Eur Respir J
1991, 4, 890–898

TECHNICAL NOTE

Increased lung deposition and biological effect of methacholine by use of a drying device for bronchial provocation tests

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ABSTRACT: A simple device aiming to increase deposition of nebulized methacholine in the lower airway was studied. The device controlled inspiratory flow and volume and dried the aerosol.

The effect of drying on deposition in the throat and lower airways was studied in six subjects given an aerosol of ^{99m}Technetium-diethylenetriamine pentaacetate (DTPA) in saline with the device and with a reference device giving the same inspiratory flow and volume but no drying. Drying reduced throat deposition from 46 (range 26–61) to 13 (range 6–22)% ($p<0.05$) of the inhaled and retained dose.

The effect of drying on the biological effect of nebulized methacholine was studied in 21 subjects who underwent three provocation tests on different days with doubling concentrations of methacholine from 0.5 to a maximum of 64 mg·ml^{-1}, one with the reference device and two with the drying device. The percentage change in forced expiratory volume in one second (%FEV_1) per cumulative dose of methacholine changed from -1.0 (2.6) %FEV_1·μmol^{-1} (geom. mean and SD) with the reference device to -1.6 (3.6) with the drying device $p<0.02$. In an additional study 20 subjects underwent four provocation tests on different days, two with a different version of the drying device and two with the reference device. The slope changed from -1.1 (3.1) to -1.6 (3.3), $p<0.02$. The reproducibility of duplicate measurements did not improve with the drying device. Thus, the drying device decreased throat deposition and increased the biological effect of nebulized methacholine.

Eur Respir J., 1991, 4, 890–898.

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Keywords: Bronchial responsiveness; methods; pharynx; radionuclide imaging.

Received: November 12, 1990; accepted after revision April 3, 1991.

Supported by grants 84-1272 and 89-0098 from the Swedish Work Environment Fund.

Two types of protocol are commonly used to assess bronchial responsiveness to methacholine or histamine, namely the continuous, tidal breathing method described by COCKCROFT *et al.* [1] and the metered dose method recommended by CHAI *et al.* [2]. Both protocols are well-documented and through long experience have become standard methods in research and in the clinical evaluation of asthma. Several other methods have been published, many for use in specific research applications, with better control of dose delivery and of volume history compared to the standard methods [3–9]. Thus, with the tidal breathing method the dose delivered to the airways is proportional to the tidal volume, which may vary considerably within and between subjects [10]. The original metered dose method uses deep inspirations, which may affect the bronchial response to the agent [11, 12]. With both methods inspiratory flow is poorly controlled, which may cause variable deposition of agents within the airways [13]. Pharyngeal deposition may account for a significant and variable fraction of inhaled mass [14], which does not contribute to the intended biological effect [15]. The purpose of the present study was to evaluate a simple method to control dose, volume history of the lungs and in addition inspiratory flow rate. The method was primarily designed for the study of normal populations exposed to environmental influences which may cause changes in airway responsiveness.

The method used in the present study was influenced by the method described by OREHEK and EISER [6], which includes drying of the nebulisate and, thus, reduced pharyngeal deposition and control of the inspired volume of nebulisate. The device used in the present study is simple in construction and use and in addition to drying the nebulisate and controlling the inspired volume of nebulisate it offers control of inspiratory flow. Thus, the dose of the agent delivered to the lower airways (below the larynx) is controlled with acceptable accuracy. Aerosol and supplementary air are delivered to a vertical column. The size of the column allows the

Table 1. – Subject's age, smoking habits, sex and spirometric values in percentages of predicted values

	Study A Isotope	Study B Column	Study C Cone
Number of participants	6	21	20
Males/females	2/4	6/15	11/9
Current smokers	0	6	0
FEV_1 % pred		98 (11) 78–116*	99 (13) 71–120*
VC % pred		95 (13) 72–118*	102 (12) 73–121*
Age yrs	37.2 24–46	37.5 25–50	36.6 23–51

*: mean (standard deviation) range; FEV_1: forced expiratory volume in one second; VC: vital capacity.

nebulisate to dry for several seconds before inhalation. Inspiratory flow is held constant at 0.4 $l \cdot s^{-1}$ and the inhalation time and number of breaths are controlled.

In order to test the assumption that throat deposition is decreased using the drying device the deposition of nebulized saline, labelled with ^{99m}Technetium-diethylenetriamine pentaacetate (DTPA), was studied using a gamma camera. The assumption that drying of the nebulisate increases the dose delivered to the lower airways was tested by comparing the biological response with and without the drying device. Finally, the absence of significant effects of orally deposited methacholine on bronchial tone was reconfirmed.

