MRI study of regional variations of pharyngeal wall compliance in cats

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1Center for Sleep and Respiratory Neurobiology, University of Pennsylvania Medical Center,
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Brennick, Michael J., Malcolm D. Ogilvie, Susan S. Margulies, Luke Hiller, Warren B. Gefter, and Allan I. Pack. MRI study of regional variations of pharyngeal wall compliance in cats. J. Appl. Physiol. 85(5): 1884–1897, 1998.—Upper airway compliance indicates the potential of the airway to collapse and is relevant to the pathogenesis of obstructive sleep apnea. We hypothesized that compliance would vary over the rostral-to-caudal extent of the pharyngeal airway. In a paralyzed isolated upper airway preparation in cats, we controlled static upper airway pressure during magnetic resonance imaging (MRI, 0.391-mm resolution). We measured cross-sectional area and anteroposterior and lateral dimensions from three-dimensional reconstructed MRIs in axial slices orthogonal to the airway centerline. High-retropalatal (HRP), midretropalatal (MRP), and hypopharyngeal (HYP) regions were defined. Regional compliance was significantly increased from rostral to caudal regions as follows: HRP < MRP < HYP (P < 0.0001), and compliance differences among regions were directly related to collapsibility. Thus our findings in the isolated upper airway of the cat support the hypothesis that regional differences in pharyngeal compliance exist and suggest that baseline regional variations in compliance and collapsibility may be an important factor in the pathogenesis and treatment of obstructive sleep apnea.

Upper airway; obstructive sleep apnea; magnetic resonance imaging; collapsibility

Compliance is an indicator of the ease with which the airway can be deformed and is usually expressed as the change in volume or airway cross-sectional area (CSA) per unit change in pressure. Compliance measurements of the upper airway are likely to be an important indicator of the potential of the airway to collapse and, thus, are relevant to the pathogenesis of pharyngeal collapse in obstructive sleep apnea (OSA) syndrome. Because compliance relates directly to the biomechanical tissue properties of the airway, a quantitative examination of compliance will offer insights that relate to maintenance of upper airway patency.

Several studies in humans, in particular in patients with sleep apnea, have examined “compliance” using methods such as acoustic reflection techniques (4) and imaging approaches such as X-ray fluoroscopy (34) or computed tomography (CT) (13). Kuna et al. (13) showed regional differences along the upper airway in compliance under continuous positive airway pressure (CPAP) in human subjects. However, measurements of upper airway compliance in human subjects undergoing voluntary glottis closure or other maneuvers are difficult to interpret, because the contribution of upper airway muscle activation to the measured compliance is undefined. Muscle activation not only varies throughout the respiratory cycle (2), but upper airway dilator muscle activity has been shown to increase because of mechanoreflexes when intraluminal pressure is negative (30) and to decrease or be abolished at positive intraluminal pressure (1). Other investigators (8–10, 14, 17) who performed studies under passive conditions using transnasal endoscopy have shown differences in the pressure-area relationship in the pharynx between normal subjects and OSA patients. These studies, however, using two-dimensional endoscopy, measured the pressure-area relationship at specific loci of narrowing or collapse in the pharynx.

To examine the passive biomechanical tissue characteristics throughout the pharynx, we used noninvasive magnetic resonance imaging (MRI) to measure the compliance of the isolated upper airway in an animal model where active muscle tone was eliminated by paralysis. Our first objective was to provide the quantitative three-dimensional (3D) analyses that would relate upper airway CSA to specific pressures, so that the compliance curve for the upper airway could be determined. We examined the effect of our interventions (tracheostomy and paralysis) on resting (zero-pressure) airway size. We also examined the effect of muscle tone by comparing the compliance measurements taken during the anesthetized-only (nonparalyzed) and paralyzed conditions. Our second objective was to determine the regional variations in pharyngeal compliance. To accomplish these objectives, pressure was controlled in the isolated upper airway segment while MRI of the pharyngeal airway was performed. The pharyngeal airway was divided into three regions: the high-retropalatal (HRP), the midretropalatal (MRP), and the hypopharyngeal (HYP) region. Thus we used computerized methods to analyze MRIs and to measure the size of the airway lumen in each region at each of a number of controlled positive or negative pressures to determine regional compliance of the pharyngeal airway and variations, if any, that may exist over the rostral-to-caudal extent of the airway.

METHODS AND MATERIALS

Overall protocol. We studied six cats (2.3–3.5 kg) of either gender. The experimental protocol was divided into three general procedures. First, we performed baseline MRI of the intact upper airway while the cat inhaled vapor anesthesia [1.5% (vol/vol) isoflurane in pure O2] through a tight-fitting...
mask. Vapor anesthesia was maintained throughout the experiment through mask breathing or through a tracheostomy tube (see Isolated upper airway surgical preparation). After imaging of the intact upper airway, the cat was taken out of the magnet and underwent a surgical procedure to create an isolated upper airway. After surgery a second series of MRls of the upper airway was performed at specific controlled pressure levels in the isolated upper airway. Finally, after neuromuscular blockade the pressure-controlled isolated upper airway MRI protocol was repeated in the paralyzed isolated upper airway. Thus we were able to measure the airway intraluminal CSA-pressure relationship for discrete regions in the isolated pharyngeal airway under nonparalyzed and paralyzed conditions and compare these changes with baseline CSA in the intact (spontaneously breathing) cat. (In the spontaneously breathing cat we did not apply controlled levels of pressure, and thus compliance was not measured.) After all testing, in each cat, euthanasia was performed, under direct observation, by barbiturate overdose (pentobarbital sodium, 300 mg/kg iv).

General imaging approach. High-resolution MRI was performed in a 4.7-T, 40-cm-diameter magnet interfaced to a General Electric Signa version 4.7 computer and console. Spin-echo images [T1 weighted, repetition-time/echo-time ratio (TR/TE) = 500/20, with 3 excitations per view] were obtained. A quadrature radio-frequency volume coil, built on polyvinyl chloride tubing with a 10-mm annular space, was employed. This coil was specially designed to fit snugly over the cat's head and was tuned to the 200-MHz resonant proton frequency.

