

## Letter

## The PD<sub>20</sub> but not the PC<sub>20</sub> in a methacholine challenge test is device independent

Methacholine challenge tests aid in the diagnosis or exclusion of asthma based on the dose (PD<sub>20</sub>) or concentration (PC<sub>20</sub>) of inhaled methacholine that provokes a 20% decrease in the forced expiratory volume in 1 second (FEV<sub>1</sub>). However, since the American Thoracic Society (ATS) standards were published in 2000,<sup>1</sup> the English Wright nebulizer (EW) recommended for the 2-minute tidal breathing method has become difficult to find; hence, many laboratories substitute other nebulizers with little standardization of the amount expected to deposit within the lungs. The recommendation for the EW was to adjust the nebulizing flow to achieve a gravimetrically determined output of 0.13 mL/min.<sup>1</sup> However, a recent in vitro study<sup>2</sup> found that 90% of the weight loss is attributable to evaporation and not output of methacholine. Another problem with the EW is environmental contamination. Even with a filter on the expiratory limb, aerosol will be lost to the environment should the patient come off the mouthpiece, usually to cough.

There have been 1 in vitro<sup>2</sup> and 2 in vivo reports<sup>3,4</sup> comparing the EW to the breath actuated AeroEclipse Breath Actuated Nebulizer (AE) nebulizer in children and adults, respectively. The expected rate of deposition from the AE is 10 fold that of the EW.<sup>2</sup> For this reason, the 2 in vivo studies use an inhalation time of 20 seconds for the AE compared with 2 minutes for the EW but still found a lower PC<sub>20</sub> for the AE. The pediatric study also quantified a cumulative effect of methacholine. They analyzed the results in terms of the PD<sub>20</sub> and found no difference between the 2 delivery systems. The purpose of this study is to do a reanalysis to determine whether the results would be similar in adults and in children based on a simpler-to-calculate noncumulative dose for PD<sub>20</sub>.

Forty-one adults were studied at McMaster University, Hamilton, Ontario, Canada, and 24 children at the Hospital for Sick Children, Toronto, Ontario, Canada. Both studies had institutional review board approval and were registered ([clinicaltrials.gov](http://clinicaltrials.gov) Identifiers: NCT01919424 and NCT01288482, respectively). All participants had stable asthma with an FEV<sub>1</sub> greater than 65% predicted and a previously measured PC<sub>20</sub> of 16 mg/mL or less using the standard 2-minute tidal breathing method with the EW.<sup>1</sup> Further details are available.<sup>3,4</sup> The protocol with the EW was identical to that described by Crapo et al,<sup>1</sup> but for the AE, the time between the end of one inhalation and the start of the next was standardized at 3 minutes 40 seconds, meaning the AE cycle was approximately 1 minute shorter than the EW cycle. The order of the test was random, and spirometry was performed at 30 and 90 seconds after inhalation for both.

The PC<sub>20</sub> was calculated by extrapolating between the 2 last concentrations given as suggested by Crapo et al,<sup>1</sup> and the PD<sub>20</sub>

was calculated in an analogous manner with dose substituted for concentration. For each device, the dose was calculated based on the in vitro model. For a concentration of 16 mg/mL, the expected pulmonary deposition was 0.19 mg/min for the EW and 2.05 mg/min for the AE,<sup>2</sup> and it was linear with concentration.

A paired *t* test was used to ascertain the difference between the logarithm of the PC<sub>20</sub> and the PD<sub>20</sub> for the adult and pediatric patients and all patients together. For the adults, the geometric mean of the PC<sub>20</sub> was 1.85 mg/mL using the EW and 0.88 using the AE, which were highly significant differences (*P* < .001). However, the PD<sub>20</sub> values were remarkably similar, 44 μg for each (*P* = .92). For the children, the geometric mean PC<sub>20</sub> was 1.24 using the EW compared with 0.55 using the AE, which were highly significant differences (*P* < .001). In contrast, the geometric means of the PD<sub>20</sub> were 29 and 23 μg (*P* = .33). Taken as a whole, the results were similar, although the *P* value for the difference in PC<sub>20</sub> was much more significant, whereas the values for PD<sub>20</sub> were similar (*P* = .37). Comparison between the 2 delivery systems is presented graphically in Figure 1 for PC<sub>20</sub> and PD<sub>20</sub> using a Bland and Altman plot.<sup>5</sup>

These results indicate that for a methacholine challenge test in both adults and children, the PC<sub>20</sub> is very dependent on the delivery system, whereas the PD<sub>20</sub> is not. The AE that was used in these studies has many advantages over the obsolete EW, with ease of acquisition being primary but also the fact that nebulization stops should the patient come off the device to cough, minimizing environmental contamination. However, other devices whose output properties have been quantified would be expected to yield similar results for PD<sub>20</sub> although possibly quite different results for PC<sub>20</sub>.

This study used a different analysis of the data that from 2 other studies.<sup>3,4</sup> Dell et al<sup>3</sup> calculated the PD<sub>20</sub> using the cumulative delivery of methacholine, whereas El-Gammal et al<sup>4</sup> found a lower PC<sub>20</sub> when using the AE compared with the EW but did not calculate delivered dose. Although there is clearly a cumulative effect, there is also a biological half-life that is less well worked out, but whether one uses a cumulative approach or the simpler noncumulative delivered dose, no solution will be perfect. This finding emphasizes the importance of standardizing the dose timing interval. A corollary is that for highly efficient delivery systems one should consider starting with a lower concentration of methacholine to avoid inadvertently causing severe bronchospasm. The protocol was written<sup>3</sup> as an attempt to shorten the test before knowing the cumulative effect.<sup>3</sup> It would have been preferable to have identical periods between the start of inhalation for both systems and perhaps a longer inhalation time (eg, 1 minute to minimize any effect of breath variability for the AE but starting at a lower concentration).

