ASIAN JOURNAL OF PHARMACEUTICAL SCIENCES 10 (2015) 501–512

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Original Research Paper

Mathematical approach for understanding deagglomeration behaviour of drug powder in formulations with coarse carrier

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ARTICLE INFO

Article history:
Received 31 May 2015
Received in revised form 17 August 2015
Accepted 18 August 2015
Available online 24 August 2015

Keywords:
Ternary agents
Deagglomeration
Cohesive powders
Non-linear regression modelling

ABSTRACT

Deagglomeration of cohesive particles in combination with coarse carrier is a key requirement for inhaled formulations. The aim of the project was to propose a mathematical approach to understand aerosolization behaviour of micronized particles alone and in formulation with carriers. Salbutamol sulphate and salmeterol xinafoate were blended separately with fine lactose (ratio 1:4) and fine and coarse lactose (1:4:63.5). Laser diffraction was employed to characterize the powder median particle size. The deagglomeration of micronized materials followed an asymptotic monoexponential relationship. When the coarse lactose was added, the relationship fitted a bi-exponential equation showing an easily and a poorly dispersed fraction. Using model hydrophobic and hydrophilic APIs, this study has demonstrated the utility of an analytical approach that can parameterize deagglomeration behaviour of carrier-free and carrier-based inhalation formulations. The analytical approach provides the ability to systematically study the effect of material, formulation and processing factors on deagglomeration behaviour.

1. Introduction

Micronized particles for inhalation with size between 1 and 5 μm form agglomerates due to the dominance of interparticulate forces. Several dry powder formulations have been marketed without the presence of carrier particles [1,2]. The absence of the coarse particle affects the performance of formulations that contain extensive agglomeration [1,2]. Budesonide (Bud), for example, is more cohesive than salbutamol sulphate (SS), showing bigger agglomerates under the SEM (scanning electron microscope) [3]. It has been suggested [3–5] that FP (fluticasone propionate), like SX (salmeterol xinafoate) and Bud, shows greater agglomeration forces than SS. However, the formation of agglomerates of micronized APIs leads to an increased entrainment of the drug particles but poor blend flowability. To

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Peer review under responsibility of Shenyang Pharmaceutical University.
http://dx.doi.org/10.1016/j.ajps.2015.08.007

address the manufacturing challenge of cohesive agglomerates, drug particles are often blended with coarse lactose carrier to improve flowability, uniformity of dosing and aerosolization. Many drug particles remain agglomerated in such carrier blends. During aerosolization, particles must be removed from the carrier particles and agglomerates must be disrupted in the airflow [6,7].

Micronized material (e.g. FP (fluticasone propionate)) shows highly-cohesive behaviour, forming big agglomerates, especially when carrier is added (median agglomerate size 40.15 ± 6.02 at 90 L min^{-1}) [8]. The authors [8] mixed coarse and fine lactose with the most cohesive batch of FP and the formulation generated a lower fine particle fraction than when the coarse carrier was not used. The authors [8] suggested that it was due to the presence of very large agglomerates of FP in the mixtures. Different types of blending, such as high- or low-shear blending, lead to the re-arrangement of the API on the surface of the carrier. The mechanism of re-distribution of the API on the carrier particles differs from drug to drug (including batch-to-batch variability of the same drug), and depends on how great the cohesive force between agglomerates is [9]. Although all micronized particles are cohesive, when coarse carrier such as lactose is added to the formulation, the micronized material starts to redistribute on the lactose surface exhibiting very different behaviour [8,10,11]. When energy is applied to the formulation (such as inhalation strength of the patient), the cohesive forces between agglomerates would be overcome and they would be released together with the fine material for lung deposition.

Salbutamol sulphate (SS), which has been widely investigated [10,12,13], presents a rectangular or a plate-like shape when analysed under the SEM [13]. Other studies [10,12] suggested that SS exhibits an adhesively balanced behaviour when added to coarse lactose and redistributes evenly on the lactose surface, with lower adhesive energy to the carrier with respect to other particles [10]. This is probably due to the relative balance of cohesive and adhesive interparticulate forces in binary (and ternary) formulations with carriers. SS showed a lower cohesive energy compared to Bud, with SX also reported to exhibit a balance in favour of cohesion when added to lactose [10,14,15]. The interaction between lactose and drug particles has to be strong enough to prevent segregation. However, upon aerosolization, the drug must be easily detached to allow lung deposition.

The design of an inhaler and the inhalation strength of the patient affect the geometries and forces available for powder entrainment and drug dispersion. The forces of interaction within the powder formulation additionally play a role in the aerosolization. Studies have demonstrated that the strong adhesion between API and coarse carrier decreased the drug dispersion [11,16]. Enhancement of the dispersion process (e.g. aerosol fraction of fine particles) and modification of interparticulate forces within the powder formulation by the presence of ternary agent have been suggested [17,18]. Adi et al. [11] showed that adding fine lactose improved the dispersion process of SX from the coarse carrier, because the interaction between the fine lactose and the SX produced mixed agglomerates. It may be the decreased drug/carrier adhesion energy or the easier break-up of mixed agglomerates with low carrier adhesion that enhances dispersion efficiency.

It has been suggested that the addition of fine lactose would increase the respirable fraction [19], whilst the presence of coarse lactose improves the flowability of the formulation. However, Behara et al. [20] showed that the presence of coarse carrier is not necessary. The aerosolization was improved by doubling the ratio of fine lactose to SS (from 1:4 to 1:8). They suggested that increasing the concentration of fine lactose reduced the agglomerate strength between SS and fine lactose [20]. Studying the impact of drug type or the properties of co-formulated excipients on deagglomeration with a focus on blend segregation into drug or drug-excipient agglomerates represents a 'blend structure' treatment of deagglomeration. However, focusing blend segregation (which can be assessed through studying the effects of content uniformity on dispersion during aerosolization [21]) ignores the fundamental causes of blend structure (or restructuring). These include the effects of the order of material addition in blending [22], of electrostatic charge [23] and tribo-electrification during blending, the blending energy and duration of blending [21,24] that are employed, and of course inhaler device design itself and tribo-electrification [25] behaviour of the aerosol that is being dispersed from the device. However, it remains of merit to consider the resultant interparticulate forces between formulation components, the balance of the agglomerative and deagglomerative forces applied during blending with respect to eventual deagglomeration performance of the blend. The adhesion forces between drug and carrier consist mainly of inter-particulate forces such as van der Waals forces, electrostatic charges, and capillary interactions and influence aerosol dispersion from dry powder inhalers (DPIs) [26].

Inverse gas chromatography, centrifugation and atomic force microscopy have been used to characterize adhesion forces between particles [27,28]. However, such techniques do not quantify the cohesion of an actual powder bed as it only measures the interaction between one drug or excipient particle and the probe of the AFM.

