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TECHNICAL NOTE

Changes of nebulizer output over the years

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ABSTRACT: The effects of long-term use and of cleaning on the output of 30 specimens of DeVilbiss 646 nebulizers were investigated in order to assess their influence on outcome and interpretation of inhalation provocation tests. Output was assessed in a standardized manner based on weight loss on four occasions one year apart.

Output was highly reproducible (intraclass correlation ≥0.90), and varied considerably between nebulizers, necessitating calibration at least once. Inadequate cleaning diminished nebulizer output by a factor of two ($p<0.01$), while long-term use was associated with a moderate increase in output of 0.28 $mg \cdot yr^{-1}$ ($p<0.0001$).

Bronchial responsiveness can be underestimated by about one doubling dose due to inadequate cleaning, while interchanging nebulizers can lead to overestimation or underestimation by up to one doubling dose. With proper care the increase in output due to wear has no consequences for clinical practice, or for longitudinal studies.

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Several aerosol delivery systems are now available for clinical and scientific studies focusing on the bronchial response to inhaled bronchoconstrictor agents and allergens. Using standardized procedures [1, 2] such devices can deliver reproducible doses of aerosolized agents, provided that the nebulizers are calibrated at least once [3, 4]. Although it has been claimed that some inter-nebulizer variability in output is due to wear on the baffles [2], we do not know of any study in which this long-term variability has been quantified. If wear occurs, it will obviously affect follow-up data from provocation tests. Therefore, it seems desirable to check nebulizer output and correct the test results for deviations in output.

In the present study, we calibrated 30 specimens of DeVilbiss 646 nebulizers four times at one year intervals. The nebulizers were being used for histamine and methacholine provocation tests using a dosimeter method in a longitudinal multicentre study, and for routine clinical purposes. Separately, we examined whether cleaning the nebulizers affected output. Hence, the results reflect the quantitative effect of regular use on the output of these nebulizers.

Material and methods

Thirty DeVilbiss 646 (DeVilbiss Inc., Somerset, PA, USA) nebulizers combined with three Rosenthal-French dosimeters (Laboratory for Applied Immunology Inc., Baltimore, MD, USA) were assembled with the baffle facing the nebulizer outlet [21] and were calibrated annually. Before calibration, all nebulizers were cleaned with tap water and dried with compressed air; the orifices were cleaned with wire furnished by the manufacturer.

Nebulization was achieved according to the standardized technique for clinical practice [5], and calibration of the output was based upon weight loss [1, 2]. Each nebulizer with closed vent was filled with 4 ml of saline at room temperature and connected to a dosimeter operating on dry air at 137.8 kPa (20 $lb \cdot in^{-2}$), with an opening time of 0.6 s. A mouthpiece was attached to the nebulizer outlet and the solution was nebulized twice ("primed"). Priming is recommended because the first aerosol discharge from the nebulizer is of inconsistent volume [6] and is, therefore, not routinely inhaled when subjects are challenged. This was followed by 30 nebulizations, achieved by suction of air through the mouthpiece manually by a 3 *l* syringe [2] in about 1 s. Weight loss due to 30 nebulizations was assessed using a Mettler precision balance (Mettler Toledo AG, Greifensee, Switzerland). Another 30 actuations later, weight was assessed again. Measurements were repeated when the weight loss of the second series of 30 actuations differed by more than 10% from that of the first series, which was an arbitrary criterion. When no reproducible data were obtained after four attempts, nebulizers were discarded. All parts of each nebulizer

were marked with an inert paint to avoid interchanging. Calibration took place in the same room with a temperature that varied between 20–23 °C. Therefore, we assumed that evaporation played no role in any long-term change of output [7, 8]. A separate dosimeter was used for each set of 10 nebulizers. This procedure was carried out in January each year, from 1988 until 1991; it was performed by the same investigators on all but the first occasion.

The effect of the cleaning procedure on nebulizer output was studied separately in three nebulizers. During a 2 week period they were not cleaned following challenges with histamine diphosphate; they were then calibrated, cleaned, and calibrated again.

