Effect of airway smooth muscle tone on airway distensibility measured by the forced oscillation technique in adults with asthma

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1Department of Medicine, Nursing and Health Sciences, Monash University, Clayton, Victoria; 2Co-operative Research Centre for Asthma and Airways, Glebe, New South Wales; 3The Woolcock Institute of Medical Research, Glebe, New South Wales; 4Department of Allergy, Immunology and Respiratory Medicine, The Alfred Hospital, Prahran, Victoria, Australia; 5Department of Medicine, Pulmonary and Critical Care Unit, Massachusetts General Hospital and Harvard Medical School, Boston; 6Division of Sleep Medicine, Brigham and Women’s Hospital and Harvard Medical School, Boston, Massachusetts

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Kelly VJ, Brown NJ, Sands SA, Borg BM, King GG, Thompson BR. Effect of airway smooth muscle tone on airway distensibility measured by the forced oscillation technique in adults with asthma. J Appl Physiol 112: 1494–1503, 2012. First published February 23, 2012; doi:10.1152/japplphysiol.01259.2011.—Airway distensibility appears to be unaffected by airway smooth muscle (ASM) tone, despite the influence of ASM tone on the airway diameter-pressure relationship. This discrepancy may be because the greatest effect of ASM tone on airway diameter-pressure behavior occurs at low transpulmonary pressures, i.e., low lung volumes, which has not been investigated. Our study aimed to determine the contribution of ASM tone to airway distensibility, as assessed via the forced oscillation technique (FOT), across all lung volumes with a specific focus on low lung volumes. We also investigated the accompanying influence of ASM tone on peripheral airway closure and heterogeneity inferred from the reactance versus lung volume relationship. Respiratory system conductance and reactance were measured using FOT across the entire lung volume range in 22 asthma subjects and 19 healthy controls before and after bronchodilator. Airway distensibility (slope of conductance vs. lung volume) was calculated at residual volume (RV), functional residual capacity (FRC), and total lung capacity. At baseline, airway distensibility was significantly lower in subjects with asthma at all lung volumes. After bronchodilator, distensibility significantly increased at RV (64.8%, P < 0.001) and at FRC (61.8%, P < 0.001) in subjects with asthma but not in control subjects. The increased distensibility at RV and FRC in asthma were not associated with the accompanying changes in the reactance versus lung volume relationship. Our findings demonstrate that, at low lung volumes, ASM tone reduces airway distensibility in adults with asthma, independent of changes in airway closure and heterogeneity.

ELEVATED AIRWAY SMOOTH MUSCLE (ASM) tone causes airway constriction and airflow obstruction in patients with asthma (4), which, in some cases, can be overcome with a deep breath (40). It follows that determining how the airway responds to lung expansion and ASM tone is fundamental for our understanding of respiratory dysfunction in asthma. High-resolution computed tomography (HRCT) imaging studies of airway diameter-pressure relationships in humans and dogs have shown that with inflation from low to high transpulmonary pressures (0 to 25–30 cmH2O), the relaxed airway dilates easily at low transpulmonary pressures (high specific compliance), reaching a plateau at pressures <10 cmH2O (5, 6, 22). Once the plateau is reached, the airway experiences minimal dilation with further lung inflation (low specific compliance). Furthermore, as demonstrated in dogs, when ASM tone is increased, the ability of the airway to dilate is reduced such that it no longer reaches a plateau in dilation at pressures <10 cmH2O (specific compliance at low transpulmonary pressures is reduced), and the airway may not reach its maximum dilation even at full lung inflation (5, 6). The effect of elevated ASM tone, in reducing specific airway compliance around FRC, may therefore be an important characteristic of airway pathophysiology in asthma.

To date, studies of airway dilation with respect to increases in lung volume in humans, namely, airway distensibility, have not been able to replicate the influence of ASM tone on the ability of the airway to dilate at low lung volumes (2, 7, 40). However, they have been able to demonstrate that airway distensibility at high lung volumes is reduced in subjects with asthma (2), and this reduction has been associated with remodeling of the airway wall (49). There are two key factors that may have contributed to the previous insensitivity of airway distensibility to alterations in specific airway compliance with varying ASM tone. First, previous studies measuring airway distensibility have focused on lung volumes above functional residual capacity (FRC). Given that the predominant influence of ASM tone on specific compliance occurs between 0 and 10 cmH2O, it is likely that the influence of ASM tone on airway distensibility occurs at lung volumes equivalent to those pressures, i.e., at and below FRC (5, 6, 22). Consistent with this notion, Ding et al. (14) has shown the bronchoconstrictor response to be enhanced at volumes below FRC. Furthermore, airway distensibility, as assessed with HRCT, is typically calculated from measures of airway lumen area at FRC and total lung capacity (TLC) (7, 40), therefore reducing the ability to detect changes in distensibility that are nonlinear with respect to lung volume. Second, airway distensibility, as measured using the forced oscillation technique (FOT), may be influenced by alterations in peripheral lung mechanics due to ASM tone. Specifically, relaxation of ASM may decrease lung tissue elastance (20) and reduce airway closure, derecruitment, and airway heterogeneity (36, 38, 42), which may alter the measured distensibility and therefore mask any true alterations due to ASM tone.

