Physicochemical characterisation of inhalation grade lactose after the removal of intrinsic fines.

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Summary

Lactose is a common excipient in Dry Powder Inhaler (DPI) formulations, used as a carrier for the micronized drug particles. The presence of intrinsic lactose fines in the formulation influences its performance and their role and interactions between the lactose carrier and the micronized drug is still not fully understood. As a first step towards this investigation, "clean" lactose, with removed fines, was produced via wet decantation. Ethanol and isopropyl alcohol have been used in wet decantation, successfully removing lactose fines from the surface of the coarse particles. Differential Scanning Calorimetry (DSC) was employed to show that the powders maintained their crystalline character. Scanning Electron Microscopy (SEM) showed tomahawk-shaped particles in all the powders and some surface alteration occurring after decantation. An airflow titration method using laser diffraction (LDA) allowed the estimation of the removal of fines as well as the particle size distributions, while the non-polar and the polar components of the surface energy of the powders were calculated via Inverse Gas Chromatography-Surface Energy Analysis (iGC-SEA). As both solvents successfully removed fines, we propose the addition of isopropyl alcohol in the list of organic solvents suitable for this purpose.

Key Message

Removal of intrinsic fines from inhalation grade lactose monohydrate can be achieved via wet decantation, using isopropyl alcohol instead of ethanol.

Introduction

Lactose is used as an excipient in a plethora of inhaled formulations; its role is that of a coarse carrier particle bulking agent where the micronized drug particles (usually in the range of 1-5 μm) are attached [1]. The physicochemical properties of lactose, as well as the addition, or absence, of lactose fines has been shown to affect the fine particle fraction of a formulation and as such, its success or failure.[2–4]. However, the underlying interactions that take place between the carrier particles and the drug fines are still not fully understood. In an attempt to shed light in these complex interactions, a step-by-step process has been proposed; clean coarse lactose particles can be prepared via wet decantation [4], in order to produce a "clean" platform where the addition of lactose fines can be achieved in a controlled manner, followed by the addition of the micronized drug.

In this present work we focus on the solvent selection for wet decantation; although Ethanol has been used before for the surface modification of lactose, the use of other organic solvents have not been reported. We investigated the use of isopropyl alcohol, as well as the established ethanol, in wet-decantation and characterised the "clean" lactose powders produced via Laser Diffraction Analysis (LDA), Scanning Electron Microscopy (SEM), Differential Scanning Calorimetry (DSC) and Inverse Gas Chromatography- Surface Energy Analysis (iGC-SEA).

Materials and Methods

Inhalation grade lactose monohydrate, namely Lactohale 100 (sieved) and Lactohale 200 (milled), was provided by DFE Pharma (Goch, Germany). Organic solvents (Isopropyl Alcohol and Absolute Ethanol) were purchased from Fischer Scientific (Loughborough, UK).

Wet decantation was adjusted following the protocol proposed by Islam *et al.*[4]. Approximately 20 g of lactose was dispersed in 500 mL of solvent (ethanol, isopropyl alcohol) in a conical flask. During the first run, the flask was sonicated for 3 mins to make a homogeneous suspension. The suspension was then allowed to settle for 15 min. The supernatant liquid was decanted through grade 1 filter paper, with extra care to minimise the disturbance on the precipitated powder at the lower part of the suspension. The supernatant was filtered and recycled back into the flask as a saturated solvent. The flask was then vigorously mixed (manually) for 2 min prior to the next settling run. The process was repeated until the supernatant liquid was clear. The decantation process was done with extra care to have minimal disturbance on lower parts of the suspension and prevent the removal of larger particles. In the final step the powders were wet-sieved (63 μm) and allowed to air-dry for 24 h.

Particle morphology was investigated via Scanning Electron Microscopy (SEM). The powders were deposited onto adhesive carbon tabs (Agar Scientific G3357N), which were pre-mounted onto aluminium stubs (Agar Scientific JEOL stubs G306). Samples were then sputter coated with gold for 1 minute to achieve a thickness of around 30 nm (Quorum SC7620). The images were acquired using a JEOL 5700 scanning electron microscope, operated at 20kV, and a working distance of 10 mm.

Differential Scanning Calorimetry (DSC) experiments were conducted in a DSC 200 (TA Instruments), calibrated with Indium as a reference material, at a heating rate of 10 ^{o}C/min. In all measurements nitrogen was used as the purge gas, flowing at a rate of 50 ml/min. Tzero sample pans with a pinhole lid were used for all the samples.

