International Journal of Pharmaceutics 431 (2012) 45–52

Contents lists available at SciVerse ScienceDirect

International Journal of Pharmaceutics

journal homepage: www.elsevier.com/locate/ijpharm

The development of a novel dry powder inhaler

Xi Zhang[a,b], Yingliang Ma[a], Liqiang Zhang[a], Jingxu Zhu[a,*], Fang Jin[b,**]

[a] Chemical and Biochemical Engineering, University of Western Ontario, 1151 Richmond Street, London N6A 5B9, Canada
[b] Shanghai Engineering Research Center of Respiratory Medicine, Shanghai Institute of Pharmaceutical Industry, China State Institute of Pharmaceutical Industry, Zhongshan No. 1 Road(North), Shanghai 200437, PR China

ARTICLE INFO

Article history:
Received 9 December 2011
Received in revised form 26 March 2012
Accepted 3 April 2012
Available online 20 April 2012

Keywords:
Dry powder inhaler
Active inhaler
In vitro evaluation
Animal test
Fluorescent image

ABSTRACT

A novel active and multi-dose dry powder inhaler (DPI) was developed and evaluated to deliver a small quantity (100–500 μg) of pure drug without any excipient. This dry powder inhaler utilized two compressed air flows to dispense and deliver drug powder: the primary flow aerosolizes the drug powder from its pocket and the secondary flow further disperses the aerosol.

In vitro tests by Anderson Cascade Impactor (ACI) indicated that the fine particle fraction (FPF) (<4.7 μm) of drug delivery could reach over a range of 50–70% (w/w). Emitted dose tests showed that delivery efficiency was above 85% and its relative standard deviation (RSD) was under 10%. Confocal microscopy was used to confirm the deposition of fluorescently labeled spray-dried powder in rabbit lungs. Also, a chromatographic method was used to quantify drug deposition. The results of animal tests showed that 57% of aerosol deposited in the rabbit lung and 24% deposited in its trachea. All the results implied that this novel active dry powder inhaler could efficiently deliver a small quantity of fine drug particles into the lung with quite high fine particle fraction.

1. Introduction

Pulmonary drug delivery is considered to be the most effective method of treating respiratory diseases, especially asthma and chronic obstructive pulmonary disease (COPD). As a relatively new method of drug delivery, dry powder inhalers (DPIs) have been increasingly accepted as treatment of asthma and COPD (Lavorini et al., 2011). When compared with pressure metered dose inhaler (pMDI), another primary pulmonary drug delivery method based on liquid propellant volatilization, most DPIs rely on patients' inhalation to disperse and drive the drug particles to avoid press-breath coordination. Better yet, the DPI does not require chlorofluorocarbons or hydrofluoroalkane as propellants so that DPI is more environmentally friendly (Newman, 2009). Hopefully, DPI may also be used for larger delivery doses, and is more chemically stable than pMDI, providing greater opportunities for pulmonary macromolecule delivery (Shoyele and Slowey, 2006).

According to package forms, DPI can be divided into 3 types: single-unit dose, multi-unit dose and multi dose reservoirs. The multi-unit dose and multi dose reservoir DPI are increasingly popular since it is convenient to administer multiple doses without charging and/or changing the inner packages. DPI can also be divided into two types in terms of its drive force, a passive DPI dependent on patients' inhalation and an active DPI dependent on external force (Daniher and Zhu, 2008; Islam and Gladki, 2008). In recent years, active DPI has become a preferred method to uniformly distribute drugs to a wide variety of people due to its independence on the inspiration flow. What is more, active DPI is particularly suitable to people who may have trouble producing enough air stream to disperse drug powder (i.e., children and elderly) (Son and McConville, 2008; Tobyn et al., 2004).

For dry powder pulmonary delivery, the drug particle size is critical. Optimally it should be smaller than 5 μm in aerodynamic diameter. As a result of high potency of respiratory delivery, the required dosage is usually very small. So, pharmaceutical companies often add excipients of much larger size, such as lactose, to facilitate flow and dispersion (Pilcer and Amighi, 2010). However, adhesion of fine drug particles to coarse carriers would decrease the delivery efficiency, and some patients may be intolerant to lactose (Saint-Lorant et al., 2007).

Consequently, a novel dry powder inhaler was developed to deliver very tiny dosage of pure active pharmaceutical ingredient (API) powder without any excipient (Zhu et al., 2011). It applies a two air flow design to produce complete dispersion of API powder with the break-up of most agglomerates of powder. The pure drug powder was metered and filled into multi-dose disks with great accuracy and reproducibility by our patented dispensing device (Zhu et al., 2004). Both ACI impaction and time-flight particle size measurement were carried out to confirm that fine drug powder could be effectively dispersed by this DPI with high fine particle fraction (FPF). In addition to in vitro tests, fluorescent imaging was

* Corresponding author. Tel.: +1 519 661 3807.
** Corresponding author. +86 21 55514600 146; fax: +86 21 33812018.
E-mail addresses: jzhu@uwo.ca (J. Zhu), jfsipi@163.com (F. Jin).

0378-5173/$ – see front matter © 2012 Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.ijpharm.2012.04.019

utilized as a fast and effective method to confirm deposition of particles with fluorescence label in animal lung. Also, assaying in animal lung further demonstrated deposition of fine particles in lung and provided quantitative information on distribution.

2. Materials and method

2.1. Materials

Spray dried insulin, phenylalanine (PHE) was provided by GlaxoSmithKline (USA); sulfate salbutamol and nitrendipine were from Nanjing Pharmaceutical Factory, China; Fluorescein Isothiocynate (FITC) and other organic solvents were from Concord Technical Co. Ltd., Tianjin, China.

Nitrendipine and PHE labeled FITC (PHE–FITC) powders were prepared by Mini Spray Dryer (Bucchi, B-290, Switzerland). 0.5% nitrendipine in ethanol solution was sprayed out with 5 ml/min feed rate. Meanwhile, fluid rate was set as 675 ml/h in Nitrogen, and inlet and outlet temperature was 130 °C and 75 °C, respectively. 10–20% FITC (based on PHE) was added to 0.5% PHE water solution which contain 10% (w/w) $NaHCO_3/Na_2CO_3$ buffer, and the whole solution was stirred for 12 h in 0–4 °C. Then, the liquid was sprayed out with 5 ml/min feed rate, and air fluid rate was 675 ml/h, inlet and outlet temperatures were 140 °C and 80 °C. Salbutamol was spray dried from a 10% (w/v), aqueous solution, with 150 °C inlet air and 75 °C outlet air, and the flow rate was 675 ml/h (Corrigan et al., 2004).