Material and methods

Subjects

The subjects were healthy and had no present or previous asthma, or other pulmonary or allergic disease. They reported no colds during the six weeks prior to the investigation. Smoking was not allowed on the day of the investigation, and coffee and tea were not allowed for at least 4 h before the investigation. The subjects gave informed consent and the experimental protocols had been approved by the Ethics Committee of the Karolinska Institute. Age, smoking habits, sex and spirometric values in percentage of predicted values [16, 17] are given in table 1.

Drying devices

In studies A and B, a column assembled from standard copper tubing was used (fig. 1). In study C, the dryer was manufactured from a stainless steel plate and shaped like a double cone (fig. 1). The angle of the upper half of the cone (30°) was chosen in order to allow a large volume within a relatively low height and yet avoid turbulence along the sides of the cone. Both devices had five openings. The aerosol was added at the top of the cone and at the side of the top portion of the column using a short, angular tube. Dry supplementary air was added through a filter above and surrounding

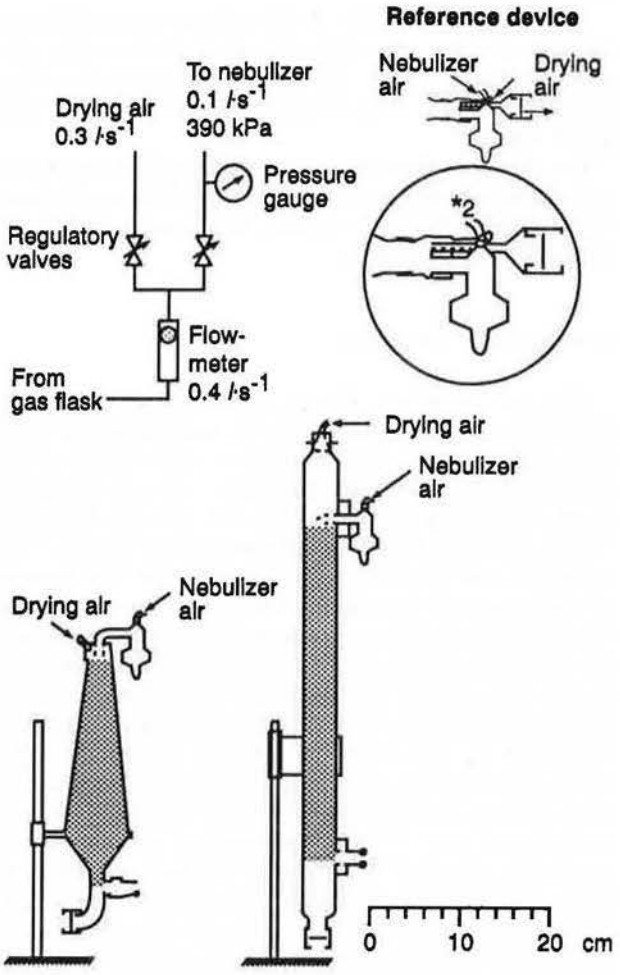

Fig. 1. – Air supply, reference device used in studies A, B and C, column used in studies A and B and cone used in study C.

the aerosol inlet. An opening with a standard spirometric mouthpiece was placed at the side of the lower part of the drying devices. At the bottom outlet a standard one-way valve (model 72064, Erich Jaeger Gmbh, Würzburg, BRD) was fitted, allowing air to escape from, but not to enter, the device. The pressure swings caused by breathing from the drying devices were monitored with an ink filled transparent U-tube. The subjects were

instructed to inhale during 2 s and exhale during 2 s for one minute, guided by the pace of the metronome. Any deviation from this instruction was easily detected by watching the pressure inside column during breathing. A negative pressure indicated that the subject inhaled all of the nebulizer plus supply air. This flow was only minimally influenced by the pressure swings, as shown by the flow monitor.

One source of dry compressed air was used for both the nebulizer and the supplementary air. Two regulatory valves (Model 11-818, IMI Norgren Enots Ltd, Warwickshire, UK) controlled the distribution of supplementary air and air to the nebulizer and the total airflow was monitored with a flow meter (Fischer and Porter, model 10A3200, Horsham, PA, USA) (fig. 1).

The effective volume of the column was arbitrarily chosen to be 3.3 *l* and the cone 3.4 *l*. Aerosol (0.1 $l \cdot s^{-1}$) plus drying air was administered at a total flow of 0.4 $l \cdot s^{-1}$. The effective drying time was thus 8.3–8.5 s.