Particle size measurements were performed on a Sympatec HELOS/RODOS Laser Diffraction (LDA) unit, using the ASPIROS dispersing system (dispersing aperture diameter 4mm, feed velocity 25 mm/s) (Sympatec GmbH, Clausthal-Zellerfel, Germany). The R5 lens (measuring range 4.5–875 µm) was fitted for the measurements. Powder was filled into the ASPIROS glass vials and was dispersed via vacuum suction. Airflow titration measurements were performed following a previously established protocol [5]. The primary pressure (PP) was manually set using the adjustment valve in the range 0.2–5.0 Bar and three measurements were taken at each pressure setting using freshly loaded powder. Particle size distributions were calculated using the Fraunhofer theory and were analysed in WINDOX 5.3.1.0 software, while further analysis was performed in MATLAB. Particle size measurements for a complete airflow titration curve were conducted on a single day.

Specific surface area (SSA_{BET}) and surface energy (SE) analysis were conducted using an Inverse Gas Chromatography Surface Energy Analyser (iGC-SEA, Surface Measurement Systems Ltd, UK). Approximately 1 g for the lactose was packed into silanised iGC glass columns (internal diameter 4mm). Prior to any measurements, the columns were conditioned using helium carrier gas at 10 scc/min for 2 h at 30 °C and 0% RH. Methane gas was injected at the start and the end of the experiments for the dead volume calculation. SSA_{BET} was calculated via the Brunauer-Emmett-Teller (BET) theory, based on the n-octane adsorption isotherm data [6]. For the surface energy, the columns were equilibrated as mentioned above. Non-polar probes (n-undecane, n-decane, n-nonane, n-octane and n-heptane) and polar probes (dichloromethane, ethyl acetate, ethanol, acetone and acetonitrile) were injected in the column at a range of surface coverages (n/n_m). Following an analysis method previously reported[5], the dispersive (γ_d, non-polar) and acid-base (γ_{ab}, polar) components of the surface energy were calculated using the Dorris-Gray method. All measurements were made in triplicate.

Results

Due to the different production method, milled Lactohale 200 has a higher percentage of fines, as can been seen in Figure 1. After wet decantation the surface fines have clearly been reduced, and a smoother surface can be seen in the larger particles that maintain their tomahawk shape. Solid bridging was observed in the initial wet decantation trials (data not shown), however it was significantly reduced thanks to the wet-sieving final step.

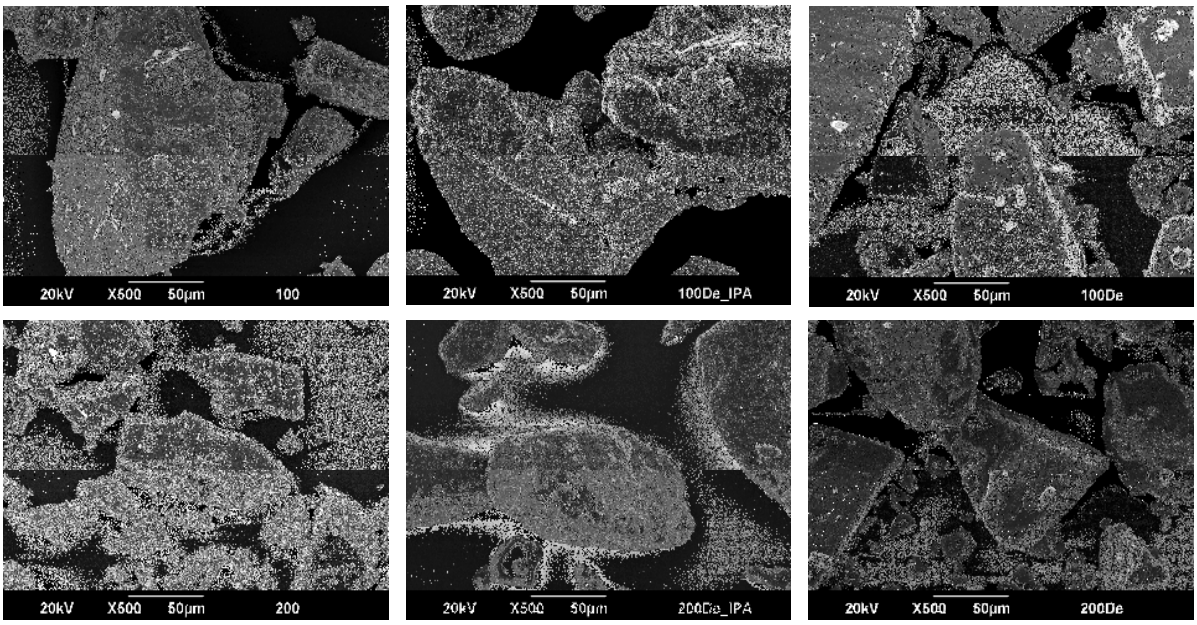

Figure 1 - SEM images of Lactohale 100 (top row left), and Lactohale 200 (bottom row left) decanted with isopropyl alcohol (IPA- middle) with ethanol (right) (x500 magnification). Some surface alteration can be seen using IPA and ethanol but they have both succeeded in the removal of the fines.