The size of the particles produced by spray drier was measured by Particle Size Distribution Analyzer (PSD) (TSI corporation model 3603, USA). The volumetric median diameter (VMD) result of aerosol for nitrendipine, PHE–FITC, insulin, salbutamol was 7.01 μm, 4.14 μm, 3.78 μm and 2.94 μm, respectively.

The shape and size of particles were also observed with scanning electron microscope (SEM) (Hitachi S-2600, Japan). SEM photos (Fig. 4a–d) confirmed that the particles were in the range of 1–7 μm. The SEM photos also clearly showed the shape of those particles: the nitrendipine particles was irregular plate, the PHE–FITC particles was irregular sphere with multi concaves, the insulin was nearly sphere with concave in the center, and salbutamol was of regular sphere.

The angle of repose and bulk density of powders were evaluated by Powder Tester (HOSOKAWA MICRON corporation, Japan). The angle of repose of insulin and PHE–FITC was 48° and 54°, respectively; and tapped bulk density was listed as following: nitrendipine 0.485 g/cm^3, PHE–FITC 0.440 g/cm^3, insulin 0.314 g/cm^3 and salbutamol 0.281 g/cm^3.

Fig. 1. The appearance of the novel dry powder inhaler.

2.2. The novel dry powder inhaler

The appearance of the novel active and multi-dose inhaler is shown in Fig. 1 and its detailed structure in Fig. 2. The inhaler provides a rotating multi-dose disk with pure drug pre-metered in small pocket holes drilled through the disk (Fig. 2b). The disk is inserted between the air tubule and compress chamber, leaving only one drug pocket in the air passage for a given time (Fig. 2a). The blister pack is arranged to have a sufficient number of doses for patient's use, 12–64 doses (Fig. 2b).

As shown in Fig. 2a, the DPI works by positive pressure from the patient pushing at the bottom button of the inhaler to produce compressed air in the sealed chamber. Then the compressed air directly goes through the drug pocket as primary air flow (solid arrow), carrying drug powder along the air tubule (diameter is 2 mm, length 35 mm) until ejecting the powder out mouthpiece. A secondary air flow is perpendicular to primary air flow above the drug pocket, produced by parallel but much smaller tubule (secondary air tubule in Fig. 2a). The secondary air flow (hollow arrow) provides an additional shear flow, and assists in entraining the fluidized powder into the primary air flow. As a conclusion, this device

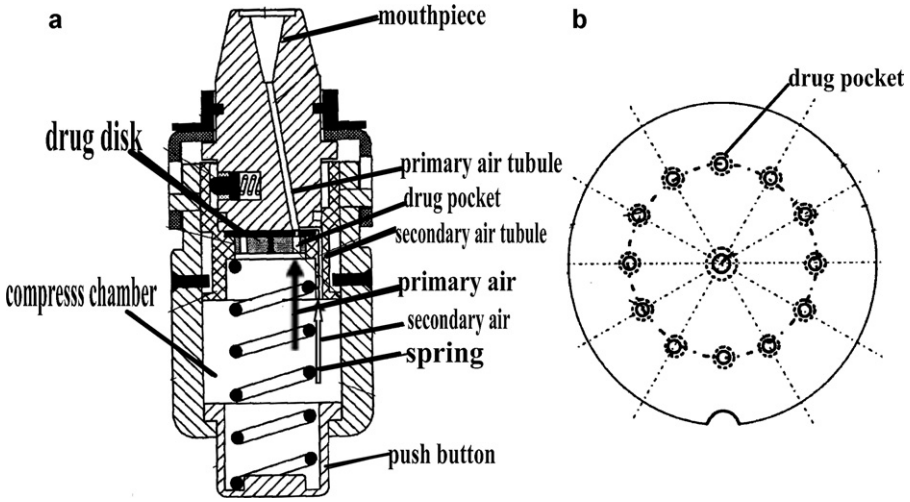

Fig. 2. Schematic diagram of the new designed dry powder inhaler (a) and multi-dose disk (b). Primary air flow (the solid arrow) goes through the drug pocket and carries drug powder along the air tubule; secondary air (the hollow arrow) goes upward directly and then becomes perpendicular to primary air flow above the drug pocket.

Table 1
Summary of evaluation tests for 4 powders.

Name of powder	TEST
Nitrendipine	SEM; sizing of aerosol by PSD analyzer; Anderson Cascade Impaction; uniformity test; distribution in rabbit lung
PHE–FITC	SEM; sizing of aerosol by PSD analyzer; Anderson Cascade Impaction; fluorescent imaging in rabbit lung
Insulin	SEM; sizing of aerosol by PSD analyzer; Anderson Cascade Impaction; uniformity test
Salbutamol	SEM; sizing of aerosol by PSD analyzer; Anderson Cascade Impaction

is an active one dependent on compressed air produced by pressing but not on inspiration flow. The volume of the push button and air tubule is calculated as 7900 mm^3 and 70 mm^3, respectively, so that the compression ratio is nearly 110. When pushing the button, the air is compressed into the tubule, resulting in high pressure and high velocity of air flow. When air flow arrives at the mouthpiece, because of the expanding mouthpiece, the velocity and pressure would drop dramatically (Zhu et al., 2011).

After one dose is delivered, the disk could be rotated to set a new dose in the air passage for the next administration. The quantity of drug is determined by the volume of holes, and as low as 20 μg extra fine and cohesive particles can be metered into those pockets (Zhu and Ma, 2006).

2.3. Method

The evaluation experiments for all 4 powders were summarized in Table 1.

2.3.1. In vitro tests for the novel dry powder inhaler

A digital camera was utilized to record the spray angle and shape of the aerosol, and pictures of initial and terminal aerosol were obtained from the video.

The emitted dose and delivery uniformity of the newly designed DPI were tested using two types of powders (PHE–FITC and insulin) by the Dose Unit Sampling Apparatus (DUSA) (Copley Scientific, UK). According to the U.S. Pharmacopoeia 30-NF25, the flow rate was set to reach 4 kPa pressure drop through the device and ensure that critical flow occurred in the flow-control valve, then 18.7 l/min sampling flow rate was determined, and the sampling time was set as 13 s to produce 4 l of air through the device. The DPI was fitted into the sampling tube and actuated. The particles deposited in the sampling tube and filter was rinsed by proper solvent and quantified using UV–visible spectrophotometer (Agilent, model 8453, USA).

The emitted aerosol size distribution was measured by Particle Size Distribution Analyzer (PSD), with the optional 3306P Impactor Inlet (TSI corporation model 3603, USA). The Impactor Inlet has a single-stage impactor, and can direct a small aerosol sample to the spectrometer for size distribution measurement. The inhaler was tested with 28.3 l/min flow rate.

