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Fluoroscopic Estimation of Thoracic Dimensional Changes in Healthy Dogs

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Background: Current methods available for assessing alterations in lung mechanics require sophisticated equipment and are of limited availability. A method that could assess lung area change with respiration might be a clinically useful surrogate for assessing lung compliance.

Objective: To use fluoroscopy to determine percent change in thoracic and lung areas in healthy dogs.

Animals: Forty-four client-owned dogs with no evidence of respiratory disease.

Methods: Prospective study. Resting respiration was recorded fluoroscopically, and peak inspiratory and expiratory frames were captured for 3 typical respiratory cycles. The number of intrathoracic pixels in the entire thoracic cavity was measured for both inspiration and expiration, and the average percent change in intrathoracic area was determined for each dog. This process was repeated by a hemithorax measurement of lung area that excluded the mediastinum and cardiac silhouette. Proposed reference ranges (and 95% confidence intervals [CI]) were computed by a nonparametric percentile distribution.

Results: Median percent change in thoracic dimension for the total thorax measurement was 12.5% (CI, 8.9–24.0%). Median percent change for the hemithorax measurement was significantly ($P < 0.001$) larger (20.8%, CI, 14.3–37.6%). Both measurement techniques were correlated with body weight but not with age, sex, thoracic conformation, body condition score (BCS), or breed.

Conclusions and Clinical Importance: Fluoroscopy allows a noninvasive and repeatable measure of lung area changes during respiration that must be corrected for body weight. Additional studies in dogs with respiratory diseases are needed to determine its utility in detecting clinically useful alterations in lung area changes.

Key words: Dog; Parenchymal disease; Pulmonary function testing; Respiratory tract; Respiratory tract; Respiratory tract.

Current methods available for assessment of lung mechanics, such as dynamic compliance and airway resistance, are limited in availability and employed primary in the research setting. One particular concern with these diagnostic procedures is that they often involve general anesthesia, which entails increased risk for patients with compromised pulmonary function.[1] Tidal breathing flow volume loops (TBFVLs) can be measured in conscious patients with a tight-fitting face mask and pneuomotachograph to measure flow, volume, and time. This technique is used most often to measure airway function, specifically for detecting bronchial and laryngeal disease.[2,3] Like TBFVLs, barometric whole-body plethysmography is an alternative procedure that can assess flow and volume changes in awake animals, but it can provoke panting and primarily is used to document airway obstruction.[4–6] More routinely available tests of pulmonary function include arterial blood analysis and the 6-minute walk test (6 MWT). The 6 MWT has proven particularly useful as a surrogate measure of cardiopulmonary dysfunction.[7–9] Direct measurement of lung compliance requires intubation and anesthesia and has a low sensitivity for mild-to-moderate disease.[10] Furthermore, advanced training in physiology and engineering is necessary to perform these evaluations.[10]

Abbreviations:

6 MWT	6-minute walk test
BCS	body condition score
CV	coefficient of variation
ICC	intraclass correlation coefficient
ILD	interstitial lung disease
TBFVL	tidal breathing flow volume loops

Interstitial lung diseases (ILDs) decrease compliance (the change in volume for a given change in pressure) and definitive diagnosis requires lung biopsy because there is a lack of well-defined noninvasive diagnostic criteria.[11] Because of the vast surface area dedicated to gas exchange within the lung, function must be severely altered before respiratory compromise can be detected. As a result, ILDs often are diagnosed at a late stage, leading to high morbidity and mortality. Hence, a sensitive and noninvasive mechanism for detecting decreased lung expansion would be a valuable tool for early

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detection of ILD, grading the severity of disease, and evaluating response to treatment.