The aim of the present study was to determine the contribution of ASM tone to airway distensibility, as assessed via
FOT, across all lung volumes with a specific focus on low lung volumes. We measured airway distensibility continuously from residual volume (RV) to TLC, before and after bronchodilator, in asthma subjects and healthy controls. To investigate the possible contributions of airway closure and heterogeneity to airway distensibility at low lung volumes, we simultaneously assessed peripheral lung mechanics through analysis of the relationship between respiratory system reactance (Xrs) and lung volume. Based on the available evidence showing that increased ASM tone reduces airway dilation with inflation at lung volumes at and below FRC (5, 6, 22), we tested the hypothesis that airway distensibility at these lung volumes in subjects with asthma would be increased after the relaxation of ASM tone by bronchodilator administration.

METHODS

Subjects. This study was approved by the Ethics Committee of The Alfred Hospital (Victoria, Australia). Subjects with well-controlled asthma, according to current guidelines (35), were recruited along with healthy control subjects (no diagnoses of respiratory or cardiac disease). All subjects were current nonsmokers (<10 pack year smoking history) and were asymptomatic for acute respiratory infection. Written informed consent was obtained from all subjects before inclusion in the study.

Experimental design. Subjects completed the following (in order): spirometry, static lung volumes via plethysmography, and measurements of respiratory system conductance (Grs) and Xrs via FOT. All tests were completed at baseline and after the administration of 300 μg of a short-acting β2-agonist (salbutamol) via a spacer. Before being tested, asthmatic subjects withheld short- and long-acting bronchodilator medications for at least 6 and 12 h, respectively.

Equipment and measurements. Spirometry and lung volumes were performed on a Medgraphics Platinum Elite Dx (Medical Graphics, St. Paul, MN) to American Thoracic Society/European Respiratory Society criteria (33, 48). Conductance and reactance (6 Hz) were recorded using a previously described in-house-built FOT device (41). Briefly, mouth pressure was recorded using a solid-state pressure transducer with a range of ±12.5 cmH2O (Sursense DCAL4, Honeywell Sensing and Control, Milpitas, CA). Flow at the mouth was recorded using a Fleisch pneumotachograph (50-mm diameter) with a similar transducer that had a reduced range (±2.5 cmH2O). Pressure and flow recordings were sampled at 300 Hz and then bandpass filtered (5–7 Hz) for analysis of respiratory impedance (Zrs). Separately, the original flow signal was integrated to obtain an estimate of breathing lung volume. Zrs was calculated every 1/6 s via the division of pressure by flow in the frequency domain, where Zrs = Grs + jXrs; respiratory system conductance (Grs) was determined as the reciprocal of resistance (Rrs), and Xrs is the component of the impedance that is 90° “out of phase” (j denotes that Xrs represents the out of phase component of Zrs).

In this study, the measurement of conductance and reactance are specific to 6 Hz. Although conductance is frequency dependent, at frequencies between 5 and 8 Hz there is very little alteration in conductance with frequency (21). Furthermore, at these frequencies, tissue resistance contributes <10% of the measured conductance (21). Therefore, conductance measured at 6 Hz predominantly measures the resistance due to the airways and, as such, is a suitable excitation frequency to use within this study for the global assessment of airway caliber. In addition, Xrs is also frequency dependent, and at frequencies between 5 and 8 Hz lung elastance has been shown to have excellent responsiveness to alterations in tissue mechanical properties (20, 28), including airway heterogeneity and closure (27), and therefore the measurement of reactance at 6 Hz is well suited for the assessment of peripheral airway heterogeneity and closure.

To obtain measurements of conductance and reactance across multiple lung volumes, a modification of the breathing protocol of Brown et al. (2) was used. First, 1 min of tidal breathing was followed by a slow vital capacity maneuver and further tidal breathing. Three incremental deflation maneuvers were then performed in succession, separated by periods of tidal breathing. Finally, a second slow vital capacity maneuver was performed (Fig. 1). Each incremental deflation maneuver began with a deep breath to TLC followed by tidal breaths with decreasing end-expiratory lung volume until RV was reached. In the previous study of Brown et al. (2), the incremental deflation maneuver was terminated at FRC. Subjects performed the entire breathing protocol twice. In the event of less than four tidal breaths during the deflation maneuvers, a third trial was performed to maximize the number of data points for analysis. Mean conductance was calculated as the average conductance over the 30-s period of tidal breathing before the initial vital capacity maneuver.