The fine particle fraction of powder delivery from the DPI was tested by Anderson Cascade Impactor (ACI) (Copley Scientific, UK). The tests were carried out under flow rate of 28.3 l/ml, with proper numbers of actuations. The particles deposited on the throat and at each stage were rinsed by proper solvent and collected, and then quantified by HPLC. PHE–FITC was delivered to a rabbit (Section 2.3.2) through an AeroChamber (AeroChamber, Trudell Medical International, Canada), so that spacer was also used for the PHE–FITC ACI test.

To compare the performance of this device to marketed product for the same powder, the Bricanyl® Turbuhaler® (Astra Pharma Inc.) was used. The terbutaline sulfate powder in the device was cleaned off by high pressure air, and then nitrendipine powder was weighed into the reservoir chamber for uniformity and fine particle fraction tests.

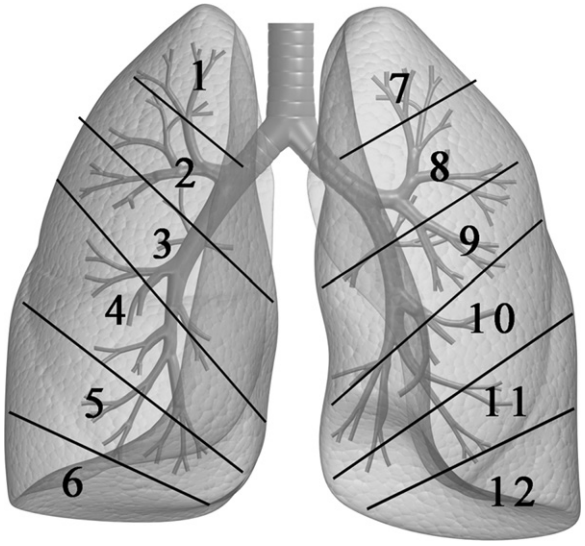

Fig. 3. Schematic diagram of the division of lung (taking human lung as illustration).

2.3.2. Tissue distribution of powder emitted from DPI in rabbits

All procedures with the animals were performed under the approval and review of the Animal Ethics Committee at the Shanghai Institute of Pharmaceutical Industry.

In this preliminary study, four male New Zealand White Rabbits (2.0–2.2 kg) were anaesthetized. One rabbit was a control and we did not administer any aerosol so that background fluorescence could be measured; one was administered PHE–FITC with a customized mask covering the nose and mouth with a mask connected to AeroChamber where DPI was inserted into. So, when actuating the DPI, the drug powder would be delivered into the spacer and then be breathed in by the rabbit's normal inhalation for qualitative confirmation of delivery of particle to lung periphery. About 30 min was taken for rabbit administration, including refilling the disks and rabbit inhaling the aerosols. To avoid the effect of moist exhaled flow on the quantitative distribution analysis, the remaining two rabbits were intubated through trachea under the larynx. Then spray-dried nitrendipine fine powder was administered through the catheter using the novel inhaler device with a custom adapter, which took about 15 min. Following administration, the rabbits were executed and the lung/trachea samples were removed and frozen/prepared immediately.

2.3.2.1. Fluorescent visualization. The control and fluorescein administered rabbit lung were frozen at −70 °C in a freezer (Sanyo, MDF-290AT, Japan). Symmetrically, the left and right lungs were divided into 6 parts (as shown in Fig. 3), and the trachea was divided into three parts (upper, middle and lower). Each section was made by freezing microtome (Leica CM1800, Germany). The fluorescent images of each section were obtained by confocal laser electrical microscope (Leica TCS-SP, Heid Elberg, Germany).

2.3.2.2. Nitrendipine analysis. The lungs of the nitrendipine administered rabbit were divided in a similar way as for the fluorescent visualization. Each tissue sample was homogenized and diluted to 10 ml with saline, with 20 μl of Nimodipine solution (2.0 μg/ml) as an internal standard. Concentrations were determined by HPLC, and the quantity of nitrendipine deposited in the trachea was measured by a similar method.

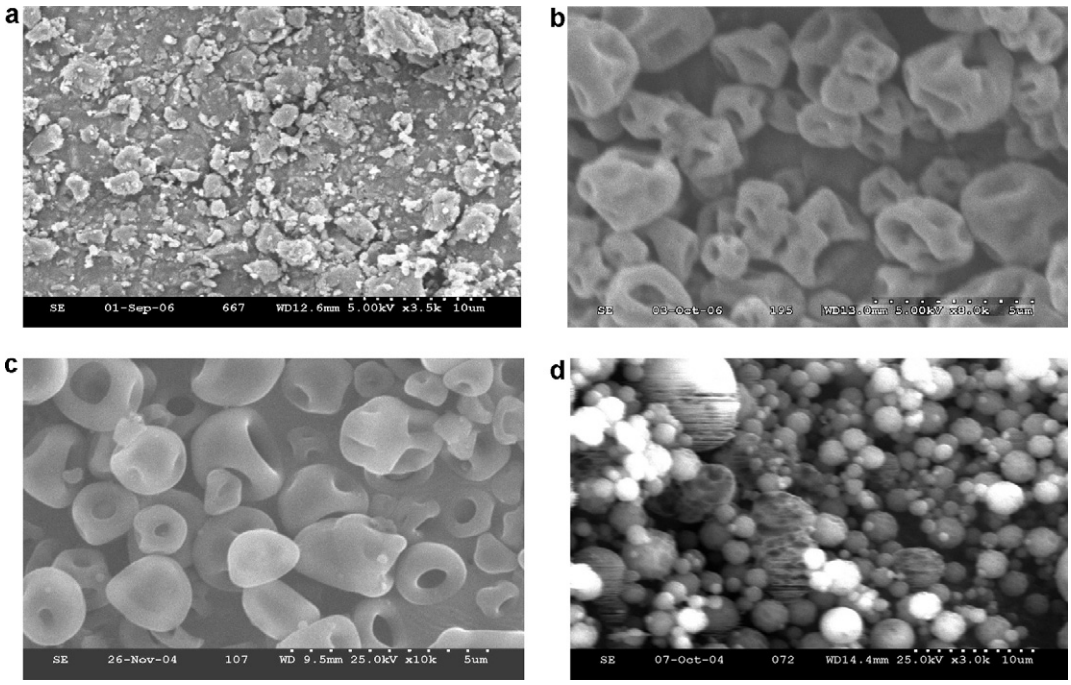

Fig. 4. SEM photos of four different powders prepared by spray dry for in vitro tests of the inhaler. (a) Nitrendipine, (b) PHE–FITC, (c) insulin, and (d) salbutamol.

