no hydrated forms identified). It was noted that equilibrium uptake was not achieved at each humidity, thus it is possible that with a longer equilibration time recrystallisation could have occurred at a slightly lower RH. The weight gain immediately prior to crystallisation was found to be 4.03% (± 0.07%, $n = 2$), which corresponds a molar ratio of 1.00:1. It is probable that the exact 1:1 required for crystallisation is not significant. However it is possible that revatropate hydrobromide could exist as a less thermodynamically stable hydrate which is transient and rapidly converts back to the anhydrous form.

Spray-dried revatropate hydrobromide was placed in the isothermal microcalorimeter and exposed to 33, 53 and 75% RH, as shown in Fig. 7. The calorimetric profile had a large co-opera-

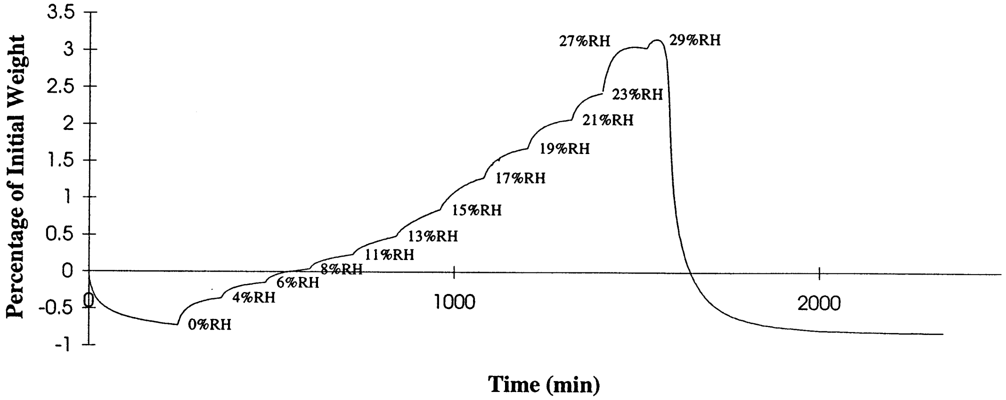

Fig. 5. Dynamic water vapour sorption profile for spray-dried revatropate hydrobromide from 0 to 29% RH in ≈2% RH steps.

tive exothermic event at each humidity. The exposure time before the exotherm occurred was dependent on the relative humidity with more rapid recrystallisation at higher humidities. Following the exothermic event, the revatropate hydrobromide was found to be crystalline by PXRD, with a pattern consistent with the material prior to spray-drying, and hence the event was ascribed to the heat of crystallisation. The more rapid recrystallisation at high humidities was ascribed to the higher thermodynamic activity of the water leading to a higher driving force for water to absorb into the bulk powder. There was also a shoulder following the sharp peak at 33% RH. The cause of this shoulder is uncertain, however this phenomena has been observed before for lactose, where it was tentatively attributed to conversion of unstable anhydrous α-lactose to α-lac-