Thoracic fluoroscopy potentially could provide a means for estimating lung volume changes during respiration. This real-time imaging modality has been recognized for its utility in interventional radiologic procedures and assessment of specific functional diseases, such as tracheobronchial collapse and swallowing disorders in dogs.[12–14] To date, this minimally invasive diagnostic test has not been evaluated for documentation of dynamic changes in thoracic dimensions or lung area. The aim of our prospective study was to quantify the percent change in thoracic dimension during resting respiration using fluoroscopic measurement of lung area and thoracic area changes. We hypothesized that the percent change in thoracic dimension between inspiration and expiration could be defined in healthy dogs represented by our client population and used to establish a reference range for dogs lacking respiratory disease. Furthermore, we investigated the influence of age, sex, thoracic conformation, body condition score (BCS), and breed on thoracic and lung area changes. This information ultimately would provide normal values for comparison with those obtained from dogs with respiratory diseases such as ILD.

Materials and Methods

Study Population

A convenience sample of 50 healthy client-owned dogs of various breed, weight, age, sex, BCS, and thoracic conformation (characterized by thoracic height-to-width ratio) were prospectively recruited for imaging studies. However, 6 dogs were excluded because of noncompliance during imaging. Body condition score was assessed with a 9-point scale[15] and was agreed on by 2 of the investigators (JC, CB). Owners provided informed consent, and the Institutional Animal Care and Use Committee at the University of California approved all procedures. All dogs resided in smoke-free households.

Dogs with a history of respiratory or cardiac disease or abnormal cardiopulmonary auscultation were excluded. Before enrollment, dogs were deemed healthy based upon results of physical examination, pulse oximetry ≥95% hemoglobin saturation with oxygen while in a standing position, and lateral cervical and 3-view thoracic radiographs (right lateral, left lateral, and dorsoventral projections). Owners obtained a resting respiratory rate at home to define the target respiratory rate for use during fluoroscopy. To do so, owners counted thoracic excursions over 1 minute when dogs were at rest but not asleep inside at a temperature of approximately 22°C.

Determining thoracic conformation

Thoracic conformation was determined by quantifying a height-width ratio by a modified version of the technique similar to that described previously.[16] Height was measured from the internal margin of the most caudal portion of the sternum within the thoracic cavity to the ventral margin of the vertebral body directly above the sternum on right lateral fluoroscopic imaging (Fig 1). For dogs whose thoracic cavity did not fit within the fluoroscopic imaging window, a right lateral radiographic projection was used to measure height. Width was derived from the dorsal-ventral fluoroscopic image used to determine the first maximal inspiration by measuring the number of pixels from the right to the left thoracic wall at its widest, most caudal portion (Fig 2). A height-width ratio of 0.86–1.05 was suggestive of typical thoracic conformation. Dogs were judged as deep chested if the height-width ratio was >1.05 and barrel chested if <0.86. These categories were used for descriptive purposes only.

Fluoroscopic Imaging Procedures

Conscious, unsedated dogs were minimally restrained in sternal recumbency and acclimated to positioning on the fluoroscopy table. Fluoroscopic imaging was performed and recorded by a commercially available fluoroscopy unit[a] once the dog's respiratory rate approximated that measured at home by the owner. Images were obtained and recorded at 30 frames per second. Machine settings were optimized based upon patient size with kV ranging from 104–125, mA ranging from 85.8 to 100, and image resolution approximated at 200 μm. Thoracic excursions were observed and recorded fluoroscopically for at least 10 full respiratory cycles, and respiratory rate was noted.

Determining Percent Change in Lung Area

Digital fluoroscopic recordings were archived and reviewed by 1 observer (JC) at a later date. Maximal inspiratory and expiratory images were selected for 3 separate representative respiratory cycles in each dog. The fluoroscopic videos were evaluated as a collection of distinct, serial images by commercially available software[b] Maximal inspiration was defined as the imaging frame in which the diaphragm was most caudally positioned and the ribs most abaxially positioned. Maximal expiration was defined as the imaging frame in which the diaphragm was most cranially positioned and the ribs most axially positioned. Inspiratory and expiratory frames were loaded into commercially available freeware[c] and percent change in lung area between the maximal inspiratory and expiratory images captured for each of the 3 breathing cycles was calculated. First, total thoracic area was measured at inspiration and expiration by drawing a region of interest around the cranial margin of the diaphragm, pleural surface of the thoracic wall, and cranial lung lobes including the cardiac silhouette and mediastinum (Fig 3A). Secondly, for the hemithorax measurement, regions of interest were drawn for the right and left lungs