Other studies investigated the interaction between individual particles using atomic force microscopy (AFM) [27,28] and suggested CAB (cohesive-adhesive balanced) approach. CAB approach calculated the ratio between API agglomerates and adhesion of API to the carrier. It suggests that CAB ratio < 1 the drug is more adhesive to the carrier than to itself, whilst a CAB > 1 indicates that more drug agglomerates can be found in the formulation, leading to a variable content uniformity [5]. However, the technique does not quantify the adhesion force of the powder bed as it only measures the interaction between one particle or excipient and the probe of the AFM. Other techniques have been proposed to study the interaction between particles. For instance, inverse phase gas chromatography [29] showed potential to understand the interaction between carrier and API, where the formulation is placed inside a packed column and a gas (e.g. helium) is passed through it. The API with higher affinity to the carrier would elute later than the one with less affinity. This is due to the higher surface energy between API particles and carrier. Studying the retention of a range of probes with different polarity and chemistry, the cohesion strength can be calculated [29]. Other techniques to study the different behaviour of blends under applied forces are powder rheometry (where flow properties are studied by studying the movement of an impeller through blends under a

specified force, such as torque) [30] and centrifugation of micronized material [31].

Laser diffraction techniques have been used for dry and liquid dispersion systems to study powder deagglomeration [32]. Airflow and pressure drop titration experiments have been performed [4,32] to assess the change in particle size when a fine powder bed is subjected to an increasing pressure drop and empirical models have been proposed for micronized particles with or without the common carrier coarse lactose [4,32,33].

When only micronized APIs are present in the formulation alone or in presence of fine lactose, studies have suggested that the liberation of fine particles <6 μm shows a sigmoidal trend with increasing airflow rate and a three-parameter sigmoidal equation has been used [32]. The latter relationship suggested that parameters such as maximum extent of deagglomeration or flow rate required to achieve 50% deagglomeration could be used to predict powder deagglomeration. However, the problem occurs when the coarse lactose is added to the formulation. Higher flow rates are required to deaggregate the mixture fully than if the lactose carrier were not present. Therefore, more energy is required to fully aerosolize micronized APIs away from the surface of the carrier. An equation similar to the Langmuir adsorption isotherm was proposed with the name of Powder Aerosol Deaggregation Equation [33]. However, the authors suggested the use of a cascade impactor together with a dry powder inhaler for the experiments that would affect the deagglomeration of the particles. The use of cascade impaction is not a rapid method to guide formulation choice in early phases of product development. Previous studies have indicated the benefits of a rapid methodology using laser diffraction to guide formulation of DPIs. However, the models proposed to explain the deagglomeration process did not show any parameter with a physical meaning. The aim of this study was to develop a data modelling approach to characterize and parameterize powder deagglomeration response to airflow for cohesive micronized particles alone or in formulation with coarse and fine carrier. This would be helpful to predict deagglomeration mechanisms of novel formulations or following different pharmaceutical processing.

2. Materials and methods

2.1. Materials

Micronized salbutamol sulphate (SS, batch number B027798) was obtained from GlaxoSmithKline Research and Development (Ware, UK). Salmeterol xinafoate (SX) was purchased from Vamsi Labs Ltd (Solapur, India). Lactose monohydrate, Span 80 and cyclohexane were purchased from FisherScientific (Loughborough, UK). Fine lactose (LH300) was donated from Friesland Foods (The Netherlands). Adhesive carbon tabs and aluminium pin stubs were purchased from Agar Scientific Ltd (UK).

2.2. Blend preparation

Lactose monohydrate (Fisher Scientific, UK) was sieved to separate the 63–90 μm particle fractions using a mechanical sieve shaker (model – AS 200 digit, Retsch, Germany) with the 90 μm sieve and the 63 μm sieve. It was operated for 5 min at 100 amplitudes. The fraction of lactose between the sieves was collected to prepare the blends. A total of 3 g of blends were prepared. The drugs were blended separately with either fine lactose (FL) in ratio 1:4, coarse lactose (CL) in ratio 1:67.5 or combination of both in ratio 1:4:63.5. The drug was sandwiched with coarse lactose (CL) in equal volumes. When the FL was used, it was first blended with the drug and the pre-mixture sandwiched with coarse lactose. Glass vials were used for the blending and vortexed (Vortex Genie 2, model G – 560E, Scientific Industries LTD, New York) for 2 min. The blending was carried out in a Turbula mixer (Turbula 2583, type +2C, Glen Mills, Clifton) for 40 min.

2.3. Particle size analysis

Salbutamol sulphate (SS), fine lactose (FL), salmeterol xinafoate (SX) and coarse lactose (CL) were tested for the homogeneity of the particle size using the laser diffraction technique. The Sympatec Rodos module (Inhaler Helos/KF, Sympatec Limited, Bury, UK) with Aspiros feeder was used. The speed of the feeder was set at 25 mm/s. The pressure drop was set between 0.1 and 5.0 Bar in order to titrate the dispersion pressure. The PSD (particle size distribution) was collected every 0.1 Bar when tested from 0.1 to 1.0 Bar and every 1.0 Bar when tested from 1.0 to 5.0 Bar. The duration of the measurements was 5 s. The optimum concentration of the powder to be detected was in the range of 1.0 and 1.1%. The technique was used to detect the particle size of the blend as a dry dispersion.

A liquid laser diffraction technique using a Malvern Mastersizer X (Malvern Instruments Ltd, UK) was also used to determine the 'true' fully dispersed particle size. The blends were suspended in 0.5% (w/v) Span 80 in cyclohexane (for SX blends, SX, FL, CL) and 1% (w/v) Span 80 (for SS blends and SS). Malvern Mastersizer X (Malvern Instruments Ltd, UK) was used with 100 mm focal length lenses (0.5–180 μm) and MS7 magnetically stirred cell. Prior to measurement, the solvents were saturated with the appropriate particles and sonicated for 30 min followed by overnight stirring. For the measurements, approximately 1 mg of powder was added to 2 mL filtered dispersant (0.2 μm cellulose acetate syringe filter, Sartorius Stedim Biotech., UK) and sonicated (Sonicleaner, DAWE, Ultrasonics Ltd, USA) for 1 min for SS, FL and CL and 5 min for SX. The stirring was set at 3 min for all the powders with sweeps of 2500 for SX and 3500 for the other powders. A background reading was taken and the suspension was added to the sample cell until the obscuration was 10–30%. Following equilibration (60 s for SX and 30 s for the others), ten individual measurements were taken for n = 3 samples. From the instruments, Dv_{10}, Dv_{50}, and Dv_{90} (corresponding to the cumulative percentage particle undersize values for 10%, 50% and 90% of the particles by volume), and the % volume <5 μm were used for analysis.

