The Dissolv*It*: An *In Vitro* Evaluation of the Dissolution and Absorption of three Inhaled Dry Powder Drugs in the Lung

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INTRODUCTION

Whereas a number of methods have been devised to simulate drug dissolution in the gastro-intestinal tract [1], fewer methods have been proposed for drug dissolution in the lungs.

Often the dissolution step [2] is studied separately from the subsequent absorption step [3] in the lungs. However, it has been shown that the dissolution step of a low-solubility solute is directly coupled to the over-all absorption process in the lungs [4].

Local saturation around deposited particles greatly affects overall absorption in the lungs. It would therefore seem possible that a diffusional barrier of physiological dimensions [5] introduced into the *in vitro* model would best simulate the overall resistance to mass transfer during the release of a particle-associated solute in the lungs.

The Dissolv*It* system was designed to simulate both the dissolution of particles into the air-blood barrier and thereafter absorption of the dissolved particles into the blood, of soluble drugs from polydisperse aerosols in the lungs.

The objective here was to investigate the Dissolv*It* system as an *in vitro* dissolution/absorption classification tool.

METHODS

The dissolution system consists of a single-use dissolution cell, a precision-controlled pump for the perfusion medium and an inverted microscope with a high resolution camera. The dissolution cell is perfused in flow-past configuration and the perfusate is collected in a fraction collector (Figure 1).

The single-use cell consists of an injection-molded polycarbonate cell with a porous polycarbonate membrane. The polycarbonate membrane mimics the basal membrane of the airway mucosa and separates the diffusion barrier of the gel from the streaming perfusate on the other side. The entire Dissolv*It* test cell rack is heated to 37ºC.

Aerosols of three dry powder drugs were generated with the PreciseInhale exposure platform [6]. The physiochemical properties of the compounds which were measured at GSK are shown in Table 1.

Compound	Solubility in SLF pH6.9 (µg/ml) (0.5-24hrs)	Permeability (nm/s MDCK, SLF pH6.9)	Chrom-logP	Ionisation at pH 7.4 (Hi/med/low)	Acid or Base
A	0.16-0.25	58.1	6.1	Neutral	Neutral
B	0.6-1.6	104.3	3.4	low	Weak base
C	400	33	3.6	High	base

Table 1: Characteristics of test compounds A, B and C.

The aerosols were deposited on circular microscope glass cover slips. The coated cover slips were then applied to an artificial, physio-chemical lung surface barrier consisting of 1.5% polyethylene oxide [7] and 0.4% L-alphaphosphatidyl choline (Sigma) and a polycarbonate membrane.

Particle dissolution and absorption were studied both by optical microscopy, and by chemical analysis of substance dissolved in the flow-past perfusion medium (phosphate buffer including 4% albumin) and by analysis of substance retained in the model barrier after the perfusion period.

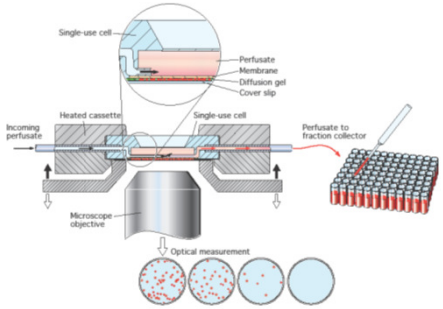

Figure 1: A schematic drawing of the Dissolv*It* system.

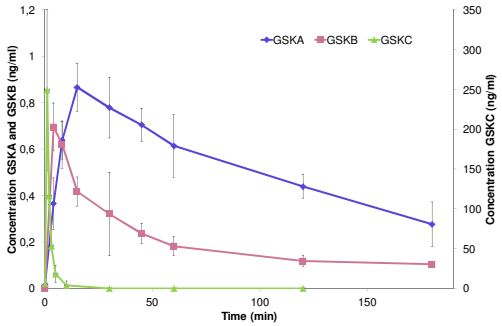

Figure 2: The concentration of test substances in the perfusate as a function of time after moment of contact between aerosol particles and the model barrier (mean ± SD, n=3).

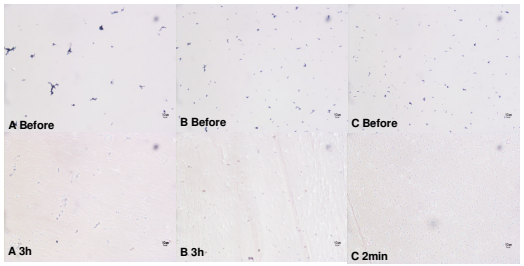

Figure 3: Microscope images of the drug particle disappearance. The photos shown are taken just before T=0 and at the end of the experiment (A, B) or at T=2 min (C).

The duration of the current experiments was 3h for A and B, and 2h for C. The perfusate samples were collected by a computer controlled fraction collector and then analyzed by LC/MS/MS. At the end of the perfusion period, the remaining substance on the cover slip and in the single-use cell with gel layer were extracted and also analyzed by LC/MS/MS.

RESULTS

The mass median aerodynamic diameter (MMAD) was measured using a Marple 8-stage cascade impactor [8]. Nine cover slips were simultaneously coated with drug particles out of which, three were randomly selected to determine the average amount of substance deposited per cover slip by HPLC. The particle sizes and applied doses are shown in Table 2.

The perfusate concentration curves for the substances are shown in Figure 2 and the C_{max} and t_{max} -values are summarized in Table 3.

	Aerosol Particle Size	Cover Slip Coating
Substance	MMAD, µm; GSD	µg, mean ± SD
A	5.07; 2.22	0.80 ± 0.08
B	2.23; 2.18	1.63 ± 0.32
C	2.34; 1.82	0.74 ± 0.09

Table 2: Aerosol particle size and the determination of the amount deposited on triplicate (for B n=5) cover slips.

	C_{max}, ng/ml	t_{max}, min
Substance	mean ± SD; n=3	mean ± SD; n=3
A	0.90 ± 0.08	20 ± 9
B	0.70 ± 0.10	4 ± 0
C	249 ± 101	1 ± 0

Table 3: Cmax and tmax during clearance of each substance, respectively.

Microscope images with indicative absorption data are shown in Figure 3. The T=0 image is taken just before particle contact with the barrier since it takes some seconds after the contact for the particles to be focused again.

CONCLUSIONS

The Dissolv*It* system successfully differentiated between three compounds with different physiochemical properties. In this study, differences in the rate of absorption between these compounds appeared to be driven by solubility with the more soluble compounds reaching the C_{max} more rapidly.

Further work is required to determine whether the Dissolv*It* profiles are predictive of absorption in vivo.

ACKNOWLEDGEMENTS
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