Research Paper

Can 'extrafine' dry powder aerosols improve lung deposition? ☆

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ABSTRACT

There is increasing interest in the use of so-called 'extrafine' aerosols to target the small airways in the management of asthma and COPD. Using previously presented deposition data, we assessed whether submicron (<1 μm) particles can improve central and deep lung deposition. Our data show instead that particles in the range 1–3 μm are much more relevant in this respect. Based on this finding the Symbicort Turbuhaler, Seretide Diskus, Rolenium Elpenhaler and Foster (Fostair) NEXThaler ICS/LABA combination DPIs were tested *in vitro* as a function of the pressure drop (2, 4 and 6 kPa) across the inhaler. Obtained fine particle fractions (FPFs) <5 μm (as percent of label claim) were divided into subfractions <1, 1–3 and 3–5 μm. Differences of up to a factor of 4 were found between the best (Turbuhaler) and worst performing DPI (Elpenhaler), particularly for the FPF in the size range 1–3 μm. The NEXThaler, described as delivering 'extrafine' particles, did not appear to be superior in this size range. The marked differences in amount and size distribution of the aerosols between the devices in this study must cause significant differences in the total lung dose and drug distribution over the airways.

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1. Introduction

Asthma and chronic obstructive pulmonary disease (COPD) are characterized by airflow obstruction and chronic inflammation of the respiratory airways. In the last few years, management of these diseases has improved considerably, as a result of the introduction of new drugs, drug combinations, drug administration devices and management strategies. Inhaled corticosteroids (ICSs) are the cornerstone of asthma and, to a lesser degree, COPD therapy because of their long-term efficacy and safety [1] but optimal effects may be expected when an ICS is administered in combination with a long acting beta$_2$-agonist (LABA) [2]. This has resulted in an increasing number of ICS/LABA inhalers becoming available. There is also a growing awareness of the importance of small airways in asthma and COPD [3,4] and the existence of a wide range of clinical phenotypes related to small airway involvement [5]. Small airways are those less than 2 mm in diameter, comprising the ducts between generation 8 and the alveoli. It has been postulated that finer aerosols than those delivered by most currently available inhalers may be needed to target these small airways more effectively and by that, to achieve a better drug distribution over the whole bronchial tree [6]. The origin of this idea may have

been the findings in the literature when chlorofluorocarbon (CFC)-based pressurized metered dose inhalers (pMDIs) containing beclometasone dipropionate (BDP) were replaced by hydrofluoroalkane (HFA)-based pMDIs, as a response to environmental concerns about the ozone layer in the 1990s [7]. It was shown that with the HFA pMDI only half the BDP dose is needed compared with CFC pMDI for effective treatment of moderate asthma [8,9]. The effect was attributed to the much finer aerosol from the HFA pMDI of which the particles had a mass median aerodynamic diameter (MMAD) of 1.1 μm versus 3.5–4 μm for the CFC pMDI. More devices delivering finer aerosols have since become available, most of them being HFA solution pMDIs [10–12]. The only ICS/LABA combination delivered so far as a fine aerosol from a pMDI and now from a dry powder inhaler (DPI) is the BDP–formoterol combination in Foster (Fostair), from Chiesi Pharmaceuticals [6]. The reported benefit of so-called 'extrafine' aerosols from HFA pMDIs has resulted in the expectation that the same improvement can be obtained with the dry small particle aerosol from this new Foster (Fostair) NEXThaler DPI compared to other DPIs with the same drug combination [6]. Several comparative studies with these new devices have recently been reviewed and it was concluded that treating the peripheral airways with smaller drug particle aerosols achieves comparable, and in some studies superior, efficacy compared with larger particles [13,14]. A reduction in the daily ICS dose was also reported, in addition to greater asthma control and quality of life in some of the real-life studies.

☆ Foster/Fostair® NEXThaler®, Symbicort® Turbuhaler®, Seretide® Diskus®, and Rolenium® Elpenhaler® are registered trademarks of the manufacturer.

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However, many questions remain to be answered before these improvements can be attributed to improved peripheral and total lung deposition from finer aerosols compared to deposition of conventional medications with larger particle size. It all starts with the size definition for 'extrafines'. Different terms have been used to describe finer aerosols, such as ultrafine [10], extrafine [6] and, more recently, small particle aerosols [13,14]. In this introduction, only the term extrafines will be used until the presentation of the term 'submicron particles'. Originally, extrafine aerosols from newly developed HFA BDP formulations were characterized as having an average diameter of 1.1 µm and a respirable fraction of approximately 60% [15]. For the Foster NEXThaler, extrafine particles are described as having a MMAD of 1.4–1.5 µm [16], while the definition for extrafine aerosols in the scientific literature has recently been widened to particles with a diameter $(D) < 2$ µm [13,14]. These different definitions partly overlap each other and do not bring clarity about which aerosols are to be considered as extrafine. Polydisperse aerosols from nearly all MDIs and DPIs may contain substantial mass fractions of particles with $D < 2$ µm. In contrast, devices producing the so-called extrafine aerosols may also deliver significant mass fractions of particles with $D > 2$ µm. Therefore, aerosols from all currently available MDIs and DPIs comprise both extrafine and non-extrafine particles according to the most recent definition ($D < 2$ µm). The difference is in the relative amounts of each of these fractions within the aerosols. Hence, for polydisperse aerosols the term extrafine has to be defined not only in terms of size, but also in the quantified mass fraction of these extrafines in the aerosol. For this reason, the rather imprecise terms extrafines and small particle aerosols will be used no further in this manuscript as an aerosol characterization term. Instead, a distinction will be made between submicron (<1 µm) and micron range (>1 µm) particles of which the micron range particles are divided into size fractions 1–3 µm and 3–5 µm to provide more detailed information about the structure of the fine particle fraction. The limit of 1 µm has been chosen because submicron particles ($D < 1$ µm) in the particle concentration of therapeutic aerosols have a significantly lower probability of total lung and alveolar deposition than micron range particles [17–19].

