INTRODUCTION

Inhalation delivery systems are a key factor in the design and development of novel therapeutics. Depending on therapeutic goal, different inhalation systems can be employed, including pressurised metered dose inhalers (pMDIs), dry powder inhalers (DPIs) and the newer generation of electrically-assisted aerosol delivery systems. These different technologies enable delivery of therapeutic agents to different areas of the respiratory system, with implications for efficacy, pharmacodynamics and pharmacokinetics.

In vitro–in vivo correlation (IV–IVC) is a crucial component in the development of new respiratory medications. However, the methods currently available for the study of absorption, distribution, metabolism and excretion (ADME) properties of inhaled therapeutics often fail to accurately model the clinical pharmacokinetic profiles observed in patients.

In order to gain a better understanding of the pharmacokinetic profiles of novel respiratory medications, a method that is more clinically relevant and predictive is required. A recent study [1] has shown that in vitro dissolution and absorption profiles can be used to predict the clinical pharmacokinetic profiles of new respiratory medications.

METHODS

Two marketed formulations of FP were tested: Flutide (GSK), (FP, strength 50 μg) and Flutiform (Mundipharma), (FP, strength 250 μg and formoterol strength 10 μg) provided as pressurized metered dose inhalers (pMDIs). The canisters were connected to the US Pharmacopeia Inhalation Aerosol Port No 1 of the Preciselinhale® aerosol delivery system and actuated into an air flow of 15 L/min. The aerosols were collected in the Preciselinhale® aerosol holding chamber and immediately dispersed to the Dissolv® and IPL modules at flow rates of 1000 mL/min and 250 mL/min, respectively.

Particle size distributions of the generated aerosols (Figure 1) were measured with an 8-stage Marple cascade impactor at a flow rate of 2 L/min. At the start of the Dissolv® experiment, the aerosol particles deposited on the cover slip (with Preciselinhale®) are brought into contact with the mucus consisting of 1.5% polyethylene oxide [4] and 0.4% L-alphaphosphatidyl choline (Sigma). The mucus had been applied to a polycarbonate membrane corresponding to the basal membrane of the airway mucosa. The mucus-solution mixture forms a diffusion barrier. On the other side of the membrane, the blood simulant (consisting of phosphate buffer with 4% albumin) is streaming.

Dissolved particles were absorbed at a perfusate flow rate of 0.4 mL/min. Dissolution was studied by observing particle disappearance using optical microscopy, and by chemical analysis of substance removed by absorption in the flow-past perfusate. The amount of FP retained in the system (comparable with the amount of drug retained in lung tissue in IPI) was analysed at the end of the 2 h experiments.

The IPL was prepared as previously described [2]. Briefly, whole lungs were isolated from female CD IGS (Sprague Dawley) rats (Charles River, Sweden). The lungs were ventilated with a negative alternating pressure and perfused in single-pass mode with a Krebs-Henseleit buffer containing 4% albumin. For the exposures to the IPL, the FP aerosols actuated via the induction port, were delivered to the lungs (n=3 per formulation) by the Preciselinhale® active dosing system, which calculates in real time the cumulative inhalated aerosol dose. The system automatically terminates the exposure when the inhaled target dose is reached. The liquid precipitated evaporated within Preciselinhale® system, and thus did not reach the lung. The perfusion was repeatedly sampled in an automated fraction collector during a 2 h period post exposure. Thereafter, the lungs and trachea were harvested for analysis of the amount of FP retained in the tissues after the perfusion period.

Analytical quantification of FP in all samples from the Dissolv® and IPL experiments was performed by LC/MS/MS, with a LOD of 100 pg/mL.

RESULTS

The concentration of absorbed FP in the perfusate/blood simulant over time is shown in Figure 2A (Flutide) and 2B (Flutiform). In Figure 2C (Flutide) and 2D (Flutiform) for both models the amount of FP cleared with the perfusate during each sample interval has been expressed as percent of the initially deposited dose cleared per second. This compensates both for the differing deposited doses of the two exposure models and for the difference between the varying perfusate flow rate of the IPL and the constant flow rate of the Dissolv® system. In Figure 2C (Flutide) and 2D (Flutiform) is shown the amount of FP still retained in the rat lung and the Dissolv® lung model at each sample time, expressed as fraction of the initially deposited dose. The retained FP fraction represents either unsolubilised particles or dissolved substance not yet absorbed by the perfusate/blood simulant.

DISCUSSION AND CONCLUSIONS

Previously published methods on in vitro dissolution testing in inhalation drug development, such as the flow through cell, paddle apparatus and the Franz cell, all give cumulative dissolution/absorption curves [5, 6] which are not easily comparable to in vivo pharmacokinetic profiles. In contrast, the Dissolv® in vitro dissolution/absorption method [1] has a dynamic flow-past perfusion strategy which generates concentration curves containing c<sub>max</sub> and AUC values. These values can be more readily compared with human clinical pharmacokinetic profiles. Here, the rat IPL, previously shown to be a good pharmacokinetic model [2, 3], was used for comparison.

In Figure 2 it is shown that the profiles in the Dissolv® are very similar to the IPL profiles, especially for Flutide. For the normalized perfusate clearance values, the curves are also quite similar in shape. The curves on fraction retained (Figure 3) in the IPL and Dissolv® models rank both FP formulations similarly. A large fraction of drug is retained in the air/perfusate barriers of both systems, as expected from the low solubility of the FP formulations. One factor contributing to the faster clearance of Flixotide compared to Flutide in both exposure models, is likely to be the finer particle size distribution of Flutide. The concentration of absorbed FP in the perfusate/blood simulant over time in the IPL (black curves) and the Dissolv® (pink curves): Panel C (Flutide) and D (Flutiform) show the percent of deposited doses (IPL black curves and Dissolv® pink curves) clearing with the perfusate clear per second.

The key exposure and pharmacokinetic parameters for the IPL and Dissolv® system are presented in Table 1.

**Table 1.** Key exposure and pharmacokinetic parameters (± standard deviation) for FP (Flutide) and Flutiform in the IPL and Dissolv® system. C<sub>max</sub> Maximum concentration of drug in the perfusate, t<sub>1/2</sub> Time to maximum concentration in the perfusate, Fraction perfused/peak values for the fraction of the deposited dose cleared with the perfusate per second, t<sub>1/2</sub> Time at which Fraction perfused occurred.

It is evident, however, that Dissolv® can generate pharmacokinetic profiles of FP that resemble those in the rat lung. This indicates that Dissolv® may be a valuable in vitro dissolution/absorption method to use for IV–IVC in the development of new and generic drugs.