For reference measurements, the nebulizer was fitted with an adaptor shown in figure 1, which allowed control of inspiratory flow and volume in the same manner as with the drying devices.

Methacholine inhalation

The jet nebulizer (Astra Meditec, model MA2, Göteborg, Sweden) was driven by compressed air (390 kPa) and produced 0.1 *l* aerosol$\cdot s^{-1}$. The output of the nebulizer was 0.38 (0.013) ml$\cdot min^{-1}$. It was measured at the beginning and end of all test days by weighing before and after a one minute nebulization period. The nebulizer was filled with 3 ml of solution. The amount of water required to humidify the dry air flowing through the nebulizer to full saturation at the temperature inside the nebulizer during the first minute was less than 20% of the total output, and decreased subsequently due to falling temperature. The concentration of the solution increased by less than 3% change during the 75 s nebulization period according to these calculations. The mass median aerodynamic diameter (MMAD) of dried nebulisate was 1.7 μm (geometric mean) with a geometric standard deviation of 1.3 μm, according to measurements with an aerodynamic particle sizer (APS-3300, TSI inc. Saint Paul, Minnesota). The distribution deviated from log normal at higher diameters, since a tail with about 20% of the mass were found in fractions with a diameter of 7 μm or larger. This is partly an artefact caused by coincidence (two or more particles may be counted as one with a combined mass). The MMAD of primary (wet) droplets from a similar MA2 jet nebulizer was 5.2 μm when measured with a Malvern Mastersizer (Malvern Instr. Ltd, Malvern, UK) [18].

Spirometry was performed with a wedge spirometer (Vitalograph®, Buckingham, UK). The highest of three reproducible measurements of forced expiratory volume in one second (FEV_1) and the highest of three slow vital capacity (VC) manoeuvres were chosen as basal values. Ten minutes later the subjects inhaled diluent (isotonic saline), followed by doubling concentrations of methacholine chloride beginning with 0.5 mg$\cdot ml^{-1}$ in isotonic saline. The test was continued until FEV_1 decreased by 20% or more compared to the value recorded after the saline provocation or after inhalation of 64 mg$\cdot ml^{-1}$. Fifteen 0.8 *l* inhalations at 0.4 $l \cdot s^{-1}$ were made, corresponding to an inhaled dose of about 0.19 ml of nebulized solution. Four minutes after the start of the provocation FEV_1 was measured. Only one forced expiration was attempted. After the end of the test the subjects inhaled 4–6 puffs of terbutaline, in order to reverse the bronchial constriction caused by the methacholine inhalations.

The cumulated inhaled methacholine dose was calculated from nebulizer output and inhalation time. The slope of the relationship between FEV_1 (expressed in percentage of the average of the pre- and post-diluent values) and the cumulative methacholine dose (linear scale) was calculated by linear regression [19, 20]. In addition the cumulated provocative dose causing a 20% decrease in FEV_1 compared to post-saline values (PD_{20}) was calculated by intrapolation on a log cumulative dose scale. In four subjects FEV_1 decreased by more than 15% but less than 20%. In these cases the PD_{20} value was extrapolated from the two last measurements. The calculations were made by use of a computer (Macintosh, EXCEL).

Scintigraphy

The subjects inhaled a nebulized solution of 500 MBq$\cdot ml^{-1}$ ^{99m}Technetium-DTPA in physiological saline. Measurements were started within 1 min of the administration of the isotope and the dose was selected such that sufficient counting precision could be obtained within 6 min after the start of the inhalation. The total inhaled dose was less than 20 MBq as indicated by the count rate of the gamma camera.

Data were collected by a dual head scintillation camera (Dual Digital Dyna Camera, Picker Inc., USA), standard fields of view (38 cm) and parallel general purpose collimators. The data were stored by a Gamma-11 system, PDP 11/34 computer. Data from the anterior view was analysed. Three regions of interest were electronically selected with a joy-stick. These were the upper airways, including nose and throat, the gastric region and the lung region. A lower level of 20% of the maximum lung region pixel counts was used to define the outer border of the lung. The activity over the lungs was expressed as a percentage of the total activity recorded in the three regions.

Experimental protocols

All repeated measurements were performed at the same time of day on different days within two weeks. Studies A, B and C were randomized with regard to the order of exposures to drying devices or reference device.

Study A. Six subjects were examined on two occasions separated by at least one week. On the first occasion three subjects were randomly assigned to inhale nebulized ^{99m}Technetium-DTPA solution using the drying column, and three inhaled the aerosol using the reference device. On the second occasion the protocol was reversed.

