Maintenance of airway caliber in isolated airways by deep inspiration and tidal strains

LaPrad AS, West AR, Noble PB, Lutchen KR, Mitchell HW. Maintenance of airway caliber in isolated airways by deep inspiration and tidal strains. J Appl Physiol 105: 479–485, 2008. First published June 12, 2008; doi:10.1152/japplphysiol.01220.2007.—Deep inspirations (DIs) are large periodic breathing maneuvers that regulate airway caliber and prevent airway obstruction in vivo. This study characterized the intrinsic response of the intact airway to DI, isolated from parenchymal attachments and other in vivo interactions. Porcine isolated bronchial segments were constricted with carbachol and subjected to transmural pressures of 5–10 cmH2O at 0.25 Hz (tidal breathing) interspersed with single DIs of amplitude 5–20 cmH2O, 5–30 cmH2O, or 5–40 cmH2O (6-s duration) or DI of amplitude 5–30 cmH2O (30-s duration). Tidal breathing was ceased after DI in a subset of airways and in control airways in which no DI was performed. Luminal cross-sectional area was measured using a fiberoptic endoscope. Bronchodilation by DI was amplitude dependent; 5–20 cmH2O DIs produced less dilation than 5–30 cmH2O and 5–40 cmH2O DIs (P = 0.003 and 0.012, respectively). Effects of DI duration were not significant (P = 0.182). Renarrowing after DI followed a monoexponential decay function to pre-DI airway caliber with time constants between 27.4 ± 4.3 and 36.3 ± 6.9 s. However, when tidal breathing was ceased after DI, further bronchoconstriction occurred within 30s. This response was identical in both the presence and absence of DI (P = 0.919). We conclude that the normal bronchodilatory response to DI occurs as a result of the direct mechanical effects of DI on activated ASM in the airway wall. Further bronchoconstriction occurs by altering the airway wall stress following DI, demonstrating the importance of continual transient strains in maintaining airway caliber.

bronchodilation; airway smooth muscle; bronchial segments

THE ABILITY of deep inspirations (DIs) to transiently reverse bronchoconstriction has gained much attention because of the vastly different responses between healthy and asthmatic subjects (4, 6, 8–10, 12, 20–22, 27, 30, 31). Specifically, asthmatic airways do not dilate to the same degree during DI (4, 21) and renarrow more rapidly following DI (20), and DI can even result in further bronchoconstriction in individuals with severe asthma (9, 25).

The mechanism(s) responsible for the initial airway dilation and renarrowing following DI remain unclear, but biochemical (7), neural (23), and mechanical events (1) have been implicated. The most intriguing hypotheses, supported by airway smooth muscle (ASM) strip studies, are those that implicate the direct mechanical effects of straining the ASM (1). Here, the large force fluctuation during DI may result in force suppression of the contractile element. It can be viewed that the large force fluctuation results in eccentric contraction of ASM according to its force-velocity relationship (2), and at the molecular level this is suggested to involve a disruption of binding between actin and myosin (13, 15) or adaptive remodeling of the contractile filaments (19, 32). This proposed mechanism would result in transient amplitude-dependent airway dilation, followed by airway renarrowing.

Indeed, the response to DI is amplitude and duration dependent in vivo with large and long-lasting DIs resulting in significant bronchodilation (6, 8), and we have recently shown that ASM force suppression following DI in isolated constricted airways is proportional to airway wall strain (28). However, DIs of small amplitude and duration in vivo have been shown to result in bronchoconstriction similar to that seen in asthmatic subjects (6, 8). This suggests that under conditions of low ASM strain and thus low ASM force attenuation, further constriction may occur by myogenic responses of the ASM (32). However, it is important to consider that the situation in vivo is further complicated by parenchymal attachments, which may govern the renarrowing characteristics after DI and may result in further bronchoconstriction (16). Thus it remains unclear if known ASM responses to stretch are the dominating mechanism of the DI response in vivo, or if and when the parenchyma contributes to the response.

