Respiratory mechanics in sedated and nonseated adult llamas

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Objective—To validate the use of noninvasive pulmonary function testing in sedated and nonseated llamas and establish reference range parameters of respiratory mechanical function.

Animals—10 healthy adult llamas.

Procedures—Pulmonary function testing in llamas included the following: measurement of functional residual capacity (FRC) via helium dilution, respiratory inductance plethysmography (RIP) to assess breathing pattern and flow limitations, esophageal-balloon pneumotachography, and a monofrequency forced oscillatory technique (FOT; 1 to 7 Hz) before and after IM administration of xylazine (0.2 mg/kg).

Results—The following mean ± SD measurements of respiratory function were obtained in nonseated llamas: FRC (5.60 ± 1.24 L), tidal volume (1.03 ± 0.3 L), dynamic compliance (0.83 ± 0.4 L/cm H2O), pulmonary resistance (Rl; 1.42 ± 0.54 cm H2O/L/s), and respiratory system resistance (2.2 ± 0.7; 2.7 ± 0.7 and 2.5 ± 0.6 cm H2O/L/s at 1, 2, 3, 5, and 7 Hz, respectively) by use of FOT. Measurements of flow limitations via RIP were comparable to other species. Sedation with xylazine induced significant increases in Rl and maximum change in transpulmonary pressure. Following sedation, a mean 127% increase in Rl and mean 116% increase in respiratory system resistance were observed across 1 to 7 Hz. The magnitude of change in respiratory system resistance increased with decreasing impulse frequency, suggesting bronchoconstriction.

Conclusions and Clinical Relevance—Noninvasive pulmonary function testing is well tolerated in untrained unseated llamas. These techniques have clinical applications in the diagnosis and treatment of respiratory tract disease, although testing should not be performed after sedation with xylazine. (Am J Vet Res 2007;230:676-684)

Respiratory diseases are recognized health problems in camels and cause for referral to specialized veterinary hospitals. The basis of high-quality care is a firm understanding of the underlying mechanisms of disease. However, a paucity of information exists about respiratory function in camels in comparison to other domestic animal species.

Respiratory function tests, commonly used in companion animal species, may improve the sensitivity and accuracy of diagnosis of respiratory diseases in camels. For example, , and are used to describe the level of airway obstruction in horses with recurrent airway obstruction, and head-out plethysmography has similar application in conscious dogs. In horses with exercise intolerance and cough, airway responsiveness can be determined with a variety of techniques including measurements of , and flowmetric plethysmography. Respiratory inductance plethysmography has further been used to describe breathing patterns in horses and more recently in a llama with dyspnea of unknown origin. In the latter report, abdominal (diaphragmatic) paradox was described, which corresponded precisely with neurologic deficits localized to the diaphragm causing paralysis.

**ABBREVIATIONS**

- Rl: Pulmonary resistance
- Cdyn: Dynamic compliance
- FOT: Forced oscillation technique
- RIP: Respiratory inductance plethysmography
- FRC: Functional residual capacity
- PIF: Peak flow during inspiration
- PEF: Peak flow during expiration
- Vt: Tidal volume
- dPPlmax: Maximum change in transpulmonary pressure
- Rsosc: Respiratory system resistance measured via forced oscillation technique
- fres: Resonant frequency
- Zrs: Respiratory system impedance
- Rlacrit: Respiratory resistance measured via forced oscillation technique
- Xosc: Pulmonary reactance measured via forced oscillation technique
- Cdynosc: Dynamic compliance measured via forced oscillation technique
The purpose of the study reported here was to examine the wider usefulness of respiratory function testing in llamas under conscious sedated and nonsedated conditions. We compared 2 methods of measuring pulmonary mechanics (ie, \( R_g \) and \( C_{dyn} \)) and evaluated the effects of sedation on these parameters. In addition, we measured the effects of sedation on RIP assessment of breathing pattern in llamas. We hypothesized that the methods to derive \( R_g \) and \( C_{dyn} \) would be correlated and that sedation would alter respiratory mechanical function and breathing pattern significantly.

**Materials and Methods**

**Animals**—The Institutional Animal Care and Use Committee at the Cummings School of Veterinary Medicine at Tufts University approved all procedures. Written client consent was obtained for all study llamas prior to enrollment in the study. Ten healthy client-owned adult llamas (3 male and 7 female) between 2 to 9 years (mean ± SD, 5 ± 2.3 years) of age with a mean body condition score of 5 out of 9 (body weight, 100 to 204 kg) were included in this study. None of the llamas had been previously exposed to pulmonary function testing or a laboratory environment. Following hospital admission in the morning, the llamas were acclimated for 4 to 6 hours indoors. All lung function testing was performed in a temperature-controlled environment (mean, 20°C and 71% humidity).

**Study design**—A complete history, physical examination, lateral radiographic views of the thorax, and a CBC were obtained prior to study enrollment to eliminate llamas with any evidence of preexisting respiratory disease. The following external body measurements were obtained by use of a flexible measuring tape with the llama in a standing position: height measured at withers, body length from point of shoulder to ischial tuberosity; and chest circumference over the eleventh intercostal space. All procedures were performed with the llama standing while lightly restrained in a commercially available chute.

