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An in vitro method for assessing particle deposition from metered pressurised aerosols and dry powder inhalers

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Summary

An in vitro model was developed to assess particle deposition from aerosol formulations. The model consisted of a glass "mouth" and "trachea", the latter being bifurcated at the lower end. These were designed to reproduce the anatomical dimensions of an average healthy human airway. One of the bifurcated 'primary bronchi' was connected to a multi-stage cascade impactor and the other to a vacuum pump. Air was drawn independently through these branches at 28.3 $l \cdot min^{-1}$ giving a combined air flow at the 'mouthpiece' of 56.6 $l \cdot min^{-1}$. This accomodated the operating flow rate of the impactor and simulates an inspiration rate typical of asthmatic patients. The model was completely enclosed in an environmental chamber in which temperature could be controlled at 37 °C. Air was drawn through the model from a second chamber, also heated to 37 °C, or taken from a humidifying chamber when the air was counterflowed over a heated water area of 1 m^2. The deposition of salbutamol as base or sulphate from a metered pressurised inhaler (MDI) and a dry powder inhaler (DPI), respectively, was examined at relative humidities of 30% and 97.5%. The percentage of the deposited dose from the MDI and DPI within the potential therapeutically active size range decreased significantly ($P < 0.02$) from 45.4 and 25.4 to 38.0 and 18.0, respectively as the humidity was increased from 30% to 97.5%.

Introduction

The direct determination of the mass distribution of an aerosol is routinely carried out using the principle of cascade impaction. This principle is employed in a number of commercially produced instruments designed to measure the size distribution and total concentration of liquid and solid particulate matter within the atmosphere. A number of such instruments have been used to determine the particle size distribution of pharmaceutical aerosols (Hallworth and Andrews, 1976; Davis and Bubb, 1978), In addition, more empirical devices have been developed to compare the likely respiratory penetrability of aerosols, without determining particle size (Kirk, 1972; Davies et al., 1976). Many of these in vitro deposition studies, however, have not controlled either temperature or relative humidity (RH). The importance of these two factors was well demonstrated by Byron et al. (1977) who placed a cascade impactor in a climatic cabinet at 37 °C and followed the changes in aerosol deposition as a function of RH. These workers showed that large and significant decreases occurred in the

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dose fraction of drug contained in droplets below 5 μm equivalent diameter when the RH was varied between 40% and 100% for propylene glycol/water and dry powder aerosols. The RH within the lungs is reported to be 99.5% (Porstendorfer, 1971) and under such conditions hygroscopic particles with an initial diameter of 1 μm would reach their equilibrium diameters within approximately 1 s which is within the effective residence time of an aerosol particle in the human lung (Ferron, 1977). The purpose of the present study was to construct an in vitro model lung, accomodating the operating requirements of a commercially available particle-fractionating sampler, which would also provide a flow rate representative of the inhalation rates found in bronchitic and asthmatic patients. Other design criteria to be included were that the model should have dimensions consistent with the anatomy of the upper airways and have the facility of controlling the temperature and relative humidity. It was intended to validate the model using well-characterised formulations of salbutamol.

Experimental

Materials

Pressurised metered dose inhalers (MDIs) of salbutamol (100 μg dose; Ventolin inhaler), capsules of salbutamol sulphate in lactose (equivalent to 400 μg salbutamol base/capsule; Rotacaps), dry powder inhalers (DPIs) (Rotahaler), salbutamol BP and salbutamol sulphate BP were provided by Glaxo, Ware, U.K. Hydrochloric acid (Analar grade) was supplied by BDH Chemicals, Poole, U.K.

Particle size analysis

An Andersen cascade impactor (Mark II, Andersen Samplers, Atlanta, GA, U.S.A.) was used. This is an 8-stage device where aerosol samples are collected on stainless steel plates. The apparatus was supplied calibrated by the manufacturer for an air flow through the apparatus of 28.3 $l \cdot min^{-1}$.