In this study, we employ an experimental system with which we can explore the effects of transmural pressure oscillations consistent with tidal breathing and DIs of varying amplitudes and durations on the caliber of a single intact airway, isolated from parenchymal attachments and other in vivo interactions. Changes in airway diameter measured by endoscopic imaging are solely due to the responses of the activated ASM and passive viscoelastic wall structures interacting in a dynamic transmural pressure environment. We specifically probe two important questions of the bronchodilatory effects of DI. First, we investigate whether the normal DI responses seen in vivo can be explained simply by the response of the active ASM embedded in the airway wall to physiological DIs, without invoking in vivo interactions other than the transmission of mechanical strain to the ASM. Second, we investigate whether physiological DIs applied to an isolated airway alone are capable of eliciting further bronchoconstriction (i.e., via a myogenic response) without a change in load contribution from the parenchymal attachments. We hypothesize that the normal bronchodilatory effects of DI will occur at the level of a single intact airway without the need for other in vivo interactions. However, further constriction following DI will not occur in...
the healthy isolated airway without invoking changes in the transmural pressure loading environment following DI.

**METHODS**

**Animal handling and bronchial segment preparation.** Animal experiments were conducted in conformity with the American Physiological Society’s Guiding Principles in the Care and Use of Animals and were approved by the Animal Ethics Committee at the University of Western Australia. Pigs, ~25 kg, were sedated with tiletamine-zolazepam (4.4 mg/kg im) and xylazine (2.2 mg/kg im), anesthetized with pentobarbital sodium (30 mg/kg iv), and then exsanguinated. Lungs were removed and placed on ice. A bronchial segment from the lower lobe of the right lung was dissected free of parenchyma, and its side branches were ligated, as previously described (17, 24). The bronchial segments were ~50 mm long and 2.5–5.0 mm in internal diameter at the proximal and distal end, respectively. The segments were cannulated at each end and mounted horizontally in an organ bath, which was open to atmospheric pressure and contained gassed (95% O2-5% CO2) Krebs solution (in mM: 121 NaCl, 5.4 KCl, 1.2 MgSO4, 25 NaHCO3, 5.0 sodium morpholinopropane sulfonic acid, 11.5 glucose, and 2.5 CaCl2) at 37°C. Segments were stretched longitudinally to 110% of their relaxed length to mimic airway lengthening during tidal breathing (24) and remained fixed at that length throughout the experiment. The lumen and adventitia were regularly flushed with fresh Krebs solution before experimentation to equilibrate each bronchial segment to organ bath conditions. Tissue viability was then confirmed with both electric field stimulation (EFS; 300 mA, 3-ms pulse width, 30 Hz) and acetylcholine (ACh; 10⁻⁵ M), as described previously (24).

**Transmural pressure oscillations.** The distal cannula was connected to a hydrostatic pressure column containing Krebs solution, which filled the airway lumen. When required, the intraluminal (thus, transmural) pressure of the bronchial segment was oscillated by cycling the height of the pressure column using a computer-controlled syringe pump. The syringe plunger was driven by a DC motor (M540, McLennan Servo Supplies) using a BioPWM sequential motor controller (V0.3) and custom-designed software (Shane De Catania, McLennan Servo Supplies) using a BioPWM sequential motor controller (V0.3) and custom-designed software (Shane De Catania, McLennan Servo Supplies). The syringe pump was calibrated to establish the linear relationship between syringe displacement and the change in fluid height in the pressure column. A calibrated pressure transducer (Motorola MPX2010DP) was placed in series immediately distal to the bronchial segment to measure intraluminal pressure. The transducer was connected to a Powerlab/20 data-acquisition system (ADInstruments), and the pressure signal was displayed and recorded on a personal computer.

