

AIM

The aim of the study was to investigate the physical stability and moisture induced phase transformations of amorphous pharmaceutical particles, *via* real time atomic force microscopy (AFM) and optical microscopy measurements.

INTRODUCTION

With the exception to the work of Zografi *et al.*, there has been no classification of the various nucleation mechanisms during the re-crystallisation processes of amorphous systems. Even though it is well known that the presence of a crystalline or a polycrystalline phase in a supersaturated solution results in nucleation and growth at a degree of supersaturation which primary nucleation would not otherwise occur. The activation energy for re-crystallisation can, therefore, be substantially lowered by the presence of secondary nuclei, which act as growth centres for nucleation and growth. With the indiscriminate nature of bulk analytical techniques, erroneous measurements of the re-crystallisation temperature (T_c) and induction time (τ) may arise, depending on the nature of amorphous materials.

In this study, we report, for the first time, the use of an atomic force microscope for *in situ* visualisation of moisture induced morphological transformations on the surfaces of individual spray dried lactose particles. The high resolution imaging capabilities and non-destructive nature of the AFM, with concurrent optical microscopy, allows subtle but significant physical changes in the surface properties of powders to be elucidated, with minimal sample preparation, under controlled environmental conditions. Such quantitative measurements of the stability of individual spray dried particles during storage may afford useful information concerning the role of surface morphological transitions in the interactions of drugs and excipients and the conditions required for long-term storage and packaging of such particles.

MATERIALS AND METHODS

Amorphous samples of lactose were produced by spray drying 12.5% w/w aqueous solutions, using a Büchi BL91 mini-spray dryer (Flawil, Switzerland). Upon collection, the powders were packed into glass ampoules and stored in desiccators, over phosphorous pentoxide. The processing conditions for spray-drying were - inlet temperature 110°C, outlet temperature 71°C, airflow 600 l/hr and feed flow rate 5 ml/min.

All AFM images were recorded in TappingMode™ operation (TM-AFM) with a Digital Instruments NanoScope® III SPM controller, a Multimode AFM head (Digital Instruments, Santa Barbara, CA, USA) and a 'J' type scanner (lateral scan range of 125µm, spatial range of 5µm).

The relative humidity was locally controlled within a small volume ca. 40cm³ of the imaging chamber of the MultiMode™ AFM by the use of a commercial HumPlug™ (BioForce Laboratory Inc., Santa Barbara, CA, USA). The partial vapour pressure of water was controlled by varying a mixture of dry nitrogen gas with the same gas humidified to 100% under constant temperature (27°C). A Thermo-Hygro™ sensor incorporated in the HumPlug™ continually measured the relative humidity (%RH) above the substrate surface. Humidity measurements were generally in good agreement ($\pm 5\%$) with tabulated values for saturated vapour environments of various salts, and, thus, are the values reported here.

RESULTS

Surface morphology of amorphous lactose particles

A representative image of the virgin surface topography of a ca. 30µm spray dried amorphous lactose particle maintained in its glass-like state is shown in Figure 1 (0%<RH<10%, T= 27.5°C). An array of elliptical shaped features of various dimensions were observed protruding from the surface. The size, heights and ratios of the major and minor axes of the elliptical features were not uniform, although a certain degree of orientation of the major and minor axes with the underlying substrate was routinely observed. Their source and ubiquitous nature was originally unknown. However, by undertaking studies on sub-10µm amorphous lactose particles and comparing aspect ratios and relative heights to the elliptical features, it was apparent that the larger spray dried particles were either co-spray dried with or incidentally blended smaller particles of spray dried lactose.

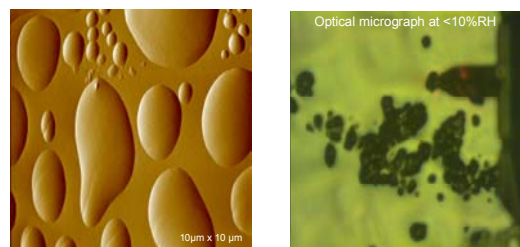


Figure 1: Ubiquitous surface topography of spray dried amorphous lactose particles.