Model lung

The pressurised aerosol pack or dry powder inhaler was discharged into a glass "mouth" and "trachea", the latter being bifurcated at the lower end. These were designed to reproduce the anatomical dimensions of an average healthy human airway (Landahl, 1950) and are shown, drawn to scale in Fig. 1. The bifurcated "primary bronchi" were connected to the sampler and to a vacuum pump. Air was drawn independently through these branches at 28.3 $l \cdot min^{-1}$. The model was completely enclosed in a thermostatted Perspex chamber (22 × 34 × 61 cm). Air, heated to 37 °C, was either drawn through the model from a second Perspex chamber (22 × 34 × 36 cm) or taken from a humidifying chamber (Fig. 2). The latter, also manufactured from Perspex, was internally divided to provide a 5 m length of water (surface area 1 m^2) heated to 37 °C by a thermocirculator (Grant Instruments, Cambridge, U.K.). Air to the model lung was drawn counter to the flow of water. Temperature control within the lung chamber and dry air reservoir was achieved by circulating water, using a second thermocirculator (Grant Instruments, Cambridge, U.K.), through glass radiators positioned around the internal walls of the chambers. Air within both of these chambers was mixed using 10 cm axial

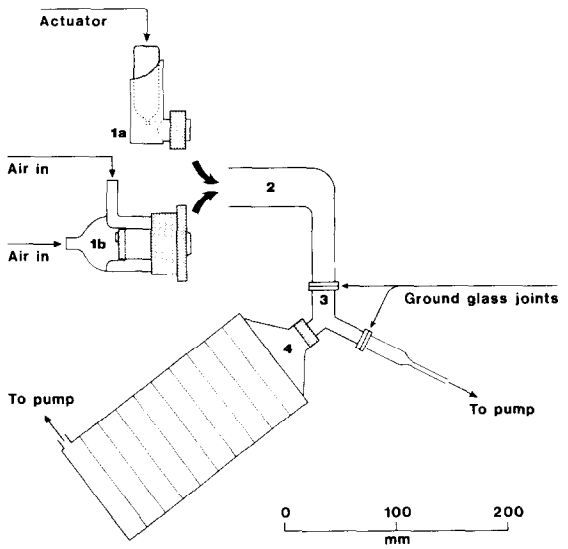

Fig. 1. Model lung. 1a = oral adaptor (MDI); Ib = oral adaptor (DPI); 2 = mouth/trachea; 3 = tracheobronchial junction; 4 = multi-stage cascade impactor.

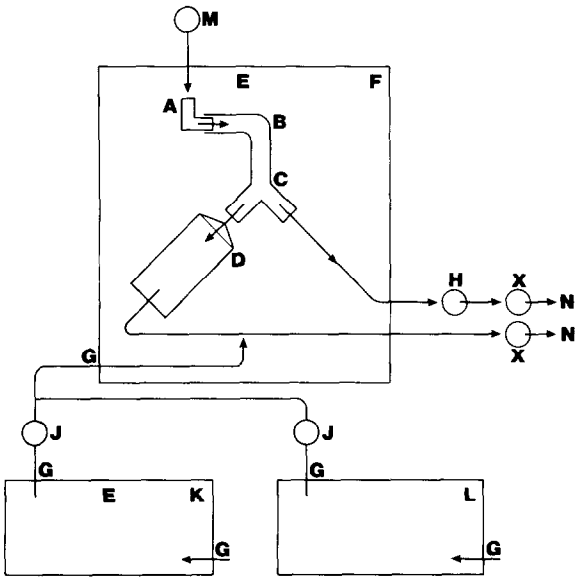

Fig. 2. Schematic layout of experimental chamber and air supply routes. A = device; B = mouth/trachea; C- = tracheo-bronchial junction; D = multi-stage cascade impactor; E = circulating fans; F = environmentally controlled chamber; G = inlet/outlet ports; H = hygrometer; J = mixing valves; K = dry, heated, air reservoir; L = humidified, heated, air reservoir; M = external actuation mechanism; N = vacuum pumps; X = flow meters.

fans installed internally. Flow rates through both branches could be adjusted independently and were monitored using flow meters (G.A. Platons, Basingstoke, U.K.). The humidity of the air drawn through the lung model was measured by an in-line hygrometer (Hygrotes Testo 6400, Testoterm, Emsworth, U.K.).