**Bronchial lumen imaging.** Methods for visualizing the bronchial lumen have been described previously (26). Briefly, a rigid fiber-optic endoscope (Olympus SES-1711D) coupled to a CCD video camera (Sony DFW-SX900, 7.5 frames/s) was inserted into the lumen through the proximal cannula. A latex seal was established around the endoscope to maintain fluid pressure. The endoscope was positioned in the lumen of the bronchial segment to visualize a suitable area between generations 10 and 13, which was determined by the number of side branches from the trachea (generation 0). Before the endoscope was inserted, the bronchial lumen was dyed with a blue ring using a steel applicator to aid in lumen visualization. Color video images of the bronchial lumen were displayed in real time and recorded on a personal computer using video acquisition software (Unibrain Fire-i 1.21).

**Experiment protocol.** Bronchial segments were initially contracted to carbachol (CCh) during tidal breathing oscillations, which consisted of sinusoidal pressure oscillations from 5 to 10 cmH2O at 0.25 Hz. The CCh dose was added to the organ bath and was chosen to elicit ~50% of maximal airway area constriction (~EC₅₀ dose, 10⁻⁵ to 10⁻⁶ M final bath concentration). The time course of contraction to CCh during tidal breathing oscillations was determined in a subset of bronchial segments (Fig. 1). During the period in which the bronchial segment was stably contracted (20–40 min after CCh addition), the segment was subjected to a single DI (described below) followed immediately by continued tidal breathing for a further 10 min. CCh was then washed out of the organ bath, and the bronchial segment was allowed to recover to its relaxed state under a static pressure of 5 cmH2O. The above protocol was repeated for a second DI of different amplitude and/or duration. Each bronchial segment received up to three DI protocols in random order. Finally, for a subset of the airways tested, the same DI was applied to the relaxed airway before constriction with CCh.

**DI amplitude and duration.** Four different DI protocols were utilized to assess the effects of pressure amplitude and duration at peak pressure on the DI response. This included DIs with amplitudes of 5–20 cmH2O, 5–30 cmH2O, and 5–40 cmH2O each performed over 6 s, and a DI of amplitude 5–30 cmH2O performed over 30 s. All DIs consisted of a 2-s linear ramp up in pressure from 5 cmH2O to peak pressure (i.e., 20, 30 or 40 cmH2O), a hold at peak pressure (2 s or 26 s for the 6-s and 30-s DI, respectively), followed by a 2-s linear ramp down to 5 cmH2O.

**Effects of reduced oscillatory stress following DI.** The above protocol was modified for a subset of bronchial segments to determine if bronchoconstriction could be elicited by ceasing tidal breathing oscillations following DI, simulating reduced elastic load after DI. Airways were contracted to CCh during tidal oscillations and a 5–20 cmH2O DI of 6 s duration was performed when the contraction was stable. Following DI, the bronchial segment was held statically at the post-DI pressure of 5 cmH2O for the remainder of the protocol. A control protocol was also performed where the bronchial segment was stably contracted to CCh during tidal breathing oscillations and then the oscillations were ceased, without performing a DI. This allowed us to determine if changes in airway caliber were due to a combined effect of DI followed by ceased oscillation or solely due to ceased oscillation.

**Data analysis.** Videos of the bronchial lumen were recorded throughout the whole experiment. The videos were played back to capture images corresponding to the following time points: pre-CCh (relaxed), pre-DI (maximal contraction just before DI), DI (end of hold at peak pressure), post-DI (at 5 cmH2O immediately after DI), and at 30 s, 1 min, 3 min, 5 min, and 10 min after the DI. Each image was captured when the bronchial lumen was visually at its smallest cross-sectional area in the cycle, corresponding to a transmural pressure of 5 cmH2O (i.e., the trough of a pressure cycle). Luminal cross-sectional area was quantified by manually tracing an area of interest with ImageJ (1.49v).
around the bronchial lumen using image analysis software (NIH ImageJ). Images were calibrated by using a probe of known diameter inserted into the lumen (26). Airway area, \( A \), at each time point was expressed as the percent recovery of contraction to CCh and was calculated by the following equation:

\[
A_{\text{recovery}} = \frac{A(t) - A_{\text{pre-DI}}}{A_{\text{pre-CCh}} - A_{\text{pre-DI}}} \tag{1}
\]