The spiromgram, recorded during the breathing protocol, was translated to absolute lung volume by equating the maximum lung volume achieved during the protocol to the TLC determined via plethysmography. Measurements of conductance and reactance during the deflation maneuvers were obtained at points of zero tidal flow, at both end inspiration and end expiration, to avoid the known effect of flow on resistance (30). Measurements of conductance and reactance made during events where there appeared to be excessive movement, leak at the mouth, or closure of the glottis, such as during swallowing, were excluded.

Conductance-lung volume relationship. Airway distensibility was defined as the slope of the relationship between conductance and lung volume (in liters) (2). Distensibility was determined from the conductance data obtained for each subject at baseline and after bronchodilator as follows: the conductance data were grouped and plotted against the lung volume (in liters) at which they were obtained. The curvilinear relationship between conductance and lung volume (Fig. 2A) (8) was then characterized by Eq. 1 using least-squares regression (Matlab, Mathworks, Natick, MA) as follows:

\[
G_{rs} = a(Vol - b)^3 + c(Vol - b) + d \quad \text{where } c \geq 0
\]

where a is the amplitude of the conductance-lung volume relationship, Vol is lung volume, b and d are the lung volume and conductance at the inflection point on the curve, and c is the slope of the curve at the inflection point. Each coefficient of Eq. 1 has a unique influence on the shape of the conductance-lung volume relationship, as shown in Fig. 2B. Curve fitting and starting and limiting conditions are shown in Table 1.

To confirm that Eq. 1 was necessary to describe the majority of the curves seen in subjects in this study, we visually compared plots of the residuals of the regression determined by Eq. 1, with the residuals obtained from both quadratic \( [G_{rs} = a(Vol - b)^2 + c] \) and linear \( (G_{rs} = aVol + b) \) regression. This analysis revealed that the residuals resulting from the linear and quadratic regression demonstrated nonrandom systematic patterns compared with those resulting from the cubic, which had a random pattern. Based on the patterns observed in the residuals in the lower order model, the quadratic was superior to the linear model in all subjects both at baseline and after bronchodilator. In control subjects, the cubic model was always considered to be superior to the quadratic model. In asthma, the cubic model was considered superior to the quadratic model in 20 of 22 subjects at baseline and in 18 of 22 subjects after bronchodilator.

The conductance-lung volume relationship allowed distensibility, the slope of the conductance-lung volume relationship, to be calculated at any lung volume. Using this approach, a measure of distensibility was made for each subject at RV, FRC, and TLC, both at baseline and after bronchodilator administration. In the event that subjects were unable to reach RV during the deflation maneuver, as determined via plethysmography, the minimum lung volume achieved was used for the determination of distensibility at RV.
Reactance-lung volume relationship. To assess peripheral lung mechanics, we determined the relationship between reactance and lung volume. $X_{rs}$ at 6 Hz reflects the balance between overall elastance and inertance properties of the respiratory system. Respiratory system elastance is a combination of both lung and chest wall elastance, but, at 6 Hz, it is dominated by the lung. Respiratory system inertance largely reflects the pressures required to accelerate intrapulmonary gas with a negligible contribution from the chest wall (34). It follows that alterations in $X_{rs}$ at 6 Hz may reflect changes in lung elastance due to alterations in tissue elastance (alveolar and surface tension), lung recruitment (13), and heterogeneity of airway caliber (27) as well as gas inertance (proportional to airway length and inversely proportional to airway lumen area). Importantly, peripheral heterogeneous airway narrowing results in airway wall shunting, which greatly increases overall lung elastance (27, 29) and therefore lowers $X_{rs}$.

Reactance data were plotted against the lung volume at which they were obtained. The bilinear reactance-lung volume relationship was segmented into two linear regions separated by a “break point,” as described by Stuart-Andrews et al. (45). Briefly, this method “breaks” the sequential data points into two data sets, separated at the break point, applies linear regression to each data set, and determines the combined sum of squares of the two regressions; the process is repeated until each data point has been the break point. The break point that represents the best overall regression for both data sets, as determined from the minimum sum of squares error, is regarded as the final break point, and the results for the bilinear regressions are determined using that break point. From the break point analysis, two indexes to describe the reactance-lung volume relationship were determined (Fig. 3) in each subject at baseline and after bronchodilator.

The reactance-lung volume relationship has a distinctive profile where as lung volume decreases from TLC, reactance remains relatively stable until a critical lung volume is reached, after which further expiration causes a dramatic reduction in reactance. We interpret this relationship as follows: since the reduction in reactance to the left of
the break point is consistent with lung derecruitment (13), we take the critical lung volume (Volcrit) to represent a surrogate measure of closing volume. At Volcrit, where reactance is at a local maximum and therefore airway closure and lung derecruitment are minimal, we interpret the reactance \( X_{\text{rs}} \) to reflect the lung in a homogeneous state. Any heterogeneous peripheral airway narrowing that persists at this volume will therefore provide a reduced \( X_{\text{rs}} \). It follows that above closing volume, within a population characterized by airway heterogeneity such as in asthma, differences in \( X_{\text{rs}} \) are taken to reflect the extent of peripheral airway heterogeneity that persists above closing volume.