Activation of aerosol devices

The drug deposition of salbutamol base from a MDI and salbutamol sulphate from a DPI was examined. The MDI was discharged into the "upper airway" a total of 5 times, allowing a 20 s interval between each activation. The deposition from the MDI was determined at 37 °C and at relative humidities of 30% and 97.5% ($n = 6$ in each case). Alternatively 4 capsules of salbutamol sulphate in lactose base were emptied into the DPI and a fifth was broken within the DPI in the manner prescribed by the manufacturer, so that only one capsule shell remained in the device along with 5 doses of drug. The DPI was installed in a glass-holder (Fig. 3) which was then sealed in position at the "mouth" by means of a rubber gasket. The glass-holder was manufactured so that air could be drawn around the sides of the DPI, until conditions within the model had reached equilibrium at 37 °C and either 30% or 97.5% RH. The DPI was activated by re-routing instantly the airstream through the back of the device (Fig. 3).

Determination of drug deposition

Salbutamol deposition upon components of the model lung and the plates of the cascade impactor

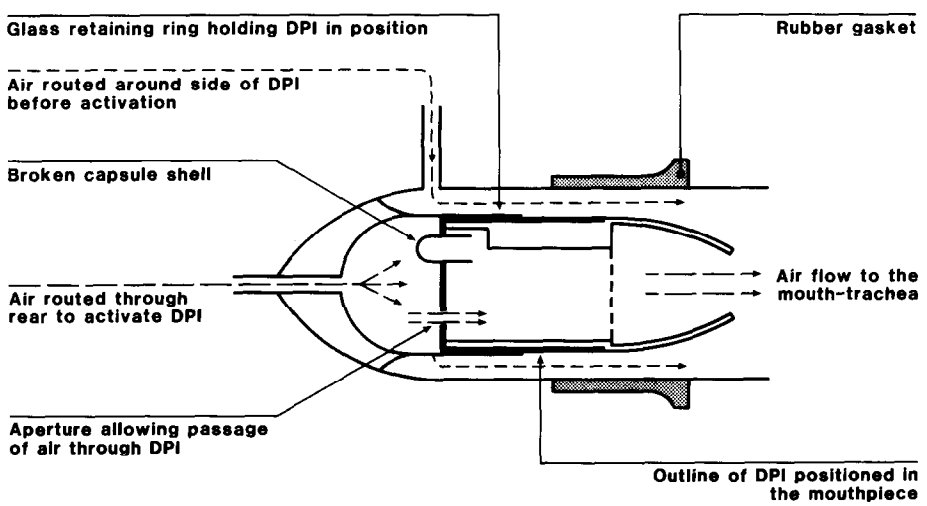

Fig. 3. Cross-section showing the air flow paths around and through a DPI in position at the mouth of the in vitro model lung.

was determined by washing with 0.1 M HCl, adjusting to a known volume, measuring the absorbance at 276 nm and the concentration obtained from a standard absorbance/concentration curve. In the case of the DPI, deposition of the salbutamol sulphate was calculated as the equivalent base.

Results were analysed for significant differences using a two-tailed Student's *t*-test.