The following tests were performed to measure pulmonary function: determination of FRC by the rebreathing technique that uses helium dilution as a test gas,\(^a\) assessment of breathing pattern (thoracic vs abdominal contribution to ventilation) by use of RIP in conjunction with pneumotachography,\(^b\) measurement of \( R_g \) and \( C_{dyn} \) during tidal breathing, and the use of monofrequency FOT.\(^c\) All tests were performed in unsedated llamas (\( n = 10 \)) and repeated following sedation in a subset (7) of these llamas for which owner consent was obtained for sedation. For sedation, llamas received xylazine hydrochloride (0.2 mg/kg, IM). All sedated llamas were tested standing while their heads were manually supported to maintain the mandible horizontal to the ground throughout the study period.

**FRC determination via helium dilution**—Functional residual capacity was measured by use of methods previously described for dogs\(^a\) and horses.\(^b\) All llamas were fitted with a low-dead space (50 mL), clear plastic face mask that was sealed around the bony portion of the muzzle approximately 10 cm behind the external nares, with a latex shroud. The face mask was connected to a low-dead space 3-way angled tap (120°) stopcock and nondiffusible gas collection bag containing breathable test gas (20 mL/kg, 0.3% carbon monoxide, 10% helium, 20% \( O_2 \), and 69.7% nitrogen). The initial and final concentrations of helium and the final concentration of CO\(_2\) following a rebreathing period of 45 to 50 seconds were determined by use of specific analyzers.\(^b\) The dilution of helium (assumed to be a nonexchangeable gas) gave a measure of FRC according to the following formula:\(^d\)

\[
\text{FRC} = \left( \frac{\text{He}}{X_{\text{He}}} - \frac{\text{He}}{X_{\text{He},f}} \right) \times \left( 1 - \frac{\text{He}}{X_{\text{He},i}} \right) \times \frac{1}{1.11} \times \text{L}
\]

where \( \text{He} \) represents the initial concentration of helium, \( \text{He}_{f} \) represents the final concentration of helium, and \( DS_{\text{m}} \) represents the instrument dead space (ie, bag volume in liters).\(^d\) Three consecutive measurements were obtained for each llama and averaged.

**RIP and pneumotachography**—The concurrent use of RIP and pneumotachography permits noninvasive assessment of breathing pattern and ventilation. The use of RIP has been described in a llama\(^a\) and has been used previously in humans,\(^d,e\) horses,\(^f,g,h\) and sheep.\(^i\) In summary, 2 elastic bands (4 cm wide, adult size), each containing a single conducting wire embedded in a sinusoidal fashion, were temporarily placed around the thorax (overlying the 11th intercostal space) and abdomen (behind the last rib) of each llama. To eliminate interference of coat, the fiber was separated to allow placement of bands in close proximity to the skin. Normal respiratory movements cause the bands to stretch (inspiration) and relax (expiration), thus altering the inductance properties of coiled wire, which runs through the center of these bands. Inductance decreases during inspiration and increases during expiration. An oscillating current produces a voltage across these bands that is linearly proportional to stretch and therefore proportional to the change in cross-sectional area of the thorax and abdomen. The voltage signals (thoracic from the thoracic band and abdominal from the abdominal band) were collected at 30 Hz, amplified, and digitized for processing by use of commercial data acquisition software.\(^d\) It is assumed that the thoracic and abdominal contributions to ventilation are separate. Thus these 2 signals can be analyzed independently to evaluate breathing pattern by examining the relative thoracic and abdominal contributions to ventilation.

The analysis of phase angle of thoracic and abdominal signals has been previously described.\(^d\) Briefly, the phase angle was measured from the \( x \)-\( y \) (ie, abdominal vs thoracic) plots by use of the following equation:\(^d\)

\[
\sin \theta = \frac{m \times s}{w}
\]

where \( m \) represents the width on the \( x \)-axis of the \( x \)-\( y \) plot at midway on \( y \)-axis and \( s \) represents the maximum width of the plot along the \( x \)-axis.\(^d\)

Additionally, thoracic and abdominal volume signals were summed to obtain a total volume signal. Flow was also derived from RIP signals. The sum of thoracic and abdominal signals was differentiated offline by use of commercial software to obtain the sum.
flow. The beginning of expiration and beginning of inspiration were defined by the upward- and downward-directed zero crossings of sum flow signals, respectively (Figure 1). Nasal flow was measured concurrently during tidal breathing by use of a heated pneumotachograph attached to the proximal port of the face mask. The pneumotachograph-transducer was calibrated by use of a precision volume syringe (3-L syringe). All measurements derived during RIP were based on the assumption that the external flow signal (ie, sum flow) equals the nasal flow signal. Because thoracic, abdominal, and sum signals are collected in an uncalibrated fashion, the gain setting for sum flow was chosen to closely match the volume signal derived from the pneumotachograph through integration (ie, volume = \int \text{flow}). External flow (ie, sum flow) and nasal flow signals were subtracted in the same time domain to obtain differences in PIP and PIF. Similarly, area differences between these signals during the first 25% of exhaled volume and first 25% of inhaled volume were computed on a breath-by-breath basis. The peak flow and area differences represent the severity of gas compression (during expiration) and expansion (ie, rarefaction [during inspiration]).