3. Results

3.1. Characterization of fine powder produced by spray drier

According to the SEM photograph, the size of nitrendipine, PHE–FITC, insulin and salbutamol was 1–6 μm, 1–4 μm, 1–4 μm and 1–3 μm, which not only further confirmed the size distribution obtained from Particle Size Distribution Analyzer (PSD), but also provided shape information for these particles as mentioned in Section 2.1. The SEM size results were somewhat smaller than the PSD results, implying that some extra fine particles may form agglomerates and were not disaggregated during the PSD test.

3.2. In vitro tests for the novel dry powder inhaler

3.2.1. Angle and shape of the spray

The aerosol from the inhaler was centralized, with <5° spray angle as shown in Fig. 5a. Fig. 5b shows that the aerosol expanded quickly to form a cloud at about 12 cm from the mouthpiece. Additionally, there was no difference in spray shape or angle between these 4 different powders. The outline of powder cloud was marked by a white line.

3.2.2. Uniformity and efficiency of drug delivery

The delivery uniformity from the inhaler was tested with PHE–FITC and insulin powder using two disks of different pocket size (small: 0.196 mm^3; large: 0.95 mm^3). The tests were repeated for 10 times, and the average emitted dose (recovered from the DUSA device) for large dose disk using PHE–FITC powder was 312.02 μg with 8.69% RSD. It showed that the average efficiency of drug delivery (emitted dose/label dose) was 87.66%, with about 13% left in the mouthpiece. The total drug recovery from the emitted dose and mouthpiece was about 100%, implying that there was little powder left in the air tubule. The diameter of air tubule is larger than the size of individual particle or agglomerates (about 400 times), plus the high velocity of air though it, minimizing the probability of blockage. Similarly, the average emitted dose for small dose disk using insulin powder was 70.54 μg with 5.7% RSD, and the delivery efficiency was 91.82%.

When nitrendipine powder was used to study Turbuhaler®, as a result of poor flowability for extra fine particles, some holes in Turbuhaler® dosing disk could not be filled properly, implying the poor uniformity and making it impossible to evaluate the efficacy directly as the metered quantity may alternate time to time.

3.2.3. Particle size distribution of powder aerosol

The size distribution of aerosol emitted from the novel inhaler by PSD was presented in Fig. 6, where only one peak existed for all four powders, suggesting that there was no particle agglomeration. The VMD result of aerosol for nitrendipine, PHE–FITC, insulin, salbutamol were 6.77 μm, 3.99 μm, 3.66 μm and 2.78 μm, respectively, all of which did not show significant difference with spray-dried particles.

The ACI impaction result was presented in Fig. 7, the fine particle fractions (ratio of mass of particle under 4.7 μm and total emitted dose) of nitrendipine, PHE–FITC, insulin and salbutamol were 50.84%, 64.73%, 69.12% and 74.0%, respectively. The corresponding mass median aerodynamic diameter (MMAD) was 4.09 μm (nitrendipine), 2.70 μm (PHE–FITC), 2.23 μm (insulin), 1.28 μm (salbutamol). Clearly, the increasing of the FPF was related to the MMAD decreasing. The ACI result (Fig. 8) of nitrendipine in Turbuhaler® device showed that the FPF value was only 21.48 ± 0.97% (n = 3), and MMAD was 8.22 ± 0.69 μm. The FPF was much lower but MMAD was higher than our device, which demonstrated that the novel device was better at dispersing the fine particles and agglomerates when compared with Turbuhaler®.

3.3. Tissue distribution of powder emitted from the novel inhaler

3.3.1. Fluorescent imaging

As shown in Fig. 9, blank trachea (Fig. 9a) did not show any fluorescent signal, while top (Fig. 9b) and middle trachea (Fig. 9c) of administered rabbit displayed stronger signal compared with the bottom part of tracheas (Fig. 9d). Similarly, as shown in Fig. 10, the blank lung (Fig. 10a) had no clear fluorescent signal, while all parts

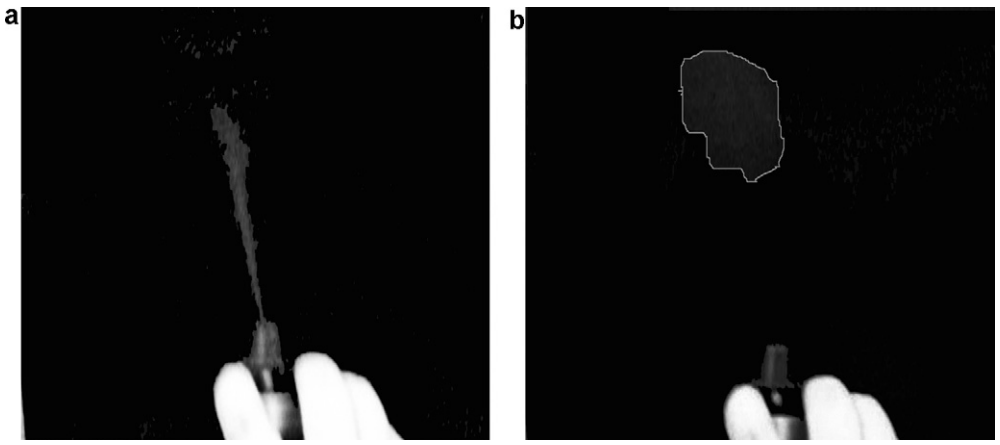

Fig. 5. Photos of aerosol spray through the DPI. (a) Initial shape of the aerosol and (b) terminal shape of the aerosol (outlined by a white line).

Table 2
Percentage of nitrendipine deposition in rabbits' lung. L1–12 stand for individual part of the lung as in Fig. 3; Tu, Tm, Td stand for the upper/middle/down part of the trachea, respectively.

	Part	L1	L2	L3	L4	L5	L6	L7	L8	L9	L10	L11	L12	Tu	Tm	Td
Rabbit 1	Percentage (%)	4.45	1.1	0.42	8.14	6.77	3.18	6.11	11.97	3.73	1.59	3.93	2.05	5.34	16.44	6.7
Rabbit 2	Percentage (%)	7.59	1.91	1.05	3.86	8.12	5.79	6.78	9.13	2.2	5.2	6.26	3.59	2.89	10.54	7.02
AVG		6.02	1.505	0.735	6	7.445	4.485	6.445	10.55	2.965	3.395	5.095	2.82	4.115	13.49	6.86

of administered lung had significant signal. Even the distal part of the lung, part 6 (Fig. 10d) and part 12 (Fig. 10f) still had fluorescent signal.