Results

The dry powder aerosol, examined in this study, consisted of a nominal 400 μg of salbutamol (as the sulphate) mixed with 25 mg of lactose in each capsule; 10% overage of drug is included by the manufacturer and the mean content of salbutamol (as the sulphate) per capsule was determined to be 435.6 μg. From the lung model 66.6-77.8% of the delivered dose was recovered (Table 1). Since the model is designed so that only half the particles reaching the tracheo-bronchial junction are sampled by the cascade impactor (Fig. 1) it is a valid exercise to multiply the percentage of the drug recovered in the impactor by two. On that basis it was possible to account for 87–103% of the administered drug (Table 1). The results of the deposition studies were therefore expressed as a percentage of the total drug recovered, after doubling the amount deposited on each plate of the cascade impactor. Fig. 4 shows the effect of humidity upon deposition of salbutamol base from a MDI in the model lung. The drug was deposited more proximal to the "mouth" at the higher humidity and significant increases ($P < 0.02$) were

TABLE 1

Percentage of delivered dose (± S.D.) recovered from the lung model

		Relative humidity (% w/v)	
		30	97.5
MDI	(i)	70.1 ± 9.9	66.6 ± 4.0
	(ii)	93.6 ± 13.6	87.0 ± 3.8
DPI	(i)	77.8 ± 6.8	72.6 ± 3.9
	(ii)	103.3 ± 8.4	93.9 ± 4.5

(i) = indicates the percentage of the dose actually recovered.
(ii) = indicates the percentage of the dose, assuming 2 cascade impactors were incorporated into the model and deposition in the two is identical.

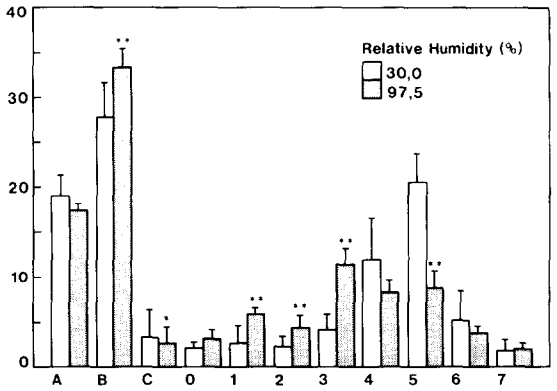

Fig. 4. The effect of humidity upon deposition of salbutamol within the model lung, administered by MDI. A = device; B = mouth/trachea; C = tracheobronchial junction; 0–7 = plates of the multi-stage cascade impactor: 0, 0–10 μm; 1, 5.8–9.0 μm; 2, 4.7–5.8 μm; 3, 3.3–4.7 μm; 4, 2.1–3.3 μm; 5, 1.1–2.1 μm; 6, 0.7–1.1 μm; 7, 0.4–0.7 μm (statistically significant differences between the amounts deposited at the two relative humidities are indicated ** $P < 0.02$, * $P < 0.05$).

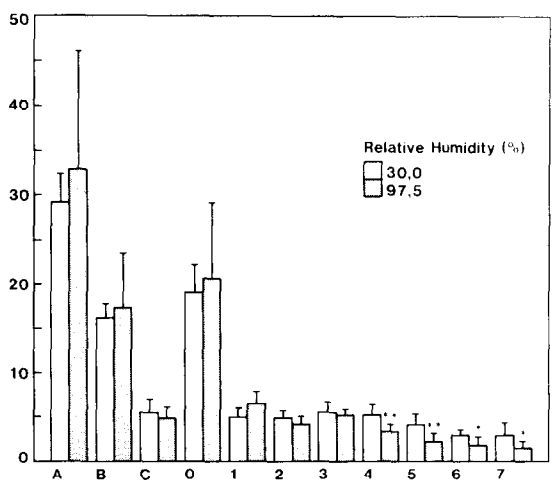

Fig. 5. The effect of humidity upon deposition of salbutamol sulphate within the model lung, administered by DPI. A = device; B = mouth/trachea; C = tracheobronchial junction; 0–7, plates of the multi-stage cascade impactor; 0, 9–10 μm; 1, 5.8–9.0 μm; 2, 4.7–5.8 μm; 3, 3.3–4.7 μm; 4, 2.1–3.3 μm; 5, 1.1–2.1 μm; 6, 0.7–1.1 μm; 7, 0.4–0.7 μm (statistically significant differences between the amounts deposited at the two relative humidities are indicated ** $P < 0.02$, * $P < 0.05$).