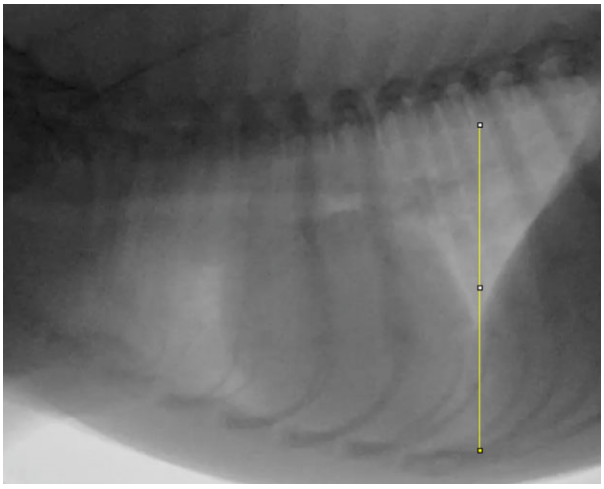

Fig 1. Height of the thoracic cavity was measured in right lateral from the internal margin of the most caudal portion of the sternum within the thoracic cavity.

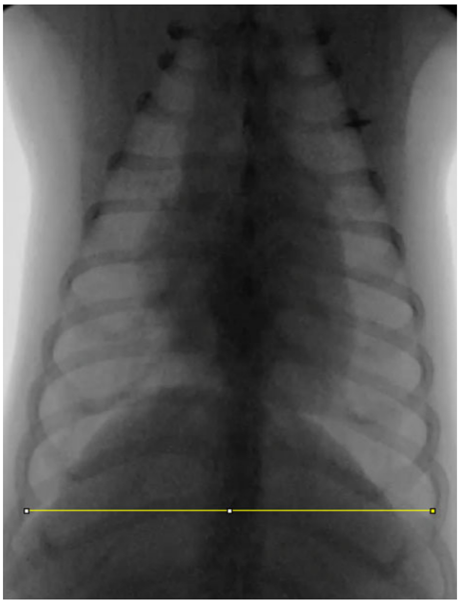

Fig 2. Width of the thoracic cavity was measured on the fluoroscopic ventral-dorsal view using the first maximal inspiratory image.

separately by tracing the cranial margin of the diaphragm, pleural surface of the thoracic wall, and cranial lung lobes but excluding the cardiac silhouette and mediastinum. Values obtained from the right and left lung were added to provide a measurement of the total lung area (Fig 3B). The percent change in total thoracic and hemithoracic area was calculated as: ([# pixels at inspiration – # pixels at expiration]/[# of pixels at inspiration])(100%), and an average value for each dog was generated from 3 respiratory cycles. Side-by-side comparison of expiratory and inspiratory frames is presented in Figure 4.

In small and medium breed dogs, the entire thorax fit within a single image, and a single frame method was used to generate percent area change. For large breed dogs that had a thoracic cavity too large to fit within a single fluoroscopic imaging field, a summated method was used to assess the change in thoracic and lung dimensions. Ten respiratory cycles were captured for the cranial thorax and 10 respiratory cycles were captured for the caudal thorax with no change in body position between the 2 imaging acquisitions. A single image of both maximal inspiration and expiration was chosen from the cranial imaging session, and the total and hemithorax lung area tracing process was performed to the caudal most completely visible rib (Fig 5). Thoracic excursion was consistent during quiet respirations, and the majority of change in thoracic dimension originated from the movement of the diaphragm in the caudal frame. Therefore, a single cranial frame was used to represent cranial thorax movement. Three frames for maximal inspiration and expiration were chosen from the caudal imaging session, and the right, left, and total lung area tracing process was performed starting at the rib that defined the caudal margin in the cranial imaging session images. The sum of traced pixels in the cranial frame and in each caudal frame was taken for right, left, and total lung area and the percent change calculated as described above. An average value for each dog was generated from 3 respiratory cycles.