**Statistical analysis.** Data were analyzed using SigmaStat (Systat Software, San Jose CA). To allow comparison of the conductance-lung volume and reactance-lung volume relationships, the coefficients \( a, b, c, \) and Volcrit), with respect to volume, were transformed from units of liters into units of percent predicted TLC (2). Two-way repeated-measures ANOVA with Student-Newman-Keuls post hoc analysis was used to assess the effects of asthma and bronchodilator administration on the conductance-lung volume relationship, airway distensibility at RV, FRC, and TLC, and the two reactance-lung volume indexes. Forward stepwise regression was used to assess the associations between baseline distensibility at each lung volume, \( X_{\text{rs}} \), and Volcrit in addition to the associations between the bronchodilator-induced increases in distensibility at each lung volume. Statistical significance was accepted at \( P < 0.05 \).

**RESULTS**

Twenty-two subjects with asthma (15 men and 7 women) and twenty healthy controls (11 men and 9 women) participated in the study. One control subject was excluded from the analysis due to inadequate test performance. There were no differences in anthropometrics between the asthma and control groups, as assessed by an unpaired Student’s \( t \)-tests (Table 2). Subjects with asthma were on standard inhaled therapy, including inhaled corticosteroids, long-acting \( \beta_2 \)-agonists, and short-acting \( \beta_2 \)-agonists as required. Pulmonary function results, including reference values (3, 10, 16), are shown in Table 2.
Subject demographics and lung function

<table>
<thead>
<tr>
<th></th>
<th>Control Group</th>
<th>Asthma Group</th>
<th>p Value (Asthma Group Vs. Control Group)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, yr</td>
<td>46.6 ± 3.5</td>
<td>53.6 ± 2.1</td>
<td>0.09</td>
</tr>
<tr>
<td>Number of subjects/group</td>
<td>19</td>
<td>22</td>
<td></td>
</tr>
<tr>
<td>Men/women</td>
<td>11/8</td>
<td>15/7</td>
<td></td>
</tr>
<tr>
<td>Height, cm</td>
<td>169.0 ± 2.1</td>
<td>170.6 ± 2.2</td>
<td>0.56</td>
</tr>
<tr>
<td>Weight, kg</td>
<td>75.9 ± 3.1</td>
<td>81.2 ± 3.8</td>
<td>0.30</td>
</tr>
<tr>
<td>Mean Grs, %predicted</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Baseline</td>
<td>86.0 ± 3.9</td>
<td>61.0 ± 4.5</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>After BD</td>
<td>94.3 ± 4.7‡</td>
<td>72.1 ± 4.7†</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>FEV₁, %predicted</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Baseline</td>
<td>104.0 ± 2.3</td>
<td>63.5 ± 3.7</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>After BD</td>
<td>106.3 ± 2.5†</td>
<td>69.8 ± 3.3‡</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>FVC, %predicted</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Baseline</td>
<td>105.1 ± 2.5</td>
<td>89.7 ± 3.6</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>After BD</td>
<td>104.4 ± 2.5*</td>
<td>92.7 ± 3.0§</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>FEV₁/FVC, %</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Baseline</td>
<td>78.4 ± 1.0</td>
<td>54.4 ± 2.1</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>After BD</td>
<td>82.9 ± 1.2‡</td>
<td>58.1 ± 1.8‡</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>TLC, %predicted</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Baseline</td>
<td>108.7 ± 2.6</td>
<td>116.5 ± 2.8</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>After BD</td>
<td>108.9 ± 2.8</td>
<td>115.3 ± 2.9</td>
<td>0.11</td>
</tr>
<tr>
<td>FRC, %predicted</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Baseline</td>
<td>93.2 ± 3.8</td>
<td>116.3 ± 5.0</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>After BD</td>
<td>91.0 ± 3.2</td>
<td>108.2 ± 4.9*</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>RV, %predicted</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Baseline</td>
<td>89.9 ± 3.9</td>
<td>142.0 ± 5.8</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>After BD</td>
<td>95.8 ± 4.3*</td>
<td>129.6 ± 5.1‡</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>RV/TLC, %</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Baseline</td>
<td>27.8 ± 1.6</td>
<td>42.9 ± 1.9</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>After BD</td>
<td>29.4 ± 1.7†</td>
<td>39.5 ± 1.6‡</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

Values are means ± SE. BD, bronchodilator; FEV₁, forced expiratory volume in 1 s; FVC, forced vital capacity; FRC, functional residual capacity; RV, residual volume. P values were determined by unpaired t-test comparison between control and asthma groups. *P < 0.05, †P < 0.01, and ‡P < 0.001 (by paired t-test comparison between baseline and after BD administration).

