Defining dosimetry & markers of inhaled exposure in drug development

Graham R. Paul[1], Graham I. Somers[1], Mark Gumbleton[2] & Glyn Taylor[2]
[1] GlaxoSmithKline, Park Road, Ware, Hertfordshire, SG12 0DP, England, UK
[2] Welsh School of Pharmacy, Cardiff University, King Edward VII Avenue, Cardiff, CF10 3XF, Wales, UK

Key objectives of studentship

To assess the extent to which changing the presentation of a pharmaceutical alters the kinetics of exposure, efficacy & toxicology of a compound following inhaled administration.

Introduction

Successful development of pharmaceuticals as dry powder formulations for treatment of respiratory diseases has significant challenges:

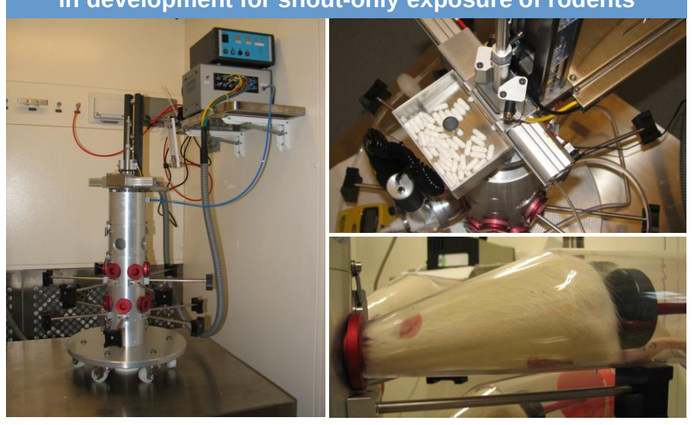
1. Adverse pathology: inflammation &/or accumulation of alveolar macrophages, & the relevance of the latter to physiological clearance as opposed to adverse toxicology.
2. Low 'safety margins' derived from:
   - no observed adverse effect levels (NOAEL), established by histopathological evaluations in inhalation toxicity studies;
   - factors applied for interspecies differences in lung dosimetry & in consideration of lesions that can't be monitored clinically.

Furthermore, development programs for inhaled drugs consume high masses of drug, relative to other exposure routes, which limits the extent to which liabilities can be discharged prior to committing significant resources to clinical development. This is especially true of conventional inhalation exposure systems for presentation of dry powder aerosols to animals. Strategies for using less test material in early respiratory drug development include:

- Progression of drugs to Phase I clinical trials using nebulised (liquid) formulations; powder formulations are developed once liabilities of unforeseen toxicity or undesirable PK/PD are discharged. This defers assessment of some liabilities such as tolerability or altered PK/PD for the clinical (particulate) formulation.
- Refining aerosol delivery systems to reduce compound requirements (Figure 1) to permit conduct of inhalation studies to discharge liabilities earlier in drug development.

Understanding differences in dosimetry, drug efficacy & toxicology of aerosols presented as powders or droplets is pivotal to such a strategy.

Figure 1: Capsule based aerosol generator (CBAG) in development for snout-only exposure of rodents



Understanding dosimetry

When administering drugs by the inhaled route to laboratory species, an understanding of the technical limitations & their implications for dosimetry is important. A dose is "a specific quantity of a therapeutic drug or agent taken at any one time or at specified intervals".①

Laboratory species are not dosed *per se*, but are exposed to an atmosphere containing the test material mixed with air (aerosol, fumes, gas or vapour). An animal breathes passively from this test atmosphere; some material is retained in the respiratory tract but most is not. It is difficult to determine the "specific quantity" of drug retained by an animal but the 'inhaled dose' (µg/kg) can be calculated as follows:

$$\text{Inhaled dose} = \frac{C \times RMV \times D \times IF}{BW}$$

C = Aerosol concentration (µg/L)

RMV = Respired minute volume (L/min); measured (for small numbers of animals) or estimated from mean body weight data② as follows:

$$RMV = 0.608 \times BW^{0.852}$$

D = Duration of exposure (min)

IF = Inhaled fraction; sometimes applied to account for the proportion of fine particle mass in an aerosol population

BW = Body weight (kg)

Deposition of particles in the respiratory tract is dependent upon:

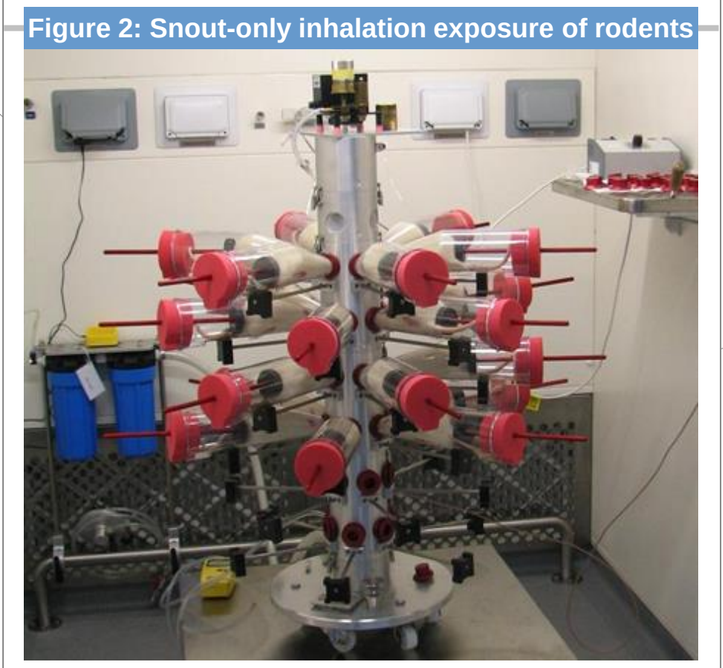
- Aerodynamic properties of the particle as a function of particle shape, mass, density & electrostatic charge (potential aggregation);
- Respiratory tract anatomy & implications for impaction of big particles (creating 'drug hot spots') & sedimentation (as air decelerates);
- Breathing pattern of the test subject.