Thus a positive percent recovery indicates bronchodilation (i.e., 100% recovery corresponds to complete reversal of constriction), while a negative percent recovery indicates additional bronchoconstriction. Percent recovery data following DI (post-DI time point to 5-min time point) were fit to a three-parameter exponential decay function \( y(t) = y_0 + a \times e^{-\tau t} \), where \( \tau \) represents the time constant of airway renarrowing following DI. For the 30-s DI, percent maximum dilation data at the peak of the DI (2-s time point to 28-s time point) were fit to a three-parameter exponential rise function \( y(t) = y_0 + a \times (1 - e^{-\tau t}) \), where \( \tau \) represents the time constant of dilation at 30 cmH2O.

**RESULTS**

**Stability of CCh contraction and bronchial segment characteristics.** The narrowing time course of control bronchial segments (\( n = 9 \)) administered an EC50 dose of CCh during tidal breathing oscillations is shown in Fig. 1. Airway area was stable between 20 and 40 min after the addition of CCh. Thus all DI and subsequent area measurements were performed during this time period.

Table 1 shows the airway area and contraction measurements for the bronchial segments in each DI group. All groups had equivalent relaxed airway areas (1-way ANOVA \( P = 0.676 \)) and exhibited similar contractions in response to CCh (1-way ANOVA \( P = 0.978 \)).

**Sample DI images.** A sample image sequence from a DI of amplitude 5–40 cmH2O and 6-s duration is shown in Fig. 2. In this example, a bronchial segment with Pre-CCh (relaxed) area of 22.2 mm² exhibited a 39.9% reduction in area at the pre-DI time point (maximal constriction) in response to an EC50 dose of CCh. At the DI time point (40 cmH2O held for 2 s), there was a 71.3% recovery from the contraction, which persisted to a 60.9% recovery when pressure returned to 5 cmH2O at the post-DI time point. The airway gradually renarrowed to a minimal recovery of 5.5% at the 5-min time point.

**Effects of DI amplitude.** To determine the effects of increasing DI peak amplitude, the bronchodilation of bronchial segments resulting from 6-s DIs with amplitudes of 5–20, 5–30, and 5–40 cmH2O were compared. In relaxed bronchial segments, DI resulted in initial dilation at the peak of DI but airway area returned immediately back to pre-DI area at the post-DI time point (data not shown). This was in striking contrast to the contracted bronchial segments, where all DI amplitudes tested resulted in sustained bronchodilation following DI (Fig. 3). The 5–20 cmH2O DI resulted in significant dilation up to 30 s after the DI (\( P = 0.008 \)) but not at 1 min (\( P = 0.614 \)). Although the mean airway recovery fell below 0% from 3 min to 10 min, no significant deviation from the pre-DI area was detected (\( P = 0.619 \) to 0.368). In comparison, the 5–30 cmH2O DI was dilated up to 1 min after the DI (\( P = 0.005 \)) but not at 3 min (\( P = 0.125 \)). Despite having the highest mean recovery at all time points, the 5–40 cmH2O DI only exhibited significant bronchodilation up to 30 s after the DI (\( P = 0.013 \)) but not from 1 min to 10 min (\( P = 0.078 \) to 0.314) due to high variation in this group.

Direct comparison between the groups shows that the 5–20 cmH2O DI exhibited significantly lower bronchodilation than both the 5–30 cmH2O DI and 5–40 cmH2O DI (2-way ANOVA \( P = 0.003 \) and 0.012, respectively). However, there was no significant difference between the 5–30 cmH2O and 5–40 cmH2O DI (2-way ANOVA \( P = 0.231 \)).

Airway renarrowing after DI followed exponential decay functions up to the 5-min time point (\( R^2 = 0.94–1.00 \); 3 of 44 data sets were excluded due to poor fits). The time constants of re-narrowing following the 5–20, 5–30, and 5–40 cmH2O DIs were 36.3 ± 6.9, 27.3 ± 4.3, and 35.4 ± 3.6 s, respectively and there was no significant difference between the groups (1-way ANOVA \( P = 0.422 \)).