Morphological changes as a function of relative humidity

The morphological changes of the surface topography of an amorphous lactose particle upon increasing the partial water vapour pressure from 10 to 30% RH is illustrated with the sequence of consecutive images contained in Figure 2. Upon increasing the relative humidity to 30%, the co-processed elliptical shaped particles decrease in size, while maintaining a relative constant aspect ratio between the major and minor axes. For extended periods, complete dissolution of the elliptical features was observed. With complete solubilisation of the co-processed particles, the surface texture of the particle under study remained unchanged.

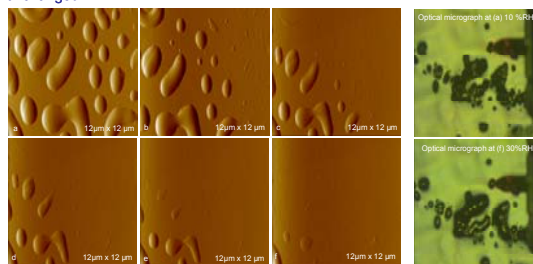


Figure 2: Surface morphological changes upon exposure to a partial water vapour pressure of 30%RH.

Surface Transformations above %RH_c

To visualise the surface transformations of amorphous lactose particles during the rubbery transition and re-crystallisation process, the %RH of the perfusion system was raised well above the critical relative humidity for re-crystallisation, at ca. 52%RH. A series of topographical images during the rubbery transition and subsequent re-crystallisation of the amorphous lactose particle upon exposure to 60%RH is shown in Figure 3. The re-crystallisation process gave rise to highly irregular, polycrystalline facets. Furthermore, a large percentage of the isolated particles did not re-crystallise even upon exposure to high humidities (>75%RH) for prolonged periods.

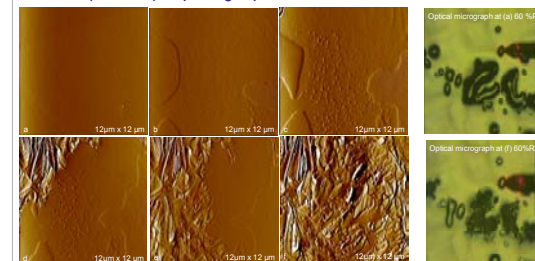


Figure 3: Surface topographical changes of an amorphous lactose particle during rubbery and re-crystallisation transformations.

Optical Microscopy Measurements

The metastable nature of amorphous particles, stored at 75%RH, to the re-crystallisation transformation, is illustrated by optical micrographs and scanning electron micrograph in Figure 4. Exposure to such humid environments, of up to six months, did not re-crystallise the rubbery like particles.

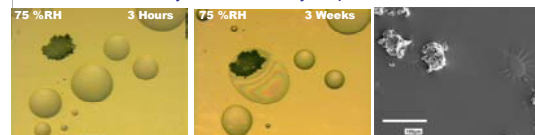


Figure 3: Incomplete re-crystallisation of isolated amorphous lactose particles upon exposures to a relatively high 75%RH.

As a result, a discrepancy exists between %RH_c and induction times of bulk analytical measurements with individual particle studies. Our studies purport that the re-crystallisation mechanism, which only occurs for a small percentage of individual particles, is the catalyst for a chain reaction process for contiguous particles under bulk measurement studies. The decrease in the degree of supersaturation requirements for secondary nucleation suggests that such a process may predominate the re-crystallisation measurements for bulk analytical techniques.

CONCLUSIONS

The combination of optical and scanning probe microscopy measurements has allowed direct visualisation of the re-crystallisation transformation of amorphous particles; and highlights possible discrepancies which may exist between bulk and individual particle measurements for the re-crystallisation processes of amorphous lactose particles.