3.3.2. Distribution of nitrendipine in rabbit lung

The amount of nitrendipine in different parts of lung and trachea was determined by HPLC, and the distribution was calculated as presented in Table 2. Total deposition in the lung (L1–12) was 57.46%, while deposition in the trachea (T_{u-d}) was 24.46%, with about 18% left in the device which was consistent with the efficiency of drug delivery in vitro tests (Section 3.2.2). The percentage of L6 and L12 deserved attention especially, since it demonstrated that particle deposited even in the lung periphery.

4. Discussion

4.1. Particle size distribution by PSD (TSI)

As mentioned in Section 3.2.3, the PSD results of aerosol emitted from the DPI seem to be similar to the initial particles size, implying that this novel DPI was able to disperse any possible agglomerates during the filling process. The mechanism of disaggregation of

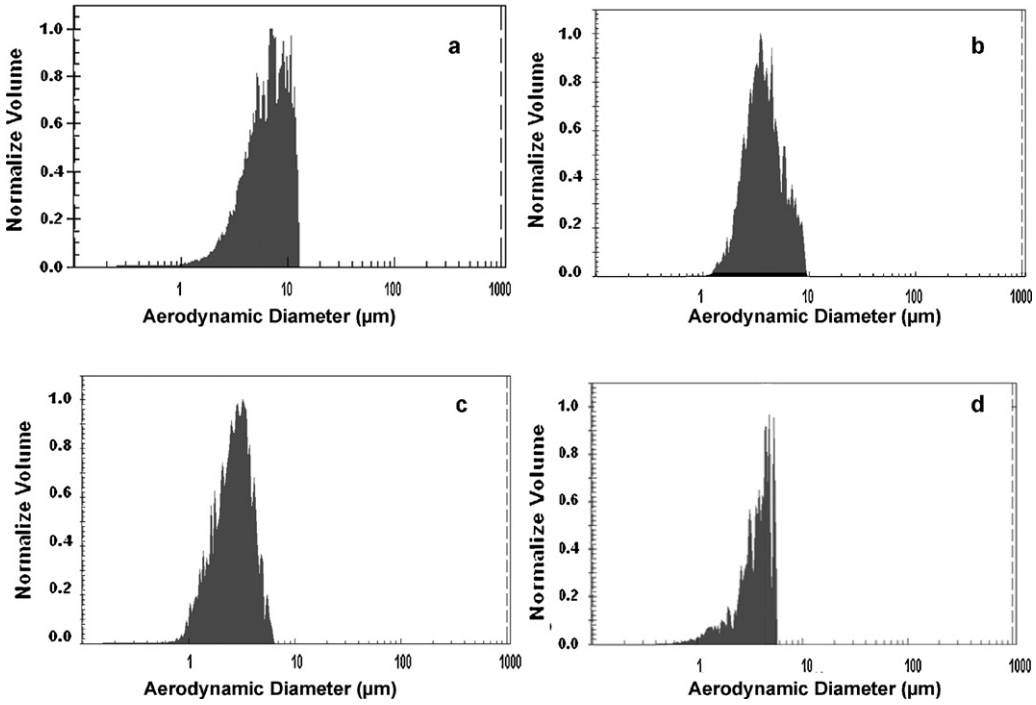

Fig. 6. Particle size of the aerosol emitted from the novel inhaler measured by PSD. (a) Nitrendipine, (b) PHE–FITC, (c) insulin, and (d) salbutamol.

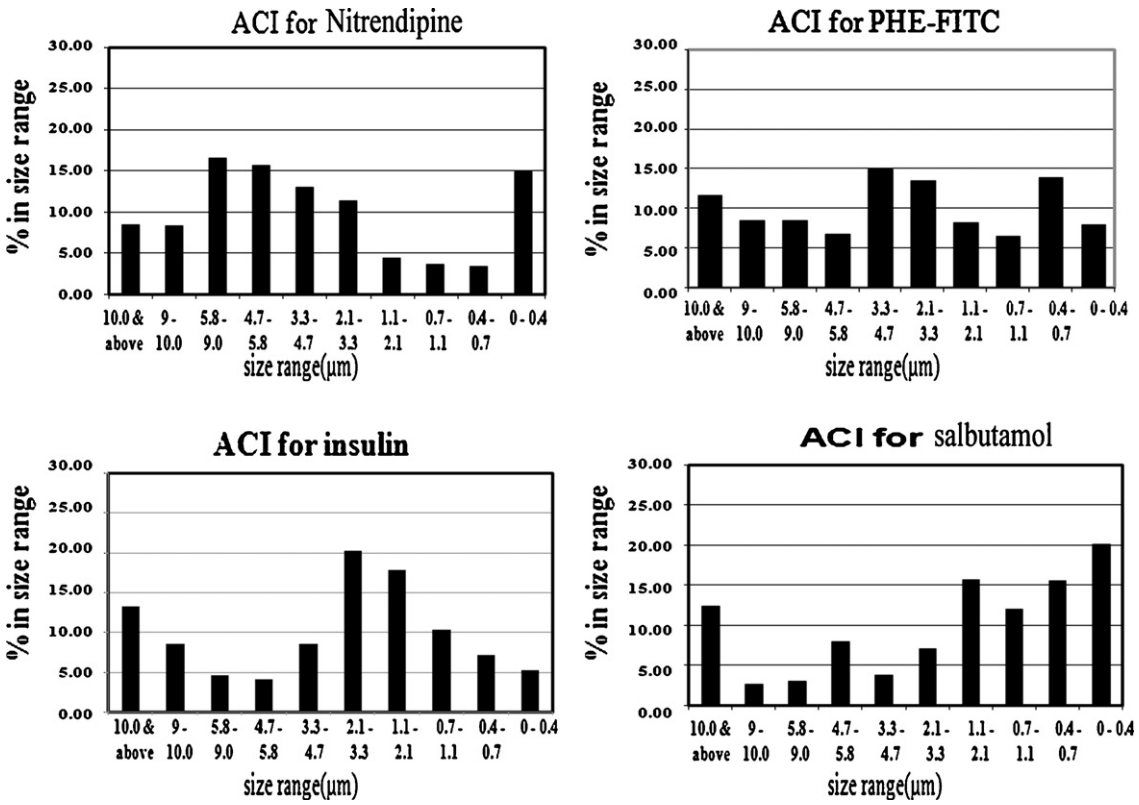

Fig. 7. Anderson Cascade Impaction for evaluation of in vitro deposition of the novel inhaler. (a) Nitrendipine, (b) PHE–FITC, (c) insulin, and (d) salbutamol.

cohesive particles may rely on the design of inhaler device. Primarily, the secondary air flow was perpendicular to the primary flow above the drug pocket, exerting lateral shear force on the upward flowing particles at the same time. Owing to the shear force and impaction on the wall of the primary air tubule, the agglomerates were easily detached. Moreover, as mentioned before (Section 2.2), the air compressibility was as high as 110 times and the time of pushing the button was very short, so that the pressure and velocity in the air tubule was very high. However, when air flow arrived at the expanded mouthpiece, the pressure would drop dramatically. Therefore, the air turbulence produced by the pressure drop also contributed to the disaggregation of particles.