2.4. Particle morphology assessment

Powder samples were transferred to adhesive carbon tabs, mounted onto aluminium pin stubs (Agar Scientific Ltd, England). To assess blend interactions, scanning electron microscopy (SEM, Jeol Carry Scope JCM 5700, Welwyn Garden City,

UK) was performed. Samples were sputter coated with gold under argon for 4 min to achieve a thickness of approx. 30 mm using an Emitec SC 7620 coater (Quorum Technologies Limited, West Sussex, England). To view the particle morphology, the SEM was operated at 15 kV in low vacuum mode and a working distance of 13 mm.

2.5. Equation of deagglomeration and mathematical model

The % relative deagglomeration for micronized particles was calculated using the model proposed by Behara et al. [32] in Equation (1).

$$\%RD = \frac{\%volume < 5\mu m(dry\ dispersion)}{\%volume < 5\mu m(liquid\ dispersion)} * 100 \quad (1)$$

where % RD is the relative deagglomeration.

2.6. Statistical analysis

Statistical analysis was performed in Minitab (version 15) using one-way ANOVA and post-hoc Tukey's test (multiple comparisons) or Student's two-tailed t test for pair-wise comparisons, both at 95% confidence intervals. Non-linear regression analysis of the powder dispersion data was performed using OriginPro (ver. 8) and MatLab software (ver. R2013a).

3. Results and discussion

The aim of the current study was to propose a model in order to understand the deagglomeration mechanisms of micronized material for inhalation in combination with fine and/or coarse carrier. The design of an inhaler and the inhalation strength of the patients allow the dispersion of inhaled formulations and lung deposition. However, upon aerosolization, the API (active pharmaceutical ingredient) might deaggregate in a different manner depending on the presence or absence of a coarse carrier or also on its distribution on the lactose surface prior to release from the inhaler [4]. Coarse lactose is well known to be used to improve aerosolization and flowability of micronized drug [12,34]. However, a strong adhesion of the API to the carrier would decrease the drug dispersion [11,16]. Therefore, ternary agents (or fine lactose) are usually added to the formulation to compete with the drug on the active sites of the carrier. Saturating high adhesive-energy surface sites, the drug can be detached easily from the carrier during aerosolization [34,35]. Behara et al. [20] showed that the presence of coarse carrier was not necessary to improve aerosolization. The addition of lactose fines in increasing concentration successfully reduced the cohesive strength of drug agglomerates, leading to better aerosolization. Moreover, Islam et al. [36] indicated that SX dispersion increased as the amount of FL increased. Fine lactose and cohesive micronized particles created agglomerates [20].

Using laser diffraction techniques, the effect of the addition of either fine lactose or coarse lactose to the API was shown [36]. On the other hand, adding coarse carrier to the mixture shifted the distribution to a larger size range [36]. Studying the deagglomeration process of the dispersed powder at different flow rates has featured widely in the literature. A few studies have investigated the deagglomeration trend of either the powder bed [4] or the aerosol emitted from a Rotahaler [32]. These studies proposed a quick methodology to assess deagglomeration of the formulation upon increasing flow rate or pressure drop. Laser diffraction analyses such as Sympatec and Malvern instrument [4,32] were used. However, the studies seem to focus on the fine particles, rather than on the formulation with coarse carrier.

3.1. Laser diffraction analysis

In order to understand the deagglomeration mechanism, blends with either SX or SS were manufactured in-house with different grades of lactose and their aerosolization was studied using laser diffraction techniques. All formulations were regarded uniformity (%CV < 10%); however, content uniformity was poorer for blends with fine lactose (Table 1). For the purpose of the study and to understand the different deagglomeration mechanism of SX and SS, the former was considered cohesively-balanced when added to the lactose, due to its tendency to create agglomerates on the carrier surface [15]. On the other hand, SS was considered due to its adhesively balanced behaviour when added to lactose as previously described [12]. The concentration of the SS and SX was 1.48% w/w and they were blended with either FL (1:4), coarse lactose (1:67.5) or both (1:4:63.5).

When the APIs were dispersed using the Sympatec/RODOS for pressure drop titration, both SX and FL showed a bi-modal

Table 1 – Particle size distribution for pure material and blends of salbutamol sulphate (SS) and salmeterol xinafoate (SX) with lactose using liquid dispersion laser diffraction (mean ± SD, n = 3) and content uniformity for the inhalation blends (coefficient of variance (%CV) of n = 6 determinations). FL = fine lactose, CL = coarse lactose.

Materials	Ratio	Dv_{10} (μm)	Dv_{50} (μm)	Dv_{90} (μm)	Volume % <5 μm	Content uniformity (%CV)
SX	n/a	0.90 ± 0.01	3.01 ± 0.05	7.40 ± 0.33	76.15 ± 1.22	n/a
SS	n/a	1.38 ± 0.03	3.63 ± 0.05	10.02 ± 0.40	66.15 ± 0.56	n/a
LH300	n/a	1.51 ± 0.01	3.40 ± 0.07	6.76 ± 0.13	75.71 ± 1.25	n/a
SS:CL	1:67.5	4.96 ± 3.11	53.42 ± 8.75	92.76 ± 2.83	13.36 ± 6.87	1.31
SS:FL:CL	1:4:63.5	4.60 ± 1.27	57.36 ± 4.07	95.24 ± 1.68	11.45 ± 2.74	1.35
SS:FL	1:4	1.38 ± 0.04	3.60 ± 0.16	9.03 ± 1.07	68.76 ± 2.69	5.94
SX:CL	1:67.5	4.79 ± 3.65	56.05 ± 3.98	89.32 ± 11.57	12.50 ± 3.98	0.32
SX:FL:CL	1:4:63.5	2.11 ± 0.43	43.35 ± 12.68	84.82 ± 8.61	22.87 ± 7.76	5.32
SX:FL	1:4	1.23 ± 0.03	3.15 ± 0.06	6.73 ± 0.23	77.89 ± 0.82	3.17

PSD at 0.5 Bar (Fig. 1A and 1B). The same trend was seen also for SS that exhibited a similar behaviour at 1 Bar (Fig. 1C), suggesting a greater energy is required for the deagglomeration mechanism to occur for SS compared to SX and FL. However SS, SX and FL showed a bell-shaped distribution at higher pressure drops as confirmed in previous studies [22,37]. When FL was added to the APIs, a more consistent distribution was seen for both drugs when the pressure was increased. However, SS–FL deagglomeration at low pressure (0.5 Bar) did not occur completely (shoulder in the large particle size range in Fig. 2A). The same trend was seen by Jaffari et al. [4] with SX, and by Behara et al. for SS:FL [32]. However, the bi-modal distribution was not present for SS:FL at 1 Bar unlike for SS alone, probably due to the addition of the FL leading to disruption of SS agglomerates and enhanced deagglomeration [20].