**Effects of DI duration.** To determine the effects of increasing DI duration, bronchodilation resulting from a 5–30 cmH2O, 30-s DI was compared with the 5–30 cmH2O, 6-s DI shown previously (Fig. 4A). Although the 30-s DI had higher mean bronchodilation at each time point, the difference between the two groups was not significant (2-way ANOVA \( P = 0.182 \)). Moreover, there was no significant difference in the time constants of renarrowing (1-way ANOVA \( P = 0.285 \)).

The lack of significant difference in bronchodilatory response between DIs of different durations was supported by the time course of dilation during the 30-s DI held at 30 cmH2O (Fig. 4B). While held at 30 cmH2O, continued airway dilation followed exponential rise functions (\( R^2 = 0.92–1.00 \)), where \( y_0 = 61.63 ± 5.60 \% \), \( a = 38.26 ± 5.70 \% \), and \( \tau = 2.11 ± 0.41 \) s. Therefore, the airway reached 85.73 ± 2.02% of maximum dilation within 4.11 ± 0.41 s from DI start. Thus the majority of maximal dilation occurs quickly and is mostly observed during a 6-s DI.

**Effects of reduced oscillatory stress following DI.** By terminating tidal breathing oscillations after a 5–20 cmH2O, 6-s DI, the profile of renarrowing was significantly altered (Fig. 5). In contrast to the standard 5–20 cmH2O DI that showed bronchodilation up to 30 s after DI and did not exhibit further bronchoconstriction at any time point, a 5–20 cmH2O DI after which tidal breathing was ceased only resulted in bronchodilation at the post-DI time point (\( P = 0.003 \)), while significant

Table 1. Airway luminal area and contraction measurements for the different DI groups

<table>
<thead>
<tr>
<th>DI Type</th>
<th>n</th>
<th>Relaxed Area, mm²</th>
<th>Contraction % Area Decrease</th>
</tr>
</thead>
<tbody>
<tr>
<td>5–20 cmH2O, 6 s</td>
<td>9</td>
<td>16.5 ± 2.2</td>
<td>39.4 ± 4.6</td>
</tr>
<tr>
<td>5–30 cmH2O, 6 s</td>
<td>9</td>
<td>16.4 ± 2.0</td>
<td>38.2 ± 4.1</td>
</tr>
<tr>
<td>5–40 cmH2O, 6 s</td>
<td>9</td>
<td>17.8 ± 1.9</td>
<td>38.3 ± 6.1</td>
</tr>
<tr>
<td>5–30 cmH2O, 30 s</td>
<td>9</td>
<td>20.0 ± 1.4</td>
<td>37.2 ± 5.0</td>
</tr>
<tr>
<td>5–20 cmH2O, 6 s (no tidal after DI)</td>
<td>8</td>
<td>15.9 ± 1.8</td>
<td>33.9 ± 4.6</td>
</tr>
<tr>
<td>No DI (no tidal after DI time point)</td>
<td>8</td>
<td>16.8 ± 2.1</td>
<td>38.9 ± 4.5</td>
</tr>
</tbody>
</table>

Values are means ± SE. DI, deep inspiration. There was no significant difference in relaxed airway area or contraction (1-way ANOVA \( P = 0.676 \) and 0.978, respectively) between any of the groups.
bronchoconstriction occurred from 1 min to 10 min \( (P = 0.028 \text{ to } 0.018) \). Overall, the absence of tidal breathing following DI significantly changed the resulting profile of renarrowing \( (2\text{-way ANOVA } P = 0.035) \), but not the time constant of renarrowing \( (1\text{-way ANOVA } P = 0.539) \).