Imaging (before and after surgery) was performed with the cat placed in the prone position. A reproducible head-to-neck angle of 135 ± 2° was provided by a wedge-shaped positioning block, which was hollowed in the center to avoid pressure on the submental and anterior midcervical region and supported the mandible from the temporal mandibular joint to the symphysis. We measured the head-to-neck angle from the sagittal images before and after surgery and then after paralysis in each cat.

Imaging of intact upper airway. The cats were preanesthetized using ketamine (15–20 mg/kg im), with diazepam (2 mg/kg im) administered for muscle relaxation and atropine (0.05 mg/kg im) to reduce airway secretions. Before place-ment of a mask, to ensure unobstructed airflow through the esophagus was ligated with umbilical tape below the larynx, and a double "T" tracheostomy tube, which combined two 5-mm-OD plastic elbows to allow for separate airflow to the esophagus and rostral tracheal segments, was inserted and secured with umbilical tape (10A cotton, Ethicon, Somerville, NJ). Mechanical ventilation (with vapor anesthesia) was then rerouted from the mask to the caudal tracheal section. The esophagus was ligated with umbilical tape below the larynx to prevent reflux or pressure loss from the upper airway. The rostrally directed portion of the "T" tube was used to control pressure in the isolated upper airway. Through this tube, a smaller tube (2 mm OD) was advanced through the vocal cords of the larynx to just below the level of the epiglottis. This prevented vocal cord adduction and allowed for air passage and pressure control of the upper airway segment above the larynx.

The isolated upper airway was sealed around the mouth using 1-0 Vicryl suture (Ethicon) and cyanoacrylate gel glue. The nares were plugged with cotton and sealed with the glue gel. Although a complete seal was sought, the pressure/vacuum control device could accommodate for slight leaks (see below).

Static pressure measurement and control in the isolated upper airway. Upper airway pressure was measured by a nonmagnetic pressure transducer (model SPC350MR, Millar Instruments, Houston, TX) connected to a catheter positioned at the mask during intact upper airway imaging or, later, to the cannula placed in the nares for measuring pressure in the isolated upper airway after surgery. The signal was amplified through a low-noise amplifier (model PM1000, CWE, Ardmore, PA), visualized on an oscilloscope (model D54, Tektronix, Beaverton, OR), and digitized (100-Hz sampling frequency, DAS-16/16F Metrabyte, Tauton, MA) and recorded on a DTK Tech 1000 computer using Asyst software (Macmillan, Rochester, NY). The digital pressure recording was averaged over a 30-s period, during steady-state, static pressure conditions (within 10 s of the initial imaging period) for each imaging series.

Pressure in the isolated upper airway segment was controlled by a mechanical device specifically designed to produce constant low-level positive or negative pressure. The
pressure control unit could be manually switched (Nupro switch valve, Whitey, Highland Heights, OH) for use at negative-pressure (vacuum input at –30.0 in. mercury; model D-25, Precision Scientific, Chicago, IL), positive-pressure (regulated tank air pressure at 20 psi), or zero-pressure (room air) conditions. Final output (to the isolated upper airway) was controlled at a predetermined pressure between –5.0 and +7.5 cmH2O (SE = ±1.0 cmH2O). Pressure was measured on a 30-cmh2O manometer. Pressure control was achieved by the adjustment of a fine needle valve located on the low-flow bleed port before the output to the airway segment, inasmuch as the system was designed for pressure (not volume) control by using a bleed port before the output to the airway segment. Therefore, we analyzed the airway dimensions only under constant, static pressure conditions recorded by the Millar catheter at the nares. In one or two cases the vacuum pressure level was below the closing pressure of the airway and caused collapse in the hypopharyngeal region. In these cases, airway dimensions in the region of airway collapse were not recorded.

Protocol for isolated upper airway imaging. After surgery to create an isolated airway segment, the cat, while under anesthesia, was placed again in the MRI head coil cradle in the same prone position as in the intact state, with the positioning block used to reproduce the head-to-neck angle. Mechanical ventilation was provided through the caudally directed T tube connection, and PEtCO2 and heart rate were monitored to maintain PEtCO2 at 28–32 Torr and heart rate (under anesthetized conditions) at 125 ± 10 beats/min during the imaging procedure.

Isolated upper airway pressure was initially set to 0.0 cmH2O during acquisition of a sagittal series of images to locates the airway. These images were examined on-line (and the series repeated where necessary) so that head-to-neck angle could be set to match the intact imaging series. Thereafter, a series of axial images was acquired during anesthetized nonparalyzed conditions within boundaries of the pharyngeal region that matched those taken during the intact upper airway imaging. The axial images were acquired under pressures produced in the isolated upper airway in the following order: 0.0, –7.5, +5.0, –5.0, +2.5, and –2.5 cmH2O.

To produce a paralyzed state, the cat was withdrawn from the magnet briefly, while neuromuscular blockade was produced by administration of gallamine triethiodide (30 mg/kg iv). Respiration rate and physiological signs were monitored while a steady state in the paralyzed cat was achieved. Anesthesia level and the respiratory rate of the mechanical ventilator were adjusted to steady-state levels as in the nonparalyzed state. After several minutes, the cat was reintroduced into the magnet and the static pressure-testing protocol described above was conducted in the paralyzed cat with the neck in the same position.

Analysis of images. We excluded from analysis initial experiments in two cats where the zero-pressure airway dimensions were noticeably increased in images obtained after surgery compared with those acquired during the intact conditions. In subsequent animals we corrected the problem, which was related to placement of the tracheostomy tube, and we used only data from the four succeeding experiments, thus providing n = 4 for analysis.