When the formulations containing either CL or FL:CL were analysed, the lactose carrier showed a distribution in the 60–90 μm range (Fig. 3A). However, in blends also containing FL, a shoulder was seen in the micronized region due to a higher amount of fine particles present in the mixture (Fig. 3B). The formulations showed a bi-modal distribution due to the presence of easily-dispersible agglomerates (small shoulder in the respirable range) and a poorly-dispersible fraction of drug still attached to the carrier (large shoulder) [4,35].

Liquid dispersion laser diffraction was used to determine the fully dispersed particle size of the API and blends. The results confirmed the dry dispersion trend of the blends as the Dv50 of the blends with the carrier were shifted to higher values than when the drugs were blended with fine lactose only (Table 1). On the other hand, the micronized API possessed a PSD within the micron size range (Table 1).

3.2. *Empirical model*

The DV_{50} was plotted for all micronized material and formulations (Fig. 4A) and the descending part of the graph prior the plateau shows different deagglomeration behaviour between particles and formulations (e.g. SS vs. SX and FL in Fig. 4A). The difference, especially at low pressure drops, suggests variability in agglomerate size and strength as suggested from the density distributions of size in Fig. 1. The % relative deagglomeration (equation 1) was also plotted (Fig. 4B) and SX reached the maximum deagglomeration earlier than SS, which on the other hand, did not reach a constant particle size at the highest pressure (Fig. 4B). When FL was added to both APIs, an increase in the degree of deagglomeration was seen at 2 Bar, which then decreased at 3 Bar (Fig. 4B). This was probably due to the instability of the formulation at high pressure drops.

The % relative deagglomeration for micronized particles and micronized formulations showed a mono-exponential trend when a pressure drop was applied to the agglomerates. The trend could be explained by Equation (2) which is a modification of the Johnson–Mehl–Avrami–Kolmogorov (JMAK) equation [38]:

$$y = m\left(1 - e^{-(x/x0)^n}\right) \quad (2)$$

Due to the high variability in the replicate measurements, an artificial error was added when normalizing the data.

The deagglomeration mechanism depends on the distribution of the APIs either alone or in combination with FL in formulation. The slope of the curve (Table 2) is the maximum gradient and represents how readily the deagglomeration process occurs before the drug reaches the plateau size (deagglomeration sensitivity). The plateau size represents the maximum deagglomeration ("m" parameter in Table 2 defines the positive asymptote of the curve) which varies with the type of particle (Table 2). This was in accordance with Jaffari et al. [4], where formulations with fine particles reached a monoexponential plateau when the pressure drop was increased. SS showed the lowest maximum gradient (indicating a deagglomeration process that is resistant to increased airflow) when pressure is applied compared to the other micronized materials (Table 2, $P < 0.05$). The difference, especially at low pressure drops, suggests variability in agglomerate size (i.e. x0 in Equation 2) and strength between agglomerates [5,14]. In order to compare the drugs and blends, the critical pressure point (CPP), the pressure point corresponding to reaching 95% of the maximum deagglomeration, was calculated. SS started to reach the plateau at 1.25 ± 0.19 Bar, at a much greater pressure than SX or FL (Table 2, $P < 0.05$). However, when FL was added to SS, the CCP_{95} was reached at lower pressure than when SS was analysed alone (0.84 ± 0.08 Bar, $P < 0.05$) and the greater maximum gradient was seen for FL than SS (Table 2, $P < 0.05$). The maximum gradient is a vector of the derivative of the function. This indicates the rate of ascent of the curve, therefore, the rate of the degree of deagglomeration. If a small tangent is drawn in a selected point (in this case x0;y0), then the maximum gradient refers to the slope of the tangent.

The enhanced degree of deagglomeration seen for SS due to addition of FL could be explained by creation of mixed SS-FL agglomerates of lower cohesive strength than SS-agglomerates [20,32]. Therefore, the cohesion forces between API (or API–FL) agglomerates are reduced. SX:FL had a poor deagglomeration compared to FL or SX, suggesting that the addition of FL to very cohesive agglomerates does not improve the deagglomeration mechanism [39]. In accordance with this theory, Adi et al. [15] showed that increasing the shear pressure to formulations of SX containing FL with different size range (3.0 and 7.9 μm) was required especially for formulation containing FL 3.0 μm. This might be a result of segregation in the formulation. The parameters 'x0' and 'n' are interdependent. The parameter 'x0' has a similar meaning to the parameter proposed by Behara et al. [32]. The authors used a Rotahaler with Spraytec to study the deagglomeration behaviour of either SS or FL. They suggested that the parameter was the dispersion energy required to overcome internal interaction of the agglomerates. In the current work we suggested that 'x0' could be seen as an inflection point and therefore the pressure at which the deagglomeration attempts to reach the maximum point. In accordance with Behara et al. [32], the blends with fine particles and FL alone showed similar 'x0' values (Table 2, $P > 0.05$) with the exception of SS. The parameter 'n' is the trend of the first part curve between 0.1 and x0 Bar. SS:FL exhibited the highest deagglomeration exponent whilst SX:FL showed the smallest. This indicates a different deagglomeration mechanism when the fine lactose overcomes SS–SS and SX–SX interactive forces. Moreover, the blend