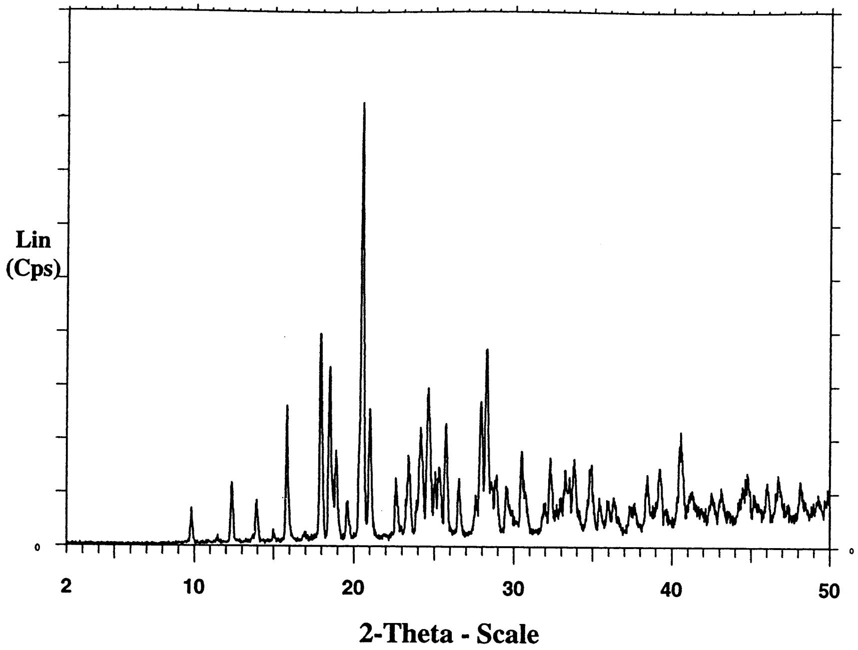

Fig. 6. Powder X-ray diffraction pattern for recrystallised revatropate hydrobromide.

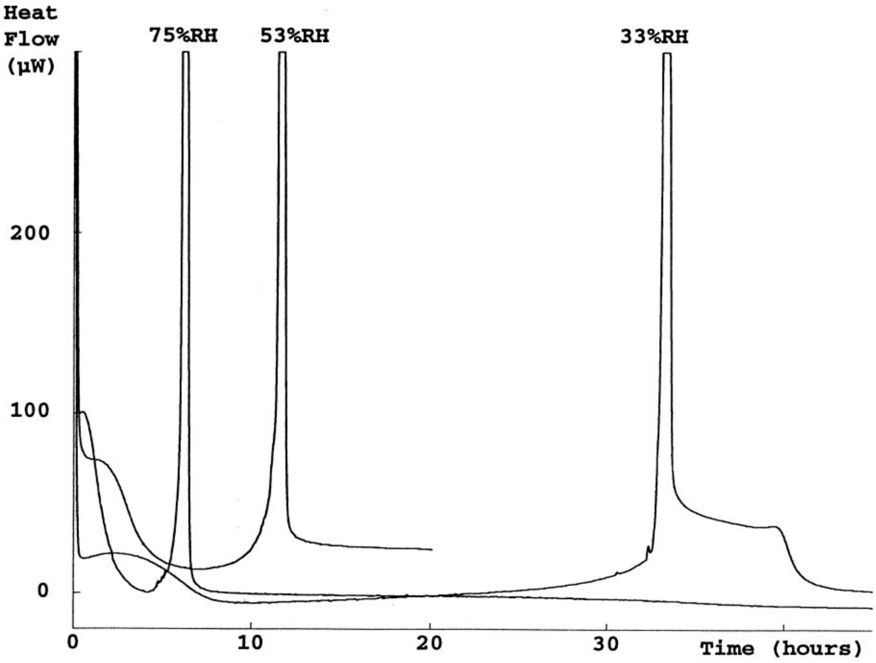

Fig. 7. Recrystallisation exotherms for amorphous spray dried revatropate hydrobromide at 30, 50, and 75% relative humidities.

tose monohydrate (Sebhatu et al., 1994). Revatropate hydrobromide has no known hydrated forms and there was no evidence of a different form after recrystallisation by PXRD, therefore, this argument does not hold. Multiple peaks have been detected for salbutamol sulphate (Buckton, 1995; Buckton et al., 1995) which were tentatively explained by the kinetics of the crystallisation process. Although the peak shape for revatropate hydrobromide is entirely different to that of the salbutamol sulphate it is possible that the extended exotherm is produced by the slower kinetics of water penetration at the lower humidity.

The heat of crystallisation (whole peak area) for a range of binary spray-dried and crystalline revatropate hydrobromide mixes containing 1–30% amorphous material, was used to prepare a calibration plot, as shown in Fig. 8. The linear profile indicated that the amorphous content of revatropate hydrobromide could be accurately quantified down to 1%. This profile was used to quantify the amorphous content of micronised samples of revatropate hydrobromide.