Data analysis

The reproducibility of duplicate measurements was assessed by studying the relationship of the difference between the two series of 30 actuations *versus* the mean of these two series [9]. Analysis of variance was

Table 1. – Assessment of output of 30 nebulizers

Year	1988	1989	1990	1991
Output mg				
Mean (SEM)	4.37 (0.07)	4.64 (0.13)*	5.21 (0.08)**	5.11 (0.15)**
Var_b	0.160	0.521	0.184	0.687
Var_w	0.018	0.022	0.015	0.016
Range	(3.15–5.08)	(2.95–6.32)	(4.42–6.05)	(3.55–6.62)
10th %	4.01	3.64	4.62	3.95
90th %	4.98	5.52	5.89	6.24
Reproducibility mg#				
Mean (SD)	-0.03 (0.19)	0.006 (0.21)	-0.012 (0.18)	-0.038 (0.18)
Range	(-0.49–0.48)	(-0.41–0.47)	(-0.32–0.27)	(-0.45–0.33)
10th %	-0.20	-0.28	-0.24	0.23
90th %	0.15	0.25	0.22	0.24
Intraclass correlation				
	0.90***	0.96***	0.92***	0.98***

*: p<0.05, **: p<0.0001 compared to 1988 output. ***: p<0.001; # reproducibility: mean (SD) difference between the mean output of the first and that of the second series of 30 actuations. Var_b: variance between nebulizers; Var_w: variance within nebulizers.

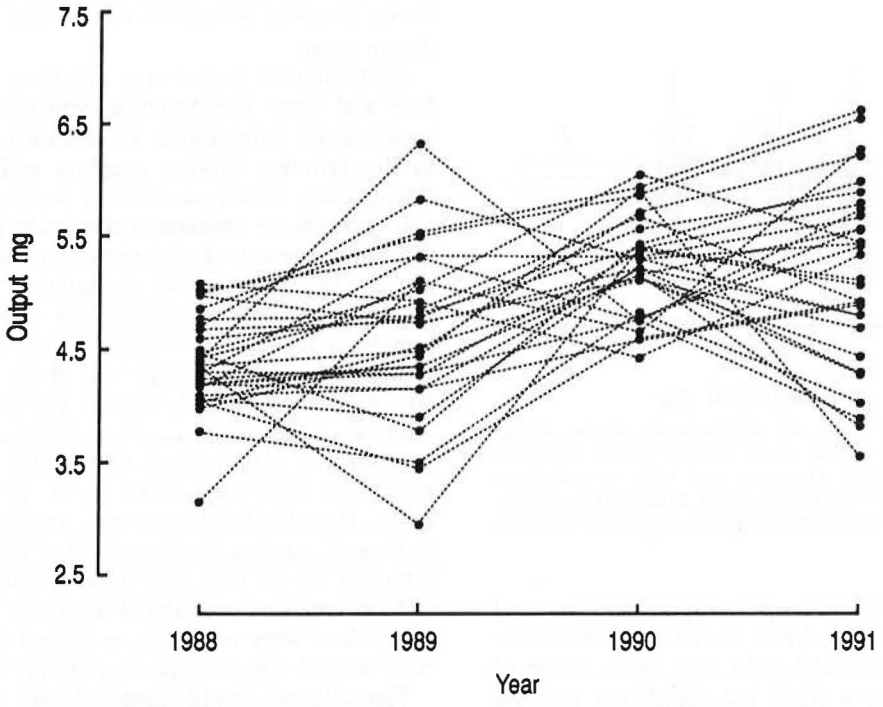

Fig. 1. – Individual output of 30 nebulizers on four calibration sessions.

used to study variance between nebulizers (Var_b), and within nebulizers (Var_w). The reliability of the calibrations was estimated by means of intraclass correlation. It quantifies the relative magnitude of Var_w as compared to Var_b, a high intraclass correlation indicating that Var_b is significantly larger than Var_w, [10]. For n replicate measurements, it is defined as:

$$(MS_b - MS_w)/(MS_b+(n - 1)MS_w)$$

where MS_b and MS_w, are mean squares between and within nebulizers, respectively. The individual and overall longitudinal trends were studied by linear regression of output on years. Individual and group results were then compared in order to test the assumption that all nebulizers behaved similarly.

The effects of cleaning on output were studied using paired t-tests. The level of significance was set at 0.05.

Results

Two nebulizers were discarded and replaced because no reproducible data could be obtained in 1988; we have no explanation for this. Because we observed no significant differences between the three sets with respect to reproducibility and change of output, the data of 30 nebulizers were pooled in the analysis. The mean weight loss per actuation in the first series of 30 actuations was comparable to that in the second series; absolute differences were uncorrelated with the means of the 2 series ($p>0.45$).