**R** and C<sub>resp</sub> measurements—Values of R and C<sub>resp</sub> were derived via the previously described method of covariance ratios and from measurements of flow, V<sub>i</sub>, and transpulmonary pressure as previously described during spontaneous breathing in 10 llamas. An esophageal balloon (10 cm in length and 3.8 cm in diameter) sealed over the distal end of a polypropylene catheter (4 mm internal diameter, 5 mm outer diameter, and 90 cm length) was passed through the nares to the midthoracic level of the esophagus and inflated with 2 mL of air. Adequate catheter placement was verified by the identification of dPPl<sup>ref</sup>. The proximal end of the esophageal balloon catheter was exited through a hole within the proximal aspect of the face mask and connected to a differential pressure transducer. Transpulmonary pressures were estimated by measuring the differences between face mask and esophageal pressures during spontaneous breathing. The nosepiece of the face mask was affixed to a heated pneumotachograph. Flow was thus detected during tidal breathing and electronically integrated to determine V<sub>i</sub> on a breath-by-breath basis. The analog signals derived from the pressure transducer and pneumotachograph were collected at 30 Hz, amplified, and digitized by use of pulmonary mechanics analysis software. Measurements of flow, V<sub>i</sub>, expired minute volume, frequency, PIF, PEF, inspiratory time, expiratory time, and dPPl<sub>max</sub> were recorded and displayed continuously. Flow, V<sub>i</sub>, and transpulmonary pressure were used to calculate R<sub>i</sub> by use of the isovolume method and C<sub>sp</sub> at 2 points of zero flow as described by AMDUR and MEAD.

**Monofrequency FOT**—Monofrequency FOT (at 1 to 7 Hz) was used to determine respiratory system impedance R<sub>osc</sub> and respiratory system reactance, as well as f<sub>m</sub>, as previously described. The R<sub>osc</sub> is comprised of the additive effects of chest wall resistance and R<sub>i</sub>.

Measurements obtained via FOT were conducted with the already described face mask and esophageal balloon catheter, but a different size pneumotachograph. The nosepiece of the face mask was affixed to the pneumotachograph, with a more rostral T-piece containing 4 ports as follows: a small port for externally derived oscillating airflow (waveform sinusoidal, 1 to 7 Hz frequencies); a large side port for spontaneous breathing through a resistive element (2 cm H<sub>2</sub>O/L/s); a large port for the pneumotachograph; and, a lateral port (4 mm internal diameter) for measurement of airway opening pressure by use of a differential pressure transducer.

A sinusoidal airflow of desired frequency was generated by use of a 3-port proportional pneumatic valve connected to a compressed air source (75 psi of pressure). Face mask pressure relative to atmospheric pressure (for measurement of respiratory system impedance) or relative to esophageal (pleural) pressure (for measurement of lung impedance) were determined by use of a differential pressure transducer. Airflow to the face mask was measured by use of a pneumotachograph and differential pressure transducer. Amplified pressure and flow signals were digitized at 25.6 Hz for 10 seconds. Signals were bandpass filtered (0.4-Hz-wide passband centered at the measurement frequency and 80-dB stopband attenuation) and divided into consecutive 5-second data segments with 50% overlap, from which resistance, reactance, and impedance were calculated by use of a personal computer, purpose-built controller, and digital processing system. A coherence value was calculated to provide an indication of signal-to-noise ratio and linearity of the system. Only values with a coherence ≥ 0.9 were used for analysis.

The Z<sub>bc</sub> was calculated on the basis of the magnitude and phase shifts between pressure and flow sinusoidal waves at a given time and frequency as follows:

\[ Z_{bc}(\omega) = \frac{P_{bc}(\omega)}{V_{ao}(\omega)} \times |\text{cos} \phi| \]

where \( \phi \) represents the phase shift between flow and pressure waves, P<sub>bc</sub> is pressure at the airway opening, V<sub>ao</sub> is flow at the airway opening, \( \omega \) represents angular frequency in radians, and \( \text{cos} \) represents the cosine function. The phase angle was used via computer analysis software to calculate the components of respiratory system or pulmonary impedance, resistance, and reactance. Respiratory system reactance and reactance (X<sub>bc</sub>) were computed as follows:

\[ R_{bc} = |Z_{bc}| \times \cos \phi \]
\[ X_{bc} = |Z_{bc}| \times \sin \phi \]

The forced oscillation system was calibrated each testing day by use of the wave tube principle. An open-ended polyvinyl chloride pipe (6.2 m long, 52.3 mm internal diameter) with a known impedance (Z<sub>0</sub>) was compared with the measured impedance by use of the forced oscillatory system. A correction factor (k) was derived from this comparison (k = Z<sub>bc</sub>/Z<sub>0</sub>) and used to correct the measured impedance (corrected impedance = kZ<sub>bc</sub>) of llamas.

The f<sub>m</sub> was defined as the impulse frequency at which impedance measurements were entirely comp
prised of resistance (ie, reactance was equal to zero). By use of the components of reactance, inerance (I) and compliance (C), $f_{res}$ can be calculated as follows:

$$f_{res} = 1/(2\pi|IC|^{3/2})$$

**Statistical analysis**—Descriptive data analyses of respiratory mechanical parameters (FRC, $V_{E}$, inspiratory time, expiratory time, expired minute volume, PIF, PEF, $R_{L}$, $C_{dyn}$, dPPl$_{max}$, $R_{osc}$, $f_{res}$, and RIP-derived variables) were reported as mean ± SD. The correlation between FRC and body dimensions (weight, height, length, and circumference) as well as between measurements obtained via FOT and esophageal balloon mechanics was evaluated by use of the Spearman rank correlation coefficient ($p$). The Bland-Altman test of agreement was applied to test agreement between measurements of $C_{dyn}$ and $R_{L}$ obtained by use of FOT and esophageal balloon mechanics. These results were reported for $C_{syn}$ and $R_{L}$ as mean difference between the 2 techniques with the 95% confidence interval. Outlier tests were detected via extreme values analysis. The effect of sedation was evaluated by use of a paired samples $t$ test, with an accepted significance level of $P < 0.05$. All analyses were performed by use of commercial software.