The influence of other variables on lung deposition involved between the different devices used in comparative studies is also relevant. Lung distribution and deposition are not governed by particle size alone, but also by particle velocity and residence time in the lung [20]. The difference between the BDP CFC and BDP HFA formulations in the previously mentioned MDI studies [7–9] is not in the particle size alone, but more particularly in the velocity with which the aerosol is released from the mouthpiece. The lower velocity of the HFA aerosol plume leads to a considerable reduction in impact force against objects in the flow direction of the plume and thus, a reduction in oropharyngeal deposition [21]. For BDP from the HFA device (MMAD ≈ 1.1 µm) developed in the late 1990s, throat deposition was found to be much lower (30%) compared with the CFC device (94%; MMAD ≈ 3.5–4 µm) [7]. Consequently, a much higher dose fraction remained available for total lung deposition, the difference being $(100-30)/(100-94) \approx 11.5$-fold. Due to these different factors, the expectation that a DPI delivering a finer aerosol (MMAD ≈ 1.5 µm for the fine particle fraction) at the same flow rate as competitor devices with only slightly coarser aerosols (MMAD ≈ 2.5–3 µm) can provide a more effective deep lung deposition may be false.

Inhalers used in various comparative studies to investigate the benefit of finer aerosols generally differ in more than particle size and velocity alone [13,14]. There may also be differences in delivered (fine particle) dose as percent of the label claim and many new inhaler types (both MDIs and DPIs) produce not only finer aerosols, but also higher fine particle doses [6,11]. In some recently reviewed studies [13,14] different types of inhalers (DPIs and MDIs) were compared with each other, and also different drugs in different strengths were involved and inhaled with different inhalation manoeuvres. In addition, many clinical studies were conducted without even recording the inspiratory flow manoeuvres and the duration of the breath hold pauses. Differences in resistance to air flow through an inhaler can lead to marked differences in flow rate at the same inspiratory effort [22]. With this variable as a major determinant for drug distribution and deposition in the respiratory tract, considerable differences in clinical effect may be expected, even if the aerosols from these devices are exactly the same *in vitro*. Several patient factors may also be involved, such as incorrect inhaler use [23], poor motivation or adherence to the therapy or to the study (for out-of-clinic studies), and severity of the disease, particularly when this affects pulmonary function and lung ventilation. As a consequence of this plurality of variables, it is virtually impossible to conclude which of them is most responsible for an improved clinical effect. Hence, clinical studies may be poor predictors for inhaler performance regarding aerosol generation and delivery. Therefore, a different approach seems necessary to investigate whether submicron particles can really contribute to improved therapeutic effects. The effects of inhaler and patient variables, including the inhalation manoeuvre, on aerosol generation, lung penetration, lung deposition and distribution and ultimately the clinical effect have to be considered separately, as well as in their interactions with each other. Judging an inhaler upon its potential to achieve a good clinical effect has to start with measuring the aerosol properties as a function of the flow rate and the emission pattern of the inhaler.

This manuscript has three aims: the first is to discuss whether submicron particles are likely to contribute to improved total and deep lung deposition. A second and equally important aim is to investigate which range of aerodynamic particle diameters is most favorable for total and deep lung deposition at the range of flow rates to be expected through a medium to high resistance DPI at moderate inspiratory effort (approx. 30–60 L/min). The third aim is to evaluate the delivered fine particle doses of four marketed ICS/LABA combination DPIs in relation to the outcome of both previous aims.

For the assessment, data from a previous deposition study in stable asthmatics were used and extrapolated towards particles in the submicron range and basic aerosol physics were used to check the validity of the extrapolations. Additionally, four DPIs, all delivering an ICS/LABA combination, were tested at three different pressure drops to measure their delivered fine particle doses (FPDs) and the structures of these FPDs as a function of the flow rate. FPFs < 1 µm were computed to obtain more detailed information about the presence and amount of submicron particles in the aerosol. Detailed information about differences in total delivered fine particle masses (FPFs < 5 µm) and the structures of the aerosols (FPFs < 1, 1–3 and 3–5 µm), as well as the flow rate at which the aerosols are delivered to the respiratory tract, is needed to decide whether differences in clinical effect are likely the result of any (or a combination) of these variables, or that of the involvement of yet unknown or overlooked parameters and mechanisms.

2. Materials and methods

2.1. Extrapolation of previously published deposition data

Usmani and co-workers measured lung deposition of radiolabelled monodisperse salbutamol particles (1.5, 3.0 and 6.0 µm) in patients with stable asthma at two different flow rates [24]. They discriminated between oropharyngeal, central plus
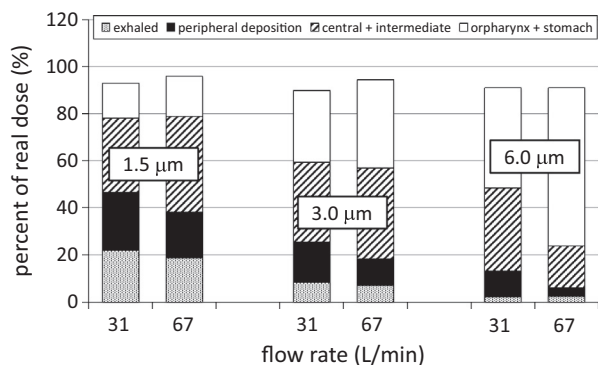
Fig. 1. Effect of particle size and inspiratory flow rate on lung deposition for monodisperse particles. Data derived from the Usmani study [24].

intermediate and peripheral deposition after a breath-holding period of 10 s to increase the lung residence time and, by that, the particle fraction deposited by sedimentation. They also measured the mass fraction of particles exhaled and their data are reprocessed into Fig. 1. Since they studied only three individual particle sizes, limited information was obtained about the deposition and distribution of particles within the submicron range ($D < 1$ μm). To further increase our understanding of particle behavior within this range, extrapolation of the Usmani data was undertaken using basic aerosol physics to derive and check the probabilities of inertial impaction and sedimentation respectively within this range. The results of this extrapolation and the procedures used for that are presented and discussed in the results and discussion sections.