Study B. Twenty one subjects performed three methacholine provocation tests. They were randomly assigned to three groups, seven in each. One group commenced with the reference device, followed by two trials using the column-shaped drying device. Another group commenced with the column, followed by the reference device and finally a second column trial. The third group began with two trials using the drying device and performed the last trial with the reference device.

Study C. Twenty subjects performed four methacholine provocations. They were randomly assigned to two groups of ten each. The first group began with the cone-shaped drying device, the second group with the reference device. On subsequent trials the procedures were reversed, so that each subject performed each two trials with and two without the drying device.

Study D. In order to test the hypothesis that orally deposited methacholine did not significantly influence bronchial tone, six subjects were given a dose corresponding to the earlier measured PD_{20} dose of methacholine as a solution which was kept in the mouth for at least 30 s, gargled and subsequently swallowed. FEV_1 was measured before and at 5, 10, 20, 40 and 60 min after administration of the dose. Thereafter, a fourfold dose was given in the same manner using the same protocol to record the effects. The subjects were chosen among those who had displayed relatively high sensitivity to methacholine.

Calculations

Statistical analysis was performed using Wilcoxon's signed rank test for paired samples (Statview® Macintosh). Results are expressed as means (standard deviation) unless stated otherwise. A p-value <0.05 was considered statistically significant.

Results

Study A. Without the drying column 54 (10.7)% of inhaled (and retained) nebulisate reached the lower airways. Using the column 87 (6.7)% reached the lower airways (fig. 2) ($p<0.05$).

Studies B and C. Individual values are given in tables 2 and 3. All subjects had a negative slope (FEV_1 decreased with increasing doses of methacholine). With

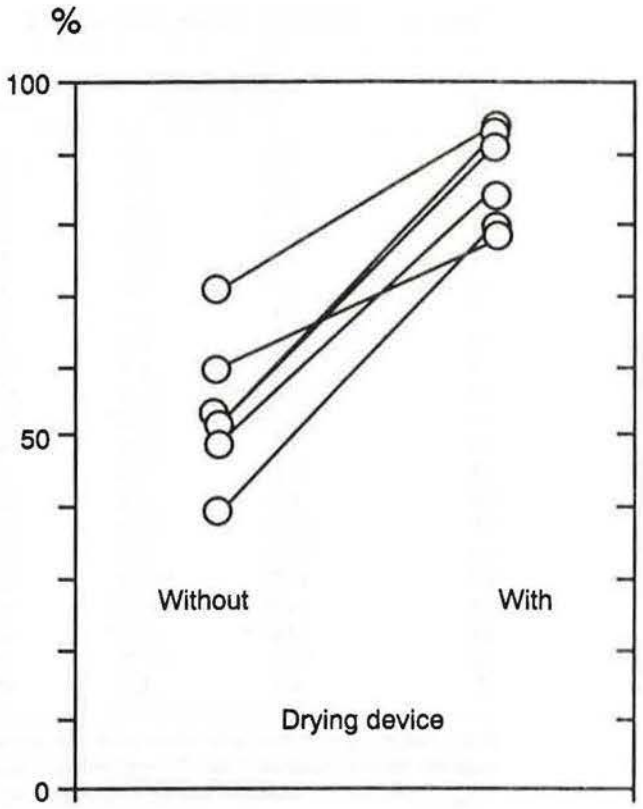

Fig. 2. – ^{99m}Technetium activity recorded over the lung in percentage of total counts (lungs, upper airways and gastric area).

the drying device the slope became more negative in both studies, indicating increased "apparent" sensitivity to methacholine (table 4). Five subjects in study B and two in study C had a FEV_1 decrease less than 20% of post-saline values following challenge with 64 $mg \cdot ml^{-1}$ in one or more of the trials. Since PD_{20} could not be determined in all subjects average values of PD_{20} are not available. As an approximation PD_{20} values were extrapolated in four subjects with a greater than 15% but less than 20% decreased in FEV_1 at the highest dose. In two of these FEV_1 at the two highest doses was unchanged. Five subjects with less than 15% decrease at the highest dose were assigned a PD_{20} value of twice the highest cumulative dose. These values were used to calculate the geometric mean values shown in table 4.