3.2. Micronised material

3.2.1. Particulate properties

The particle size distributions of powders prepared at 0.24, 0.31, 0.41, and 0.52 MPa, collected from the collection pot and the mill bag, and stored over phosphorous pentoxide, are displayed in Table 1. The grind pressure is the pressure of the air entering the milling chamber and hence the

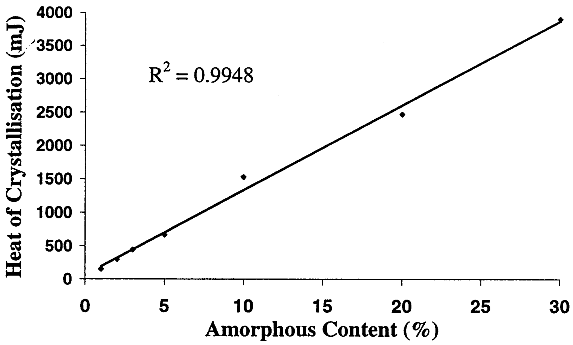

Fig. 8. Calibration plot of heat of crystallisation versus amorphous content for revatropate hydrobromide.

Table 1
Particle size distribution of micronised revatropate hydrobromide taken from the mill chamber and mill bag

Grind pressure (MPa)	Particle size of powder sample from mill container (μm)			Comments	Particle size of powder sample from mill bag (μm)			Comments
	90%<	50%<	10%<		90%<	50%<	10%<	
0.24[b]	2.1	1.6	1.1	Not visually agglomerated	ND[a]	ND[a]	ND[a]	ND[a]
0.31[c]	3.7	2.1	1.1	Not visually agglomerated	4.0	2.2	1.1	Not visually agglomerated
0.41[b]	6.2	3.1	1.4	Not visually agglomerated	4.0	2.1	1.0	Not visually agglomerated
0.52[b]	2.2	1.2	0.5	Not visually agglomerated	4.7	1.6	0.5	Not visually agglomerated

[a] ND, not determined.
[b] Measured using dry powder feed.
[c] Measured using a 'wet' (suspension) method.

higher the pressure the greater the energy input into milling. Scanning electron micrographs of a typical non-agglomerated micronised batch is shown in Fig. 9a and b.

The data in Table 1 shows that all the micronised batches are within the respirable range (typically defined as < 5 μm). However there is no trend in terms of particle size distribution versus grind pressure. This confirms that at above 0.24 MPa grind pressure the majority of the energy associated with the micronisation process is not being used to fracture particles and produce size reduction.

Micronised material sampled from the collection chamber was physically stable when stored immediately over phosphorous pentoxide. However samples from the collection chamber prepared at two of the grind pressures (0.31 MPa and 0.52 MPa) stored over activated clay desiccant were found to form large agglomerates. When viewed with the SEM, these agglomerates were observed to be made up of fused particles, similar to that shown in Fig. 1a and b. The smooth surface of the fused particles also differs from the non-agglomerated particles (Fig. 9a and b) which have a more jagged appearance. Therefore, the smoothing of the edges is attributed to the recrystallisation process. The particle size distributions of the agglomerated revatropate hydrobromide, demonstrate the size enlargement when compared to the non-agglomerated samples in Table 2. These results indicate that activated clay is not always an adequate desiccant system for micronised revatropate hydrobromide taken from the collection chamber.

In contrast the micronised material taken from the mill bag was found to be physically stable when stored with activated clay for all the batches. The reason for differences between the physical stability of the micronised material from the same batch taken from different mill locations is difficult to explain. However, extra drying of powder in the mill bag by the extremely dry microniser grind air (dew point $< -40°C$) is a possible explanation. In order to explore this hypothesis, freshly micronised samples from the mill container and mill bag were analysed by Karl–Fischer titration. The moisture contents measured by Karl–Fischer were 0.19% ($\pm 0.01\%$, $n=3$) and 0.24% ($\pm 0.03\%$, $n=3$) for the mill bag and mill container, respectively. It is possible that the difference in the water content of the two materials could be due to a physical difference between the materials sampled from the two locations such as particle size and levels of disorder. For these reasons the hypothesis that the difference in moisture content is due to drying of disordered regions can at best be considered tentative.