**Results**

**Physical characteristics of llamas and hematologic results**—Measurements of weight (150 ± 40.3 kg), height at the withers (113.9 ± 12.1 cm), body length from point of the shoulder to ischial tuberosity (108.7 ± 12.6 cm), and chest circumference (135.5 ± 18.6 cm) were within the reference range for healthy adult llamas. Similarly, hematologic analysis revealed a mean total WBC count of 12.76 X 10³ cells/µL (range, 9.5 X 10³ cells/µL to 19.4 X 10³ cells/µL) and mean Hct of 26% (range, 24% to 34%), values which were within the reference range for healthy adult llamas.

Thoracic radiography did not reveal any changes consistent with respiratory disease in the study llamas.

**Pulmonary mechanics in nonsedated llamas**—Statistical analysis revealed a normal distribution for all data for nonsedated and sedated llamas. Baseline data for ventilatory parameters before sedation in 10 llamas were determined (Table 1). Mean FRC in nonsedated llamas was 5.60 ± 1.24 L, which translated to 39.18 ± 11.45 mL/kg for this group. A positive correlation was observed between FRC and body length in our study llamas ($P = 0.016; p = 0.733$). However, FRC values were not significantly correlated with body weight, height, or chest circumference. Breathing patterns were assessed by RIP (Figure 1). Peak differences between RIP and pneumotachographic flows were 2.48 ± 1.04 L/s during inspiration (ie, differences in PIF), and 2.61 ± 1.02 L/s during expiration (ie, differences in PEF). When flow was integrated over the first 25% of the inspired or expired breath, the derived RIP and pneumotachographic volume differences were 0.05 ± 0.04 L and 0.04 ± 0.14 L, respectively.

Measured parameters of pulmonary mechanics ($R_{L}$, $C_{dyn}$, dPPl$_{max}$, $R_{osc}$, and $R_{osc}$) were determined (Table 2). Values of $R_{L}$ (conventional $R_{L}$ and $R_{osc}$) were significantly correlated ($P = 0.025; p = 0.733$) at 1 Hz (the input frequency closest to the spontaneous breathing frequency). The Bland-Altman test of agreement between

<table>
<thead>
<tr>
<th>Variable</th>
<th>Nonsedated llamas</th>
</tr>
</thead>
<tbody>
<tr>
<td>$f$ (breaths/min)</td>
<td>42.38 ± 12.12</td>
</tr>
<tr>
<td>$V_{E}$ (L/s)</td>
<td>1.03 ± 0.3</td>
</tr>
<tr>
<td>PIF (L/s)</td>
<td>2.14 ± 0.65</td>
</tr>
<tr>
<td>PEF (L/s)</td>
<td>2.01 ± 0.72</td>
</tr>
<tr>
<td>PEF:PIF</td>
<td>0.95 ± 0.24</td>
</tr>
<tr>
<td>$Te$ (s)</td>
<td>0.89 ± 0.36</td>
</tr>
<tr>
<td>$Ti$ (s)</td>
<td>0.67 ± 0.15</td>
</tr>
<tr>
<td>$Te$:$Ti$</td>
<td>1.31 ± 0.4</td>
</tr>
<tr>
<td>$V_{E}$ (L/min)</td>
<td>40.84 ± 12.95</td>
</tr>
</tbody>
</table>

$P$ = Respiratory frequency. $Te$ = Inspiratory time. $Te$ = Expiratory time. $V_{E}$ = Minute ventilation.

**Figure 1**—The RIP and pneumotachographic signals. A—Diagram depicting combined matching nasal flow ($V’_{n}$; blue line) and sum flow ($V’_{sum}$; red line) signals derived from elastic band measurements (sum flow = abdominal + thoracic components) in a healthy llama. The beginning of expiration (E) and beginning of inspiration (I) were defined by the upward- and downward-directed zero crossings (over horizontal double arrowheads) of sum flow signals, respectively. B—Thoracic (RIB) versus abdominal (ABD) contribution to breathing in a healthy llama. Both signals are completely in phase, and the llama has a much larger abdominal relative to thoracic contribution to ventilation.
between Rₛ and R₁ osc revealed a mean difference of 0.83 ± 0.69 cm H₂O/L/s with a 95% confidence interval of 0.3 to 1.35 cm H₂O/L/s. The Cdyn obtained via conventional pulmonary mechanics was compared with an index of Cdyn calculated from X₁ osc values at 1 Hz (X₁ osc values at 1 Hz, −0.54 ± 0.39; n = 9) as previously described.¹³ These compliance values (ie, Cdyn and Cdyn osc) were positively correlated (P = 0.03; p = 0.667). However, mean compliance calculated from X₁ osc (Cdyn osc; 0.32 ± 0.43 L/cm H₂O) was significantly (P = 0.05) lower than mean Cdyn measured via conventional pulmonary mechanics. The Bland-Altman test of agreement between the calculated compliance and measured Cdyn revealed a mean difference of 0.43 ± 0.22 L/cm H₂O with a 95% confidence interval of 0.26 to 0.60 L/cm H₂O.