The cumulative distributions of the PD_{20} values in studies B and C are shown in figure 3. Duplicate determinations were averaged. In study B median values were reversed compared to expected, probably due to random variation, as illustrated in figure 3. The middle portion of the curve can be approximated with a straight line, which was estimated by linear regression. From this regression an "intrapolated value" of the median was calculated. In study B the intrapolated median was 2.0 with the drying device and 2.6 without. In study C corresponding values were 2.1 and 3.1

Table 2. – Individual values from study B

Subject no.	PD_{20} µmol cumulated dose			Slope $\%FEV_1 \cdot \mu mol^{-1}$		
	With column		Without	With column		Without
1	3.5	4.5	9.8	-5.1	-3.9	-2.0
2	1.5	1.1	2.0	-15.6	-4.0	-11.2
3	1.4	2.1	5.0	-11.5	-8.5	-4.2
4	9.1	23.3	6.7	-2.0	-0.8	-2.8
5	3.6	10.1	5.1	-6.5	-1.9	-3.9
6	1.7	3.5	4.0	-13.0	-4.3	-4.9
7	3.4	1.2	4.9	-6.5	-16.4	-4.3
8	6.2	6.4	6.1	-3.1	-2.9	-3.1
9	1.2	4.3	10.4	-8.2	-3.9	-2.0
10	51.2	21.0	31.5	-0.4	-0.8	-0.4
11	41.4	69.0*	20.4	-0.3	-0.3	-0.9
12	7.2	10.8	14.7	-2.4	-1.7	-1.4
13	8.8	7.3	9.7	-1.8	-2.4	-2.1
14	8.9	7.4	12.1	-2.3	-2.5	-1.7
15	24.2	33.4	67.9	-0.4	-0.5	-0.2
16	17.1	3.2	11.8	-1.1	-6.7	-0.8
17	>127	107.8	>127	-0.11	-0.12	-0.11
18	>127	151.6*	>127	-0.11	-0.16	-0.07
19	>127	>127	>127	-0.08	-0.04	-0.08
20	63.1	147.7*	>127	-0.19	-0.16	-0.13
21	4.0	3.9	32.0	-4.3	-4.6	-0.5

Each subject performed two trials with the drying column and one with the reference device on separate days in random order. *: extrapolated value; FEV_1: forced expiratory volume in one second; PD_{20}: provocative dose producing a 20% fall in FEV_1.

Table 3. – Individual values from study C

Subject no.	PD_{20} µmol cumulated dose				Slope $\%FEV_1 \cdot \mu mol^{-1}$			
	With cone		Without cone		With cone		Without cone	
1	11.1	6.2	7.9	8.9	-1.8	-3.3	-2.9	-2.3
2	4.4	5.9	12.3	23.6	-4.5	-3.7	-1.4	-0.9
3	3.3	6.6	8.0	10.1	-3.4	-2.7	-1.7	-1.8
4	27.5	13.1	127.0	82.7	-0.6	-1.3	-0.2	-0.2
5	8.8	3.3	6.2	7.2	-2.1	-5.6	-2.7	-2.2
6	12.5	22.3	7.8	14.0	-1.7	-0.7	-2.5	-0.3
7	6.8	13.9	15.3	34.6	-3.1	-1.3	-0.9	-0.4
8	1.5	1.3	1.3	3.0	-9.3	-13.0	-16.1	-6.7
9	47.5	21.7	57.1	41.1	-0.4	-0.7	-0.3	-0.5
10	19.0	12.8	30.5	18.8	-0.9	-1.6	-0.5	-1.0
11	17.6	23.0	10.7	17.4	-1.1	-0.8	-1.7	-1.0
12	13.9	22.1	75.3	15.9	-1.8	-1.1	-0.2	-1.1
13	2.6	5.4	3.9	3.7	-6.3	-3.5	-5.8	-4.6
14	5.9	29.1	12.7	14.9	-3.2	-0.6	-1.4	-1.0
15	26.6	13.7	49.4	23.2	-0.8	-0.7	-0.4	-0.9
16	>127	>127	>127	>127	-0.02	-0.07	-0.03	-0.08
17	65.4*	16.0	120.4	59.5	-0.1	-0.9	-0.1	-0.3
18	3.6	1.4	3.6	1.3	-5.4	-14.8	-5.2	-16.4
19	37.7	84.1	52.0	75.7	-0.4	-0.2	-0.3	-0.3
20	2.9	3.7	4.2	3.8	-6.7	-4.2	-3.1	-4.9

Each subject performed two trials with the drying cone and two with the reference device on separate days in random order. *: extrapolated value. For abbreviations see legend to table 2.

The "reproducibility" of the method was calculated from the standard deviation of the differences in log PD_{20} on those subjects who had a measurable PD_{20} at all trials. With the drying device the 95% confidence interval of the difference between duplicate measurements was 2.4 and 2.3 doubling concentrations of methacholine (study B and C, respectively) and without the drying device (study C) 2.0 doubling.