2.2. In vitro evaluation of four marketed DPIs

2.2.1. Materials

The four DPIs tested in this study were Symbicort 160/4.5 μg Turbuhaler (SY-TU; AstraZeneca), Seretide 250/50 μg Diskus (SE-DI, GlaxoSmithKline), Rolenium 250/50 μg Elpenhaler (RO-EL, Elpen) and Foster/Fostair 100/6 μg NEXThaler (FO-NE; Chiesi). Three different batches were tested and all inhalers were obtained from the market in different European countries depending on their availability. Solvents, eluents and chemicals (high-performance liquid chromatography [HPLC] grade) were supplied by Biosolve Chimie (Dieuze, France), Merck (Darmstadt, Germany), VWR International (Fontenay-sous-Bois, France) and Fischer Scientific (Loughborough, UK). Ultra-pure water of Milli Q quality (Millipore, Amsterdam, the Netherlands) was used for preparing drug solutions and for rinsing the filter holder and impactor parts. Delivered doses were collected on 50 mm glass fiber filters type A/E (PALL Corporation, USA) and passed through 0.2 μm membrane filters Whatman, FP 30/0.2 CA-S (Dassel, Germany) for water or Phenomenex RC 0.20 (Utrecht, the Netherlands) for organic solvents prior to HPLC analysis. Active pharmaceutical ingredients for the HPLC assays were provided by DFE Pharma (Goch, Germany), Teva Pharmachemie (Haarlem, the Netherlands), Almirall Sofotec (Bad Homburg, Germany) and AstraZeneca (Mölndal, Sweden).

2.2.2. Air flow resistance

For inhalers, the air flow resistance (R) is the proportionality constant in the relationship between the square root of the pressure drop (dP) across the device and the corresponding flow rate (Φ): dP = $R \cdot \Phi$ [25,26]. For all DPIs, the pressure drop was recorded for the range of flow rates between 0 and 90 L/min with incremental steps of 10 L/min. In this manuscript, values for L/min given are for ambient conditions (1013 mbar and 20 °C). Calibration was for three devices per batch and each device was measured in triplicate, yielding 27 recordings per type of inhaler. The inhalers were

connected through a coupling flange with a seal ring fitting closely around the mouthpiece to a thermal mass flowmeter (5863S; Brooks Instruments, the Netherlands). The pressure drop was measured immediately downstream of the mouthpiece with a differential pressure gauge (HBM PD1; Hottinger Baldwin Messtechnik, Germany). Triplicate series per device were averaged to evaluate the inter-batch variation.

2.2.3. Consistency of delivered dose (DD)

Consistency of DD at each pressure drop was determined for 10–20 doses per device, depending on the labelled number of doses. For RO-EL 10 doses (blisters) were taken randomly per batch. For the multi-dose (including multiple unit-dose) inhalers, doses were taken from the beginning, middle and end of labelled contents. Doses from the multi-dose inhalers were delivered with an interval time of at least 30 s to prevent excessive tribocharge of the device. Delivered doses were measured in 4 L of air drawn through the inhalers at 2, 4 and 6 kPa. The inhalers were connected to a filter system similar to that described by the US Pharmacopeia (USP) 30 [31]. Delivered doses collected on 50 mm glass fiber filters were dissolved by submersion of the filters in a 100 mL beaker. The collection tube and coupling flange were thoroughly rinsed with the same solvents and the washings were added to the beaker for HPLC analysis. For all drugs and drug combinations checks were made to ensure that no adsorption occurred to the filters and that the filters did not release leachables to affect the analysis. Delivered doses were expressed as percent of label claim.

2.2.4. Consistency of delivered fine particle dose (FPD)

Next Generation Impactors (NGIs) were used according to the procedures given in the USP 30 [27] after mensuration by the supplier (Copley Scientific, Nottingham, UK). Impactor cups were coated using a mixture of Brij 35-p in ethanol with glycerol as described previously [28]. The pre-separator was filled with 15 mL of a suitable solvent for the HPLC analysis. For the dispersion tests, the inhalers were connected to a coupling flange on the USP induction port of the impactor with silicone rubber seals fitting closely around their mouthpieces. Delivered FPDs in 4 L air were measured at 2, 4 and 6 kPa. After the impactor test, drug deposits on the impactor stages were dissolved and the USP induction port and pre-separator were rinsed thoroughly. FPDs presented are the mean of 20–30 doses per kPa per device, depending on the labelled number of doses per device and each test comprised 5 or 10 successive doses analysed together to obtain sufficient accuracy. For the multi-dose DPIs, series of 5–10 doses for a test were taken from the beginning, middle and end of labelled contents. Doses from the multi-dose inhalers were delivered with an interval time of at least 30 s to prevent excessive tribocharge of the device. For RO-EL three series of 10 doses were taken randomly per batch. FPDs were expressed as percent of the label claim (yielding fine particle fractions: FPFs < 5 μm). Each FPF < 5 μm was divided into subfractions < 1, 1–3 and 3–5 μm.

2.2.5. HPLC procedures

Filter deposits (from consistency of DD measurements) and impactor stage and induction port deposits (from consistency of delivered FPD measurements) were analysed on an Agilent 1100 series HPLC (Waldbronn, Germany) using different solvents, mobile phases (isocratic or gradient elution), columns and settings for column temperature, pump flow rate, injection volume and wavelength, depending on the drug combination to be measured. All assays were previously validated. Gauge lines were prepared from pure drug combinations. For FO-NE the effect of magnesium stearate on the assay was checked before starting the analyses.

It must be mentioned that different manufacturers use different label claims for their products. Label claims may refer either to

masses measured by dose measuring principles, respectively, masses measured into blisters, or to delivered masses (ex-mouthpiece doses). To complicate the situation even further, some multi-dose DPIs measure higher powder quantities than the label claim indicates. On the European market there seems to be no consensus or good regulation in this respect and therefore, DDs and FPFs expressed as percent of label claim are not fully comparable.