To validate the summation methodology applied to large dogs, the following method was used to perform total and hemithorax measurements in 10 randomly selected small-medium dogs for comparison to the original measurements. In the absence of a partition for the thoracic cavity, the sixth rib was chosen as the division between the cranial and caudal frames for small-medium dogs, because this rib approximated the caudal most visible rib in the cranial frames of large dogs. The cranial image was selected from a new respiratory cycle, and the respiratory cycle frames from the initial analysis were used for the 3 caudal measurements.

Intra-Observer Variation

Ten dogs were randomly selected and percent area change measurements were performed a second time by the same observer to assess intra-observer variation.

Statistical Analysis

Data were assessed for normality by the D'Agostino & Pearson Omnibus method.[d] Normally distributed values are presented as mean ± standard deviation, and statistical analysis was performed

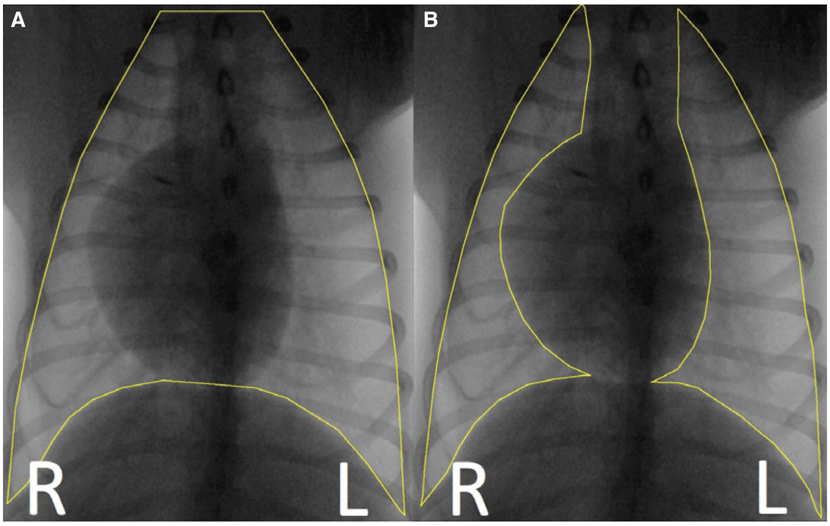

Fig 3. **(A)** Total measurement includes the entire thoracic cavity. **(B)** Hemithorax measurement excludes mediastinum and cardiac silhouette.

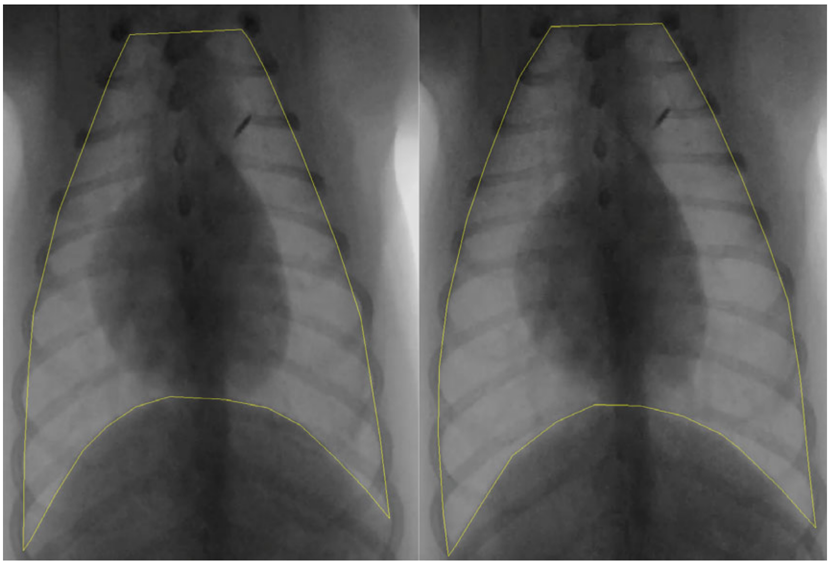

Fig 4. Side-by-side comparison of maximal expiratory (left) and inspiratory (right) frames for a given respiratory cycle by the total thorax method.