We sought to analyze the region of the nasopharynx bounded ventrally by the soft palate and, therefore, chose the aponeurosis of the hard and soft palate as the rostral boundary of the overall pharyngeal region to be examined. A unique reference point (a bony landmark above the dorsal roof of the nasopharynx) was evident in the axial images of all cats, and this point, approximately and no less than 10 mm caudal to the aponeurosis of the hard and soft palate, was used to align the airway measurements. Thus we defined an overall pharyngeal region in each cat, which began 10 mm rostral to the bony reference point and extended 60 mm (caudally) along the airway centerline to a location near the tip of the epiglottis and the free margin of the soft palate.

An axial data set was recorded at each pressure level in the nonparalyzed and paralyzed states. In addition, one axial data series was acquired from the cat before surgery while the cat was spontaneously breathing through the intact upper airway. The axial images were analyzed using VIDA software (University of Pennsylvania) installed on a Sun computing network. VIDA contains a number of analysis modules that provide 3D reconstruction and image analysis tailored for upper airway image analysis. Details relating to the VIDA 3D analyses are described by Schwab et al. (25). Briefly, a bilinear interpolation algorithm was used to reconstruct each series of contiguous axial images into a volume, so that in 3D space each unit volume (voxel) had dimensions of 1 pixel3 (1 pixel = 0.391 mm). A threshold edge detection program was used to segment the pharyngeal airway tube from the surrounding tissues. A tube geometry analysis program obtained the location of the airway centroids of the pharyngeal airway and then reconstructed from the 3D volume axial slices that were orthogonal to the computer-generated centerline of the airway. Finally, for each cat, under each condition (intact, nonparalyzed, and paralyzed), and for the latter two conditions, under each pressure, airway CSA and anteroposterior (A-P) and lateral dimensions were measured (using VIDA) from the reconstructed axial images, orthogonal to the airway centerline, at points ~1 mm apart, along the 60-mm rostral-to-caudal length of pharyngeal airway (see above).

The overall pharyngeal region was then divided into three parts, each 20 mm long (Fig. 1C). We identified these regions as HRP, MRP, and HYP. The HRP region is comparable to the velopharynx in humans, and the MRP region is comparable to the middle and lower portion (near the uvula) of the retropalatal region in humans. The HYP region, as we have defined it in the cat, approximates the same region in humans. We expressed the dimensional data as a function of the distance along the airway centerline and used linear interpolation to determine the airway dimensions at the midpoint of each for the three 20-mm pharyngeal regions (HRP, MRP, and HYP). The mean dimensions were calculated using data from all four cats. For the nonparalyzed condition, we examined five static pressure levels [-7.37 ± 0.37, -4.23 ± 0.48, 0.0, 2.1 ± 0.28, and 4.23 ± 0.31 (SE) cmH2O, n = 4] and, for the paralyzed condition, six pressure levels [ -7.61 ± 0.44, -4.05 ± 0.43, -2.66 ± 0.48, 0.0, 2.17 ± 0.32, and 4.67 ± 0.28 cmH2O, n = 4].

We excluded from our analyses retropalatal airway measurements at positive pressures >7.5 cmH2O. When pressures >7.5 cmH2O were applied to the isolated upper airway, a large air pocket of variable size was formed in the oral cavity that displaced the tongue from its natural position bordering the inferior surface of the soft palate. Thus, at high pressures, distension of the oral cavity caused pharyngeal mechanics in some regions to change from a one- (single tube) to a two-compartment system. Therefore, because we sought to compare the regional compliance of the retropalatal and hypopharyngeal airways under conditions where pharyngeal mechanics were comparable under all pressures, we limited our measurements to pressures ≤7.5 cmH2O in all cats.
The regional values of the CSA and A-P and lateral dimensions obtained at each pressure level for four cats were averaged for that pressure level across all cats. To compare compliance in the nonparalyzed condition with compliance in the paralyzed condition, we used five pressure levels ($-7.5 \pm 0.3$, $-4.1 \pm 0.3$, $0.0 \pm 0.0$, $2.1 \pm 0.2$, and $4.3 \pm 0.2$ cmH$_2$O), where one CSA value measured at the pressure closest to each given level from each cat under each condition was used in the mixed-model ANOVA described below.

Data analysis. To examine the effect of experimental conditions on regional CSA, we compared dimensions in the intact condition, during spontaneous breathing, with the zero-pressure results for the nonparalyzed and paralyzed conditions. We tested the hypothesis that there was no effect of condition on CSA using two-way ANOVA with repeated measures ($n = 4$ random cats) with CSA as the dependent variable and fixed factors: condition (intact, nonparalyzed, and paralyzed) and region (HRP, MRP, and HYP). For significant $F$ value ($P < 0.05$), we tested whether a significant region $\times$ condition interaction existed ($P < 0.05$), and where the interaction term region $\times$ condition was not significant, we used Student-Newman-Keuls multiple comparisons test to compare regional CSA values among conditions (Sigma Stat, Jandel, San Rafael, CA).

Next, we tested the primary stated hypothesis that the effect of pressure on compliance was different among regions using a mixed-model ANOVA with fixed factors (pressure, region, condition, and pressure $\times$ region) and random factors (cat, cat $\times$ pressure, cat $\times$ region, condition $\times$ cat, condition $\times$ pressure, and condition $\times$ region). The three-way interaction term pressure $\times$ region $\times$ condition was also tested. Significance was assumed to be $P < 0.05$. However, these data did not conform to the assumptions of two-way ANOVA, inasmuch as the cell standard deviations for $n = 4$ were positively linearly associated with the cell means (nonparalyzed: Pearson’s $r = 0.55$, $P < 0.0001$, Spearman’s $r = 0.66$, $P < 0.0001$; paralyzed: Pearson’s $r = -0.34$, $P < 0.02$, Spearman’s $r = -0.31$, $P < 0.03$). Logarithmic transformation of the dependent variable CSA removed the relationship between cell standard deviation and cell means, and thus we performed analyses of regional CSA data on logarithmically transformed data (12).