3. Results and discussion

Different *in vitro* evaluation studies with some of the currently marketed DPIs are known. The DPIs tested in various studies may produce rather extreme differences in aerosol properties, but the results from different studies usually cannot be compared with each other because of the differences in testing conditions, data processing and data presentation used. To make a comparative evaluation regarding the most favorable size distribution for total and deep lung deposition possible, we tested four ICS-LABA combination DPIs under precisely the same conditions (2, 4 and 6 kPa) and used the same data presentation for all devices by computing mass fractions of the delivered aerosols in the size ranges < 1, 1–3 and 3–5 μm. This is breaking with a tradition according to which only MMADs of delivered aerosols are given. We chose this approach because MMADs do not provide information about the size distribution of the aerosol or about the mass (or the size) fraction of the dose for which the MMADs were computed. We computed MMADs only for the FPFs < 5 m obtained at 4 kPa (Table 2).

Also, many different lung deposition simulation studies in the past have shown which particle diameters have the highest deposition probability in simplified lung models, as described by Weibel and various others [29] as a function of the most relevant ventilatory parameters, including particularly the inhaled volume, flow rate and residence time [30,31]. Various empirical relationships and deposition parameters have been derived to mathematically fit the deposition behavior of inhaled particles [17,20]. However, many of these relationships and parameters were derived for deposition during tidal breathing instead of deep inhalation through a DPI, whereas the human lung is much more complex than the simplified Weibel cascade of bifurcating tubes. The approach in this study is different, using experimental deposition data for monodisperse aerosol particles from inhalation at a moderate flow rate, similar to inhaling through a DPI [24]. These data are explained and extrapolated towards the submicron range using basic aerosol physics. For our computations and discussion we assumed that no uncontrolled or unknown effects such as condensation or hygroscopic particle growth and/or tribocharge effects in the *in vivo* deposition study occurred. Several studies have shown that such effects can significantly affect lung delivery for orally inhaled aerosols [32–34].

3.1. Evaluation of previously published deposition data

The drug dose deposited in the lungs depends on the delivered lung dose and the dose fractions lost by deposition in the oropharynx and by exhalation. The probability of a particle being deposited in the oropharynx can be predicted with the impaction parameter (IP) which is the product of a particle's density (ρ), the square of its diameter (D) and its velocity (U). In practice, the flow rate (Φ) can be used instead of the velocity for the computations when the same inhaler is used for all experiments and a linear proportionality exists between U and Φ [32]. Usmani et al. used monodisperse particles ($\rho = 1$) with aerodynamic diameters of 1.5, 3 and 6 μm [24] and because they presented the mean flow rates at which these particles were inhaled, mean IP-values for their aerosols

can be computed. The relationship in Fig. 2 between the computed impaction parameters and the experimentally obtained oropharyngeal deposition values (from Fig. 1) enables a realistic estimation of oropharyngeal depositions for smaller particle diameters towards zero. Using this relationship oropharyngeal deposition values were assessed for particle diameters 0.6, 0.8, 1.0 and 1.25 μm at 31 and 67 L/min.

Small particles ($D < 1$–2 μm) deposit primarily by sedimentation in the periphery of the lung [30]. The probability of sedimentation depends on the particle's terminal settling velocity (U_{TS}), the distance (H) of a particle to an airway wall (below the particle) and the residence time (t) given for settling. Improving the settling of small particles in the respiratory tract can thus be achieved by increasing their residence time in the peripheral airways (i.e. by elongating the breath hold pause after deep inhalation). If the distance (H) is greater than the product of time and settling velocity ($H > U_{TS} \cdot t$), the particle may be exhaled again. Therefore, for a particle at a given distance from the airway wall and a fixed residence time in the lungs, the chance of being deposited depends on the terminal settling velocity, which is achieved when the force of gravity is in equilibrium with the Stokes' drag or resistance force. The terminal settling velocity decreases exponentially with decreasing particle diameter {$U_{TS} = Fu(D^2)$} and the time needed to travel a fixed distance increases correspondingly. Because the chance of being exhaled for the smallest particles in the aerosol is inversely proportional to the chance of being deposited by sedimentation, the fraction exhaled shows basically the same dependence on the particle diameter as the time to fall a certain distance. Particles of 3 μm require a residence time of only 1.6 s to fall across the diameter of a respiratory bronchiole (0.45 mm), but this time increases to 12.8 s and 511 s for 1.0 and 0.1 μm particles respectively (with correction of the Stokes' drag force for the slip flow) [20]. In Fig. 3 the time to fall a distance equal to the diameter of a respiratory bronchiole is plotted for particles in the size range from 0.4 to 6 μm. The relationship is shown to illustrate the steep increase in this time for submicron particles. For such small particles the time to travel across the diameter of a respiratory bronchiole becomes significantly longer than the (average) attainable breath hold pause. Fig. 3 also shows the exhaled fractions of 1.5, 3 and 6 μm particles from the Usmani study [24] and the relationship between the fraction exhaled and particle diameter shows more or less the same trend as the settling velocity for these particle diameters. This trend can be extended into the range of submicron particles by roughly following the trend for the settling time and using real *in vivo* (and simulated) deposition data presented in the literature for refinement [20,29–31]. Literature data show a minimum lung deposition efficiency for
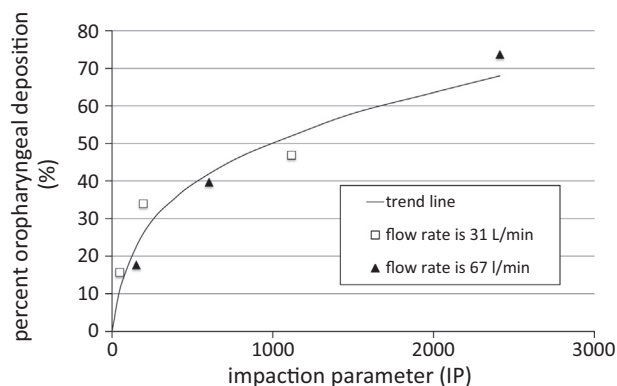

Fig. 2. Percent oropharyngeal deposition as a function of the impaction parameter (IP = ρ·D²·Φ). Data points derived from the Usmani study [24].
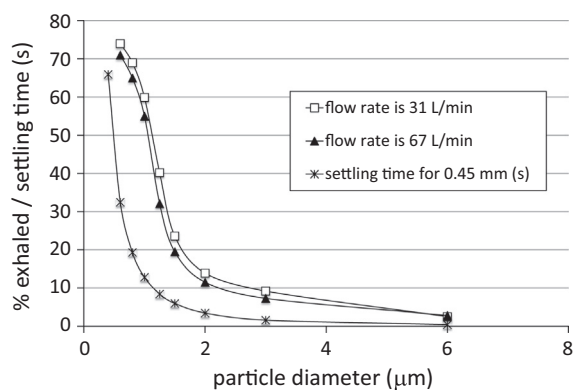
Fig. 3. Percent exhaled and time to fall a distance of 0.45 mm (equal to the diameter of a respiratory bronchiole) as a function of the particle diameter. Percentages exhaled for 1.5, 3 and 6 μm particles are derived from the Usmani study [24].