Overall, the asthma group had mild gas trapping (RV/TLC above the upper limit of normal) and airflow obstruction (forced expiratory volume in 1 s (FEV₁)/FVC below the lower limit of normal, low mean Grs) that was only partially reversed by bronchodilator (18).

Conductance-lung volume relationship. The conductance-lung volume relationships at baseline and after bronchodilator (BD) administration (dashed lines). Circles represent RV, FRC, and TLC. Note that conductance was lower at all lung volumes in the asthma group before and after BD administration.

Airway distensibility at RV, FRC, and TLC. The airway distensibility at RV, FRC, and TLC at baseline and after bronchodilator in subjects with asthma and healthy controls are shown in Fig. 5. At baseline, airway distensibility was lower in the asthma group compared with the control group at each lung volume, with a progressively greater effect at lower lung volumes; airway distensibility was lower than control subjects by 28%, 38%, and 60% at TLC, FRC, and RV. In the asthma group, the administration of bronchodilator increased airway distensibility by 61% at FRC and by 65% at RV but had no significant effect at TLC. In contrast, bronchodilator had no effect on airway distensibility at any lung volume in control subjects.

To investigate the potential that the increase in distensibility at RV and FRC in the asthma group was, in part, due to the significant decrease in the RV and FRC lung volumes after bronchodilator (Table 2), an isovolume analysis of distensibility was also performed. Specifically, postbronchodilator distensibility was calculated at baseline RV and FRC lung volumes, denoted as RV* and FRC*. In the cases where baseline RV was lower than postbronchodilator RV, RV* was taken as the lowest lung volume observed in both conditions to avoid extrapolation. The effect of bronchodilator on the isovolume distensibility at RV* and FRC* in asthma and control subjects

### Table 3. Effects of asthma and BD administration on the conductance-lung volume relationship

<table>
<thead>
<tr>
<th>Coefficient</th>
<th>Control Group</th>
<th>Asthma Group</th>
<th>p Value (Asthma Group vs. Control Group)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Baseline</td>
<td>After BD</td>
<td>Baseline</td>
</tr>
<tr>
<td>a</td>
<td>6.08 ± 1.05</td>
<td>6.75 ± 1.08</td>
<td>3.68 ± 0.58</td>
</tr>
<tr>
<td>b</td>
<td>64.58 ± 2.75</td>
<td>70.25 ± 2.38</td>
<td>73.46 ± 4.80</td>
</tr>
<tr>
<td>c</td>
<td>3.84 ± 0.58</td>
<td>3.07 ± 0.62</td>
<td>3.38 ± 0.47</td>
</tr>
<tr>
<td>d</td>
<td>0.338 ± 0.024</td>
<td>0.395 ± 0.023</td>
<td>0.191 ± 0.016</td>
</tr>
</tbody>
</table>

Values are means ± SE (in %predicted TLC). *P < 0.05 and †P < 0.001, significant difference between asthma and control groups using post hoc analysis; ‡P < 0.01 and §P < 0.001, significant difference between baseline and after BD using post hoc analysis.
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Fig. 5. Airway distensibility at RV, FRC, and TLC for the asthma and control groups at baseline and after BD administration. Bars are means ± SE. Airway distensibility was significantly lower under baseline conditions in the asthma group compared with the control group at RV, FRC, and TLC. BD increased airway distensibility at RV and FRC in the asthma group. After BD, distensibility remained lower in the asthma group compared with the control group at RV and TLC but not at FRC. **P < 0.01 and ***P < 0.001, baseline vs. after BD; †P < 0.05, ††P < 0.01, and †††P < 0.001, asthma group vs. control control (by two-way repeated-measures ANOVA post hoc comparisons).

was assessed using two-way repeated-measures ANOVA. This analysis showed that after bronchodilator, distensibility in the asthma group increased twofold at FRC* (baseline vs. after bronchodilator: 0.09 ± 0.01 vs. 0.19 ± 0.02, P < 0.05) to become no different from the control group (0.17 ± 0.02). There was a trend toward an increase in airway distensibility at RV* in the asthma group, but this trend no longer reached significance (baseline vs. after bronchodilator: 0.14 ± 0.02 vs. 0.19 ± 0.02, P = 0.07); however, distensibility remained significantly lower compared with the control group (0.35 ± 0.03, P < 0.001). In the control group, bronchodilator had no effect on airway distensibility at FRC* (baseline vs. after bronchodilator: 0.14 ± 0.02 vs. 0.17 ± 0.01) or at RV* (baseline vs. after bronchodilator: 0.31 ± 0.04 vs. 0.35 ± 0.03).