Given that the hypothesis is correct, the drier micronised powder from the mill bag would need

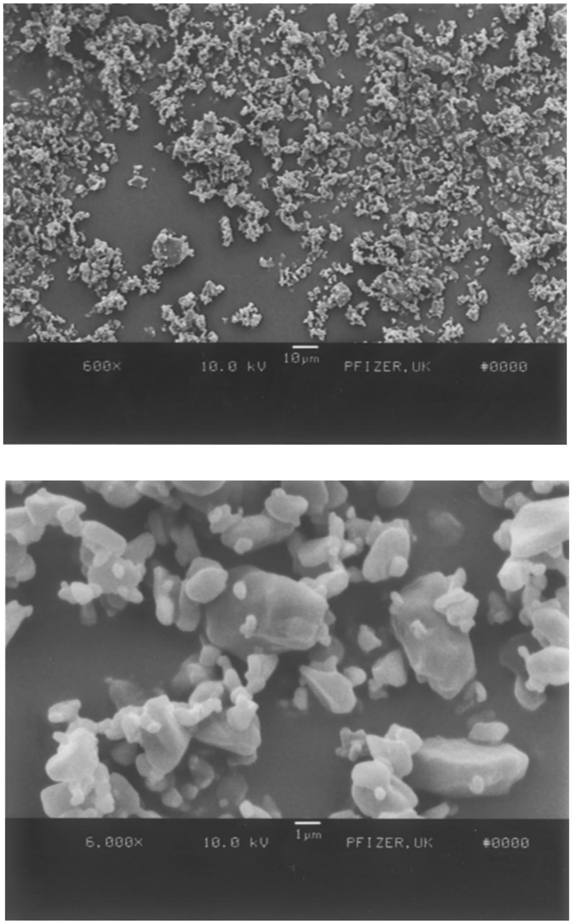

Fig. 9. (a) Low magnification scanning electron micrograph illustrating non-agglomerated revatropate hydrobromide. (b) High magnification scanning electron micrograph illustrating non-agglomerated revatropate hydrobromide.

more water to reach a threshold moisture content required for recrystallisation. In a sealed system, a larger amount of atmospheric water would be absorbed reducing the relative humidity. Thus the dried revatropate hydrobromide could act as a 'self-desiccant' enhancing the performance of the activated clay and maintaining the humidity below the threshold required for recrystallisation. Assuming equal levels of disorder in the mill bag and mill container material, the small but measurable difference in moisture content means that the drier mill bag material is required to absorb an extra 50 mg of water/100 g of powder. Although this is a low water volume, in liquid terms, it corresponds to approximately 2 l of 100% RH air at 25°C/100 g of powder[1]. Indeed drying of a micronised powder post micronisation and prior to storage may provide a good way of improving its physical stability.

3.2.2. Bulk properties

There were no significant differences between the XRPD patterns of the micronised powders and the feed material either before or after agglomeration. However PXRD is typically only sensitive to changes in crystallinity of the order of 10% (Sebhatu et al., 1994; Saleki-Gerhardt et al., 1994). Therefore, the PXRD patterns can only be taken to indicate that micronisation causes no

Table 2
Particle size distributions of micronised powders taken from the collection chamber and stored over phosphorous pentoxide (non-agglomerated) and activated clay (agglomerated)

Grind pressure (MPa)	Particle size of powder stored over phosphorous pentoxide (μm)			Observation	Particle size of powder stored over activated clay (μm)			Observation
	90%<	50%<	10%<		90%<	50%<	10%<	
0.31	3.7	2.1	1.1	Not visually agglomerated	9.0	3.6	1.7	Visually agglomerated
0.52*	2.2	1.2	0.5	Not visually agglomerated	18.4	4.7	1.7	Visually agglomerated

---

[1] One mole of water (18 g) equals 24.47 dm^3 at 101.32 kPa (1 atmosphere). At 25°C the partial pressure of water is 3.17 kPa (Lide et al., 1997), therefore 1 mole (18 g) equals 782.1 dm^3. Therefore 50 mg of water at 25°C equals 2.17 dm^3.