Fig. 4. (A) Aerosol losses due to oropharyngeal deposition and exhalation as a function of the particle diameter computed for 31 (A) and 67 L/min (B). The figures are based on the experimental data of Usmani et al. (1.5–6 μm) [24] and the extrapolated data from Figs. 2 and 3 for the particle size range 0.6–1.5 μm.

0.5 m particles of only 20% and this seems in good agreement with what can be expected on the basis of an exponentially increasing settling time (Fig. 3). It must be recognized that there is no constant proportionality between settling time and fraction exhaled in Fig. 3, as the deposition efficiency of 1.5 μm particles is less influenced by inertial deposition than that of 3 and 6 μm particles. Moreover, when the particle size approaches that of the surrounding air molecules, the settling time becomes infinite whereas deposition efficiency increases again due to diffusion or Brownian motion [20,30]. For the assessment of the percent exhaled in Fig. 3, we took account of this minimum in deposition efficiency. Mass fractions of particles in this size range are extremely low in therapeutic aerosols however, and, therefore, hardly contribute to the lung dose. For that reason, our extrapolations in Fig. 4A and B do not encompass this size range: we stopped at 0.6 μm.

The fractions deposited in the oropharynx together with the fractions exhaled comprise the total aerosol fractions not available for lung deposition, referred to as 'losses'. Figs. 4A and B show the sum of these losses from oropharyngeal deposition and exhalation as a function of the particle diameter at 31 and 67 L/min respectively, based on the Usmani study [24]. Obviously, the percentages complementary to 100% represent the fractions deposited in the lungs. Fig. 4 shows that the deposition fraction decreases dramatically for particles in the submicron range. Such particles in therapeutic aerosols are mostly exhaled again and, therefore, it is highly unlikely that they contribute to improved total and peripheral lung deposition compared to aerosols consisting of particles in the micron-range (D = 1–5 μm). The result is in agreement with various theoretical deposition modeling studies showing that there is minimum deposition for particles within the size range 0.1–1 μm [30,31]. Figs. 4A and B show nearly the same fraction exhaled, as sedimentation deposition is largely independent of the flow rate (Fig. 3), but significantly higher oropharyngeal deposition fractions and the increase in oropharyngeal deposition are largest for the largest particles. This makes the peak of % lung deposition more pronounced, whereas the particle size for which the peak is computed is almost the same: at 67 L/min it shifts to 1.5 μm. Therefore, the lung deposition percentages in Figs. 4A and B, complementary to the exhalation and oropharyngeal deposition percentages, have the highest values in the range of diameters between 1.25 and 3.5 μm at low to moderate flow rate, and because the precise peak depends on the inhalation manoeuvre and changes slightly towards lower diameters both at higher flow rates and at longer breath hold pauses we decided to adhere to the proposed division into subfractions of <1, 1–3 and 3–5 μm.
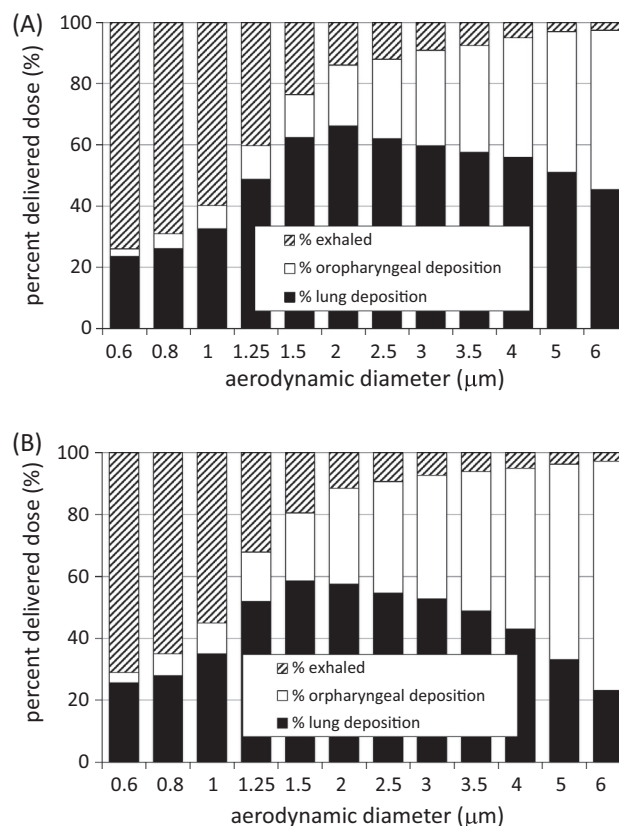
There is a compelling reason for not taking the values in Fig. 4 as absolute. They are only indicative for the effect of particle size on lung deposition behavior. Although they were derived from real deposition data [24], aerosol deposition fractions from marketed inhalers, particularly dry powder inhalers, may be completely different. They are most likely to be lower than those shown in Figs. 4A and B for a number of different reasons. First of all, aerosols from marketed inhalers are polydisperse and total lung deposition for such aerosols is the sum of the deposition fractions for the individual particle sizes in these aerosols. Total lung deposition also depends on the mass fractions of each of these particle sizes in the aerosol. Nearly all marketed dry powder inhalers, except the Turbuhaler, make use of adhesive mixtures and their dispersion during inhalation is rather incomplete. Only a fraction of the total drug mass is liberated from the carrier particle surface within the appropriate size range for effective lung penetration and deposition. Therefore, the mass fraction of fine particles in the aerosol may be even more important than their MMAD. High total mass fractions of particles (FPFs < 5m) may contain more particles in the optimal size range for lung deposition than low FPFs < 5 m, even if the lower FPF < 5 m has a considerably lower MMAD. Furthermore, lung deposition of the same aerosol may vary with the inhaler design. Different mouthpiece designs may result in different jet effects and circulations, even at the same flow rate and this can affect the oropharyngeal deposition in particular [35]. Nevertheless, and irrespective of the absolute deposition values, the effect of particle size on deposition can well be estimated from Figs. 4A and B and it may be clear that high mass fractions of submicron particles in the aerosol are not likely to contribute to improved lung deposition.