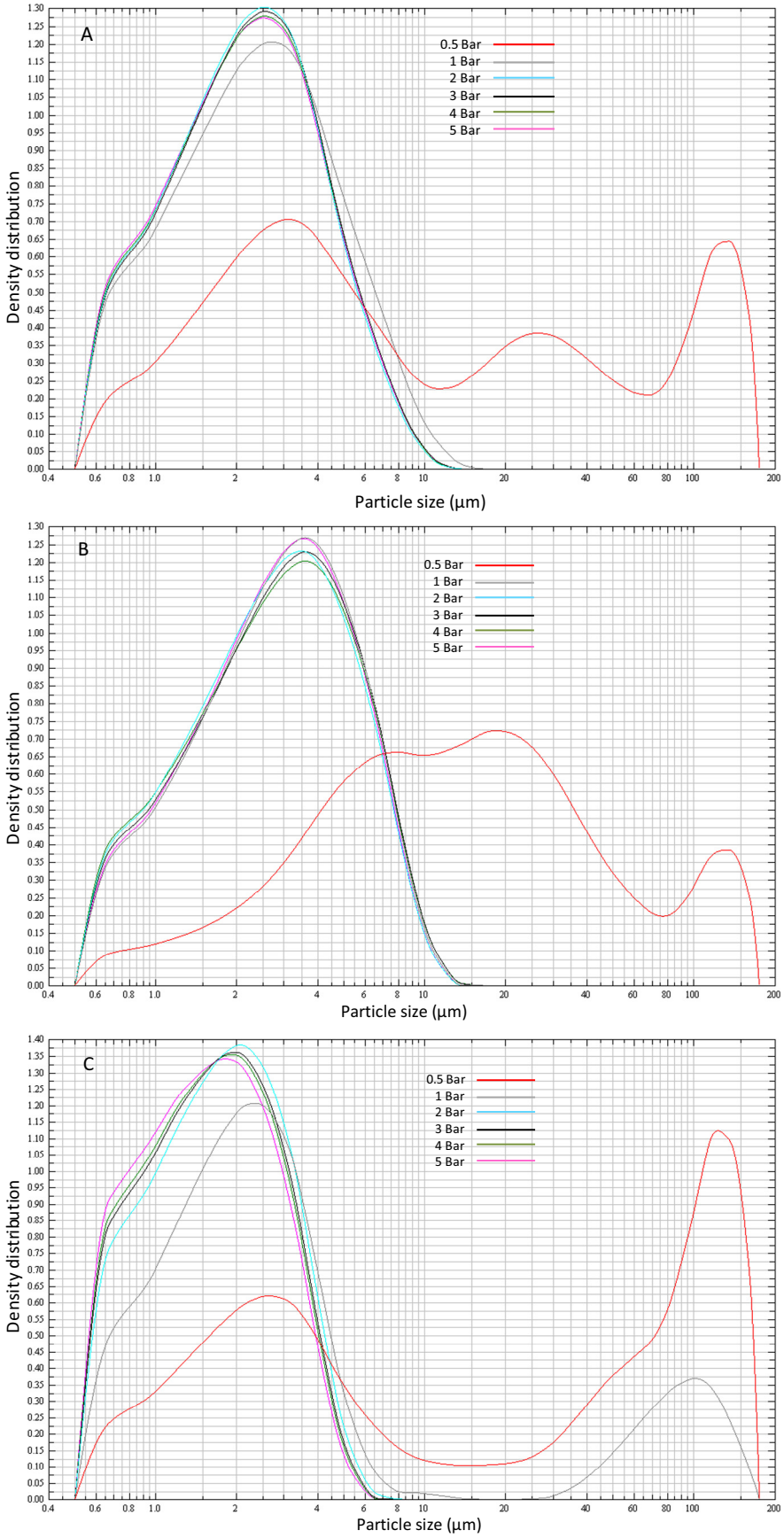

Fig. 1 – Density distribution of size of (A) salmeterol xinafoate, (B) fine lactose LH300 and (C) salbutamol sulphate between 0.5 and 5 Bar.

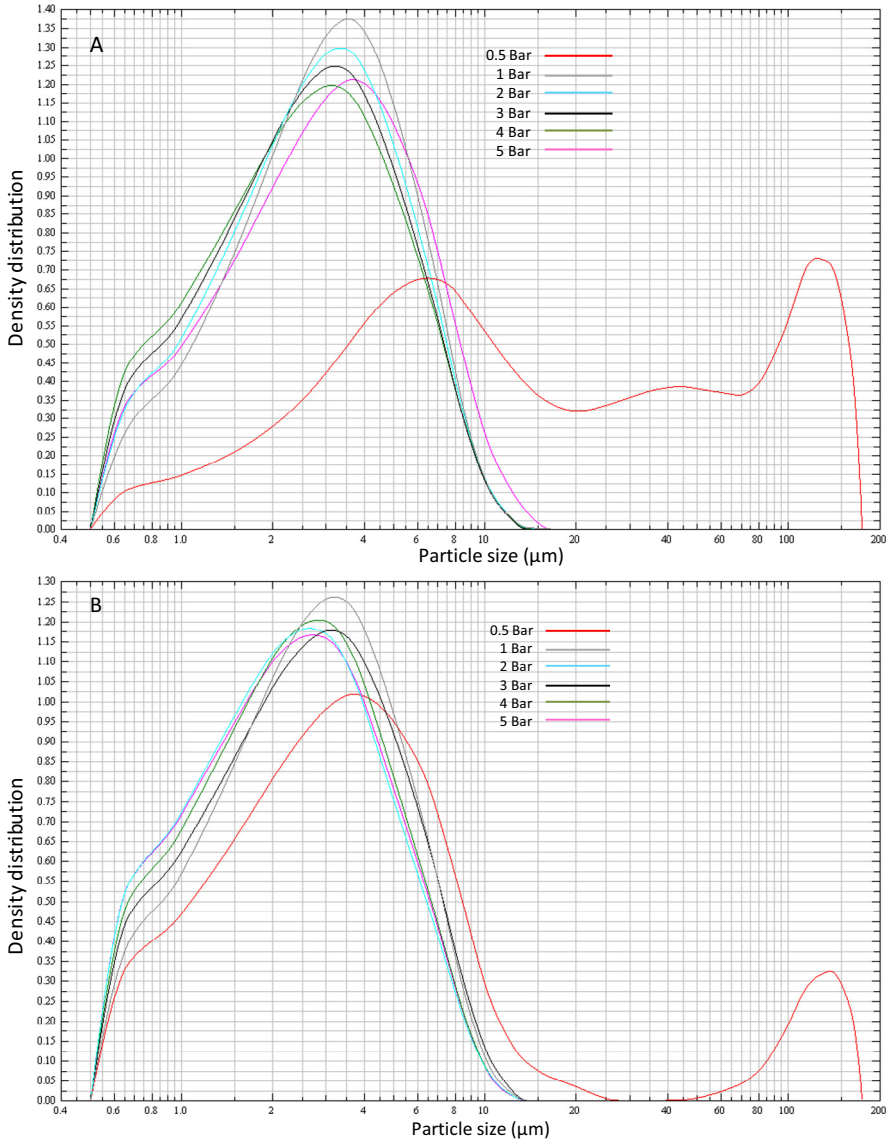

Fig. 2 – Density distribution of size of (A) salbutamol sulphate:fine lactose blends and (B) salmeterol xinafoate:fine lactose blend between 0.5 and 5 Bar.

structure plays a role. Adding the fine lactose would modify the adhesive strength between particles and enhance the detachment of the API (Table 2, maximum gradient was greater for SS:FL than SX:FL).

When CL was added to the formulation, the plateau was not reached for the blends containing CL. Instead, the formulations showed a bi-exponential trend (Fig. 5) highlighting an easily dispersed fraction and a second poorly dispersible fraction of respirable particles. This response is described by Equation (3).

$$y = A\left(1 - e^{-(x/x1)}\right) + H(x - x_w)(100 - A)\left(1 - e^{-(x/x2)}\right) \quad (3)$$

'A' represents the amplitude of the easily dispersed fraction (i.e., extent of deagglomeration), whereas 100 – 'A' represents the deagglomeration extent of the poorly dispersed fraction; 'x_w' is the pressure when the poorly dispersible fraction starts to deagglomerate; 'H' is the Heaviside-function (1 if $x > x_w$, 0 if $x < x_w$); x_1 and x_2 are pressure points when the deagglomeration response is at maximum for the easily and poorly dispersed fractions, respectively.