Pulmonary mechanics in sedated llamas—The mean FRC in sedated llamas (n = 5) was 4.17 ± 0.60 L with a calculated FRC/kg of 30.35 ± 10.56 mL/kg. This represents a significant (P = 0.02) mean decrease in FRC of 0.83 ± 0.23 L (17%) following sedation with xylazine. Differences in PEF (n = 6) increased by 76% following sedation, with a mean difference in PEF of nonsedated llamas of 2.61 ± 1.02 L/s versus differences in PEF of sedated llamas of 4.99 ± 2.8 L/s. Differences in PIF values (n = 6) increased by 34% following sedation, with a mean differences in PIF of nonsedated llamas of 2.47 ± 1.04 L/s versus differences in PIF of sedated llamas of 3.74 ± 1.69 L/s.

Effects of sedation with xylazine on ventilatory parameters and tidal breathing mechanics were determined (Tables 3 and 4). Although measurements obtained via FOT and conventional pulmonary mechanics were attempted in 7 sedated llamas for which owner consent was available, results were excluded for 1 llama that did not remain standing following sedation with xylazine and 1 llama that developed epistaxis resulting from passage of the naso-esophageal balloon during conventional pulmonary mechanics. Data were also excluded if acceptable test coherence (ie, signal-to-noise ratio) was not met during the performance associated with the llama’s normal respiratory frequency, glottic closure, cardiogenic oscillations, or abnormal signal transduction.

A significant increase was found in dPPl asc (P = 0.03) and R₁ (P = 0.015) in sedated llamas. Although Table 2—Mean ± SD respiratory function variables obtained via conventional pulmonary mechanics and FOT in nonsedated adult llamas.

<table>
<thead>
<tr>
<th>Method</th>
<th>Variable</th>
<th>Oscillation frequency (Hz)</th>
<th>No. of llamas</th>
<th>Non-sedated llamas</th>
<th>Sedated llamas</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Conventional*</td>
<td>dPPl asc (cm H₂O)</td>
<td>NA</td>
<td>10</td>
<td>6.55 ± 2.06</td>
<td>8.80 ± 2.79</td>
<td>0.004</td>
</tr>
<tr>
<td></td>
<td>R₁ (cm H₂O/L/s)</td>
<td>NA</td>
<td>10</td>
<td>1.42 ± 0.54</td>
<td>3.18 ± 1.65</td>
<td>0.015</td>
</tr>
<tr>
<td></td>
<td>Cdyn (L/cm H₂O)</td>
<td>NA</td>
<td>10</td>
<td>0.83 ± 0.40</td>
<td>0.62 ± 0.15</td>
<td>0.119</td>
</tr>
<tr>
<td>FOT†</td>
<td>R₁ osc (cm H₂O/L/s)</td>
<td>1</td>
<td>9</td>
<td>2.24 ± 0.90</td>
<td>3.93 ± 2.45</td>
<td>0.021</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2</td>
<td>10</td>
<td>2.08 ± 0.72</td>
<td>4.34 ± 1.91</td>
<td>0.005</td>
</tr>
<tr>
<td></td>
<td></td>
<td>3</td>
<td>10</td>
<td>2.13 ± 0.61</td>
<td>3.99 ± 2.18</td>
<td>0.034</td>
</tr>
<tr>
<td></td>
<td></td>
<td>5</td>
<td>10</td>
<td>2.33 ± 0.68</td>
<td>3.76 ± 1.49</td>
<td>0.009</td>
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<tr>
<td></td>
<td></td>
<td>7</td>
<td>10</td>
<td>2.56 ± 0.39</td>
<td>3.78 ± 1.11</td>
<td>0.023</td>
</tr>
<tr>
<td>FOT†</td>
<td>R₁ osc (cm H₂O/L/s)</td>
<td>1</td>
<td>5</td>
<td>2.14 ± 0.84</td>
<td>5.14 ± 2.91</td>
<td>0.112</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2</td>
<td>7</td>
<td>2.14 ± 0.61</td>
<td>5.39 ± 2.86</td>
<td>0.022</td>
</tr>
<tr>
<td></td>
<td></td>
<td>3</td>
<td>7</td>
<td>2.10 ± 0.64</td>
<td>4.41 ± 1.85</td>
<td>0.015</td>
</tr>
<tr>
<td></td>
<td></td>
<td>5</td>
<td>7</td>
<td>2.55 ± 0.58</td>
<td>4.06 ± 1.28</td>
<td>0.019</td>
</tr>
<tr>
<td></td>
<td></td>
<td>7</td>
<td>7</td>
<td>2.51 ± 0.54</td>
<td>4.03 ± 1.02</td>
<td>0.019</td>
</tr>
</tbody>
</table>

*Use of esophageal balloon and pneumotachography. †Only values with a coherence ≥ 0.9 were used for analysis via FOT. NA = Not applicable.

Table 3—Mean ± SD lung mechanic values of adult llamas (n = 7) before (nonsedated) and after (sedated) IM administration of xylazine.

<table>
<thead>
<tr>
<th>Llamas</th>
<th>Variable</th>
<th>Nonsedated</th>
<th>Sedated</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>f (breaths/min)</td>
<td>41.32 ± 12.9</td>
<td>17.67 ± 6.05</td>
<td>0.004</td>
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<tr>
<td></td>
<td>Vt (L)</td>
<td>1.06 ± 0.19</td>
<td>1.45 ± 0.2</td>
<td>0.002</td>
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<tr>
<td></td>
<td>PIF (L/s)</td>
<td>2.24 ± 0.56</td>
<td>1.32 ± 0.48</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td></td>
<td>PEF (L/s)</td>
<td>2.17 ± 0.84</td>
<td>1.27 ± 0.5</td>
<td>0.019</td>
</tr>
<tr>
<td></td>
<td>PEF:PIF</td>
<td>0.95 ± 0.27</td>
<td>0.97 ± 0.27</td>
<td>0.917</td>
</tr>
<tr>
<td></td>
<td>Te (s)</td>
<td>0.96 ± 0.45</td>
<td>2.08 ± 0.48</td>
<td>0.004</td>
</tr>
<tr>
<td></td>
<td>Ti (s)</td>
<td>0.66 ± 0.14</td>
<td>1.57 ± 0.38</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>Te/TT</td>
<td>1.39 ± 0.49</td>
<td>1.35 ± 0.13</td>
<td>0.054</td>
</tr>
<tr>
<td></td>
<td>Vt (L/min)</td>
<td>43.95 ± 14.42</td>
<td>24.62 ± 9.31</td>
<td>0.012</td>
</tr>
</tbody>
</table>