3.2. In vitro deposition data

The four DPIs tested in this study all contain a similar combination of a bronchodilator drug (LABA) and an ICS, but they are different in design and many other respects. All are originator devices, except for the Rolenium Elpenhaler, with different formulations of soft spherical pellets (Turbuhaler) and carrier-based adhesive mixtures using different carrier products (the other three inhalers). The Rolenium Elpenhaler was selected for this study because this DPI is marketed as being equivalent to the Seretide Diskus and contains the same drug combination in the same strengths as the Diskus. Some of the DPIs tested (Turbuhaler and NEXThaler) are multi-dose inhalers with a dose measuring system to be operated by the patient; the Diskus is a multiple-unit dose inhaler with the doses pre-metered in blisters. The Elpenhaler makes use of separate formulations for both drugs stored in separate blisters whereas all other devices contain one formulation with both drugs. The formulation in the NEXThaler has been prepared with magnesium stearate as a force control agent [36]. The use of force control agents enables the separation of the drug particles more easily from the carrier crystals during inhalation and part of this water insoluble excipient is inhaled. Only the Turbuhaler and NEXThaler have a specific powder dispersion principle for the aerosol generation energized by the inspiratory flow.

The resistances to air flow and the flow rates corresponding to a pressure drop of 4 kPa of the four DPIs are presented in Table 1. The data show that Turbuhaler and NEXThaler are of medium to high resistance (4 kPa corresponds with 59 L/min) whereas Elpenhaler (68.3 L/min) and Diskus (75.2 L/min) are of medium resistance [37]. Nevertheless, these differences in resistance between all four devices are relatively small and hardly of any influence on the deposition pattern of particles with the same size (distribution). Their *in vitro* delivered doses (DDs) are illustrated in Figs. 5A and B. These DDs are expressed as percentage of the label claim for ICS (Fig. 5A) and LABA (Fig. 5B) for three different pressure drops: 2, 4 and 6 kPa. Values measured for both drugs are from the same inhaler and same dose numbers at the same pressure drop and generally well within the expected range of 75–90% of the label claim. The differences between individual doses from the same inhaler (at all pressure drops) are quite considerable: maximum and minimum individual doses indicated by the spread bars may differ by as much as a factor of 2.

While the proportion of delivered dose is important, the fine particle fraction (FPF < 5 µm) within the delivered dose is considered a direct measure of the inhaler's potential for lung deposition. Fig. 6 shows these fractions as a percent of the label claim for ICS and LABA respectively with the spread bars indicating the extremes obtained. The Diskus and Elpenhaler delivered the lowest proportions of particles <5 µm. FPFs from the Turbuhaler and NEXThaler were 2–3 times higher at 4 and 6 kPa compared to Diskus and Elpenhaler. The difference in fine particle fractions
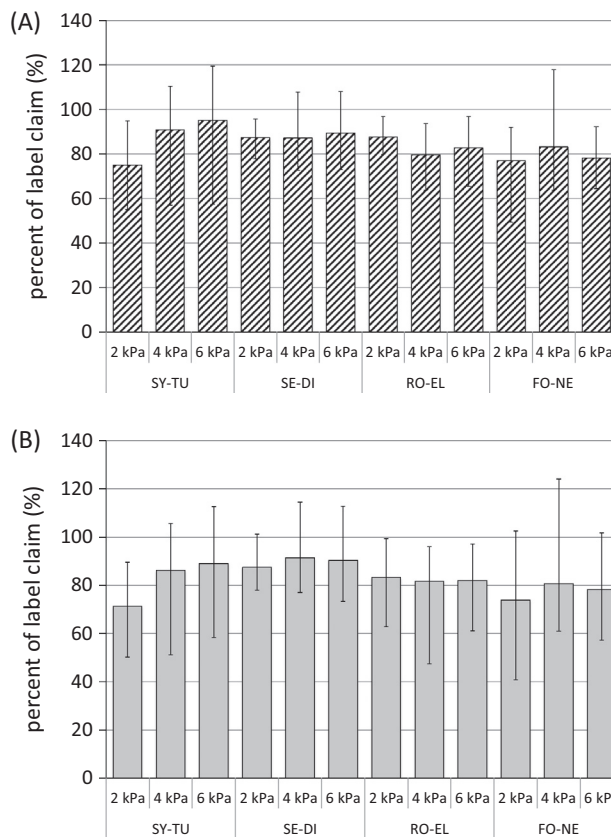

Fig. 5. Delivered doses (as percent of the label claim) from Symbicort Turbuhaler (SY-TU), Seretide Diskus (SE-DI), Rolenium Elpenhaler (RO-EL) and Foster NEXThaler (FO-NE) as function of the pressure drop. Mean of 30–60 doses per kPa (from three batches). The spread bars indicate the batch variation (highest and lowest individual doses measured). (A) corticosteroid and (B) bronchodilator fractions.
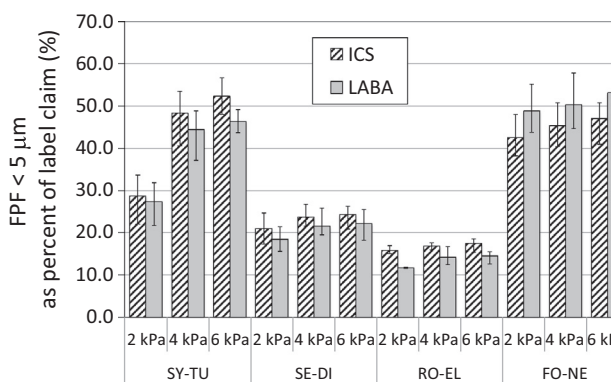

Fig. 6. Delivered fine particle doses (FPFs < 5 µm as percent of the label claim) from Symbicort Turbuhaler (SY-TU), Seretide Diskus (SE-DI), Rolenium Elpenhaler (RO-EL) and Foster NEXThaler (FO-NE) as function of the pressure drop. Mean of 20–30 doses per kPa (from three batches). The spread bars indicate the highest and lowest individual batches measured.

between the Diskus and Elpenhaler shows furthermore that these devices, presented to the market as equivalent, are in fact not comparable. The Diskus, Elpenhaler and NEXThaler show an almost pressure drop-independent output for the fine particle dose. In contrast, FPF increases with increasing inspiratory effort for the Turbuhaler. The increase is most pronounced between 2 and 4 kPa and is desirable to compensate for oropharyngeal losses and a shift in deposition towards larger airways at higher flow rates [22].