In conclusion, PD<sub>20</sub> but not PC<sub>20</sub> will be independent of the delivery system, providing the delivery characteristics of the device are known.

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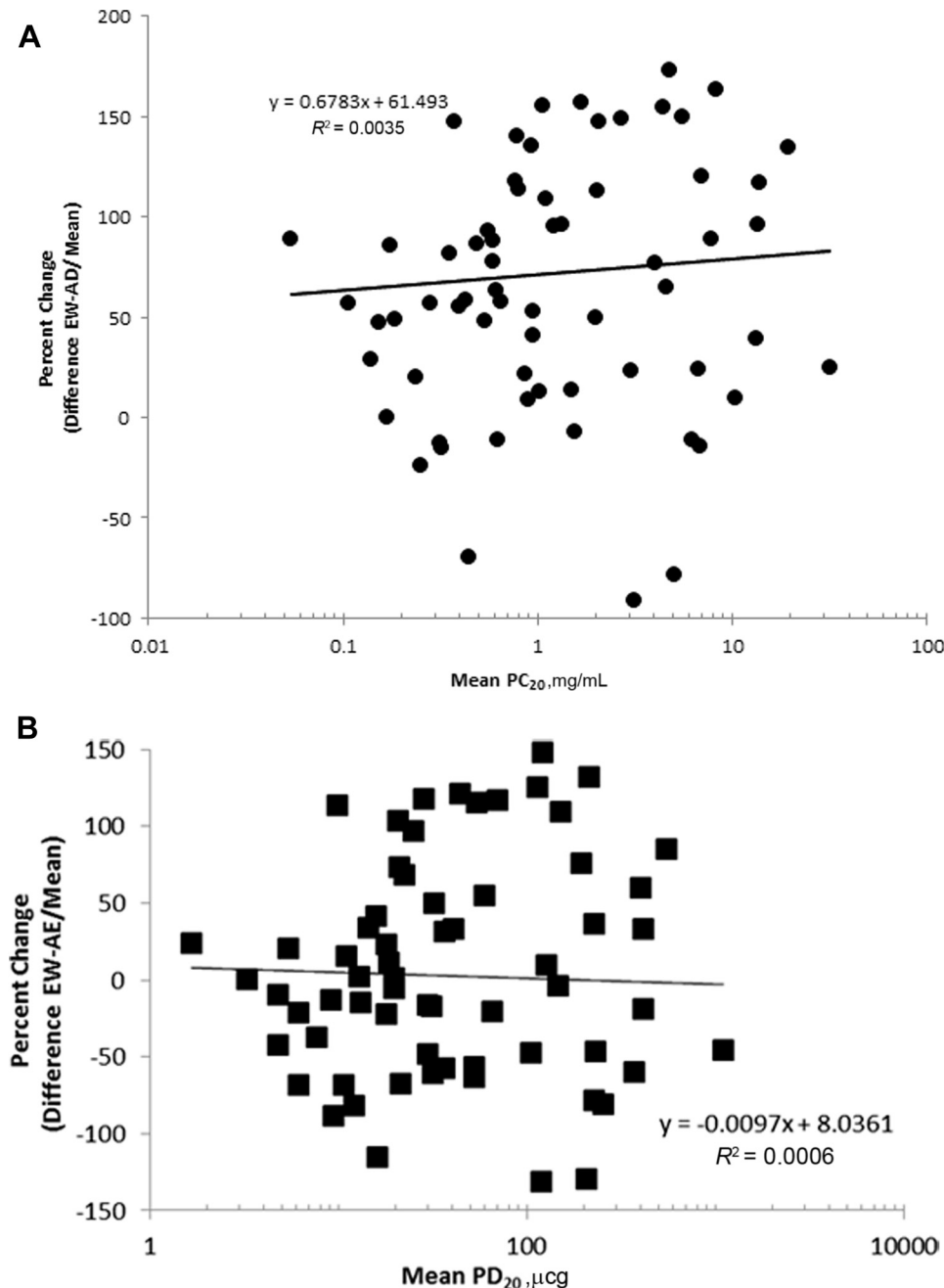


**Figure 1.** Bland and Altman plots showing the percentage of difference between the English Wright nebulizer (EW) and the AeroEclipse Breath Actuated Nebulizer (AE) delivery systems plotted against the mean concentration of inhaled methacholine that provokes a 20% decrease in the forced expiratory volume in 1 second (PC<sub>20</sub>) (A) and dose of inhaled methacholine that provokes a 20% decrease in the forced expiratory volume in 1 second (PD<sub>20</sub>) (B). Values above zero indicate a difference that is present for PC<sub>20</sub> but not PD<sub>20</sub>. The magnitude of PC<sub>20</sub> or PD<sub>20</sub> did not influence the degree of difference. The low values for  $R^2$  indicate a very poor correlation between the percentage of difference in PC<sub>20</sub> or PD<sub>20</sub> and the means of PC<sub>20</sub> and PD<sub>20</sub>, respectively.

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