Study D. Five minutes after oral administration of a dose corresponding to the previously measured PD_{20} dose, FEV_1 decreased by less than 2%. Five minutes

Table 4. – Bronchial reactivity with and without drying device

	Use of drying device	Study B (Column) n=21 geometric mean (SD)	Median		Study C (Cone) n=20 geometric mean (SD)	Median	
Slope	Yes	-1.6 (3.6)	-2.4		-1.6 (3.1)	-1.7	
$\%FEV_1 \cdot \mu mol^{-1}$	No	-1.0 (2.6)	-1.7	p<0.02	-1.1 (3.3)	-1.2	p<0.05
PD_{20}	Yes	12.9 (67.2)	8.2		12.5 (45.6)	16.7	
µmol	No	18.0 (79.6)	11.8	p<0.05	17.8 (50.1)	16.0	p<0.02

For abbreviations see legend to table 2.

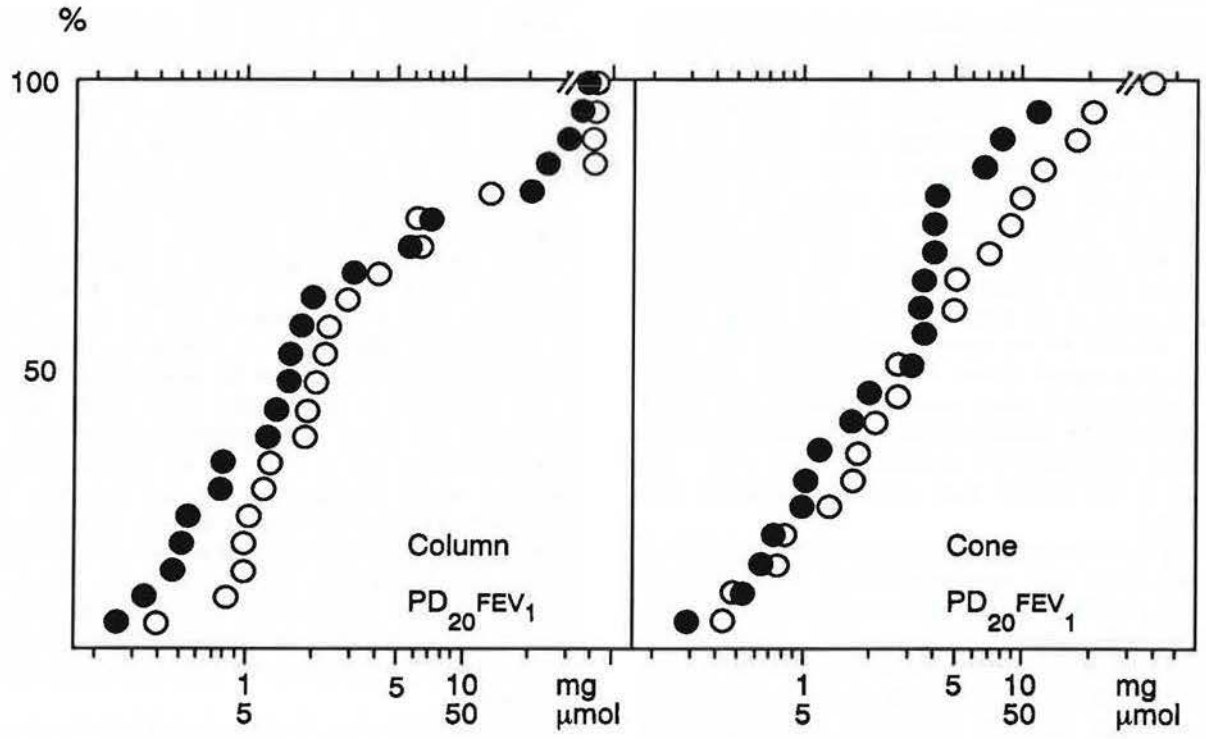

Fig. 3. – Cumulative distribution of PD_{20}. The points show average values of duplicate measurements of PD_{20}, obtained with the drying device (●) or with the reference device (O). $PD_{20}FEV_1$: provocative dose producing a 20% fall in forced expiratory volume in one second.

after oral administration of four times this dose, FEV_1 decreased by less than 2.5%. At other times responses were even smaller.