We calculated numerical values for compliance (mm$^2$/cmH$_2$O) for each region by least-squares linear regression on the CSA-pressure relationship obtained from the data in all four cats. From these analyses, performed separately for the nonparalyzed and paralyzed conditions, we provide the slope, Pearson’s $r$, and significance ($P$) of measured regional compliance. We used slope and intercept in the negative-pressure range for paralyzed and nonparalyzed data to determine a linear extrapolated collapse point ($P_c$, i.e., pressure at which zero CSA was predicted) for each region.

For the paralyzed condition, we used one-way ANOVA to compare the mean regional dimensions (A-P and lateral dimensions and elliptical ratio) as a function of pressure ($-7.61 \pm 0.44$, $-4.05 \pm 0.43$, $-2.66 \pm 0.48$, $0.0$, $2.17 \pm 0.32$, and $4.67 \pm 0.28$ cmH$_2$O). The A-P and lateral dimensions were determined from the computerized analyses described above as the length of the axes through the centroid located at the airway centerline in the pharyngeal cross section orthogonal to the airway centerline. The elliptical ratio was defined as the ratio of the lateral to the A-P dimension. An elliptical ratio of 1.0 would describe a perfect circle, and an elliptical ratio $>1.0$ would be oval shaped, having the long axis in the lateral dimension. We further compared, by one-way ANOVA in each region, changes in A-P dimensions with changes in the lateral dimension from 0.0 cmH$_2$O to the most negative or most positive points in the pressure range ($-7.6$ and $-4.7$ cmH$_2$O).

RESULTS

Zero-pressure conditions: overall position head-to-neck angle. Complete sets of sagittal and axial images were taken at three time points in the experimental protocol. Figure 1 shows, in a representative cat, three sagittal views of the pharyngeal airway under the three (anesthetized) experimental conditions: before surgery during spontaneous breathing in the intact airway (intact, Fig. 1A), after creation of the isolated upper airway (nonparalyzed, Fig. 1B), and after paralysis (paralyzed, Fig. 1C). These images were acquired under baseline conditions. Thus, in the isolated upper airway (Fig. 1, B and C) upper airway pressure was 0.0
cmH₂O, but in the intact preparation (Fig. 1A) during spontaneous breathing there were respiratory-related changes in upper airway pressure ranging from −1.8 to +1.0 cmH₂O. The overall airway size and the position of relevant structures such as the soft palate, in bright contrast above the genioglossus and ventral to the pharyngeal airway, and bony structures such as the mandible were essentially unchanged in location in these representative images among experimental conditions. Before surgery, head-to-neck angle averaged 134.5 ± 2.7° (mean ± SE, n = 4) and was not significantly different after surgery (135.5 ± 0.95°, F = 0.12, P = 0.74). Thus the surgical preparation did not alter the basic geometrical dimensions or position of relevant structures in the area of interest in the four animals for which data are reported here.

CSA at zero pressure. To quantify airway size, we divided the airway into three regions. Radial lines perpendicular to the airway tube are drawn in Fig. 1C to delineate these regions in a representative animal: HRP, MRP, and HYP regions (quantitative determination of the regions from the reconstructed 3D data are detailed in METHODS AND MATERIALS). CSA for each region for four cats, compared for the different experimental conditions (intact, nonparalyzed, and paralyzed), are shown in Fig. 2. There was no significant effect of experimental condition (P = 0.9) or any significant interaction between region and experimental condition on CSA (P = 0.4). However, mean CSA was significantly different among regions (ANOVA, P < 0.05). CSA, averaged over all three conditions among four cats, were 14.5 ± 2.6, 21.1 ± 2.6, and 25.9 ± 2.6 mm² for HRP, MRP, and HYP, respectively.

CSA vs. pressure. Pressure changes in the isolated upper airway segment produced large changes in airway size and shape. They are demonstrated in the midsagittal views of a representative cat in Fig. 3. These images were acquired in the paralyzed condition and are for the same cat shown in Fig. 1. In these T1-weighted images the soft palate is visible as a bright layer above the genioglossus and ventral to the darkened air space, probably because of a saliva coating. The largest changes due to pressure were in the HYP region, which was partially occluded at −6.8 cmH₂O (Fig. 3A) yet greatly expanded at −5.1 cmH₂O (Fig. 3C). Thus comparison of these representative images shows the difference in the compliance along the airway, with the caudal airway being the most compliant.

Regional CSA from axial 3D reconstructed slices. A plot of the CSA for each tested pressure level vs. position along the pharyngeal airway centerline is shown for each cat in the nonparalyzed (Fig. 4) and paralyzed states (Fig. 5). The pharyngeal CSA is plotted for several specific pressure levels, and CSA measured for each isopleth is spaced in a graded fashion, from negative to positive isobaric pressure values. Each data set was aligned to the bony reference point (equal to 10 mm, defined in METHODS AND MATERIALS), and the data are shown for 0–60 mm, where 0 mm was approxi-
Fig. 4. CSA measured at rostral-to-caudal pharyngeal airway locations at 5 pressure levels in each cat (A–D) during nonparalyzed conditions in isolated upper airway. CSA was measured by VIDA software in plane sections orthogonal to computer-generated airway centerline. CSA measurements for each isobaric condition were aligned using a unique bony landmark near junction of soft to hard palate, then data were indexed on x-axis according to locations (measured in mm) along airway centerline. Raw data were smoothed by calculating a 5-point moving average for these plots.

Fig. 5. CSA measured at rostral-to-caudal pharyngeal airway locations at 6 pressure levels in each cat (A–D denote same cat as in Fig. 4) during paralyzed conditions in isolated upper airway. CSA was measured by VIDA software in plane sections orthogonal to computer-generated airway centerline. CSA measurements for each isobaric condition were aligned using a unique bony landmark near junction of soft to hard palate, then data were indexed on x-axis according to locations (measured in mm) along airway centerline. Raw data were smoothed by 5-point moving average for these plots.
mate to the aponeurosis of the soft to hard palate. In some cases (Figs. 4, C and D, and 5, B and C) plots for CSA at negative pressure values are discontinuous at the point where occlusion occurred, because airway CSA was zero at these locations. Overall, the graphs show that changes in intraluminal pressure produced much larger differences in CSA in the HYP than in the HRP region.