Reactance-lung volume relationship. The mean reactance-lung volume relationships at baseline and after bronchodilator are shown in Fig. 6, and the indexes determined from the reactance-lung volume relationship (Vol\textsubscript{crit} and Xr\textsubscript{scrit}) are shown in Table 4. At baseline, subjects with asthma exhibited a reactance-lung volume relationship that was significantly shifted rightward and downward compared with the control group (Fig. 6), with a 49% greater Vol\textsubscript{crit} and a 124% greater magnitude of Xr\textsubscript{scrit} in asthma subjects (Table 4). After bronchodilator, asthma subjects demonstrated a modest but significant shift in the reactance-lung volume relationship toward the control curve (Fig. 6), with an 8% decrease in Vol\textsubscript{crit} and a 15% decrease in Xr\textsubscript{scrit} (Table 4). This shift occurred in combination with a significant decrease in RV and FRC (Table 2). Despite these effects, Vol\textsubscript{crit} and Xr\textsubscript{scrit} remained significantly lower in the asthma group versus the control group after bronchodilator. There was no effect of bronchodilator on the reactance-lung volume relationship in the control group, although there was a significant increase in RV.

Airway distensibility and reactance in asthma. No univariate associations were found between baseline airway distensibility at RV or FRC and baseline reactance-lung volume indexes in asthma (Vol\textsubscript{crit} and Xr\textsubscript{scrit}). In addition, we observed no associations between the increase in airway distensibility at RV and FRC after bronchodilator and the changes in the reactance indexes. However, baseline airway distensibility at TLC was inversely associated with baseline Xr\textsubscript{scrit} (R\textsuperscript{2} = 0.29, P < 0.05). Moreover, coefficient c (minimum distensibility) was also inversely associated with baseline Xr\textsubscript{scrit} (R\textsuperscript{2} = 0.48, P < 0.001).

DISCUSSION

The present study demonstrated that at low lung volumes, airway distensibility is lower in adults with asthma compared with control subjects. Reducing ASM tone via bronchodilator increases airway distensibility at RV and FRC, effectively normalizing airway distensibility at FRC in asthma. In contrast, distensibility is unaffected by ASM tone in control subjects. Importantly, such changes are independent of alterations in peripheral lung tissue mechanics as determined by FOT at 6 Hz (Vol\textsubscript{crit} and Xr\textsubscript{scrit}). Thus, our study shows that in asthma, elevated ASM tone not only reduces airway caliber (5, 7) but also reduces the distensibility of airways in the tidal breathing range.

Methodological considerations. Rather than assessing lung conductance and reactance using a relatively invasive esophageal pressure measurement, we report total respiratory system values, which means that the mechanical properties of the chest wall will be included in our measurements. In relation to resistance, Black et al. (1) demonstrated that chest wall resistance, which decreased from ~0.5 to 1.0 cmH\textsubscript{2}O·l\textsuperscript{−1}·s to zero with inflation from FRC to TLC, was not different between asthma subjects and healthy controls and was not affected by ASM tone. Furthermore, Nagels et al. (34) demonstrated that, at 4-Hz, chest wall resistance, elastance and inertance are similar in control and obstructed subjects at both high and low lung volumes. It follows that, in the present study, chest wall properties likely contribute to the increase in conductance between FRC and TLC but are unlikely to explain the differences in airway distensibility between the groups or the changes in airway distensibility with ASM tone.

Our conclusion that changes in airway distensibility with ASM tone are independent of peripheral lung mechanics...
hinges on the suitability of reactance as a surrogate measure of peripheral lung mechanics. When airway heterogeneity and closure are absent or minimal (e.g., above closing volume in healthy controls), reactance at 6 Hz reflects lung tissue elastance. However, reactance is also reduced in the presence of heterogeneous airway narrowing and closure (26, 27, 29). It follows that we and others (12, 13) interpret the sharp reduction in reactance at a critical volume during deflation (Figs. 3 and 6) as effective lung derecruitment due to the onset and progressive development of peripheral airway narrowing and closure below closing volume. Direct evidence showing that decreasing reactance at low lung volumes is a measure of airway closure has been provided in a recent study (13) demonstrating that FOT compliance (a transform of reactance) was strongly correlated with the percentage of nonaerated lung tissue in pigs ($\rho^2 = 0.89$). In addition, Dechman et al. (12) demonstrated increased lung elastance at low lung volumes in dogs and attributed this increase to the gradual development of airway closure.

Furthermore, the higher Vol$_{crit}$ (FOT closing volume) in asthma versus control subjects reflects the greater degree of airway heterogeneity and the elastic properties of lung; assuming lung tissue elastic properties are comparable in asthma versus control subjects, we suggest that the less than twofold greater magnitude of Xr$_{crit}$ in asthma versus control subjects reflects the greater degree of airway heterogeneity that is characteristic of the disease.

Our conclusions regarding the effect of ASM tone on distensibility at low lung volumes also rely on the degree to which conductance reflects all airway calibers, specifically when some of those airways close at low lung volumes. When peripheral airway closure occurs at lower lung volumes, we considered the possibility that conductance may also begin to reflect the proportion of closed peripheral airways (17, 46). However, the independent effects of ASM tone on distensibility and peripheral mechanics are consistent with conductance at 6 Hz being dominated by central airway characteristics and reactance at 6 Hz being dominated by peripheral airway characteristics (21).