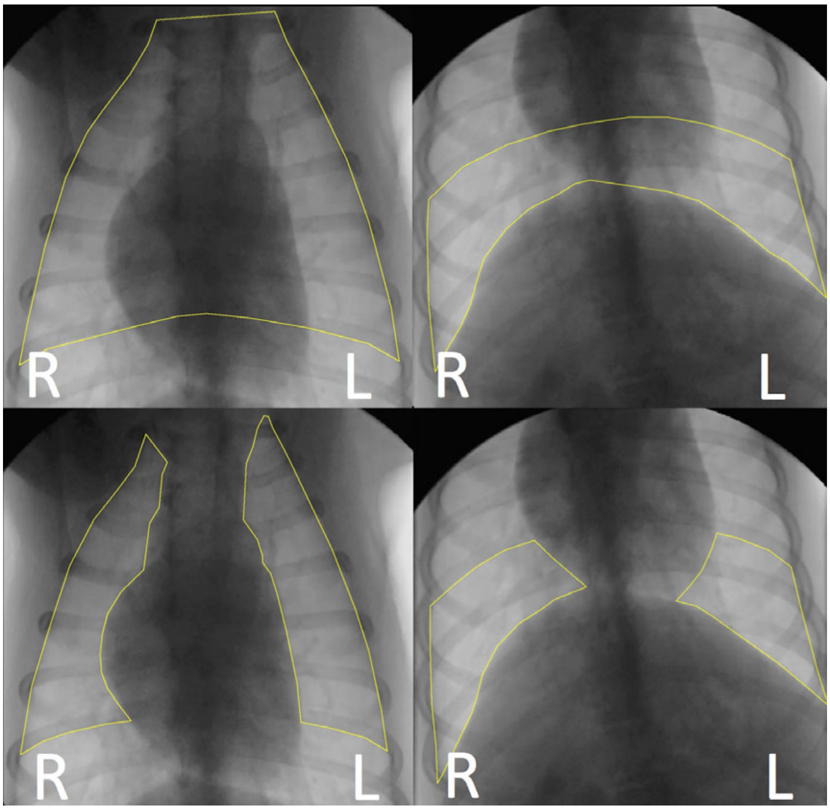

Fig 5. Summated method was used for dogs whose thoracic cavities did not fit within the imaging window to determine the total measurement (above) and hemithorax measurement (below).

by Student's *t* test or analysis of variance. Data that were not normally distributed are presented as median with range and analyzed with nonparametric statistics. To compute reference ranges of 95% confidence, both parametric (normal and log normal) and nonparametric methods (percentile, bootstrapping statistics) were evaluated initially.[e] Use of percentiles provided the best goodness of fit for this dataset.

Variability in the 3 measurements used to average the percent area change for each method is reported as coefficient of variation (CV) with 25 and 75% percentiles.

Percent change for total and hemithorax measurements of the intrathoracic area and lung area, respectively, was compared by the Wilcoxon matched-pairs signed rank test. Median values for percent change in the left versus right hemithorax were compared with Mann-Whitney test. Correlation between the 2 methodologies was assessed by Spearman correlation for nonparametric data. Lung area changes calculated from the single frame and summated methods applied to images from 10 small- to medium-sized dogs were compared for both total and hemithorax measurements by the paired Wilcoxon test.[d]

Allometric models were fitted to the relationship of percent change in thoracic and lung areas with weight and with height-width ratio, by linear least squares regression to fit the model $\ln(Y) = a + b \ln(X)$ and then exponentiating both sides of the equation. Proposed reference ranges were obtained by exponentiating the 95% prediction intervals from each model.[e]

Intraclass correlation coefficients (ICCs) were used to assess intra-observer variation from repeat measurements in 10 dogs.[17,e] For all analyses, P was set at <0.05.