Regional compliance. The data in Figs. 4 and 5 were used to determine the average CSA for each of three regions (HRP, MRP, and HYP) for the nonparalyzed and paralyzed condition in each cat. Regional compliance was defined as the change in regional CSA per unit change in pressure and is shown as the regional CSA-pressure relationship for each cat. Figures 6 and 7 show that in every case the change in CSA per unit change in pressure was greatest in the HYP region, whereas CSA changes in the MRP and HRP regions were less sensitive to pressure. Figure 8 shows mean regional compliance for four cats as the regional CSA vs. pressure for nonparalyzed and paralyzed conditions.

In our mixed-model ANOVA (performed on results from paralyzed and nonparalyzed conditions) a statistically significant region × pressure interaction was observed, indicating that the effect of pressure on CSA was different among the three regions (F = 16.61, df = 66, P < 0.0001). This result strongly supports the primary hypothesis that significant differences in compliance among the three defined pharyngeal regions exist in this cat model. In the model the fixed-factor condition (paralyzed or nonparalyzed) was not significant (F = 0.01, P = 0.92), nor were condition × pressure (F = 0.35, P = 0.84) and condition × region (F = 1.52, P = 0.22) interactions significant. The three-way interaction terms condition × pressure × region (F = 0.28, P = 0.97) and condition × pressure × cat (F = 0.28, P = 0.97) were also not significant.

The full random mixed-model ANOVA results showed that although cat (n = 4) as a variable was not significant (F = 0.69, P = 0.92), the interaction term condition × cat reached significance (df = 74, F = 7.09, P < 0.003). Thus, although there was no major difference in the results obtained for the paralyzed and nonparalyzed state (condition, condition × pressure, and condition × region terms were not significant), the significant cat × condition term would suggest that the paralyzed or nonparalyzed condition may have had some effects in individual cats. Although compliance cannot be accurately computed when there is muscle

![Fig. 6. Pharyngeal CSA plotted vs. static upper airway pressure for each cat (A–D denote same cat as in Fig. 4) under nonparalyzed conditions. Compliance in each region is slope of CSA-pressure relationship.](image-url)
tone, i.e., the nonparalyzed state, we performed the same calculations in this state as when the animal was paralyzed. Thus we computed what might be called the “effective compliance” or the change in CSA per unit change in pressure for all three regions in the nonparalyzed condition. This proved to be almost identical to the true compliance determined in the paralyzed animal. Compliance varied in the rostral-to-caudal direction, with the most rostral HRP region being the least compliant, the HYP region the most compliant, and the

Fig. 7. Pharyngeal CSA plotted vs. static upper airway pressure for each cat (A–D denote same cat as in Fig. 4) under paralyzed conditions. Compliance in each region is slope of CSA-pressure relationship.

Fig. 8. Pharyngeal CSA (mean ± SE, n = 4) plotted vs. static upper airway pressure (mean ± SE) for nonparalyzed (A) and paralyzed (B) conditions. Compliance in each region is slope of CSA-pressure relationship. Mean pressure SE values were < 0.50 cmH2O for all measurements; thus horizontal error bars were smaller than symbols and are not shown. Symbols same as in Figs. 6 and 7.
MRP region intermediate. The compliance values (for the overall range of positive and negative pressures) for the paralyzed and nonparalyzed state for the three regions were 1.0 and 1.1 cm²/cmH₂O, respectively, for the HRP region, 2.3 and 2.5 cm²/cmH₂O, respectively, for the MRP region, and 3.4 and 3.4 cm²/cmH₂O, respectively, for the HYP region. This confirms the visual impression obtained by comparing the data from each cat in Figs. 4 and 5. Thus in this preparation the degree of muscle tone in the nonparalyzed state had a variable effect on individual cats but did not alter the overall relationship between applied pressure and dimensional changes when the data from both conditions were combined.

For the nonparalyzed and paralyzed condition, we also determined for each region the extrapolated $P_c$. $P_c$ for nonparalyzed and paralyzed conditions was $-8.78 \pm 1.37$ and $-11.95 \pm 1.68$ (SE) cmH₂O, respectively, for the HRP region, $-8.29 \pm 0.88$ and $-7.95 \pm 0.83$ cmH₂O, respectively, for the MRP region, and $-5.63 \pm 1.49$ and $-6.62 \pm 0.81$ cmH₂O, respectively, for the HYP region. Not surprisingly, in both conditions, $P_c$ for the least compliant region (HRP) was the most negative and $P_c$ for the most compliant region (HYP) was the least negative.

Regional differences in airway dimensions and shape. The axial images at the same pressure level in each cat demonstrated a marked similarity in shape, whereby the same regions in all cats had a similar cross-sectional shape. At zero pressure, each airway region had a distinct shape, which could be characterized by the ratio of the lateral to A-P dimensions (the elliptical ratio). Figure 9 shows images from three series of axial images in the three different regions that were acquired in a representative cat at $-6.8$, $0.0$, and $5.0$ cmH₂O. Specifically, the HRP region was elliptically shaped with the long axis in the lateral direction, the MRP region was less elliptical with less pronounced increase in the lateral dimension, and the HYP region was more circular than the HRP and MRP regions.

Table 1 shows the airway dimensions (A-P and lateral dimensions and elliptical ratios) for four cats in the paralyzed condition. When we compared the dimensional values among regions at specific pressure levels, we found that, at $0.0$ and $-4.7$ cmH₂O, mean A-P dimensions were significantly different among all three regions (HRP < MRP < HYP, one-way ANOVA, $P < 0.05$) and the mean elliptical ratios were significantly greater in the most rostral region than in the more caudal regions (HRP > MRP and HYP, one-way ANOVA, $P < 0.05$). In contrast to regional differences in the A-P dimensions and elliptical ratios, there were no significant differences in the lateral dimensions among regions at any specific pressures (Table 1).