The bimodal distribution of certain blends was attributed to the presence of coarse carrier in the formulation that, when pressure was applied led to a release of easily dispersible agglomerates (fine particles) and a poorly dispersible fraction of large cohesive agglomerates. The trend of the latter seemed to be the same for all the formulations. However, differences were observed in the initial exponential region of the deagglomeration curves of the blends. SS:CL seemed to have the steepest curve compared to the other API:CL blends. As expected, API:FL:CL blends showed a steeper slope in the first fraction than API:CL blends (Fig. 5). They showed also a greater % of deagglomeration expressed by the parameter "A" in equation (3). SX:FL:CL for example showed 170.97 ± 12.74 bar^{-1} and SX:CL presented 71.64 ± 7.42 bar^{-1}. At high pressures there was an increase in the release of fine particles. The presence of a

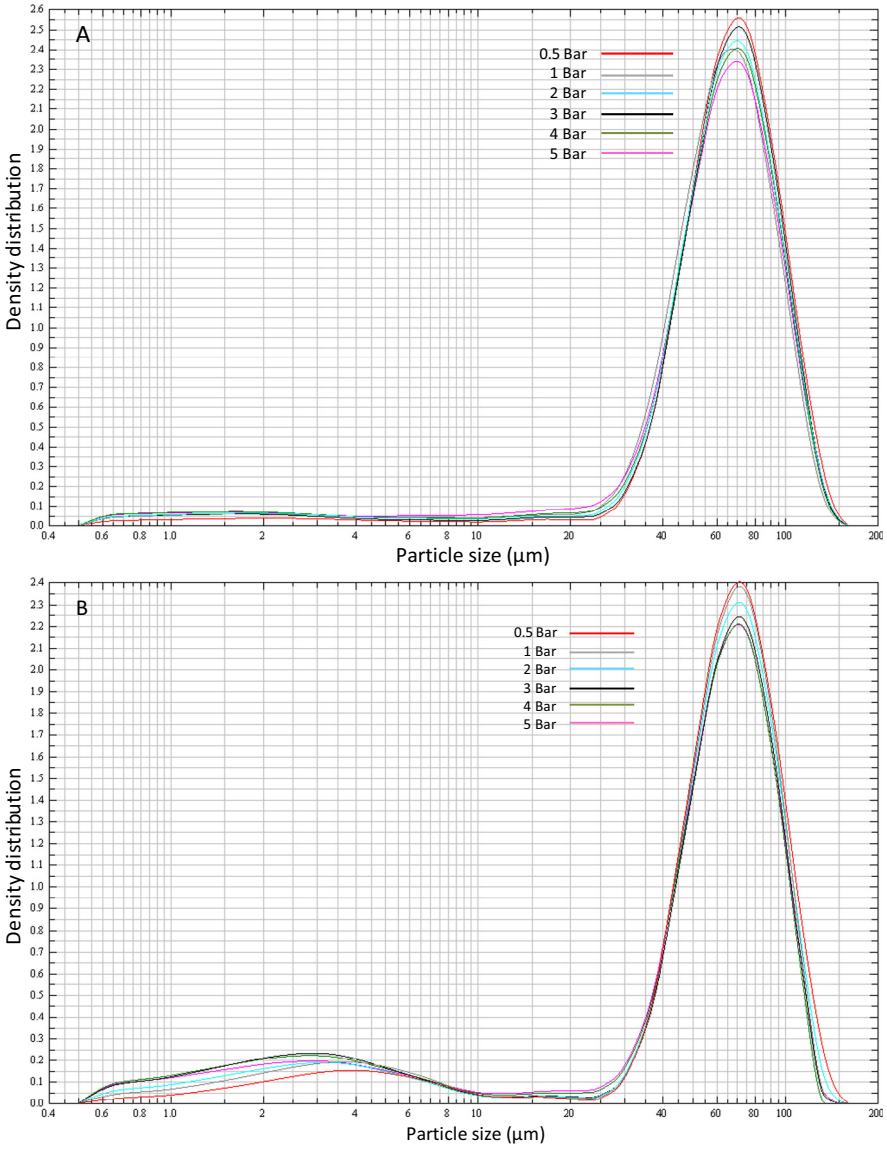

Fig. 3 – Density distribution of size of (A) salbutamol sulphate:coarse lactose blend and (B) salmeterol xinafoate:fine lactose:coarse lactose blend between 0.5 and 5 Bar.

ternary agent would help the detachment of the micronized API from the surface of the carrier, resulting in greater respirable fraction for the patient than when the formulation was composed of CL only [39] (higher 'A' parameter, Table 3). The greatest 'A'-value was observed for SS:FL:CL (Table 3). Jones et al. [39] also confirmed improved detachment when alternative ternary agents were added to binary formulations by showing an increased mode in the respirable fraction for Bud ternary erythriol-lactose formulations.

Equation (3) shows the Heaviside function (H) which is usually used in engineering to represent a parameter that changes abruptly at specified values of time t. In the current model the step function (or Heaviside function) was applied to the bi-exponential trend. A small plateau can be observed in Fig. 5, between 1.0 and 2.0 Bar, between the end of the deagglomeration of the easily dispersed fraction and the beginning of the deagglomeration of the poorly dispersed fraction [40]. 'H' was applied to the deagglomeration mechanism of the blends with coarse carrier, where the "H" parameter of the function was 1 if $x > x_w$, with x_w being the pressure when the poorly dispersed fraction starts to deagglomerate (the delayed unit), or 0 if $x < xw$. All the formulations with the exception of SX:CL showed the plateau between the easily and the poorly dispersed fraction, with SS:CL showing the highest pressure value. This suggested that this particular formulation required the highest pressure to deagglomerate. The addition of FL led to a greater deagglomeration as the pressure (x_w) decreased by 1 unit. Furthermore, the corresponding % deagglomeration to xw was the highest between all the formulations (Table 3).