Results

Fifty dogs were recruited for the study. Six dogs were excluded because of poor cooperation with restraint or failure to achieve a respiratory rate <3 times the resting respiratory rate at home so that the final study population consisted of 44 dogs. Age of dogs in this study ranged from 0.67 to 14 years (median, 3.5 years). Body weight ranged from 2.6 to 31.8 kg (median, 18.1 kg). Body condition score was between 4 and 6 in all dogs with a mean of 4.8 ± 0.62 of 9. There were 16 of 44 (36%) spayed females and 28 of 44 (64%) neutered males. Pure breed dogs included 2 Labrador Retrievers, 2 Schipperkes, 1 Chihuahua, 1 Norfolk Terrier, 1 Border Collie, 1 Australian Cattle Dog, 1 Standard Poodle, 1 Australian Shepherd, and 1 Papillion. The majority of dogs (33/44, 75%) were mixed breed dogs. Typical thoracic conformation was recorded for 25 of 44 dogs (57%), 8 of 44 (18%) were considered deep chested, and 11 of 44 (25%) were considered barrel chested. There was no significant difference in body weight or BCS among different thoracic conformations (Table 1). The single frame method was possible in 24 dogs, and the summated technique was required in 20 dogs.

The coefficient of variation (CV) for repeated measurements (with 25% and 75% percentiles) by the total thoracic method was 7.6% (4.6–11.1%). For the hemithorax measurement of lung area, CV was 8.7% (4.8–12.4%).

Median percent change for the left hemithorax was not significantly different from the value determined for the right hemithorax ($P = 0.14$).

Median percent change in thoracic and lung area between inspiration and expiration is presented in Table 2. The total thoracic measurement yielded a median of 12.5% (range, 8.9–26.1), and percentile distribution generated a CI of 8.9–24.0%. When considering all dogs, the hemithorax measurement, which excluded the mediastinum and cardiac silhouette, yielded significantly higher values for percent area change ($P < 0.0001$) than the total thoracic method, with a median percent change of 20.8% (range, 12.8–39.2) and CI of 14.3–37.6%. Values for percent area change computed by the 2 methods were highly correlated ($P < 0.0001$, Spearman $r = 0.89$; Fig 6). Two of 44 dogs had an area change outside the confidence intervals including a Papillon using the hemithorax method (39.2) and a Norfolk Terrier using the total thorax method (26.1; Fig 7).

Change in thoracic and lung areas by either the total or the hemithorax measurement method was not correlated with age, sex, or BCS. The percent change in area by both methods decreased significantly with increasing body weight ($P < 0.001$, Table 3, Fig 8). Percent change in area by either method was not significantly associated with height-width thoracic ratio (Table 3).

Thoracic area measurements obtained by the single frame method were repeated with 10 randomly chosen small-medium dogs by the summated method and results were compared. Single frame and summated methods provided equivalent measurements of change in thoracic dimension ($P = 0.43$ for total, $P = 0.56$ for hemithorax), validating the use of the summated method when a single frame measurement could not be obtained (data not shown).

Finally, images from 10 dogs were randomly selected for repeat measurements to assess intra-observer variation. A strong intraclass correlation (ICC) with the original measurements was found for both total (ICC = 0.97, 95% CI = 0.89–0.99) and hemithorax values (ICC = 0.95, 95% CI = 0.82–0.99), indicating that the methodology was comparable and repeatable.

Discussion

Results support the hypothesis that fluoroscopy can be used to noninvasively quantify the percent change in thoracic and lung dimension of healthy dogs during

Table 1. Thoracic conformation vs. median body weight

Thoracic Conformation	Height-Width Range	Number of Dogs	Median (range) Body Weight (kg)	Median (range) BCS
Barrel Chested	<0.86	11	9.5 (2.6–28.4)	4 (4–6)
Typical	0.86–1.05	25	18.4 (3.2–30.2)	5 (4–6)
Deep Chested	>1.05	8	24.0 (6.6–31.8)	5 (5–6)

Table 2. Comparison of the total and hemithorax measurements for assessing percent change of thoracic dimension in all 44 dogs

	Total	Hemithorax
Median	12.5*	20.8*
Range	8.9–26.1	12.8–39.2
CI	8.9–24.0	14.3–37.6