4.2. Drug aerosol spray shape and emitted uniformity from the DPI

The centralized fume emitted from the inhaler and small spray angle <5° were mainly caused by the relatively high velocity of air

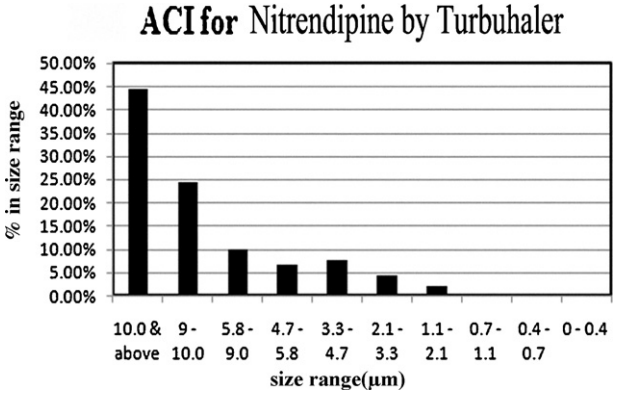

Fig. 8. Anderson Cascade Impaction result for nitrendipine delivered by Turbuhaler®.

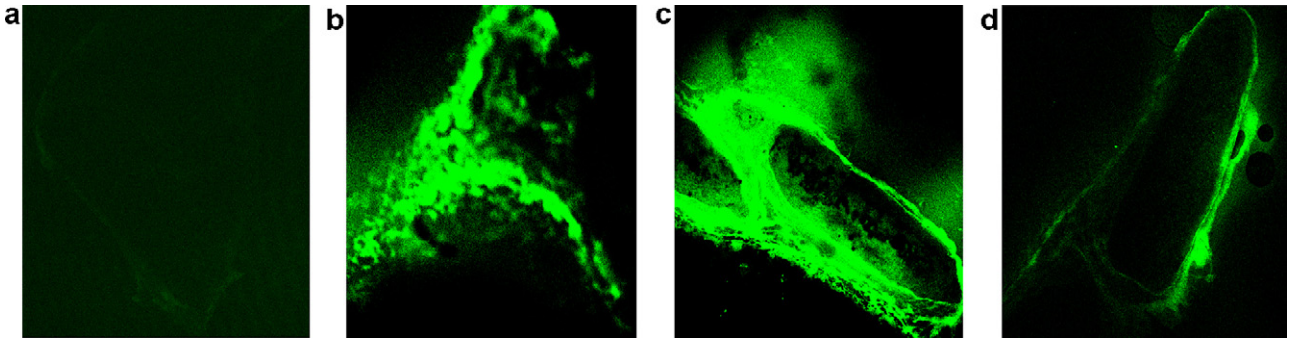

Fig. 9. Fluorescent images for trachea part of rabbits. (a) Blank trachea of the control rabbit, (b) top trachea, (c) middle trachea, and (d) bottom trachea of rabbits administrated with fluorescein-labeled FITC powder by the novel inhaler.