Discussion

The device was designed to ensure delivery of a known amount of agent to the lower airways. For this purpose inspiratory flow was held low and constant and the nebulisate was dried. Constant inspiratory flow was achieved by use of valves and was monitored by a flowmeter. The air supply to the nebulizers was 0.1 $l \cdot s^{-1}$. The supplementary air flow (0.3 $l \cdot s^{-1}$) was not let through the nebulizer. Thus, the supply air did not dehydrate the solution in the nebulizer, but only the nebulisate. Hydration of air passing through the nebulizer can cause considerable water loss from the solution [21]. Since the "output" of the nebulizer is estimated by weighing after a period of nebulization, the solute output from the nebulizer may be seriously overestimated. In the present study this error was less than 20% and constant.

By controlling inspiratory flow, one source of variation was reduced. Inspiratory time was kept constant and, thus, also inspired volume at each breath (0.8 *l*), which therefore reduced variability of the amount of inhaled methacholine and the volume history of the lungs. The subjects experienced no difficulties in inhaling at the controlled air flow and the pressure monitor (a U-shaped tube with ink) indicated that the subjects inhaled all the prescribed volume.

Inspiratory flow and volume (0.4 $l \cdot s^{-1}$, and 0.8 *l* respectively) were the same as those used by OREHEK and EISER [6]. Control of the inspired amount of

nebulisate is also possible with the dosimeter method [2], but it was considered as a disadvantage that the original method prescribes deep inhalations, which may influence the biological response [11]. Recently, however, dosimeters have been described which allow tidal breathing [8].

The two most commonly used nebulizers produce aerosols which differ considerably with regards to particle size (median MMAD or original droplets of Wright nebulizers 1.4±2.0 μm and deVilbiss 646 4.4±2.2 μm [22]). The nebulizer used in the present study produces slightly larger primary droplets than the deVilbiss nebulizer.

A large fraction of the inhaled nebulisate was impacted in the throat when challenge was made without drying of the nebulisate, in spite of the controlled, low inspiratory flow rate. This proportion varied between 29–61% in six subjects (average 46%). These data are compatible with the findings of RYAN and co-workers [13] who found that 36% of the inhaled and retained dose (range 16–60%, average of duplicate measurements) was deposited in the throat with tidal breathing inhalations from a Wright nebulizer. The slightly larger average value in the present study could be due to the higher MMAD of the aerosol produced by the MA2 nebulizer compared to the Wright nebulizer. Findings with radiolabelled teflon particles indicate that throat deposition can be predicted from aerodynamic diameters (squared) multiplied by inspiratory flow [23]. The findings in the present study are also compatible with these data.

Using the column, a significantly smaller fraction of the nebulisate was impacted in the throat and the variability of this fraction was smaller (average 13%, range 6–22%). Theoretically, particles which would otherwise have been deposited in the throat may have been deposited on the walls of the drying device instead. However, the apparent sensitivity to methacholine increased by an amount corresponding to the improved deposition in the lungs by use of the dryer. Thus, the proportion of inhaled aerosol which reached the lower airways (below the vocal cords) increased from 53 to 87% with the drying device. If the inhaled amount of methacholine is unchanged when using the dryer, and given the change in throat deposition, a theoretical value of 61% reduction in median PD_{20} would be expected. This value is similar to the observed 68% reduction of intrapolated median value in study C. The most probable explanation is, therefore, that the drying column has permitted particles which would otherwise end up in the throat to reach below the vocal cords and the sites of measured biological activity.

There are also theoretical reasons why deposition in the drying device should be low compared to deposition in the throat. Thus, the main factors influencing impaction on a surface are velocity and mass of a particle. In the upper bend connecting the nebulizer with the drying device airflow is low (0.1 $l \cdot s^{-1}$). The drying devices were designed to promote laminar flow. The lower bend where the patient inhaled dry nebulisate has a large diameter compared to the throat and, thus, the velocity of the particles is low compared to velocities in the throat. In addition, rehydration of the particles in the mouth may contribute to increase deposition in the throat [24]. The throat deposition of dried aerosol is compatible with data from other studies using hygroscopic [25] or nonhygroscopic particles [26] and with theoretical calculations [27]. With increased methacholine concentration the size of the dried particles increases, which could affect deposition patterns. This change is, however, probably limited. Thus, the dry MMAD of an aerosol of physiological saline was 1.5 μm and increased to 1.8 μm with an 80 $mg \cdot ml^{-1}$ methacholine solution [5].