In control airways in which tidal breathing was terminated without DI, significant bronchoconstriction was observed from 30 s to 10 min after ceasing tidal breathing \( (P = 0.004 \text{ to } 0.013) \). Interestingly, the bronchoconstriction resulting from the termination of tidal breathing was virtually identical in both the presence and absence of a 5–20 cmH2O DI \( (2\text{-way ANOVA } P = 0.919) \), suggesting that the absence of tidal breathing following DI, and not the DI itself, was the major contributor to the further bronchoconstriction after DI.

DISCUSSION

DIs in healthy humans are a powerful mechanism of maintaining airway caliber following induced bronchoconstriction \( (6, 8–10, 12, 20, 21, 27, 30, 31) \), but their effectiveness is often reduced in asthma and can even result in transient bronchoconstriction \( (9, 25) \). Previous studies on ASM strips and airway segments showed reduction in force after transient strains \( (14, 18, 28, 32) \), suggesting that the bronchodilatory effects of DI result from the direct mechanical effects of straining the ASM. However, it is unclear how and to what degree such changes in ASM force alter the actual caliber of an airway. Furthermore, the effects of DI on ASM length and airway caliber are complicated by the varying mechanical load contributions of both the passive viscoelastic properties of the airway wall and the parenchymal attachments under dynamic pressure conditions. The present study investigated the degree and dynamics of bronchodilation and subsequent airway renarrowing produced by transient stress on single intact contracted airways, isolated from parenchymal attachments and other in vivo interactions. Our findings show that the caliber of contracted airways is significantly altered by DI, but the extent of this alteration is dependent on the specific conditions and parameters of the DI.

Fig. 2. Sample image sequence from a 5–40 cmH2O deep inspiration (DI). The pre-CCh (relaxed) area of 22.2 mm² was reduced by 39.9% at the pre-DI time point (maximal contraction). There was a 71.3%, 60.9%, and 5.5% recovery of contraction at the DI, post-DI, and 5-min time points, respectively.

Fig. 3. Effects of DI amplitude on bronchodilation from 6 s DIs. Both the 5–30 cmH2O and 5–40 cmH2O DI result in significantly higher bronchodilation than the 5–20 cmH2O DI \( (2\text{-way ANOVA } P = 0.003 \text{ and } 0.012, \text{ respectively}) \), but there was no significant difference between the 5–30 cmH2O and 5–40 cmH2O DI \( (2\text{-way ANOVA } P = 0.231) \). There was no significant difference in the exponential time constant \( \tau \) between the groups \( (1\text{-way ANOVA } P = 0.422) \).

Fig. 4. Effects of DI duration on bronchodilation from 5–30 cmH2O DIs. A: the 30-s DI exhibited higher mean bronchodilation than the 6-s DI at all time points, but the difference was not significant \( (2\text{-way ANOVA } P = 0.182) \). There was no significant difference in the exponential time constant \( \tau \) between the groups \( (1\text{-way ANOVA } P = 0.182) \). B: the time course of the 30-s DI followed an exponential rise function with a fast time constant, showing that the majority of dilation occurred within 4 s of the DI start.
airways is increased for up to 1 min after physiological transmural pressures replicating DI. The magnitude of dilation by DI increased with increased pressure amplitude, consistent with the effects of DI in vivo (6). These results bridge the gap between ASM strip studies and in vivo studies, showing that the normal bronchodilatory effects of DI observed in vivo can be explained simply as a direct mechanical effect of physiological DIs on activated ASM embedded in the airway wall. However, unlike in vivo DI studies (6, 8), low-amplitude DIs did not result in further bronchoconstriction after DI (Fig. 3), suggesting that bronchoconstriction after DI may arise from changing airway-parenchymal interactions, as discussed below.