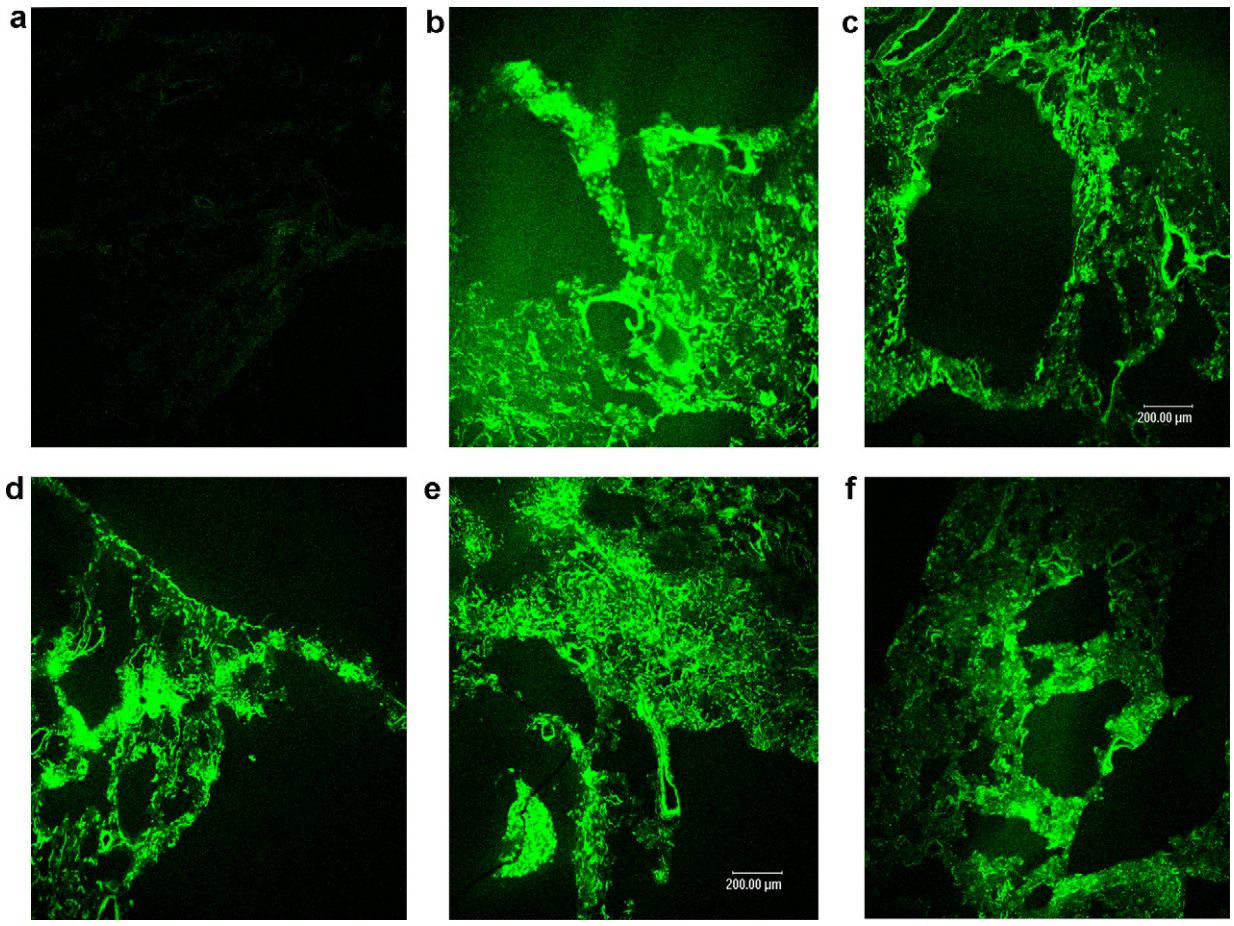

Fig. 10. Fluorescent images for divided part of rabbits lung. (a) Blank lung part of the control rabbit (b), (c), (d), (e), and (f) stand for parts 1, 5, 6, 7, and 12 of the rabbit administrated with fluorescein-labeled FITC powder by the novel inhaler.

flow at the mouthpiece. The drug aerosol emitted from our DPI formed a cloud at around 12 cm from the mouthpiece, where the particle velocity was much slower compared to its initial velocity. It may be understood that the aerosol was composed of very tiny and extra fine particles, consequently, the inertia was too low to maintain velocity, resulting in a dramatic velocity drop. Compared with pMDI (data not shown here), the aerosol spray angle, the distance and volume of aerosol cloud of this DPI were much lower, implying less deposition in the mouth.

The uniformity of drug delivery was very important for pulmonary drug delivery. As mentioned in Section 3.2.2, PHE–FITC and insulin powders were tested for evaluation of the novel DPI delivery uniformity. The delivery efficiency from our DPI was above 85%, with RSD < 10%, which was much better than a lot of commercialized products, such as the very successful Turbuhaler® (Budesonide) with efficiency 58.1% and RSD 18.3% (Newman and Busse, 2002). However, the evaluation of uniformity and efficiency for Turbuhaler® in this study could not be carried out for poor flowability of extra fine particles and unsatisfying filling, which further confirmed that dry powder inhaler was a combined product of formulation, device and filling technology. When considering the different performance of the novel device for insulin and PHE–FITC, the flowability was also very important. Since the angle of repose of insulin (48°) was smaller than PHE–FITC (54°), implying that the flowability of insulin was better than PHE–FITC. As a result of better flowability, the uniformity of filling the insulin powder into the pockets was better, and then the emitted dose of insulin was more uniform than PHE–FITC. As in Section 3.2.2, the delivery efficiency of PHE–FITC was somewhat lower than insulin, indicating that the tendency of PHE–FITC powder adhering the device wall was more pronounced, which may be explained by the fact that PHE–FITC was more sticky with larger angle of repose.

4.3. Fine particle fraction of drug delivery from the DPI

Although there is no direct relation between the FPF and the fraction entering the lung in vivo, FPF value is a very important parameter in evaluating the performance of the inhaler product. Usually, it is believed that higher FPF means better pulmonary drug delivery efficiency.

All four powders were tested with the novel inhaler for evaluating the performance of dispersing different powders (Section 3.2.3). ACI results demonstrated that FPF value from our DPI for all 4 powders were over 50%, much higher than the Turbuhaler® when using the same extra fine particles (about 21%), imply the potential of the novel device to disperse extra fine particles. The FPF value for Turbuhaler® was somewhat lower than the results (25% under about 30 l/min flow rate) summarized by Frijlink and De Boer (2004), which may be explained by the different powders used. The better FPF performance of the device compared to Turbuhaler® may lie in the device design: a secondary air flow was perpendicular to the primary air flow, providing a strong lateral shear force for disaggregation besides the turbulent air flow on which most other DPIs are dependent. Besides, the lateral air flow may result in the impaction of particles on the tubule wall, which was also helpful for disaggregating agglomerates. Compared with common DPI products which add larger excipient particles like lactose to increase the flowability and accuracy of dosing, this novel device only had fine

drug particles. Consequently, this device avoids strong interactive force between fine particles and carrier, and subsequently improves FPF by decreasing impaction of drug particles on the oropharynx with coarse carrier.

The different FPF value for four powders may be related to their size and shape. When comparing plates particles (nitrendipine powder) with nearly sphere particles (left 3 powders), plate particles had larger contact surface and interactive force, and therefore clearly lower FPF (Hassan and Lau, 2009a,b). Usually, it was believed that $D_{\text{aerodynamic}} = (\rho)^{0.5} * D_{\text{p}}$, which meant that if D_{p} was almost the same, and aerodynamic diameter decreases when density decreases (Bosquillon et al., 2001; Edwards et al., 1998). Consequently, the ACI results showed that salbutamol had the smallest MMAD and largest FPF, followed by insulin, PHE–FITC and nitrendipine, which may be explained by the bulk tap density for those 4 powders: salbutamol 0.2805 g/cm^3, insulin 0.314 g/cm^3, PHE–FITC 0.44 g/cm^3 and nitrendipine 0.485 g/cm^3.

4.4. Animal testing for aerosol deposition

Although there were several reports (Lombry et al., 2002; Pinkerton et al., 1993) about the application of fluorescent imaging for studying the distribution of particles in the lung, the confocal fluorescence microscopy was not a reliable quantitative tool to study tissue distribution for its photo bleaching, depth limitation and light scattering (Helmchen and Denk, 2005; Svoboda and Yasuda, 2006). Hence, in this work, the fluorescent imaging was solely utilized for qualitative confirmation of pulmonary drug delivery. When considering the fluorescent results, from Fig. 10, it can be initially inferred that even when the rabbit was anaesthetized, the rabbit could still breathe in the particles from the spacer. In other words, the aerosol could suspend in air for a while before being inhaled or settlement. Meanwhile, the fluorescent signal was detected in the lung periphery, which meant that the particles size was small enough to pass through fine bronchiole.