Small droplets (1–2 μm) dry quickly, within a fraction of a second. However, in a humid atmosphere larger droplets require a much longer time to dry [25, 28]. The water content in the saturated air from the nebulizer and in the primary droplets is sufficient to fully saturate air at a temperature of 18°C and will give 79% relative humidity at a temperature of 22°C. During evaporation the temperature decreases. Thus, the primary droplets are exposed to gradients of temperature and humidity which are difficult to estimate. FERRON and SODERHOLM [28] have presented data on drying times for particles of different sizes at different humidities. Their calculations indicate that the larger droplets present in the nebulisate in the present study require several seconds to dry at the high humidity prevailing in the drying devices, but in view of the many unknown variables exact calculations are not possible. The drying time (8 s) was arbitrarily chosen, but appears to have been sufficient for most of the desired biological effects. The drying times could be in excess of what is required. Shorter drying times, and thus small drying devices, would probably be sufficient if a lower water output relative to total air flow is used or if the nebulisate is warmed.

Several other studies have employed dried nebulisate [6, 15]. Thus, for example "settling bags" or spirometers have been used [4–6] and warming of aerosol has been advocated for special purposes [3]. In a study with a similar design to the present study apparent PD_{20} was increased (rather than decreased) following exposure to aerosol which had been dried with an diffusion dryer [15]. Loss of nebulisate in the drying device could, however, have influenced these results. When challenged with a small monodisperse dried aerosol, throat deposition can be essentially avoided [29]. On the other hand the biological effect of such particles may be limited and a significant fraction may be exhaled [5]. Variability in the fraction of exhaled particles explains some of the variability in the biological response in normal subjects [30]. By increasing the breath-holding time and by use of forced expiration [31, 32], the exhaled fraction may be decreased, but this was outside the scope of the present study.

The reproducibility of the present study was not improved by use of the drying device and was in the upper range of values published from other studies [13, 33–38]. It should, however, be pointed out that the reproducibility in population based samples with

mostly healthy subjects appears to be larger [38] than in patients with hyperresponsive airways. This may reflect a greater variability in biological response, measured as FEV_1 decrease, of normal subjects compared to patients with hyperresponsive airways. The variability in throat deposition differs considerably between subjects, but appears to be rather constant in each person (M. Svartengren, personal communication). Thus, reducing the throat deposition does not necessarily increase the reproducibility, but should reduce the variability between individuals.

Acknowledgements: The authors gratefully acknowledge the generous help of Astra Meditec for supplying the nebulizers and M. Dahlbäck, Draco AB (a subsidiary of Astra AB) for determinations of the MMAD of the aerosol. They also thank B-M. Sundblad for expert technical assistance.

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Accroissement des dépôts pulmonaires et effets biologiques de la methacholine par l'emploi d'un appareil dessiccatoire pour les tests de provocation bronchique. P. Malmberg, K. Larsson, S. Thunberg.

RÉSUMÉ: Un appareil simple, visant à diminuer les dépôts de methacholine nébulisée a été étudié. L'appareil contrôle le débit inspiratoire et le volume, et assèche l'aérosol.

L'effet de la dessiccation sur les dépôts dans la gorge et les voies aériennes inférieures a été étudié chez six sujets, au moyen d'une aérosol de ^{99m}Technetium-diéthylenetriamine penaccétate (DTPA) dans la solution saline isotonique, avec l'appareil et un appareil de référence donnant les mêmes débit inspiratoire et volume, mais sans dessiccation. La dessiccation a réduit les dépôts dans la gorge de 46 (26–61) à 13 (6–22) % ($p<0.05$) de la dose inhalée et retenue.

L'effet de la dessiccation sur les effets biologiques de la methacholine en aérosol a été étudié chez 21 sujets, qui ont subi trois tests de provocation à des jours différents l'un au moyen de l'appareil de référence, et deux au moyen de l'appareil dessiccant, avec doublement des concentrations de methacholine depuis 0.5 jusqu'à un maximum de 64 $mg \cdot ml^{-1}$. Les pourcentages de modification du VEMS (% FEV_1) par dose cumulative de methacholine ont changé de -1.0 (2.6) % de FEV_1 μmol^{-1} (moyenne géométrique et déviation standard) avec l'appareil de référence jusquà 1.6 (3.6) avec l'appareil dessiccant ($p<0.02$). Dans une étude additionnelle, 20 sujets ont subi quatre tests de provocation à des jours différents, deux avec une version modifiée de l'appareil dessiccant, et deux avec l'appareil de référence. La pente a changé de -1.1 (3.1) à - 1.6 (3.3), $p<0.02$. La reproductibilité des mesures dupliquées ne s'est pas améliorée au moyen de l'appareil dessiccant.

Donc, l'appareil dessiccant a diminué les dépôts dans la gorge et a augmenté l'effet biologique de la methacholine en aérosol.

Eur Respir J., 1991, 4, 890–898.