The nature of breathing also greatly influences the site of deposition, e.g. rodents breathe nasally, humidifying an aerosol & filtering out larger particles, but human patients inhale medicines orally, bypassing the nasal cavities.

Methods

Male Crl:CD(SD) rats were administered GSK-CMPD, by inhalation (1 hour daily), at 'doses' of 1 or 45 mg/kg for up to 28 days. Aerosols were generated from dry powder formulations of 5% or 40% (w/w) micronized GSK-CMPD in lactose using Mk II Wright Dust Feeds③ at target concentrations of 24 & 1080 µg/L to achieve the respective doses. The aerosol was directed into a snout-only (flow-through) inhalation exposure chamber; rats restrained in polycarbonate tubes were attached to exposure ports (Figure 2) such that snouts projected into the chamber environment.

Figure 2: Snout-only inhalation exposure of rodents



Chambers were operated at ambient pressure using airflows of 10 L/min for the aerosol generator output & 16 L/min for chamber air extract; air balance was drawn via a tangential inlet in the chamber top section. Aerosols were sampled (2 L/min) for analysis of concentration (filters twice daily) & particle size distribution (Marple 296 cascade impactor once weekly); GSK-CMPD was analysed by HPLC/UV.

Plasma samples were obtained 0, 0.5, 1, 2, 4, 8 & 23 hours post 'dose' on Days 1 & 28 for toxicokinetic analysis. Lungs harvested 0 & 23 hours after a single 'dose' & 23 hours post 'dose' on Day 28 were homogenised. GSK-CMPD plasma & lung concentrations were analysed by HPLC/MS/MS.

Results

Sub-proportional increases in exposure (C_{max} & AUC_{0-t}; Table 1) & lung concentrations (Table 2) were evident for increases in inhaled 'dose'. Lung concentrations at 0 & 23 hours post exposure (Day 1) were similar for each 'dose' & increases in lung concentrations (>7-fold) & AUC_{0-t} (>3.6-fold) were seen from Day 1 to 28.

Table 1: Aerosol characterisation & toxicokinetics (plasma)

Estimated Inhaled Dose (mg/kg/day)[A]	Aerosol concentration (µg/L)	MMAD (GSD)[B]	Period (Day)	AUC_{0-t} (ng.h/mL)[C]	C_{max} (ng/mL)[C]	T_{max} (h)[D]
1.06	22.5	2.22	D1	6.34	1.60	1
	25.6	(2.39)	D28	32.5	2.28	0
42.3	1101	2.23	D1	38.6	5.20	0
	1026[d]	(2.59)	D28	140	12.7	0

A. Mean value for Days 1 to 28 of treatment (used for dose normalisation of AUC & C_{max})
B. MMAD = mass median aerodynamic diameter; GSD = geometric standard deviation; parameters within acceptable range (MMAD 1 to 3 µm & GSD 1.5 to 3) for rodents④
C. Calculated by dose-normalising for individual toxicokinetic sampling occasion & re-normalising with the overall estimated inhaled dose (Days 1 to 28).
D. Median time presented relative to the end of the 1-hour exposure period.

Lung concentrations were extremely variable, particularly after a single exposure (up to ±59% of mean; n=3). RMV, estimated from body weight, was less variable (mean ±5%; n=3) indicating contributory factors in addition to experimental error (nominally mean ±10%).

Table 2: estimated RMV vs lung homogenate concentration

Inhaled dose (mg/kg/day)	Parameter	RMV (litres)	GSK-CMPD lung concentratCion (ng/g)		
			Day 1-0h	Day 1-23h	Day 28-23h
1.06	Mean	0.303			684
	(sd)	(0.045)			(59.1)
	Range (± mean)	16%			9.6%
42.2	Mean	0.315			11000
	sd	(0.014)			(1999)
	Range (± mean)	4.6%			19%
0.77	Mean	0.342	69.2		
	sd	(0.020)	(30.6)		
	Range (± mean)	6.1%	45%		
0.77	Mean	0.337		67.8	
	sd	(0.231)		(2.2)	
	Range (± mean)	4.0%		3.5%	
44.1	Mean	0.364	1176		
	sd	(0.015)	(422)		
	Range (± mean)	4.8%	41%		
44.3	Mean	0.353		1498	
	sd	(0.007)		(797)	
	Range (± mean)	2.1%		59%	

Conclusions

Plasma & lung concentrations for a micronized material indicated accumulation of GSK-CMPD in lung tissue with repeated administration. Further experiments will evaluate systemic & lung exposure data after inhaled administration of GSK-CMPD using alternate aerosol forms. A higher variation in lung concentrations vs 'estimated RMV' infers potential for respiratory parameters to increase variability in achieved lung doses. Direct measurement of RMV will permit investigation of this phenomenon.

References

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Acknowledgements

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