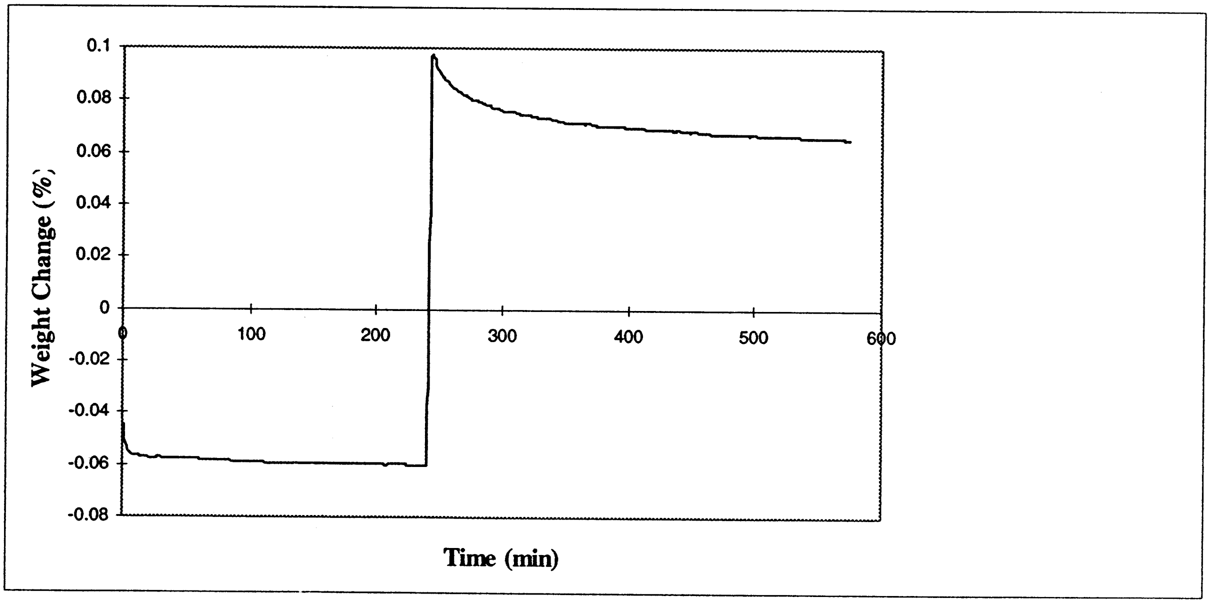

Fig. 10. Representative DVS Profile of micronised revatropate hydrobromide dried then exposed to 75% RH.

gross changes in crystallinity of revatropate hydrobromide.

3.2.3. Dynamic water vapour sorption

Samples of micronised powder were analysed by DVS at a range of humidities. Fig. 10 shows a typical profile for micronised powder at 75% RH. A repeat exposure of the same sample at 75% RH produced a flat profile with no characteristic spike. The characteristic spike on the profile is attributed to absorption of moisture, which plasticises the amorphous regions, triggering spontaneous recrystallisation and expulsion of water. This type of peak has been detected down to 25% RH for some batches. All samples which had not visually agglomerated showed a characteristic peak on the DVS. In some cases even samples which had visually agglomerated showed a very small, but detectable, peak.

Micronised revatropate hydrobromide (0.24 MPa grind pressure sampled from the collection chamber) was analysed by DVS immediately after micronisation (within 30 min) and after 39 days stored over phosphorous pentoxide, as shown in Fig. 11a and b. The freshly micronised and aged samples had different DVS profiles. The DVS profiles suggest that a higher humidity and/or a longer period of time is required to produce crystallisation of the disordered regions in the aged sample. Hence, it can be inferred that the aged sample would be more physically stable to recrystallisation and subsequent agglomeration. The reason for the change is unclear and from these data it is not possible to speculate whether it is a change in kinetic or thermodynamic stability of the disordered regions.

The presence of characteristic spikes at RHs above the threshold crystallisation peaks in Fig. 11a and b suggest that further crystallisation is occurring at these higher humidities. This is most likely attributed to the recrystallisation not being complete at each RH.