3.3. *Particle morphology*

Microscopy was employed to examine blend structure. Unlike what some studies have suggested [4,10], SS did not exhibit a plate-like shape (Fig. 6A), whereas both APIs created elongated agglomerates without clear differentiation in their shape

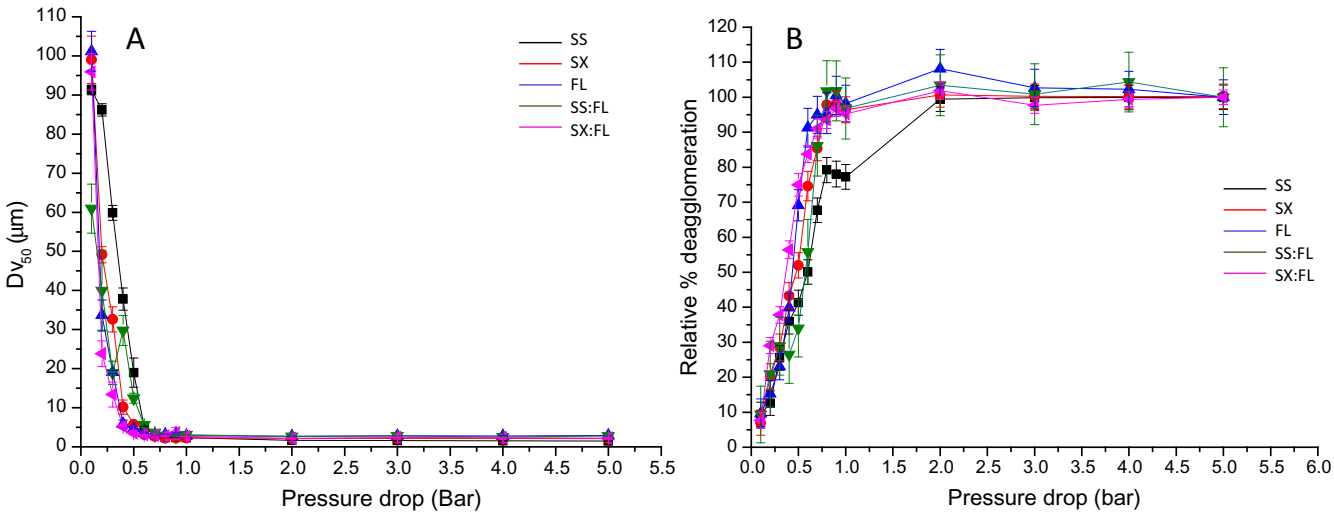

Fig. 4 – Graph of (A) median particle size and (B) deagglomeration behaviour as a function of pressure drop for micronized salmeterol xinafoate (SX, red), salbutamol sulphate (SS, black), fine lactose (FL, blue), salbutamol sulphate:fine lactose (SS:FL, green) and salmeterol xinafoate:fine lactose (SX:FL, pink) (mean ± SD, n = 3).

Table 2 – Mono-exponential deagglomeration parameters for salbutamol sulphate (SS), salmeterol xinafoate (SX), fine lactose (FL) and SS–FL or SX–FL blends.

Materials	m (%)	x0 (bar)	n	Maximum gradient (bar^{-1})	CPP_{95} (bar)	R^2
FL	101.99 ± 3.00	0.49 ± 0.02	2.65 ± 0.35	289.05 ± 32.08	0.74 ± 0.05	0.98274
SX	101.18 ± 2.19	0.53 ± 0.03	2.03 ± 0.27	194.27 ± 9.67	0.90 ± 0.05	0.98606
SS	99.89 ± 2.61	0.69 ± 0.04	1.53 ± 0.17	108.88 ± 5.06	1.25 ± 0.19	0.98587
SS:FL	103.40 ± 5.40	0.61 ± 0.04	2.71 ± 0.71	230.20 ± 34.34	0.84 ± 0.08	0.93928
SX:FL	99.40 ± 1.60	0.43 ± 0.01	1.68 ± 0.12	207.35 ± 6.1	0.82 ± 0.04	0.99360

m = positive asymptote.
x0 = pressure value in the x axis which takes a value of $n \geq 0$.
n = deagglomeration exponent.
Maximum gradient = slope of the curve.
CPP_{95} = critical primary pressure where the degree of deagglomeration has reached the 95% of its final value.
R^2 = value of the best fitted line.

(Fig. 6). Moreover, when added to coarse lactose, no difference in the API distribution was seen on the surface of the carrier (Fig. 7). This particle behaviour is different to what was proposed previously in the literature. Begat et al. suggested that SS distributes evenly on the surface of the carrier [10]. SX created agglomerates due to the fact that SX–lactose blends are dominated by SX cohesive interactions [11]. However, this was not shown in the current work. Nevertheless, different redistribution behaviour of the two different APIs was clearly seen when FL was added to the API–CL mixtures (Fig. 8). The addition of FL to SX:CL (Fig. 8B) led to more agglomerates of FL–SX being heterogeneously distributed on the lactose surface, presenting larger agglomerates on the extremities of the carrier compared to when FL was absent in the formulation. This was probably due to the weak adhesion of FL to the carrier surface, and therefore, its tendency to agglomerate with SX [35]. This suggests that SX–FL have stronger adhesion than FL:FL. Adi et al. [35] showed that when the concentration of fines (i.e. SX and FL) was increased, a better detachment of agglomerates from the lactose was observable because of saturation of the active

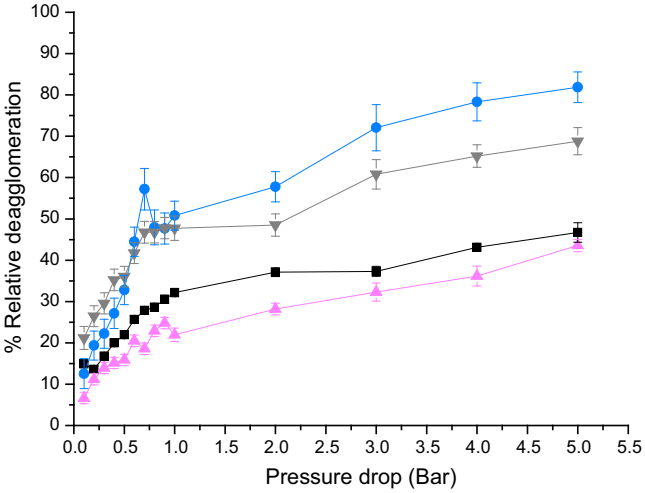

Fig. 5 – Bi-exponential distribution of the % relative deagglomeration for salbutamol sulphate and salmeterol xinafoate blended with coarse lactose (SS:CL black, SX:CL pink) and with fine lactose (SS:FL:CL blue, SX:FL:CL grey) (mean ± SD, n = 3).

Table 3 – Parameters from a bi-exponential equation for salbutamol sulphate (SS) blends (FL = fine lactose, CL = coarse lactose) and salmeterol xinafoate (SX) blends.