The solid-state of the powders was evaluated via DSC (Figure 2). The thermographs are in accordance with the expected α-lactose monohydrate behaviour, exhibiting an endothermic dehydration peak at 147 °C while the melting endotherm commenced at about 207°C [7].

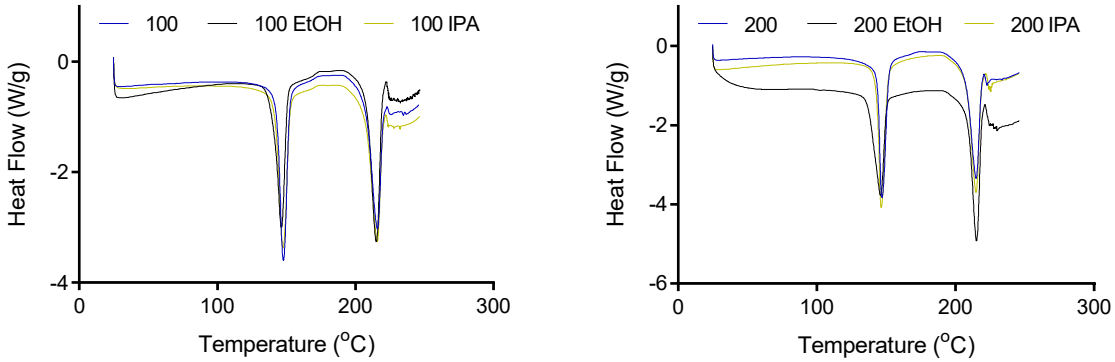

Figure 2 – DSC thermographs of the raw (blue line) Lactohale 100 (left) and Lactohale 200 (right), decanted with ethanol (light brown line) and isopropyl alcohol (black line). The powders exhibit the same thermal behaviour indicating that their crystalline state has not been altered.

LDA measurements were conducted to investigate the particle size distribution of the powders under flow (Figure 3, top row). D_{10} was found to be 55.1 ± 1 for Lactohale 100, 47.9 ± 1.1 for Lactohale 100 – Ethanol, and 66.2 ± 2.4 for Lactohale 100 – IPA. The same trend was observed for the Lactohale 200 powders as well with D_{10} = 13.2 ± 0.8 for Lactohale 200, 19.7 ± 0.5 for Lactohale 200 – Ethanol and 28.2 ± 0.7 for Lactohale 200 – IPA. In order to have a better understanding of the removal of fines, D_2, reporting the particles in the lower 2% of the PSD was measured in the whole pressure range (Figure 3, bottom row). As the pressure increases, D_2 decreases, as any smaller particles, loosely bound to the surface will de-agglomerate a result consistent with literature[5,8]. However, IPA has in both cases, the highest D_2, suggesting a better removal of fines.