Attempts to use the magnitude of the characteristic spike on the DVS to quantify bulk disorder in micronised powder have, at best, shown trends versus energy input. It is hypothesised that the crystallisation event monitored on the DVS is not strictly quantitative because the size of the peak does not reflect the crystallisation of the whole powder mass; rather it reflects the recrystallisation of a small region of the powder from which the expelled water serves to catalyse further recrystallisation. Therefore, the size of the peak is dependent on a number of variables, other than the level of disorder, including bed packing and particle size.

3.2.4. Isothermal microcalorimetry

Micronised samples were exposed to 30% RH in the microcalorimeter immediately after processing. The samples were found to give a characteristic co-operative exothermic event, as shown in Fig. 12, which was attributed to recrystallisation. The recrystallisation of the micronised powder is more rapid than for the mixtures of crystalline and spray-dried amorphous material (≈ 7 h for the 1% sample). This is considered to be due to the higher surface area of the disordered component in the micronised powder.

The magnitude of the heat of crystallisation was compared to the calibration plot (Fig. 8) to determine the amorphous content. Although the disorder of the micronised powder is presented in terms of amorphous content, this is for the sake of simplicity. It is understood that there may be a continuum of crystallinities within the micronised powder which may not be best defined by amor-

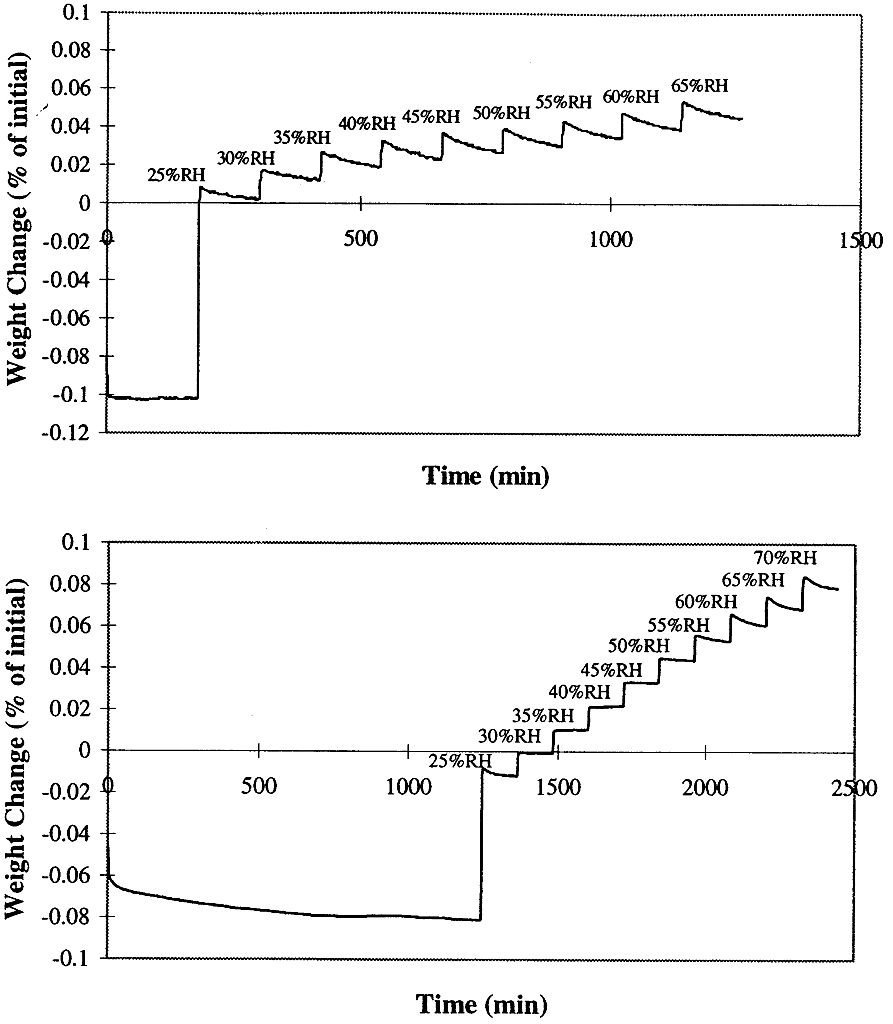

Fig. 11. (a) A DVS profile for freshly micronised revatropate hydrobromide. (b) A DVS profile of micronised revatropate hydrobromide aged for 39 days.