In the present study, we assessed the response of bronchial segments to DI in an environment that simulated in vivo conditions as much as possible. Each airway segment was stretched to 110% of its resting length at atmospheric pressure, which we previously determined as the airway length in vivo at total lung capacity (20). On the contrary, there was no bronchodilatory effect in relaxed airways, and the airways returned immediately to their pre-DI area (data not shown). Thus a direct effect of the activated ASM is necessary to cause sustained bronchodilation to DI in the intact airway. The low-amplitude DI (5–20 cmH2O) resulted in significantly less bronchodilation following DI than the higher amplitude DIs (5–30 and 5–40 cmH2O). The observed amplitude dependence of DI-induced bronchodilation is likely caused by the magnitude of airway wall stress and the resulting strain on ASM, which is governed heavily by the intrinsic viscoelastic properties of the airway wall and increased airway wall stiffness from ASM contraction. At higher pressures (5–30 cmH2O and 5–40 cmH2O), the viscoelastic airway is presumably stiffer, and further increases in pressure would result in little additional strain, explaining why bronchodilation following these DIs was not significantly different.

Compared with changes in DI amplitude, increasing the duration at peak pressure had a much weaker effect (not significant) on the amount of bronchodilation produced by DI. We expected that viscoelastic creep and prolonged ASM strain would occur by increasing the duration at peak pressure, resulting in increased bronchodilation following DI. However, our analysis indicated that the increase in airway caliber with DI was ~85% complete after ~2 s at peak pressure. Consequently a “breath hold” (an additional 24 s at peak pressure) produced only slightly greater distension of the airway wall than a normal DI, explaining why duration of DI was far less important than amplitude in determining the extent of bronchodilation following DI. These findings are consistent with observations in healthy humans whereby periods of breath hold at total lung capacity do not produce significantly more bronchodilation than a normal transient DI (12).

Some investigators have sought additional insight on the impact of transient stretch on ASM dynamics at various length scales via modeling (e.g., 2, 15). For example, at the level of ASM, it has been shown that a disruption in binding between actin and myosin can bias the ASM toward lengthening (15). Our experiments were not designed to probe the mechanisms underlying ASM force attenuation at the cellular level, but instead, our data reflect ASM behavior in the context of a complete airway system. At this macroscopic level, one of the intuitively simplest modeling approaches uses the Hill hyperbolic force-velocity (F-V) curve to describe the change in ASM length based on the forces opposing the ASM (2). We take an even simpler extrapolation by examining if a single linear F-V relationship is sufficient to describe the renarrowing characteristics of isolated airways following DIs of various amplitudes. We used the following equation:

\[
V = \frac{dr}{dt} = K \times (F_0 - F_a)
\]

where \(r\) (in cm) is the airway radius, \(K\) [in (cmH2O·s)−1] is a constant, \(F_0\) (in cm·cmH2O) is the maximum force the ASM can generate under the prescribed loading condition, and \(F_a\) (in cm·cmH2O) is calculated as the total opposing force on the ASM. In Fig. 6, we present an example of the model’s ability
Fig. 6. Feasibility of describing airway renarrowing data following DI with a computational airway model, in which airway renarrowing is described by a linear F-V equation \([dF/dt = K \times (F_0 - F_a)]\). Here, airway radius decreases when \(F_a\) (the total opposing force on the ASM, \(F_a = F_t - F_p\)) is less than \(F_0\) (the maximum force the ASM can generate at the prescribed loading condition). \(F_t\) is the total force created by the transmural pressure, which we represent as a fixed mean of 7.5 cmH\(_2\)O during tidal breathing oscillations following DI. \(F_p\) is the passive force determined by a quasi-static passive radius-\(F_p\) curve obtained from similar-sized porcine airways. \(F_0\) was determined as the steady-state force required to describe the equilibrated constricted airway’s radius before DI. The best-fit value for the one free parameter, \(K\), was found using a grid search optimization algorithm inversely weighted to the standard errors of the data. Here, \(K\) equals 0.001117 \(1/(\text{cmH}_2\text{O} \cdot \text{s})\), \(F_0\) equals 2.084 cm-cmH\(_2\)O, and the exponential time constant equals 39.5 s.