Table 1
Air flow resistances and flow rates corresponding to 4 kPa pressure drop across the inhaler. Mean of three devices per batch [a] and three duplicate measurements per device. Min and max represent the lowest and highest mean batch value.

DPI	Air flow resistance ($kPa^{0.5}\ min\ L^{-1}$)			Flow rate at 4 kPa ($L\ min^{-1}$)		
	Mean	Min	Max	Mean	Max	Min
SY-TU	0.0340	0.0338	0.0343	58.8	58.3	59.2
SE-DI	0.0266	0.0240	0.0295	75.2	83.3	67.8
RO-EL	0.0293	0.0291	0.0295	68.3	68.7	67.8
FO-NE	0.0339	0.0334	0.0345	59.0	59.9	58.0

[a] For Symbicort Turbuhaler (SY-TU), Seretide Diskus (SE-DI), Rolenium Elpenhaler (RO-EL) and Foster NEXThaler (FO-NE).

Differences between the inhalers were obtained not only for the total fine particle fractions (<5 µm), but also for the structures of these fractions. Figs. 7–9 show the rather extreme differences in these structures. The high fraction of particles <1 µm in the NEXThaler (Fig. 7), on average more than one-third of the FPF <5 µm, contributes to the low MMAD of the aerosol produced by this device (Table 2) [16]. Submicron fractions in the aerosols from the other inhalers are much lower and those in the aerosols from the Diskus and Elpenhaler are almost negligible. Differences in FPF <5 µm between the inhalers (Fig. 6) are also strongly reflected in the differences in the fraction 1–3 µm (Fig. 8). For this fraction, most relevant to total lung deposition (Fig. 4), Turbuhaler and NEXThaler score best. Fractions within this size range from the Elpenhaler are less than 25% and from the Diskus less than 50% of those from the best scoring DPIs at 4 and 6 kPa. Differences in the coarsest fine particle subfraction 3–5 µm are least pronounced (Fig. 9). This fraction may be more relevant to the LABA component than to the ICS component as particles in this size range are known to have a good bronchodilating effect when they are inhaled at a moderate flow rate [24]. The sum of the fractions 1–3 and 3–5 µm at 4 kPa (the total fine particle fraction without the submicron particles: FPF 1–5 µm) is given in Table 2.

The comparative *in vitro* evaluation part of the study shows that the DDs of all four DPIs tested were fairly comparable in spite of the fact that label claims are not defined unambiguously (Figs. 5A and B). In contrast, the delivered FPFs (<5 µm) differed considerably (Fig. 6) and so did the structures of these FPFs (Figs. 7–9). From DPIs with a higher FPF <5 µm, a higher total lung deposition may be expected at the same pressure drop, particularly when the resistances to air flow are comparable and the flow rates at which the aerosols are delivered to the respiratory tract are more or less the same (Table 1). Differences in the structure of FPF are more likely to influence drug distribution in the airways, including the oropharynx. It could be shown that DPIs delivering finer aerosols, such as the NEXThaler, contain particularly high fractions of submicron particles in the aerosol. Such particles <1 µm do pass the oropharynx effectively and travel into the bronchial tree. However, they have only a small chance of deposition there and are predominantly exhaled again (Fig. 4) as a result of the exponentially decreasing settling velocity with decreasing aerodynamic particle diameter. Particles in the size range 1–3 µm are much more relevant to total lung deposition (Fig. 4) and the extreme differences in this size fraction between the inhalers, which all have comparable air flow resistances, could be a good indicator for a choice between the devices. For the efficacy of inhaled therapy, other variables are also important, however. For instance, the differences in flow dependence between the
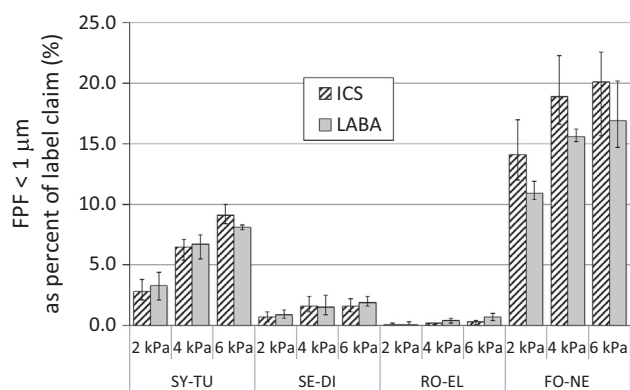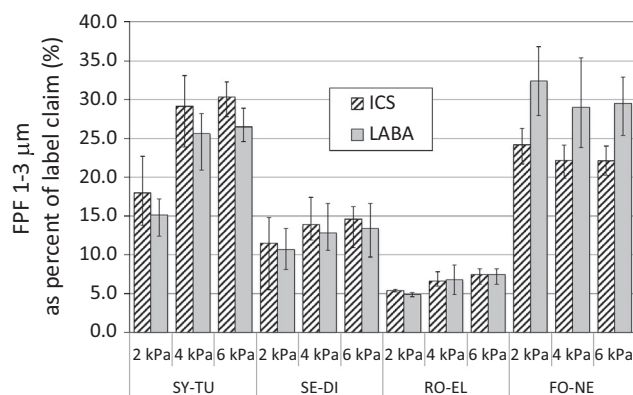

Fig. 8. Fraction of 1–3 µm particles as percent of label claim. For abbreviations and numbers of doses, see Fig. 6.