**Increased airway distensibility after reduced ASM tone in asthma.** Our finding that airway distensibility is increased after bronchodilator at low lung volumes but not high lung volumes is supported by previous in vivo investigations into the relationship between ASM tone and airway compliance in animals (5, 6) and in adults with asthma (22). In these studies, airway compliance, measured as the airway diameter-pressure relationship, was shown to increase at low lung volumes after a decrease in ASM tone. In contrast, greater airway compliance at higher transpulmonary pressures was not seen after bronchodilator, as airways were already largely dilated. This relative insensitivity of airway compliance to ASM tone at high transpulmonary pressures, and thus high lung volumes, readily explains the progressive insensitivity of distensibility to ASM tone at higher lung volumes.

Despite the increase in airway distensibility at RV and FRC after bronchodilator, distensibility remained reduced in the asthma group at RV, which suggests that ASM tone only partially contributes to reduced airway distensibility at RV. Furthermore, distensibility at TLC was not influenced by ASM tone. Reduced airway distensibility at RV and TLC in asthma may be explained by airway inflammation, airway remodeling, or disrupted airway-parenchymal interdependence. However, airway inflammation contributes only in part to reduced airway distensibility in asthma subjects, since the resolution of inflammation with a 12- wk inhaled corticosteroid treatment does not completely normalize airway distensibility, as measured between 75% TLC and TLC (23). The effect of inflammation on airway distensibility at RV remains unknown. Airway remodeling, by stiffening the airway wall, may also contribute to reduced airway distensibility at RV and TLC (49). In addition, a reduction in the transmission of parenchymal distending forces to the airways, altered airway parenchymal interdependence (32), may act to reduce airway distensibility, but it is unclear how this would preferentially reduce distensibility at RV and TLC, with no effect at FRC.

**Lower baseline airway distensibility in subjects with asthma.** Using a curvilinear model to describe changes in FOT conductance with lung volume, we demonstrated that airway distensibility in subjects with asthma is lower at RV, FRC, and TLC compared with healthy controls. Our finding that airway distensibility at TLC was 28% lower in subjects with asthma compared with control subjects is consistent with our previous finding that airway distensibility between 75% TLC and TLC is 52% lower in asthma (2). Conversely, our finding of lowered airway distensibility at FRC may appear inconsistent with our previous observation that the mean airway distensibility between FRC and 75% TLC is similar in asthma and control subjects (2). This apparent inconsistency lies with the differences in ranges of lung volumes assessed. Since the conductance-lung volume relationship exhibits a local plateau between FRC and 75% TLC (Fig. 4), the observation of reduced airway distensibility at low lung volumes is only seen when using a sensitive measure that captures the curvilinear profile of the conductance-lung volume relationship in combination with obtaining measures below FRC.

**Table 4. Effects of asthma and BD administration on the reactance-lung volume relationship**

<table>
<thead>
<tr>
<th>Index</th>
<th>Control Group</th>
<th>Asthma Group</th>
<th>Two-Way Repeated-Measures ANOVA</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Baseline</td>
<td>After BD</td>
<td>Baseline</td>
</tr>
<tr>
<td>Vol$_{crit}$</td>
<td>51.1 ± 2.4</td>
<td>51.1 ± 2.3</td>
<td>76.3 ± 3.0†</td>
</tr>
<tr>
<td>Xr$_{crit}$</td>
<td>−0.72 ± 0.10</td>
<td>−0.62 ± 0.11</td>
<td>−1.61 ± 0.20†</td>
</tr>
</tbody>
</table>

Values are means ± SE. Vol$_{crit}$, closing volume (in % predicted TLC) using the forced oscillation technique; Xr$_{crit}$, reactance at Vol$_{crit}$ (in cmH$_2$O·L$^{-1}$·s$^{-1}$).

* $P < 0.01$ and † $P < 0.001$, significant difference between asthma and control groups using post hoc analysis; ‡ $P < 0.01$ and § $P < 0.001$, significant difference between baseline and after BD using post hoc analysis.
In two recent studies, lowered airway distensibility in subjects with asthma was not found when the caliber of relatively large airways was assessed via HRCT (40) and anatomical optical coherence tomography (50). This disparity may be because conductance varies with diameter to the fourth power, meaning that small changes in lumen area may be sensitively detected when measured via conductance (19). As an alternative explanation, Williamson et al. (50) also reported a leftward shift in the diameter-pressure curve in subjects with asthma compared with control subjects, an effect that reduced the extent of airway dilation when transpulmonary pressure was increased from 0 to 20 cmH₂O, which is consistent with reduced airway distensibility between FRC and TLC.