The model predicts the exponential decay in airway area with a time constant on the same order as that reported in our data and in vivo (20). Thus we show that a single F-V equation with only one free parameter \((K)\) can describe the dynamics of renarrowing following a transient stretch. This parameter presumably captures both ASM reshortening dynamics at the cellular level in unison with passive visceroelastic dynamics and would be ubiquitous for any transient stretch of a given airway. We speculate that the asthmatic airway would have a larger \(K\) value, resulting in a faster renarrowing following DI and thus a smaller time constant, as reported in vivo (20). Although this model captures the transient airway renarrowing response following DI, it is important to note that it does not describe the underlying cellular mechanisms that may be occurring and are still debatable.

An important second aim of our study was to assess whether bronchoconstriction after DI, as can occur in asthmatic subjects (9, 25), could arise as a result of an intrinsic response of the airway wall to stretch. The occurrence of this phenomenon at the airway level would require alternative/additional mechanisms than those proposed for the normal bronchodilatory effect, such as a myogenic response where stretch of the ASM may cause further constriction (32). Our results clearly show that such a myogenic response does not occur following DI in healthy airways, in that all the DIs tested were seen to produce significant bronchodilation for at least 30 s. Previous computed tomography studies by Brown and Mitzner (6, 8) have demonstrated the potential for bronchoconstriction after DI in contracted healthy dogs subjected to small DI amplitudes (positive inflation pressures of 35 cmH\(_2\)O and less, corresponding to \(\sim 20 \text{ cmH}_2\text{O } P_{up}\)) or short-duration DI (10 s and less). The absence of further bronchoconstriction following DI in isolated airways compared with in vivo may suggest that mechanisms other than the airway wall alone produce bronchoconstriction. For instance, the hysteretic properties of lung parenchyma may cause a decrease in lung elastic recoil pressure after DI (i.e., a reduced load against the ASM), favoring bronchoconstriction in vivo (11, 16). Consequently, if airway dilation is small (e.g., after a low-amplitude DI), then parenchymal changes may dominate and allow bronchoconstriction after DI.

A complementary explanation may be that methodological constraints in the studies by Brown and Mitzner (6, 8) inadvertently caused decreased load against the ASM following DI, mimicking the conditions described above. Due to imaging constraints of computed tomography, ventilation of animals temporarily ceased immediately after DI. Ceasing ventilation has been shown to result in further constriction (5) and is probably due to a twofold effect: a reduced mean load on the ASM and a loss of oscillatory load on the ASM. In our present study, we also mimicked these conditions and found that an absence of tidal oscillations after DI resulted in significant airway constriction within 1 min of DI. Interestingly, in airways where tidal oscillations were ceased without a DI, the bronchoconstrictor response was virtually identical to the protocol in which a DI was included (Fig. 5). This suggests that the further airway constriction is not initiated by the DI and is instead solely due to the removal of tidal oscillations. Thus further constriction can occur in isolated airways with reduced oscillatory stress, exactly as reduced elastic recoil pressure after DI in asthmatic subjects is believed to cause further constriction. This suggests that maintaining consistent stress against the ASM before and after DI is vital to prevent further constriction of the ASM, and it may become important to advance the understanding of if and how this is affected in asthmatic subjects by examining airway-parenchymal interactions.

In summary, we show the intrinsic response of the contracted airway to DI in terms of changes in airway caliber. The bronchodilatory response observed here at the level of an individual airway has many similarities with the responses seen in animals and humans in vivo. Thus the normal bronchodiatory effect in vivo can be explained simply as a response of activated ASM embedded in the airway wall to physiological DIs. Using computational modeling approaches, the dynamics of airway renarrowing following DI can be explained by ASM reconstriction following a simple linear F-V relationship. In asthmatic subjects, reduced or absent bronchodilation from DI can be attributed to a stiffer remodeled airway wall that cannot be strained with physiological pressures, in unison with ASM that can renarrow more quickly following DI. However, to understand the asthmatic phenomenon of bronchoconstriction following DI, it may be important to further examine airway-parenchymal interactions following stretch to determine if and how conditions of reduced airway wall stress occur.
REFERENCES