See Table 1 for key.

Table 4—Mean ± SD respiratory function variables obtained via conventional pulmonary mechanics and FOT in adult llamas before (nonsedated) and after (sedated) IM administration of xylazine.

<table>
<thead>
<tr>
<th>Llamas</th>
<th>Method</th>
<th>Variable</th>
<th>Oscillation frequency (Hz)</th>
<th>No.</th>
<th>Nonsedated</th>
<th>Sedated</th>
<th>Change (%)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Conventional*</td>
<td>dPPl asc (cm H₂O)</td>
<td>NA</td>
<td>6</td>
<td>6.34 ± 1.24</td>
<td>8.80 ± 2.79</td>
<td>46</td>
<td>0.030</td>
</tr>
<tr>
<td></td>
<td></td>
<td>R₂ (cm H₂O/L/s)</td>
<td>NA</td>
<td>6</td>
<td>1.29 ± 0.5</td>
<td>3.18 ± 1.65</td>
<td>147</td>
<td>0.015</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Cdyn (L/cm H₂O)</td>
<td>NA</td>
<td>5</td>
<td>0.96 ± 0.44</td>
<td>0.62 ± 0.15</td>
<td>-33</td>
<td>0.119</td>
</tr>
<tr>
<td>FOT†</td>
<td>R₂ osc (cm H₂O/L/s)</td>
<td>1</td>
<td>6</td>
<td>2.04 ± 0.97</td>
<td>5.39 ± 2.45</td>
<td>175</td>
<td>0.021</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>2</td>
<td>7</td>
<td>1.92 ± 0.69</td>
<td>4.34 ± 1.91</td>
<td>130</td>
<td>0.005</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>3</td>
<td>7</td>
<td>1.95 ± 0.54</td>
<td>3.99 ± 2.18</td>
<td>176</td>
<td>0.034</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>5</td>
<td>7</td>
<td>2.06 ± 0.51</td>
<td>3.76 ± 1.49</td>
<td>80</td>
<td>0.009</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>7</td>
<td>7</td>
<td>2.57 ± 0.43</td>
<td>3.78 ± 1.11</td>
<td>53</td>
<td>0.023</td>
</tr>
<tr>
<td>FOT†</td>
<td>R₂ osc (cm H₂O/L/s)</td>
<td>1</td>
<td>5</td>
<td>2.14 ± 0.84</td>
<td>5.14 ± 2.91</td>
<td>181</td>
<td>0.112</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td>2</td>
<td>7</td>
<td>2.14 ± 0.61</td>
<td>5.39 ± 2.86</td>
<td>161</td>
<td>0.022</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>3</td>
<td>7</td>
<td>2.10 ± 0.64</td>
<td>4.41 ± 1.85</td>
<td>118</td>
<td>0.015</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>5</td>
<td>7</td>
<td>2.55 ± 0.58</td>
<td>4.06 ± 1.28</td>
<td>64</td>
<td>0.019</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>7</td>
<td>7</td>
<td>2.51 ± 0.54</td>
<td>4.03 ± 1.02</td>
<td>55</td>
<td>0.019</td>
</tr>
</tbody>
</table>

See Table 2 for key.
Figure 2—Box plots of X_{Losc} versus oscillatory frequency in xylazine-sedated (white boxes) and nonsedated (gray boxes) llamas. Box represents the 25th to 75th percentile, horizontal line represents the median, and whiskers represent the range.

Discussion

To our knowledge, this study presents for the first time the application of respiratory mechanical function tests in adult llamas. Some of the respiratory function tests discussed here have been used in other domestic species, results of which warrant comparison where possible. The ease with which respiratory function tests were performed in conscious llamas may provide an opportunity to use these tests as ancillary diagnostic tools. However, the significant alteration caused by sedation with xylazine may prohibit the use of α_{1}-adrenergic agonists for this purpose.

The mean FRC per kilogram of body weight in llamas from our study (40 mL/kg; 5.60 ± 1.24 L) falls within the range of values obtained in horses^{11,12,20} (44 mL/kg), cattle^{25} (39 mL/kg), dogs^{10,27} (34 to 98 mL/kg), and sheep^{28-30} (25 to 52 mL/kg). Numerous factors influence FRC including abdominal fill, thoracic wall versus lung compliance, thorax size, active component of expiration, posture, and position^{10,30}. In this study, a positive correlation (P = 0.016) between body length and FRC was observed but not between FRC and body weight or circumference. Functional residual capacity has previously been positively correlated to body weight in horses^{13}. Earlier studies evaluating FRC in dogs^{25} and sheep^{29} found positive correlations between FRC and body weight, circumference, and length. Variation in body condition may explain some of the failure of allometric scaling on the basis of body weight and shape within species. The most extreme example of how body weight fails to predict FRC is in dogs, where body shape varies tremendously with breed^{25,10,27}.