**Physiology of the reactance-lung volume relationship.** Here, we describe the reactance-lung volume relationship at 6 Hz in its entirety for the first time. We found that the FRC of subjects with asthma typically lies below \( V_{\text{olcrit}} \), suggesting that airway closure is present during normal tidal breathing in this subject group; in contrast, FRC lies above \( V_{\text{olcrit}} \) in healthy control subjects. This new evidence that asthma is associated with airway closure at FRC is supported by imaging studies (11, 47) demonstrating that ventilation defects are present in asthma at FRC. Airway closure at FRC may also explain the clearly elevated reactance found in a subset of asthma subjects (20, 29). Moreover, our finding that \( X_{\text{rs}} \text{crit} \) remains lower in asthma versus control subjects, even after bronchodilator is likely to reflect a “persistent” (lung volume independent) peripheral airway impairment, such as heterogeneous narrowing, that requires further investigation. Furthermore, in asthma subjects at lung volumes above \( V_{\text{olcrit}} \), where airway closure and lung derecruitment are minimal, the gradual decrease in reactance toward TLC may reflect an increased “overall lung elastance” as the parenchyma stiffens with alveolar dilation and as surface tension rises (43, 44) or an increased intrapulmonary gas inerterance with airway lengthening (39).

**Clinical implications.** An increase in airway distensibility at low lung volumes after a reduction in ASM tone is likely to be a mechanism of improved airflow in patients with asthma. In confirmation of this view, further analysis revealed a modest significant correlation between the percent improvement in FEV₁ after bronchodilator and the percent change in distensibility at FRC in asthma (\( R^2 = 0.29, P < 0.05 \)); that is, those that have a greater improvement in airflow obstruction with reduced ASM tone also have a greater improvement in distensibility at FRC. Notably, no association was found between the improvement in FEV₁ and that in distensibility at RV. An increase in airway distensibility at FRC is likely to relieve airflow obstruction by promoting increased airway conductance at tidal volumes. It is also possible that increased airway distensibility at FRC amplifies the tidal oscillations in ASM length during tidal breathing that may act to further reduce ASM tone and stiffness (15), although this notion remains controversial (25, 37).

Recent evidence has demonstrated that reduced airway distensibility (between FRC and TLC) is associated with the ineffectiveness of a deep breath to cause subsequent bronchodilation in asthma (deep breath response). In addition, the minimum resistance achieved within a deep breath is elevated with increasing asthma severity (presumably reflecting lowered distensibility) in association with a diminished deep breath response (19). Notably, the association between distensibility and the deep breath response is abolished after the administration of a bronchodilator (39), suggesting that reducing distensibility with ASM tone renders a deep breath ineffective. Taken together with our observation that airway distensibility at RV, FRC, and TLC are reduced in asthma and increased at RV and FRC with bronchodilator, the present evidence suggests that the maintenance of airway distensibility within the tidal breathing range may be a contributing factor to the effectiveness of the deep breath response.

Our study demonstrates that airway distensibility measured at TLC and RV are reduced in subjects with asthma, but only distensibility at TLC is independent of ASM tone. It follows that the airway distensibility measured at TLC remains a potential noninvasive method for assessing changes to the properties of the airway wall due predominantly to airway remodeling in asthma (2, 23). Since, in asthma, more severe airflow obstruction is associated with greater airway remodeling (9), we further examined the relationship between distensibility at TLC and airflow obstruction at baseline in asthma. As expected, we found a trend for the more obstructed asthma subjects (baseline FEV₁ <75% predicted) to have lower baseline distensibility at TLC than less obstructed subjects (\( 0.34 \pm 0.05, n = 16, \) vs. \( 0.52 \pm 0.09, n = 6, \) means ± SE, \( P = 0.07 \)), although this trend did not reach the level of statistical significance. Examination of a larger group of asthma subjects, including those with milder disease, would be required to confirm this trend. Similarly, a significant relationship between reduced distensibility (measured between FRC and TLC with HRCT) and increased obstruction has been reported by Pygros et al. (40). It follows that reduced airway distensibility at TLC likely reflects alterations to the airway wall, and therefore this particular measure may be useful for assessing the efficacy of new treatments to prevent or reverse the remodeling process.

**Conclusions.** In conclusion, we demonstrated that ASM tone reduces airway distensibility in adults with asthma, a contribution that is specific to low lung volumes and occurs independently of changes in peripheral lung mechanics, namely, airflow closure and heterogeneity.

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**AUTHOR CONTRIBUTIONS**

Author contributions: V. J. K., G. G. K., and B. R. T. conceived and designed the research; V. J. K. performed the experiments; V. J. K. analyzed the data; V. J. K., N. J. B., S. A. S., and B. R. T. interpreted the results of the experiments; V. J. K. prepared the figures; V. J. K. drafted the manuscript; V. J. K., N. J. B., S. A. S., B. M. B., G. G. K., and B. R. T. approved the final version of the manuscript.
REFERENCES