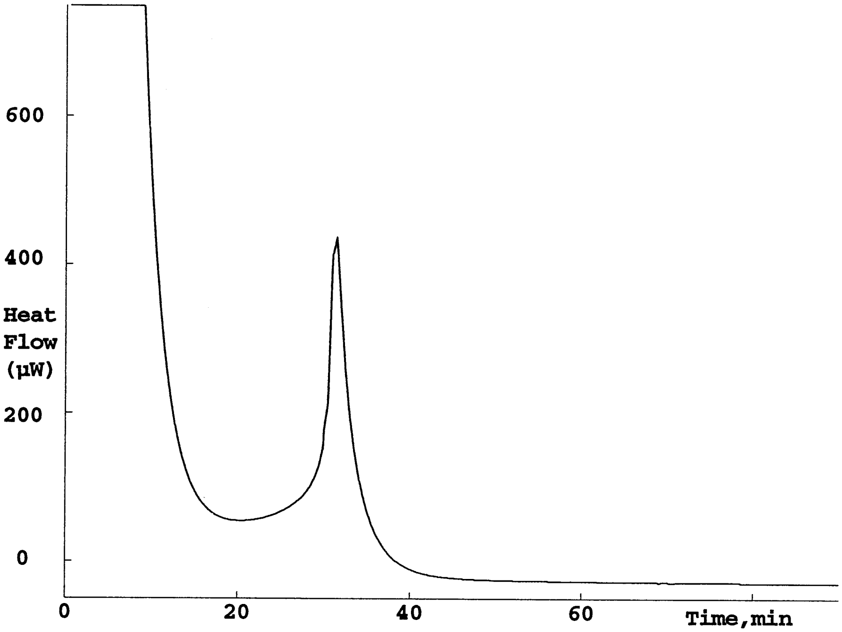

Fig. 12. Recrystallisation exotherm for revatropate hydrobromide micronised at 60 psi.

phous content. Not withstanding this, the amorphous content of micronised material prepared at different grind pressures, which were sampled from the collection chamber, are shown in Table 3.

There is, as expected, a correlation between the energy of micronisation (as defined by grind pressure) and the amorphous content of the micronised powder. In addition there is a tentative link between the amorphous content and the magnitude of the agglomeration of the micronised powder as shown in Table 2. That is the micronised powder with the higher amorphous content exhibits a greater extent of agglomeration

Interestingly, the heat of crystallisation could only be detected for samples immediately after processing. This suggests that there may be some reordering of the disorder regions within the powder even at very low humidity without visual agglomeration. This is in accord with the change in threshold humidity for crystallisation detected by the DVS.

4. Conclusions

This work has again highlighted that the characterisation of micronised materials is critical to the understanding of their physical stability. Isothermal microcalorimetry and DVS have proved to be important techniques in the characterisation and quantification of low levels of disorder in processed material. For revatropate hydrobromide the level of disorder after micronisation has been shown to be dependent on intensity of micronisation, whilst the particle size does not show a dependence on grind pressure above 0.24 MPa. This indicates that a relatively low energy is required to produce brittle fracture and efficient size reduction. With excess energy, at higher grind pressures causing disruption of the crystal lattice. In the case of revatropate

Table 3
Amorphous content of micronised revatropate hydrobromide samples as determined by microcalorimetry

Micronisation grind pressure (MPa)	Equivalent amorphous by microcalorimetry (%)
0.24	Not detected (<1%)
0.31	2.0
0.41	3.3
0.52	6.7

hydrobromide it is possible to optimise the energy of micronisation at around 0.24 MPa to get an apparent amorphous content of $<1\%$ whilst still achieving respirable particles.