Changes in FRC may serve as an index of pulmonary mechanical dysfunction. For example, loss of lung volume caused by pneumonia or space occupying masses in the chest, or decreased lung compliance secondary to pulmonary fibrosis, may all reduce FRC. In our study we demonstrate that FRC is obtained easily in conscious llamas.

Llamas have a greater abdominal (ie, diaphragmatic and abdominal muscle) than rib cage (ie, intercostal muscle) contribution to ventilation at rest, as evidenced by their larger abdominal signal, compared with thoracic signal. These findings are in contrast to those for humans and healthy horses. In humans, the contribution of rib cage motion predominates over abdominal movement during quiet breathing^{15}. Healthy horses have a relatively equal thoracic to abdominal contribution to ventilation at rest^{10}. This discrepancy may be explained by the relatively smaller chest size and possibly greater passive motion of the llama rib cage during respiration, in comparison with the adult horse^{10}. Abdominal and thoracic signals were synchronous during inspiration and expiration in all our study llamas, as expected for healthy llamas. Previous publications in veterinary medicine in which RIP was used include the measurement of breathing pattern during exercise^{32}, recurrent airway obstruction (heaves) and histamine challenge^{8}, proximal and distal airway obstructions evoked in foals^{13}, and diaphragmatic paralysis in a young llama^{8}. Results of our study expand our knowledge about the normal breathing pattern in llamas, which shows a remarkable dominance by the abdominal compartment. This may suggest that quiet breathing is dominated by the diaphragm, but does not imply that this relationship will persist during periods of increased ventilatory demand. Future studies are warranted to examine the llama breathing pattern under conditions of resistive load and hyperpnea.

In our study, mean V_{E} in llamas was 7 mL/kg (1.03 L). This is less than the mean V_{E} measured in horses (12 mL/kg)^{25,31,30} and dogs (12 mL/kg),^{3} but falls within the range observed in domestic ruminants such as cattle (8 mL/kg)^{23,34} and sheep (6 to 7 mL/kg).^{28} Variation in V_{E} across species is related to absolute differences in lung volume as well as other anatomic features such as size and shape of the abdomen and relative abdominal mass^{35} and metabolic demand. For example, although similar in body size to the horse, cattle have a smaller lung volume and V_{E} relative to body size^{25}. Although total lung volume was not measured in our study, lla-
mas share certain anatomic similarities to ruminants because of their large abdominal compartment relative to thoracic compartment. Therefore, it could be speculated that $V_r$ relative to body size in llamas would be more similar to ruminants than monogastrics. Measurements of $R_\text{res}$ and $C_{dyn}$ in llamas are comparable to values observed in other species once corrections for differences in body size, lung volume, and breathing patterns have been made. Values of $R_\text{res}$ in llamas from our study are greater than those found previously in horses (1.16 ± 0.65 cm H$_2$O/L/s) and less than values recorded in sheep (2.19 cm H$_2$O/L/s). The $R_\text{res}$ is expected to decrease with increasing body size as a result of an overall increase in airway diameter, and comparisons of specific $R_\text{res}$ (R$_\text{max}$ and FRC) among llamas, horses, and sheep are similar.

In contrast, mean $C_{dyn}$ in llamas is less than mean values recorded in horses (1.76 ± 0.33 L/cm H$_2$O), and calculated specific $C_{dyn}$ ($C_{dyn}$ and FRC) is also lower than in horses. As $C_{dyn}$ is related to lung volume, it should scale directly to body size. However, it is also determined by chest wall compliance. Results of a previous study comparing cattle and horses of similar body size and breathing patterns have been made. Values of $R_\text{res}$ in llamas from our study are greater than those found previously in horses (1.16 ± 0.65 cm H$_2$O/L/s) and less than values recorded in sheep (2.19 cm H$_2$O/L/s). The $R_\text{res}$ is expected to decrease with increasing body size as a result of an overall increase in airway diameter, and comparisons of specific $R_\text{res}$ (R$_\text{max}$ and FRC) among llamas, horses, and sheep are similar.

A significant correlation ($\rho = 0.025; \rho = 0.73$) between $R_\text{res}$ measured by noninvasive FOT (ie, R$_\text{osc}$) and the conventional, isovolume method was observed, as expected. However, the mean R$_\text{res}$ at 1 Hz (2.0 cm H$_2$O/L/s) was higher than the ‘llamas’ conventional R$_\text{res}$ (1.4 cm H$_2$O/L/s), measured during spontaneous breathing frequencies (mean, 0.7 Hz). Although these results complement a report comparing R$_\text{osc}$ to R$_\text{res}$ in horses, this observation stands in general contrast to expected higher resistance values obtained via conventional techniques because R$_\text{res}$ tends to increase at lower frequencies. Measurements of $C_{dyn}$osc and conventional $C_{dyn}$ were also significantly correlated ($P = 0.03; \rho = 0.67$). However, values obtained via FOT were lower, compared with conventional measurements, with a mean difference of 0.43 ± 0.22 L/cm H$_2$O (95% confidence interval, 0.26 to 0.6 L/cm H$_2$O). A similar close correlation but disparity in absolute agreement between forced oscillation and conventional techniques has been previously reported in horses and calves and attributed to biological factors, including frequency dependence of compliance. Values of $C_{dyn}$osc in the study reported here were obtained at 1 Hz, which is higher than the spontaneous breathing frequencies of our llamas, and might partially account for the differences between $C_{dyn}$osc and conventional $C_{dyn}$ measurements. Additionally, a decrease in $C_{dyn}$osc and increase in $R_\text{res}$ may reflect presence of airway obstruction and consequent unequal time constants in the lung. Although all llamas tolerated the pulmonary function procedures well, they were not accustomed to FOT and may have experienced glottic narrowing in response to airflow oscillation, similar to reports in humans and horses.