Blends	A (%)	x_1 (bar)	x_2 (bar)	$x_w;y_w$ (bar;%)	R^2
SS:CL	37.6 ± 0.7	0.5 ± 0.0	11.2 ± 2.8	3.0 ± 0.3; 37.4 ± 0.8	0.81143
SS:FL:CL	61.4 ± 2.6	0.6 ± 0.1	3.7 ± 1.0	2.0 ± 0.3; 59.8 ± 6.4	0.91714
SX:CL	17.8 ± 2.8	0.3 ± 0.1	14.0 ± 1.0	0;0	0.88214
SX:FL:CL	48.7 ± 1.8	0.3 ± 0.0	5.6 ± 1.4	1.9 ± 0.4; 48.6 ± 1.6	0.91714

sites on the carrier. This led to a better interaction between SX and FL causing agglomeration and surface detachment. The authors proposed that this was due to the fact that adhesion force between SX and CL was lower than SX–SX and SX–FL [35]. This potentially shows the strong cohesive behaviour of SX particles that is likely to create agglomerates.

The same tendency to create agglomerates was seen for FL–SS, and "free" agglomerates of FL:SS were visible in the space between carrier particles (Fig. 8B). When energy is applied, these agglomerates would be dispersed more easily compared to those adhered to the surface of the carrier (e.g. in SX:FL:CL), leading to the higher % deagglomeration for the SS:FL:CL compared to SX:FL:CL ("A" parameter, Table 3). In fact, the maximum gradient value for SS:FL (Table 2) was greater than SX:FL, suggesting that the aerosolization of SS was enhanced in presence of the FL. However, the adhesive strength predicted (Table 3) of the CL–SS interactions in the agglomerates would require high pressure to achieve full deagglomeration (x_w = 3.0 ± 0.3 for SS:CL in Table 3).

4. Conclusion

The study revealed a rapid technique to characterize the deagglomeration behaviour of micronized particles during formulation development. The micronized particles showed different deagglomeration processes to each other suggesting variability in agglomerate size and strength between agglomerates. The empirical parameterization was descriptive of agglomerate and powder structure and deagglomeration mechanisms. The micronized particles (including one model hydrophobic API and one model hydrophilic API) showed different deagglomeration processes to each other suggesting

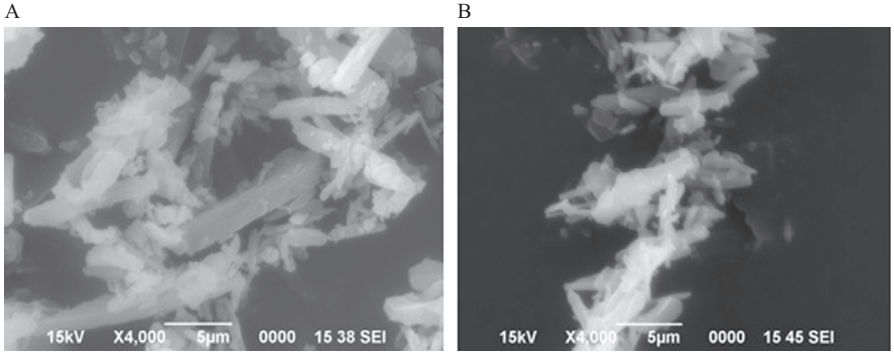

Fig. 6 – Scanning electron microscopy images for (A) salbutamol sulphate and (B) salmeterol xinafoate.

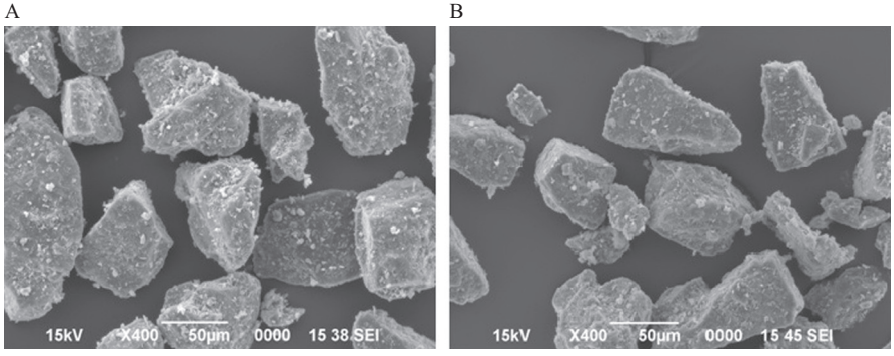

Fig. 7 – Scanning electron microscopy images for (A) salbutamol sulphate:coarse lactose and (B) salmeterol xinafoate:coarse lactose.

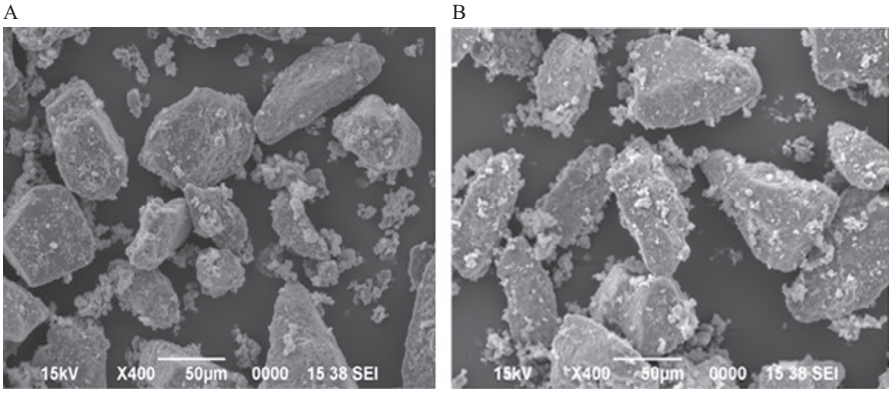

Fig. 8 – Scanning electron microscopy images for (A) salbutamol sulphate blended with fine and coarse lactose (SS:FL:CL) and (B) salmeterol xinafoate blended with fine and coarse lactose (SX:FL:CL).

variability in agglomerate size and strength between agglomerates. Co-formulating with a coarse carrier and fine lactose did modify the deagglomeration process suggesting that two different types of fractions of dispersed particles can be identified when increasing dispersing airflow. Moreover, SEM studies suggested a greater number of free agglomerates for SS:FL:CL than SX:FL:CL, leading to higher dispersion of the micronized material. Inhaler devices play a major role in the aerosolization of the formulation. Previous powder deagglomeration models have been proposed with specific devices (such as the Rotahaler) and additional work is required to expand our understanding of the implications of adding lactose (or other excipients) for the aerosolization of blends when used in other device platforms. However, we believe that the outcomes of the current study (albeit using a limited range of API and excipient materials and low shear blending) provide for good starting point for future rapid screening experiments into the effects of multiple material, formulation, blend and processing properties and provide functionally-relevant parameters of deagglomeration performance for DPI blends.

Acknowledgements

The authors would like to acknowledge the financial support of the University of Hertfordshire and the Society of Electron Microscopy Technology.

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