For data from the paralyzed condition A-P lateral dimensions and elliptical ratio were significantly related to pressure (one-way ANOVA, $P < 0.05$). Regional differences in A-P and lateral dimensions and elliptical
ratios among pressure levels are noted in Table 1 (comparisons vertically, in each column). At the lowest pressure, −7.6 cmH2O, for a specific region, the values of the elliptical ratio were significantly different from values at 0.0 and −4.7 cmH2O. In specific regions we further compared the magnitude of changes in the A-P dimensions (ΔA-P) from 0.0 to −7.6 cmH2O and from 0.0 to 4.7 cmH2O with changes in the lateral dimensions (Δlateral). We found no significant differences between ΔA-P and Δlateral during negative pressure application (P = 0.63, 0.40, and 0.80 for HRP, MRP, and HYP, respectively). However, at positive pressure, ΔA-P was significantly greater than Δlateral in the more rostral HRP and MRP regions (P < 0.05) but not significantly different in the HYP region (P = 0.43).

**DISCUSSION**

Airway occlusion can occur when negative pressure overcomes the active dilating force of upper airway muscles (11, 26). It is difficult, however, to predict where, along the rostral-to-caudal extent of the pharyngeal airway, occlusion is likely to occur (6). The location of collapse will be determined, at least in part, by the fundamental biomechanical characteristics of the soft tissues that support and surround the upper airway. We have therefore examined the properties of the passive upper airway in an animal model to test our hypothesis that regional pharyngeal compliance varies along the rostral-to-caudal length of the airway. We have made three new findings in our study. First, we found that the average radial compliance, measured as the relationship between intraluminal CSA and pressure, was significantly different along the rostral-to-caudal extent of the airway. Compliance was lowest in the HRP region, highest in the HYP region, and intermediate in the MRP region. Second, we found that Pc was least negative in the most compliant regions, which, paradoxically, also had a significantly larger baseline (zero-pressure) CSA. Third, we analyzed the differences in the A-P and lateral dimensions among regions and the regional dimensional changes that occurred with negative or positive pressure. We found that significantly different A-P dimensions by region at the zero-pressure level contributed to the differences in elliptical shape among regions. In all regions, A-P and lateral dimensions changed significantly in relation to negative or positive static pressure application. However, in the HRP and MRP regions, increases in the A-P dimensions were greater at positive pressures than the respective increases in the lateral dimensions.

The paralyzed isolated upper airway cat preparation and the MRI protocol we employed were designed to acquire physiologically relevant measurements of the upper airway under controlled static pressure levels to determine regional compliance variations. Only a few studies have used imaging approaches to study the pharyngeal patency or upper airway musculature in animals. CT and MRI have been used with a bulldog model of sleep apnea (23, 32), and an acute cat preparation has been studied by Wasiacko et al. (33) in a 0.6-T MRI. In all these studies an endotracheal tube was placed to provide ventilation during the anesthetized procedure. However, endotracheal intubation, in contact with portions of the pharyngeal walls, larynx, and trachea, might potentially alter the mechanical behavior of the airway walls, whereas it also poses difficulties for the creation of a sealed upper airway segment. We eliminated the encumbrance of endotracheal intubation by the use, initially, of a fitted mask for breathing during intact upper airway MRI and, in the later part of our protocol, through the use of a surgical tracheostomy to create an isolated upper airway segment. Thus our model enabled us to examine the singular effects of controlled levels of negative or positive static pressure in the absence of pressure fluctuations and with minimal changes to the natural anatomy and physiology of the pharyngeal airway.

We chose cats because they have been extensively used as a model to study the upper airway muscle structure (5), upper airway neuromuscular control (3, 7), and effects of sleep states on neural control of respiration (31). In addition, the upper airway of the cat is of the optimal scale (for a 10 × 10 cm FOV) to produce detailed images in our 4.7-T high-resolution (1 pixel = 0.391 mm) 40-cm-diameter research magnet. We examined the cats in the natural, prone position so that upper airway structures would be under normal gravitational forces, and we controlled the head-to-neck angle to approximately the middle of the range of flexion to extension (90–145°), where normal upper airway muscle activity has been reported in cats (3). Other investigators who have studied upper airway mechanics in cats (27, 28) have positioned the animals
in the supine position but sutured the tongue in place or positioned it at the anterior mandible to prevent prolapse. These investigators (27, 28) report that the flow-limiting segment or collapsible portion of the pharynx was located in the distal retropalatal region. Their finding of greatest collapsibility in the MRP region, as opposed to our finding of greater compliance in the more caudal HYP region, probably relates to the pharyngeal mechanical differences between the prone and the supine position, including the degree of head-to-neck flexion or extension and positioning of the tongue. Our choice of MRI in the prone position enabled us to acquire MRI of the spontaneously breathing cat, then reproduce upper airway geometry in subsequent MRI of the isolated upper airway. By this method, the tongue position was essentially unchanged between conditions before surgery and after surgery. In addition, by reproducing the head-to-neck angle (controlled to 135.3 \pm 1.3\degree) and maintaining the caudal tracheal length the same for intact and postsurgical conditions, we were able to limit the introduction of changes in airway dimensions due to changes in head-to-neck angle or tracheal tension (19, 28, 29) that could affect compliance. We used midsagittal images of the airway (locator images) acquired at baseline (zero-pressure) conditions to reproduce initial conditions in each trial. Thus in postexperimental analysis we were able to compare regions from the same anatomic location among all the cats, and we found that surgical isolation of the airway did not affect its baseline (zero-pressure) dimensions.