Nitrendipine, a calcium channel blocker with marked vasodilator action, was selected to study particle deposition in lung tissue. There were two reasons for the selection of nitrendipine instead of salbutamol, one common inhalation drug, as a model drug for tissue distribution study. First, nitrendipine was not easily absorbed into systematic circulation at lung tissue for higher log P (Patton and Byron, 2007), while salbutamol was rapidly absorbed after inhalation and entered systematic circulation (Lipworth, 1996), which may cause the deposition was lower than real value. Second, the half time of nitrendipine was 8 h, much longer than salbutamol with 4 h (Eichelbaum et al., 1988; Goldstein et al., 1987). At last, because the FPF value of nitrendipine was the lowest, as long as nitrendipine particles could be dispersed to the lung periphery, it can be inferred that other particles with higher FPF would have the same or better results. In general, the nitrendipine particles were distributed in the whole lung uniformly, and even the distributed quantity at the distal end, like L 5, 6, 11, 12, had similar results as other parts in the center. This confirmed that enough extra fine particles were emitted by the inhaler and can easily reach the lung periphery. When compared to ACI and the tissue distribution result of nitrendipine, it was found that the percentage of deposition in trachea and lung reached 81%, much higher than the FPF value (50%). It was caused by the animal administrative method and that powder was directly delivered to the trachea by intubation under larynx.

5. Conclusion

Through this work, a novel active and multi dose dry powder inhaler was developed and evaluated. This dry powder inhaler was able to utilize the compressed air to deliver a small quantity of extra fine particles without any excipient, which could be applied to any high potential or very expensive drugs. In vitro tests showed that the FPF values of this inhaler for different powder ranged from 50 to 70%, with good delivery uniformity and acceptable device residue. The image of powder aerosol spray showed that the active inhaler had better spray properties than pMDI due to less probability of deposition in the oropharynx. The animal tests of fluorescent imaging and tissue distribution confirmed that this inhaler was especially suitable to deliver fine particles to deep lung tissue for systematic therapy.

References

Bosquillon, C., Lombry, C., Preat, V., Vanbever, R., 2001. Influence of formulation excipients and physical characteristics of inhalation dry powders on their aerosolization performance. J. Control. Release 70, 329–339.

Corrigan, D.O., Corrigan, O.I., Healy, A.M., 2004. Predicting the physical state of spray dried composites: salbutamol sulphate/lactose and salbutamol sulphate/polyethylene glycol co-spray dried systems. Int. J. Pharm. 273, 171–182.

Daniher, D.I., Zhu, J., 2008. Dry powder platform for pulmonary drug delivery. Particuology 6, 225–238.

Edwards, D.A., Ben-Jebria, A., Langer, R., 1998. Recent advances in pulmonary drug delivery using large, porous inhaled particles. J. Appl. Physiol. 85, 379–385.

Eichelbaum, M., Mikus, G., Mast, V., Fischer, C., Kuhlmann, U., Machleidt, C., 1988. Pharmacokinetics and pharmacodynamics of nitrendipine in healthy subjects and patients with kidney and liver disease. J. Cardiovasc. Pharmacol. 12 (Suppl. 4), S6–S10.

Frijlink, H.W., De Boer, A.H., 2004. Dry powder inhalers for pulmonary drug delivery. Expert Opin. Drug Deliv. 1, 67–86.

Goldstein, D.A., Tan, Y.K., Soldin, S.J., 1987. Pharmacokinetics and absolute bioavailability of salbutamol in healthy adult volunteers. Eur. J. Clin. Pharmacol. 32, 631–634.

Hassan, M.S., Lau, R.W., 2009a. Effect of particle shape on dry particle inhalation: study of flowability, aerosolization, and deposition properties. AAPS PharmSciTech 10, 1252–1262.

Hassan, M.S., Lau, R.W., 2009. Pollen shape particles for pulmonary drug delivery: in vitro study of flow and deposition properties. In: 13th International Conference On Biomedical Engineering IFMBE Proceedings, pp. 1434–1437.

Helmchen, F., Denk, W., 2005. Deep tissue two-photon microscopy. Nat. Methods 2, 932–940.

Islam, N., Gladki, E., 2008. Dry powder inhalers (DPIs) – a review of device reliability and innovation. Int. J. Pharm. 360, 1–11.

Lavorini, F., Corrigan, C.J., Barnes, P.J., Dekhuijzen, P.R., Levy, M.L., Pedersen, S., Roche, N., Vincken, W., Crompton, G.K., 2011. Retail sales of inhalation devices in European countries: so much for a global policy. Respir. Med. 105, 1099–1103.

Lipworth, B.J., 1996. Pharmacokinetics of inhaled drugs. Br. J. Clin. Pharmacol. 42, 697–705.

Lombry, C., Bosquillon, C., Preat, V., Vanbever, R., 2002. Confocal imaging of rat lungs following intratracheal delivery of dry powders or solutions of fluorescent probes. J. Control. Release 83, 331–341.

Newman, S., 2009. Respiratory Drug Delivery: Essential Theory and Practice, first ed. Davis Healthcare International Publishing, Richmond.

Newman, S.P., Busse, W.W., 2002. Evolution of dry powder inhaler design, formulation, and performance. Respir. Med. 96, 293–304.

Patton, J.S., Byron, P.R., 2007. Inhaling medicines: delivering drugs to the body through the lungs. Nat. Rev. Drug Discov. 6, 67–74.

Pilcer, G., Amighi, K., 2010. Formulation strategy and use of excipients in pulmonary drug delivery. Int. J. Pharm. 392, 1–19.

Pinkerton, K.E., Gallen, J.T., Mercer, R.R., Wong, V.C., Plopper, C.G., Tarkington, B.K., 1993. Aerosolized fluorescent microspheres detected in the lung using confocal scanning laser microscopy. Microsc. Res. Tech. 26, 437–443.

Saint-Lorant, G., Leterme, P., Gayot, A., Flament, M.P., 2007. Influence of carrier on the performance of dry powder inhalers. Int. J. Pharm. 334, 85–91.

Shoyele, S.A., Slowey, A., 2006. Prospects of formulating proteins/peptides as aerosols for pulmonary drug delivery. Int. J. Pharm. 314, 1–8.

Son, Y.J., McConville, J.T., 2008. Advancements in dry powder delivery to the lung. Drug Dev. Ind. Pharm. 34, 948–959.

Svoboda, K., Yasuda, R., 2006. Principles of two-photon excitation microscopy and its applications to neuroscience. Neuron 50, 823–839.

Tobyn, M., Staniforth, J.N., Morton, D., Harmer, Q., Newton, M.E., 2004. Active and intelligent inhaler device development. Int. J. Pharm. 277, 31–37.

Zhu, J., Ma, Y., 2006. A new breath-activated, excipient-free dry powder inhaler and a rotating fluidized bed powder dispenser for pulmonary drug delivery. Respir. Drug Deliv., 925–930.

Zhu, J., Wen, J., Ma, Y., Zhang, H., 2004. Apparatus for volumetric metering of small quantity of powder from fluidized beds, US Patent 6,684,917, February 3.

Zhu, J., Wen, J., Ma, Y., Zhang, H., 2011. Dry powder inhaler, US Patent 8,037,880, October 18.