found in the amounts of drug recovered from the glass portion of the model lung and from those plates of the cascade impactor corresponding to particles in the size range 3.3–9.0 μm. There was a concomitant decrease from 20.4% to 8.7% ($P < 0.02$) in particles within the size range 1.1–2.1 μm. The percentage deposited on the lowest plates of the cascade impactor (0.43–2.1 μm) were unaffected by humidity. A similar qualitative change in the deposition profile upon increase in the relative humidity was found for salbutamol sulphate administered from a DPI, with more drug being recovered from the upper portions of the model at 97.5% RH than at 30% RH (Fig. 5). However, only the decrease at the higher humidity in percentage drug deposited in the size range 0.4–3.3 μm could be shown to be significant ($P < 0.05$).

Discussion

For normal healthy adult males the peak inhalation flow rate increases from 30 $l \cdot min^{-1}$ when sedentary to up to 300 $l \cdot min^{-1}$ when undergoing vigorous exercise (Hatch and Gross, 1964). In bronchitic or asthmatic patients the inhalation rate has been found to be within the range 50–400 $l \cdot min^{-1}$ (Coady et al., 1976). The air flow used in the lung model of 56.6 $l \cdot min^{-1}$ at the mouthpiece is thus comparable with that of a severe asthmatic. The use of such a flow rate enabled the incorporation of a commercially available cascade impactor which operated at 28.3 $l \cdot min^{-1}$, by utilising a vacuum pump which drew air at the same rate. A simple twin-stage liquid impinger system, proposed for use in routine control and comparative evaluation purposes, employs a similar airflow rate (60 $l \cdot min^{-1}$) at the mouthpiece (Meakin and Stroud, 1983). The model lung incorporates a mouth and tracheal portion of typical anatomical dimensions and these are usual components of both empirical lung models (Kirk, 1972; Davies et al., 1976) and liquid impinger systems (Hallworth and Andrews, 1976). Most of the throat deposition is as a result of impingement of the spray cone on the inner walls of the mouth and throat section. Some particle size fractionation occurs in this region with the larger particles being more efficiently removed than the finer (Hallworth and Andrews, 1976).

One of the advantages of this lung model compared with many others is that the temperature and RH can be adequately controlled. The latter, through suitable adjustment of the mixing valves, could be varied between 25 and 98%. The maximum time interval between activation of a device at the mouthpiece to deposition of any drug on the lower plates of the cascade impactor is about 1 s. Any effects of particle growth or shrinkage under prevalent humidity conditions upon drug deposition within the model is therefore being examined under realistic time intervals. Table 2 shows that the percentage salbutamol of size 5.8 μm and below which was deposited from an MDI at 30% RH was 45.4 ± 4.0%. It is particles of this

TABLE 2

Percentage of deposited dose (± S.D.) of size 5.8 μm and below

	Relative humidity	
	30	97.5
MDI	45.4 ± 4.0	38.0 ± 3.9 *
DPI	25.4 ± 3.0	18.0 ± 3.1 *

* Indicates a significant difference ($p < 0.02$) between the percentage of dose deposited at the two humidities.