An integral part of our methodology was the pressure control system we used to apply specific levels of pressure or vacuum to the isolated upper airway segment. Our pressure control system did not rely on a complete seal in the upper airway to maintain pressure or vacuum. Small leaks could be accounted for by the equilibrating action of the side-stream bleed valve, in parallel to the airway application. We used gallamine triethiodide (30 mg/kg) to produce neuromuscular blockade (28), since we wanted to study compliance of the airway in the absence of muscle tone. Statistical results indicated that paralyzed or nonparalyzed condition did not have a significant effect on the overall compliance results, and this suggests that, even before paralysis, there was very little upper airway muscle tone in the isoflurane-anesthetized cats. This is not surprising, since upper airway muscle tone in humans and animals is particularly sensitive to general anesthesia (7, 22). Because PetCO2 was maintained in the normocapnic range during paralyzed and nonparalyzed conditions, increases or decreases in muscle tone secondary to hypercapnia or hypocapnia (2) probably did not occur in this anesthetized preparation.

Our first finding was that variations in pharyngeal compliance exist in the paralyzed isolated upper airway. Compliance was progressively greater from rostral to caudal regions, and this would imply that passive support to the pharyngeal airway is essentially heterogeneous, being greatest in the more rostral regions. Olson et al. (18) reported on upper airway compliance in anesthetized rabbits, and they modeled the upper airway as if it had a single value of compliance. Their results were based on singular pressure-volume measurements of the airway and, thus, could neither validate nor invalidate the hypothesis that regional variations in compliance exist in the pharyngeal airway. They correlated P, to the baseline volume (at zero pressure) and suggested that the airway behaves as a singular unit, similar to a hole in an elastic medium (18). However, we have used 3D image analysis to examine the pressure-area relationship throughout the pharynx, and although we would agree that the overall collapsing behavior could be characterized by the single (most collapsible) region, there are significant variations in compliance along the rostral-to-caudal extent of the airway that suggest a model more complex than that suggested by Olson et al.

Kuna et al. (13) obtained results compatible with ours in a study where CPAP was applied in unanesthetized humans during CT imaging. They found that compliance increased progressively from the nasopharynx to the hypopharynx. Moreover, compliance was greater in OSA patients than in normal subjects. Kuna et al. did not, however, measure true compliance, since they measured pharyngeal CSAs in unanesthetized subjects during tidal breathing, during which dynamic changes in pressure and upper airway muscle activation affect pharyngeal CSA (24). Furthermore, changes in measured CSA may vary between subjects because of differences in dilator muscle action, inasmuch as studies report greater upper airway dilator muscle activity in OSA patients than in normal subjects (16). Thus our study, in which we measured true passive tissue compliance, extends the results of Kuna et al. by showing that the rostral-to-caudal variation in compliance is a property of the passive pharyngeal tissues.

Other investigators who have made use of invasive endoscopic methods have measured the CSA-pressure relationship at discrete loci under hypotonic, passive conditions in the velopharynx, oropharynx, and hypopharynx in OSA patients (8, 14, 17). Morrison et al. (17) showed that although the majority of patients (80%) had primary narrowing in the velopharynx, a similar number (82%) had two or more sites of narrowing. Isuno et al. (9) extended these studies to compare pharyngeal mechanics in OSA patients and normal subjects under neuromuscular blockade. The overall results (8, 14, 17) indicate that the velopharynx was the most compliant region in OSA patients and normal subjects, with evidence of heterogeneity in compliance throughout the airway. Although species differences may account for variations in the loci of greatest regional compliance, our results, which showed that pharyngeal compliance in the cat was greatest in the more caudal regions and lowest in the high retropalatal region, are similar to those of some other human studies (13, 34). Other factors, in particular those relating to posture or the position of the mandible (10), affecting airway geometry are likely to contribute to differences in pharyngeal mechanics and should be considered when comparisons are made between studies.
We found that the pressure-area relationship was described by a linear function for the overall range of pressure values (−7.5 to −4.3 cmH2O) we measured. Although our methods, wherein dimensions were averaged within a region at each pressure, may have smoothed some curvilinear effects, it is likely that we have measured compliance in a pressure range in the cat where linear compliance predominates. In studies in humans (9) there is a linear relationship between pressure and area in the pressure range close to 0.0 cmH2O or near the $P_c$, for that curve but not at high pressures. For technical reasons, we did not study the pressure-area relationship at high pressures. Our finding that, in the range of pressures we examined, compliance in the passive pharynx was described by a linear relationship would indicate that the surrounding soft tissues that support the pharyngeal airway walls do not show a significant increase in passive tension in response to greater collapsing forces.

Because the pharyngeal regional CSA-pressure relationship was well described by a linear relationship, we used this relationship to determine an extrapolated $P_c$ equal to $\frac{-7.5}{\text{baseline CSA/compliance}}$. We found that the most compliant region (HYP) was also the most collapsible; i.e., the HYP region had the least negative $P_c$. If compliance was the same in all regions, then increased baseline CSA would result in decreased $P_c$. However, we found that compliance was significantly greater in the caudal than in the rostral regions. Thus in our analyses, where $P_c$ depended on the baseline CSA and compliance, the increase in compliance in the HYP region resulted in an increased $P_c$, despite the fact that baseline CSA of the HYP region was larger than that of the more rostral regions. The regional differences in collapsibility that we determined from our compliance data were confirmed in MRIs obtained at negative pressures, which showed that collapse occurred in the compliant HYP region under the same negative pressure conditions during which the HRP and MRP regions were still patent (Fig. 9, a–c).