The location from which micronised revatropate hydrobromide is sampled and the immediate storage conditions significantly affect the physical stability of the micronised material. In the case of revatropate hydrobromide, it appears that the sample needs to be stored at less than 25% RH immediately after micronisation to prevent agglomeration. DVS data suggests that the disordered regions within aged material required a longer period of time and/or a higher humidity to recrystallise. Thus controlled storage prior to processing may allow an opportunity to improve the apparent stability of micronised revatropate hydrobromide. However, the potential for recrystallisation of material at ambient humidity, even after storage, indicates that careful choice of final packaging of bulk drug substance and drug product is required to control or prevent moisture permeation and to ensure physical stability.

Although DVS and microcalorimetry allow the physical stability of disorder regions of micronised material to be determined they do not specifically measure changes in surface energy. Inverse gas chromatography has recently been used to assess the effect of milling and micronisation on propranolol hydrochloride (York et al., 1998) and could be a complimentary technique to DVS and microcalorimetry providing information on changes in surface chemistry.

References

Briggner, L.-E., Buckton, G., Bystrom, K., Darcy, P., 1994. The use of isothermal microcalorimetry in the study of changes in crystallinity induced during processing of powders. Int. J. Pharm. 105, 125–135.

Buckton, G., 1995. Surface characterisation: understanding sources of variability in the production and uses of pharmaceuticals. J. Pharm. Pharmacol. 47, 265–275.

Buckton, G., 1997. Characterisation of small changes in the physical properties of powders of significance for dry powder inhaler formulations. Adv. Drug Del. Rev. 26, 17–27.

Buckton, G., Darcy, P., 1995. The use of gravimetric studies to assess the degree of crystallinity of predominantly crystalline powders. Int. J. Pharm. 123, 265–271.

Buckton, G., Darcy, P., 1996. Water mobility in amorphous lactose below and close to the glass transition temperature. Int. J. Pharm. 136, 141–146.

Buckton, G., Darcy, P., Greenleaf, D., Holbrook, P., 1995. The use of microcalorimetry in the study of changes in crystallinity of spray-dried salbutamol sulphate. Int. J. Pharm. 116, 113–118.

Bystrom, K., Briggner, L.-E., 1994. Microcalorimetry-a novel technique for characterisation of powders, Respiratory Drug Delivery IV, pp. 297–302.

Carstensen, J.T., Van Scooik, K., 1990. Amorphous-to-crystalline transformation of sucrose. Pharm. Res. 12 (7), 1278–1281.

Elamin, A.A., Sebhatu, T., Ahlneck, C., 1995. The use of amorphous model substances to study mechanically activated materials in the solid state. Int. J. Pharm. 119, 25–36.

Lide, D.R., (Editor in chief), 1997–1998. Handbook of Chemistry and Physics, seventy-eighth ed, CRC Press.

Parrott, E.L., 1990. Comminution. In: Swarbrick, J., Boylan, J.C. (Eds.), Encyclopaedia of Pharmaceutical Technology, vol. 3. Marcel Dekker Inc, New York.

Saleki-Gerhardt, A., Ahlneck, C., Zografi, G., 1994. Assessment of disorder in crystalline solids. Int. J. Pharm. 101, 237–247.

Sebhatu, T., Angberg, M., Ahlneck, C., 1994. Assessment of the degree of disorder in crystalline solids by isothermal microcalorimetry. Int. J. Pharm. 104, 135–144.

Ward, G.H., Schultz, R.K., 1995. Process induced crystallinity changes in salbutamol sulphate and its effect on powder physical stability. Pharm. Res. 12, 773–779.

York, P., Ticehurst, M.D., Osborn, J.C., Roberts, R.J., Rowe, R.C., 1998. Characterisation of the surface energetics of milled dl-propranolol hydrochloride using inverse gas chromatography and molecular modelling. Int. J. Pharm. 174, 179–186.

Zografi, G., Hancock, B., 1994.Water-solid transitions in pharmaceutical systems, fifty-third ed. International Congress of Pharmaceutical Sciences, Tokyo.