Llamas had a higher $f_m$ (3.37 ± 0.68 Hz) relative to values reported in the horse (2.46 ± 0.02 Hz), and lower mean $f_m$, compared with values recorded in calves (5 to 6 Hz). Beagles (6.17 ± 0.5 Hz), and humans (6 to 11 Hz). This is expected as a result of differences in body size where $f_m$ increases, with decreases in body size similar to increases in respiratory resistance. Results of numerous studies have previously revealed that $f_m$ increases in response to bronchoconstrictive respiratory diseases such as human chronic obstructive pulmonary disease and asthma and equine heaves and is related to higher airway resistance.

Sedation with xylazine significantly altered ventilatory parameters in llamas. Respiratory frequency and minute ventilation decreased, in conjunction with increased $V_r$, a reduction in peak flows (ie, PIF and PEF), and prolongation of inspiratory and expiratory times, as found in horses. Measurements obtained via conventional pulmonary mechanics further revealed significant increases in $R_\text{res}$ ($P = 0.013$) and dPPl peak ($P = 0.02$), which was corroborated via FOT. The $C_{dyn}$ decreased in sedated llamas, although this change was not significant. The effect of xylazine on respiratory mechanical function has been previously evaluated in studies on horses and calves. Potential adverse effects of sedation with xylazine include increased upper airway resistance, decreased respiratory rate and minute volume, increased $V_r$, bronchodilation or bronchoconstriction, and increased work of breathing. In several earlier studies, horses had increased upper airway resistance, primarily caused by nasal edema developing secondary to vascular congestion in the nares when the head was allowed to drop during sedation. Many of the adverse effects of xylazine have since been attenuated by controlling head and neck position and administering the lowest possible dose for sedation. Because of the anticipated potential for nasal edema in llamas, similar precautions were performed in our study.

Sheep appear to have a response to xylazine comparable to that of llamas, including evidence of bronchoconstriction, increased dPPl peak, decreased $C_{dyn}$, and increased $R_\text{osc}$; similarly, in xylazine-sedated calves, analysis via FOT revealed a higher $f_m$ and R$_\text{osc}$, compared with controls, with R$_\text{osc}$ demonstrating negative frequency dependence (ie, R$_{osc}$ was highest at lower frequencies) as a result of increased peripheral airway resistance. These reported results in ruminant species are qualitatively the same as our findings in llamas, where airway narrowing (increased resistance) was evidenced by significantly increased R$_{osc}$, nega-
tive frequency dependence of $R_{osc}$, and increased $f_{res}$ in sedated llamas for which calculation was possible ($n = 3$ for $f_{res}$). The 4 llamas for which $f_{res}$ could not be calculated under sedation had higher $R_{osc}$ and $R_{osc}$ values, compared with the other llamas. Results of a previous study\(^{10}\) evaluating FOT in swine and calves revealed similar difficulties in calculating $f_{res}$ that were attributed to poor model fit secondary to shifting from increased face mask dead space and interference by soft tissue structures of the nasopharynx. Thus it is possible that in several of our sedated llamas, the degree of increased $R_{osc}$ impeded measurement of accurate impedance values (and thus $f_{res}$). The magnitude of the observed airway closure in healthy sedated llamas strongly suggests that sedation with xylazine is contraindicated during respiratory function testing in llamas. On the basis of the excellent cooperation of untrained nonsedated llamas in our study, sedation should not be necessary, assuming proper chute restraint is available. Individual case reports have also reported good test compliance in sick llamas,\(^{9}\) although further experience needs to be gained under clinical conditions.

The respiratory mechanical function of llamas in our study was comparable to other animal species, and observed differences may be attributed to variations in body size or conformation. However, further evaluation of anatomic adaptations to a dry, high-altitude environment (ie, increased minute ventilation, narrowing of the nasal passage, and long neck) based on the native origin of camelids should be considered. In addition, a concise understanding of the gross and histologic lung anatomy of llamas and how this may influence the type and distribution of lung disease is important. Clear anatomic differences in lung anatomy exist between species. Although the gross lung anatomy of llamas is similar to horses,\(^{39}\) a description of the histologic anatomy is currently not available. Grossly, equine lungs have simple lobation, compared with the well-developed lobe divisions found in cattle, sheep, pigs, and dogs. Furthermore, cattle, pigs, and sheep have well-developed lung lobulation, which is incomplete in horses and nonexistent in dogs.\(^{36}\) Because cattle and pigs lack collateral ventilation, lung disease, atelectasis, and airway obstruction generally localize to lobules. In horses with incomplete lobulation and dogs with absent lobulation, collateral ventilation is more developed and lung disease more diffusely distributed.\(^{50}\)

In conclusion, noninvasive pulmonary function tests (ie, FOT and RIP) were well tolerated in untrained nonsedated llamas and validated through comparison to conventional (gold standard) techniques. Noninvasive pulmonary function testing has extensive clinical applications in the diagnosis and treatment of respiratory disease in llamas, although testing should not be performed with the use of sedation with xylazine to avoid bronchoconstriction.

References