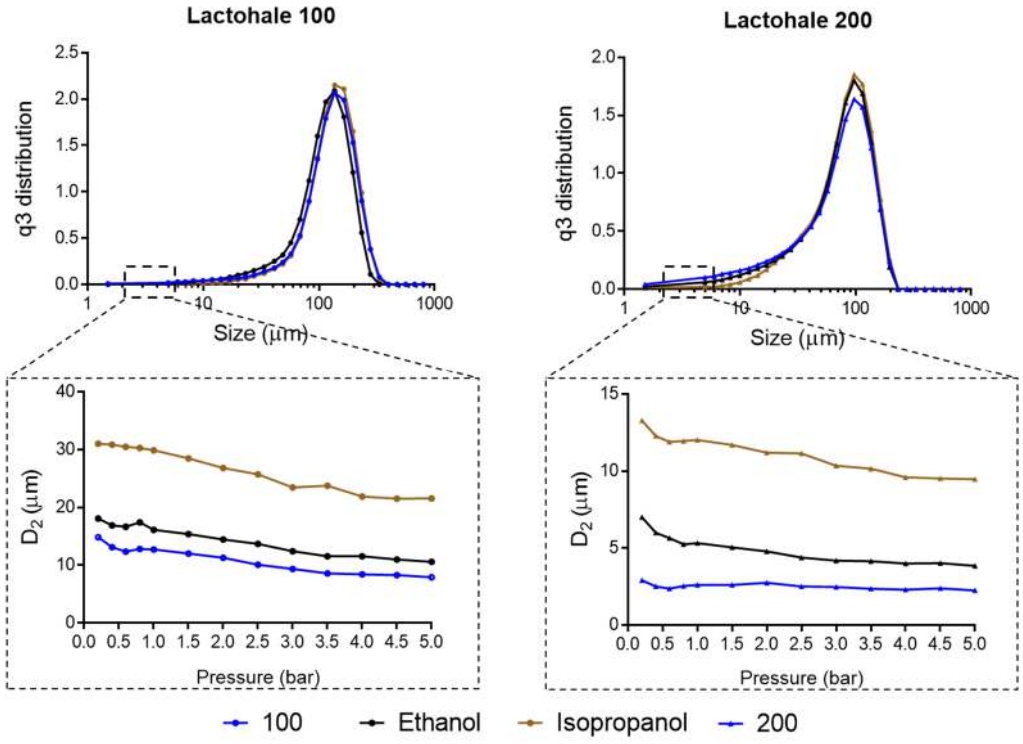

Figure 3 – Top row: Particle Size Distribution of the raw materials and the decanted powders when dispersed at 2.0 bar primary pressure. Bottom row: Airflow titration measurements in the range 0.2-5.0 bar, reporting the amount of particles smaller than the 2% of the distribution (D_2), for Lactohale 100 (left) and Lactohale 200 (right). In both cases, the IPA treated lactose (light brown) has the highest D_2, indicating the highest removal of fines. (70.3% for Lactohale 100 and 84.2% for Lactohale 200).

Surface Energy (SE) analysis was conducted for the Lactohale 200 powders using iGC (Table 1). An increase in the BET_{SSA} of the decanted powders was observed; this could be attributed to surface modification as an effect of the solvents. The highest contribution in the total surface energy is through dispersive forces (45.2 ± 0.2 for Lactohale 200) as has been reported previously[9]. Wet decanted powders exhibit lower SE values; 39.2 ± 0.3 for Lactohale 200-Ethanol and 41.4 ± 0.1 for Lactohale 200 – IPA. The reduction in the SE indicates the successful removal of the high-energy fines from the surface.

Table 1 – Surface Energy measurements via Inverse Gas Chromatograph (iGC-SEA) at 3% surface coverage (n=3, ± SD)

	Lactohale 200	Lactohale 200 Ethanol	Lactohale 200 Isopropyl Alcohol
BET_{SSA} (m^2/g)	0.3071 ± 0.0008	0.391 ± 0.003	0.3703 ± 0.0008
γ_d (mJ/m^2)	45.2 ± 0.2	39.2 ± 0.3	41.4 ± 0.1
γ_{ab} (mJ/m^2)	6.2 ± 0.1	2.59 ± 0.06	3.73 ± 0.05

Discussion

It has been shown in the past that addition of fines in coarse lactose samples results in an overall increase in surface energy [10]. In a similar note, the removal of fines via wet decantation as presented in this work, has resulted in the decrease of the surface energy of the coarse lactose particles. The lower SE of the ethanol-treated lactose indicates that fewer high-energy fines are present in the powder i.e. a better removal of fines in comparison to IPA. This comes in contradiction with what was observed via Laser Diffraction, where the D_2 values of IPA-treated lactose were consistently higher than the others. This could be explained by the remaining fines in the IPA-treated powders not being as readily dispersed under the airflow as the ethanol ones. An alternative explanation is the potential for surface-modification (e.g. localized surface etching and/or dissolution) due to dispersion in IPA solvent. The latter issue is currently being addressed through further surface characterization studies. The importance of iGC-SEA is highlighted, as it can facilitate the understanding of the absence/presence of fines in a formulation. Nevertheless, IPA has demonstrated a similar behaviour to ethanol, and is recommended from the authors as a second solvent of choice for wet-decantation.

Conclusion

This study investigated the use of isopropyl alcohol (IPA) instead of ethanol in wet decantation for the removal of intrinsic lactose fines. Both solvents have successfully removed lactose fines and in doing so a decrease in the Surface Energy of the lactose carriers was observed. Although LDA measurements suggested that IPA had removed more fines than ethanol, the significant difference in the Surface Energy measurements between the two implies that ethanol still removed more fines, while IPA-remaining fines were more strongly adhering to the particles' surface, and were not as easily dispersed under the airflow.

References

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