Fig. 9. Fraction of 3–5 µm particles as percent of label claim. For abbreviations and numbers of doses, see Fig. 6.
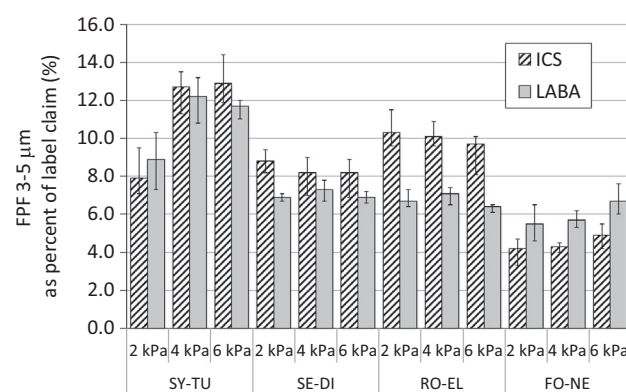
Table 2
Fine particle (sub)fractions < 1, 1–3; 3–5 and 1–5 µm as percent of the label claim at 4 kPa. MMADs are for the fraction < 5 µm (at 4 kPa).

ICS				
Fraction	SY-TU budesonide	SE-DI fluticasone	RO-EL fluticasone	FO-NE BDP
<1 µm	6.5	1.6	0.2	18.9
1–3 µm	29.1	13.9	6.6	22.2
3–5 µm	12.7	8.2	10.1	4.3
1–5 µm	41.8	22.1	16.7	26.5
<5 µm	48.3	23.7	16.9	45.4
MMAD (µm)	2.07	2.54	3.33	1.21

LABA				
Fraction	SY-TU formoterol	SE-DI salmeterol	RO-EL salmeterol	FO-NE formoterol
<1 µm	6.7	1.5	0.4	15.6
1–3 µm	25.6	12.8	6.8	29.0
3–5 µm	12.2	7.3	7.1	5.7
1–5 µm	37.8	20.1	13.9	34.7
<5 µm	44.5	21.6	14.3	50.3
MMAD (µm)	1.96	2.36	2.88	1.57

devices may have an effect on consistency of the therapy. An increasing fine particle dose with increasing flow rate is desired for compensating at least partly for the increased oropharyngeal losses (Fig. 4) and a shift in deposition towards larger airways [22]. From this viewpoint, none of the inhalers tested should be used at a higher pressure drop than 4 kPa. FPFs 1–3 µm and 3–5 µm are hardly further increased at higher pressure drops, in

Fig. 7. Fraction of submicron (<1 µm) particles as percent of label claim. For abbreviations and numbers of doses, see Fig. 6.

contrast to oropharyngeal deposition, particularly for the largest of the aerosol particles up to 5 μm (Fig. 1). Therefore, lung deposition will not further be increased between 4 and 6 kPa. The increase in FPF 1–3 and 3–5 μm between 2 and 4 kPa is highest for Turbuhaler and even slightly negative for NEXThaler. Aerosol emission patterns are also relevant to lung distribution but they have not been investigated in this study.

For lung deposition other variables are also important however. For instance, the differences in flow dependence between the devices may have an effect on the consistency of the therapy. An increasing fine particle dose with increasing flow rate is desired to compensate at least partly for the increased oropharyngeal losses (Fig. 4) and a shift in deposition towards larger airways [22]. Only the Turbuhaler delivers an increasing FPF <5 m (between 2 and 4 kPa) and this device should, therefore, not be operated at pressure drops >4 kPa to obtain the most consistent therapy. For the Diskus, Elpenhaler and NEXThaler maximal lung deposition may be obtained at 2 kPa, but for Diskus and Elpenhaler, this is considerably lower compared to Turbuhaler and NEXThaler. Not only the amount and quality of the aerosol, but also several patient factors are important. Patient factors are quite complex, particularly for DPI use, and include gender, age and disease related factors as well as compliance with correct inhaler use and adherence to the therapy. Therefore, *in vitro* assessments may overestimate the quantity of drug deposited in the lungs. There is also a wider variability in lung deposition *in vivo* than would be predicted by *in vitro* measurements [38], and the relationship between deposition and effect, as a function of the particle diameter, can only be studied when all other controllable variables are kept the same. This will be one of the challenges for future research.

4. Conclusions

The steep increase in the fraction exhaled with decreasing particle diameter for submicron particles shown in Fig. 3 suggests that particles <1 μm are not suitable for inhalation. High mass fractions of submicron particles contribute to a lower MMAD of the aerosol, but such particles are also largely exhaled again as can be concluded from previously presented *in vivo* deposition data for monodisperse particles, extrapolated towards the submicron range. Therefore, previously presented size definitions for so-called 'extrafine' particles seem to be irrelevant; from the viewpoint of clarity, 'submicron' is proposed as a well-defined and much more meaningful alternative. Lung deposition percentages in Fig. 4 confirm various lung deposition modeling studies that the particle size range 1–3 μm is most favorable for total and deep lung deposition, when such particles are inhaled at a moderate flow rate (approx. 30–60 L/min) and given sufficient time for settling in the most distal airways (preferably 10 s, following a deep inhalation after maximal exhalation).

Considerable differences exist between the delivered fine particle doses (FPF <5 μm), and their structures, from Symbicort Turbuhaler, Seretide Diskus, Rolenium Elpenhaler and Foster NEXThaler. The differences are most pronounced for the less favorable submicron ($D < 1$ μm) particle fractions and the more relevant fractions 1–3 μm. The FPFs 1–3 μm vary by a factor of 4 between the extremes in this study with Turbuhaler and NEXThaler being much better than Diskus and Elpenhaler. In contrast, most mass fractions of particles 3–5 μm are of same order of magnitude. On the basis of these differences in fine particle output, significant differences in delivered lung dose and drug distribution over the respiratory tract between the inhalers may be expected when they are operated correctly. The fine particle fractions at the same pressure drop in this study are directly comparable as they are delivered at roughly the same flow rate, due to the comparable resistances to air flow for all four DPIs.

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