$P_c$, defined under static conditions, can be considered analogous to the critical pressure ($P_{\text{crit}}$), defined as the pressure at which flow limitation occurs, in the Starling resistor model of Schwartz et al. (26) for pressure and flow in the upper airway of patients with OSA. Thus $P_{\text{crit}}$, as an indicator of collapsibility, has been determined by measuring the flow-limiting velocity ($V_{\text{max}}$) in animal and human studies (26, 28). Although $P_{\text{crit}}$ is a measure of airway collapsibility, a change in $P_{\text{crit}}$ could imply changes in compliance or changes in baseline airway geometry, because the measured variable, $V_{\text{max}}$, is a function of $P_{\text{crit}}$ and upper airway resistance. We found that, under static conditions, increased compliance was the determining factor for increased collapsibility ($P_c$). Thus our results offer some evidence to suggest that $P_{\text{crit}}$, which is analogous to $P_c$, may be determined primarily by fundamental tissue characteristics such as regional pharyngeal compliance.

Our quantitative analysis of airway dimensions showed that within the same region there was a significant relationship with pressure in A-P and lateral dimensions ($P < 0.05$). We found that, during positive-pressure application, A-P dimensions increased more than lateral dimensions, and these differences occurred in the more rostral regions (HRP and MRP), whereas the A-P and lateral increases were not different in the caudal (HYP) region. Negative-pressure application caused decreases in regional A-P and lateral dimensions that were of the same magnitude at −4.1 and −7.6 cmH2O. In a study where CPAP was applied during tidal breathing in normal subjects and OSA patients, Kuna et al. (13) found that although A-P and lateral dimensions were increased significantly with pressure compared among regions, the lateral dimensions were significantly increased in the more compliant (more caudal) regions. OSA patients showed even greater lateral dimension increases. Similar findings by Schwab et al. (25) showed that, during application of CPAP in normal subjects, increases in the lateral dimensions were significantly greater than increases in A-P dimensions. However, Schwab et al. also reported that significant increases in A-P dimensions were observed under several positive-pressure conditions in some, but not all, pharyngeal regions measured.

Although the differences between our results and those in humans might be due to species differences, there are also significant methodological differences. We used a paralyzed preparation, where active muscle tone in upper airway muscles was abolished, and thus the passive tissue characteristics were examined. In the study of Kuna et al. (13) in awake unanesthetized humans, electromyogram activity of upper airway dilator muscles was normally reduced to varying degrees in different subjects during positive-pressure application. Therefore, although the positive-pressure application in awake humans causes, in general, greater lateral than A-P expansion, it is also possible that some degree of upper airway muscle tone in awake humans underlies the support of A-P-directed structures compared with lateral structures. We do not know whether, in an unanesthetized cat, active muscle tone would favor lateral or A-P expansion with positive pressure. Thus although our results indicate that greater compliance in the A-P dimensions may exist in the paralyzed cat upper airway, the human studies have shown that the lateral walls are more compliant in the more caudal regions and in OSA patients. Our study is in agreement with the overall findings of others (13, 25), in showing that the pharyngeal airway is supported around the circumference in a heterogeneous fashion, since changes in A-P and lateral dimensions under applied pressures vary among different regions along the rostral-to-caudal extent of the airway.

The changes in airway dimensions that we observed during static negative-pressure application showed that negative pressure caused equal reductions in A-P and lateral dimensions within each region and caused a significantly more flattened elliptical cross-sectional shape at the most negative pressures. Our findings are similar to the results obtained by Wheatley et al. (34), who used fluoroscopy to measure the A-P and lateral dimensions in humans at rostral-to-caudal locations in
the upper airway and examined the difference between the “active” and “passive” control of upper airway dimensions during maneuvers produced with or without voluntary exertion of upper airway muscles. They found that although voluntary active effort (inspiratory effort against an occlusion) could prevent decreases more effectively in the A-P dimensions, during the passive maneuvers (ramped negative pressure, while the glottis was closed), A-P and lateral dimensions in the oropharyngeal and HYP regions were reduced by a similar magnitude (34). Thus, under conditions where muscle tone was reduced, A-P and lateral dimensions were reduced equally under negative-pressure application in the human upper airway in the same manner as we found in the paralyzed upper airway in cats.

Our results showed that airway shape varies in a characteristic fashion throughout the airway in the cat, with the elliptical ratio becoming greater, i.e., cross-sections were more elliptical in the rostral (HRP) than in the caudal (HYP) regions. Thus we suggest that, along with factors such as pharyngeal wall thickness, wall flexural rigidity, and the nature of the soft tissue and bony tissue support surrounding the pharyngeal tube, intraluminal shape could be considered as one of the factors that may contribute to the collapsibility and compliance of the pharyngeal tube. Other investigators have examined the intraluminal cross-sectional shape of the airway in normal subjects and patients with OSA (20, 21, 24, 25) and have reported that the airway shape in OSA patients, particularly in the retropalatal area, is more circular or even oval shaped, i.e., with the long axis in the A-P direction, as opposed to normal subjects, whose airways at the same level were elliptical, with the longer axis in the lateral direction.

Interestingly, shape may also play a role with regard to airway stability when active muscle contraction is factored into the pathogenesis of OSA (15). Leiter (15) contends that some dilator muscles, e.g., the genioglossus, which operate primarily in the A-P direction, can more efficiently dilate an airway with a laterally directed elliptical shape and has suggested that the pathogenesis and surgical treatment of OSA would benefit from studies that include airway shape and orientation measurements along with measured changes in airway CSA and muscle activity.

In conclusion, we have established a model for MRI studies in the paralyzed isolated upper airway in cats, and we have provided results that show the heterogeneous nature of support along the rostral-to-caudal extent, as well as in the radial cross-sectional aspect, of the paralyzed pharyngeal airway in cats. We found that pharyngeal compliance was greatest in the caudal HYP region and lowest in the more rostral HRP region. We also found that pharyngeal compliance differences among regions were directly related to collapsibility (Pc), whereas baseline (zero-pressure) regional CSA was inversely related to Pc. Our overall findings, which describe the regional variations in pharyngeal compliance in cats, point to the existence in the airway of a heterogeneous muscle support system designed to provide maximum active support to the more compliant regions as well as to the regional differences in pharyngeal compliance that may underlie predisposition to pharyngeal collapse in OSA.

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