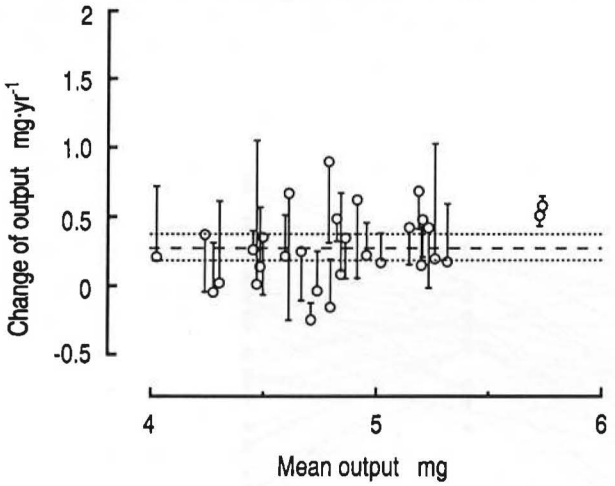

Fig. 2. – Mean output related to individual and overall change of output (n=30). Mean output: mean of four annual individual calibrations. Open symbols and error bars refer to individual change of output and 95% confidence limits. Dashed line and dotted lines refer to the overall (n=30) change of output and 95% confidence limits.

Summary statistics of output, reproducibility and intraclass correlation are listed in table 1. Nebulizer output varied between 2.95–6.32 mg, *i.e.* a factor of 2 (fig. 1). There was a slight but significant increase in nebulizer output (mean 0.28 mg, or 6% of 1988 output) per year ($p<0.0001$). In 26 nebulizers the individual increase in output was not significantly different from the overall trend (fig. 2); the increase was lower in one, and higher in three nebulizers ($p<0.05$).

The three nebulizers that had not been cleaned during 2 weeks of regular use were calibrated, cleaned as described above, and calibrated again. This was associated with a near doubling of nebulizer output ($p<0.01$): mean (SD) output was 2.93 (0.53) mg before cleaning and 5.64 (0.24) mg after cleaning. Depositions of salt crystals near the orifice were visible prior to cleaning and removed by the cleaning procedure.

Discussion

In order to correct data from provocation tests for possible changes, we calibrated DeVilbiss 646 nebulizers on four occasions one year apart, and investigated changes of output to cleaning and due to regular use. We found that the variability in output between nebulizers was considerable, and much larger than variability within nebulizers, as illustrated by significant intraclass correlations. Hence, calibrations were carried out reliably, and reflect true differences between nebulizers, that remained relatively stable during the years. Inadequate cleaning resulted in visible crystal formation near the orifice and an output that was almost halved, while the long-term use was accompanied by a slight but significant increase of output that averaged about 6% per year. Output increased similarly in almost all nebulizers. The nebulizers involved in this study were used for routine and scientific purposes between 200–400 times per year, which makes the explanation of wear a plausible one. However, the variability between nebulizers and that due to the cleaning procedure were much larger than that due to wear.

Calibration of output was based on weight loss before and after nebulization, while conditions were standardized with respect to assembly, position of the baffle, priming, driving pressure and dose duration. Evaporation, which can lead to overestimation of output, depends on ambient temperature [2, 7, 8] and on the water content of the pressurized air [4]. We used dry air, and as the room temperature was comparable for each calibration session, this cannot explain any long-term increase in output. "Inspiratory" flow generated with a large syringe was about 3 $l \cdot s^{-1}$ and similar on each calibration session. For provocation tests, a lower inspiratory flow is usually recommended because it is an important determinant of aerosol deposition [1]. However, RYAN *et al.* [11] showed that, when the inhaled dose was standardized with a dosimeter, varying inspiratory time from 1–5 s had no influence on the total lung dose. Cleaning of nebulizers and orifices was considered adequate when all visible depositions were removed, as judged by visual inspection, and all water evaporated by dry compressed air.

The clinical implications of our findings are: 1) hyperresponsiveness can be underestimated by about

one doubling dose when nebulizers are regularly used but not cleaned; 2) when nebulizers are interchanged, the dose delivered can be almost doubled or halved in comparison with the original dose, in the most extreme cases. This would lead to underestimation or overestimation of hyperresponsiveness of up to one doubling dose. As the magnitude of these errors is about equal to the 95% confidence interval of repeated measures of the response to inhaled histamine or methacholine [12], we recommend that nebulizers be cleaned immediately after use according to a protocol that involves pricking the orifices. When several nebulizers are used, care should be taken to see that nebulizers are not interchanged or their assembly altered.

For quality control we recommend that nebulizers are calibrated at regular intervals. Incorporation of differences in output between nebulizers and changes of output during the years can conveniently be implemented with the aid of computer programs that adjust the dose-response curves before provocative doses are calculated by log-linear interpolation. This procedure was also carried out in our studies. Although it has been claimed that nebulizer output is affected by wear [2], we found that with proper care the long-term increase in nebulizer output is relatively small and unlikely to affect the interpretation of test results.

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