TABLE 3

The potentially therapeutic dose of salbutamol determined using other lung models

Device	Percentage	Sizing device	Air flow ($l \cdot min^{-1}$)	Reference
MDI	46.2	Model lung	58	Davies et al., (1976)
MDI	49.7	Multistage liquid impinger	60	Hallworth (1977)
MDI	53.0	Twin stage liquid impinger	60	Meakin and Stroud (1980)
DPI (400 μg capsule)	24.1	Multistage liquid impinger	60	Hallworth (1977)

size that may be considered to form the therapeutic component of the administered dose. This value compares favourably with other values found in the literature for the same dosage form employed in different lung models (Table 3). Some caution must always be used in making such comparisons, since the deposition models vary in construction and operating conditions. In addition the size range of the particles suggested to be of therapeutic benefit tend to differ slightly between studies depending upon the fractionating capabilities of the particle sizing apparatus employed. When the RH was increased to 97.5% then the proportion of the dose depositing with a particle size of 5.8 μm or less decreased significantly ($P < 0.02$) to 38% (Table 2). The MDI used in this study contains salbutamol base with oleic acid (10% w/w of drug) as surfactant (Malton et al., 1982). Both of these materials are relatively hydrophobic and therefore particle growth under conditions of high RH may be expected to be slight. Yoshida et al. (1976), however, showed that small non-hygroscopic particles of carbon black and particles of stearic acid grew within milliseconds of being transferred to a supersaturated water atmosphere. In the present study it would appear that condensation of water at the surface of the particles of salbutamol base occurs within the model lung at the higher RH, causing the found change in particle size distribution. Similar results have recently been reported for micronised salbutamol formulated with Span 85 (0.5% w/v) as an MDI (Kearney and Kinrade, 1987), where the fraction of the dose below 4.7 μm was found to decrease significantly ($P < 0.05$) from 38.9% at 35% RH to 34.4% at 95% RH.

The lactose contained in the capsules of salbutamol sulphate has a mean diameter of about 80 μm, with most particles being greater than 10 μm whereas the drug particles are mostly less than 2 μm (Hallworth, 1977). Examination by electron microscopy revealed that some lactose particles had one dimension of over 220 μm. Much of this larger-sized lactose was deposited in the mouth/trachea and first plate (Stage 0) of the cascade impactor. Much smaller-sized drug particles were observed, adhering to the lactose and this accounts for the large amount of drug deposited within the upper regions of the model. The proportion of the potentially 'therapeutic dose' fell significantly ($P < 0.02$) from 25.4 to 18% when the RH was increased from 30 to 97.5% (Table 2). Gonda et al. (1982) have calculated from vapour pressure osmometry measurements a growth ratio of aerodynamic diameter at 99.5% RH of 2.4 for salbutamol sulphate. The influence of growth of the lactose particles upon the resultant drug deposition is likely to be minor because of the relatively large initial particle size of the diluent. The percentage of the dose deposited with a particle size of 5.8 μm and below at 30% RH (Table 2) agreed well with that proportion reported to be therapeutically effective using a different particle sizing technique (Table 3) and an earlier design of the DPI.

This study shows that deposition from the DPI can be considered inefficient when compared to the MDI in terms of the percentage dose deposited within the potential therapeutic size range (Table 2). However, one 400 μg capsule provides a similar quantity of drug (equivalent to 101 μg salbutamol at 30% RH; 72 μg at 97.5%) as a 200 μg dose administered form an MDI (91 μg at 30% RH: 76 μg at 97.5%). These results support clinical studies which show no significant differences in bronchodilation responses induced by salbutamol when administered from the two dosage forms at these concentrations (Duncan et al., 1977; Hartley et al., 1977).

Conclusions

The inclusion of precision particle sizing equipment within a model lung which combines upper airways of anatomical dimensions with a realistic inspiration rate would appear to provide a suitable means by which the effect of formulation variables of an aerosolised product on the deposition profile can be assessed in vitro. Since the size of aerosolised products can change considerably between delivery from an appropriate dosage device to arrival at the site of deposition, some attention should be given to the adequate control of humidity and temperature. The model lung developed here appears to fulfil these require-

ments and enables a commercially available cascade impactor to be employed to assess any humidity-related change in particle distribution, which may occur over appropriate deposition time intervals.

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