Total, measurement of entire thoracic cavity; Hemithorax, measurement excludes mediastinum and cardiac silhouette; CI, confidence interval. Values obtained by the total thorax measurement method were significantly lower than those obtained by the hemithorax method, *$P < 0.0001$.

tidal respiration. The percent change obtained from 42 of 44 healthy dogs fell within a reproducible reference range with acceptable variability. We aimed to characterize lung excursions in healthy dogs representative of the general population, and as a result, a group of dogs with variable demographic characteristics was enrolled. The proposed reference ranges generated from this diverse group of dogs appear to be reliably independent of age, sex, thoracic conformation, BCS, and breed. However, body weight had a significant influence on these measurements such that smaller dogs had a higher percent change in thoracic and lung area during resting respiration.

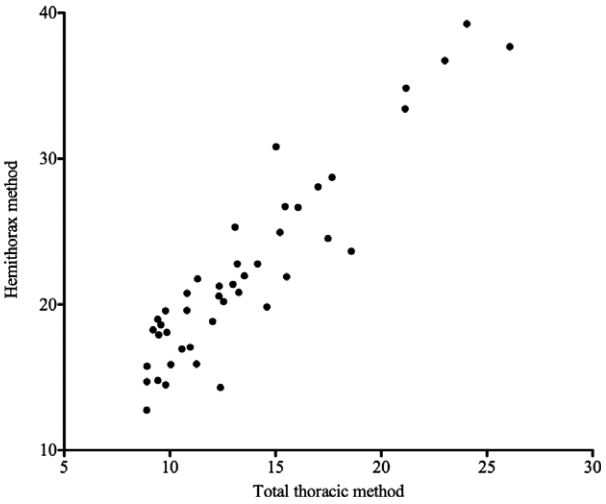

Fig 6. Thoracic area change as determined by the total thoracic measurement and hemithorax measurement methods. Results were highly correlated ($P < 0.0001$, Spearman $r = 0.89$).

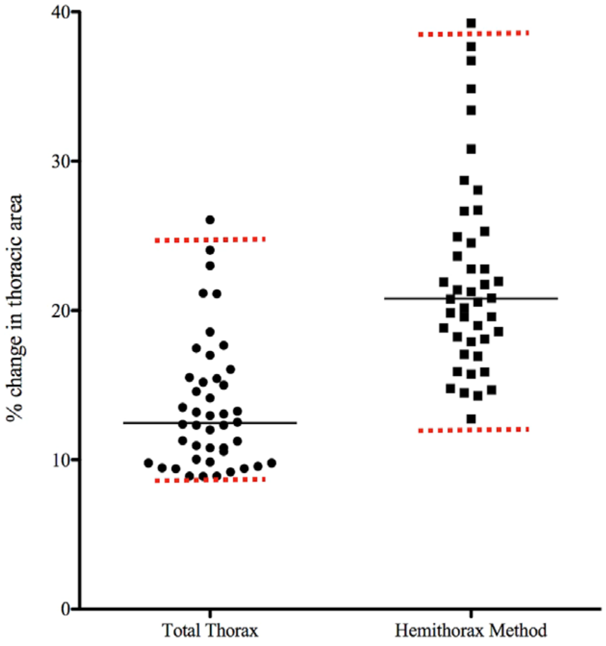

Fig 7. Percent change in thoracic area calculated by the total measurement method (filled circles) and the hemithorax measurement (squares), which excluded the mediastinum. Bars represent median values. Dashed lines represent 95% confidence intervals.

Percent change in area generated by the total measurement technique was significantly less (median, 12.5%; reference range, 8.9–24.0%) than that generated by the hemithorax technique (20.8%; reference range, 12.8–39.2%). Elimination of the heart and mediastinum from the measurement is slightly more time consuming but likely gives a better representation of change in pulmonary area because thoracic area not contributing to lung area has been excluded. Although not assessed in this study, the influence of cardiac cycle on cardiac, and consequently overall mediastinal area could have impacted hemithoracic measurements. Literature in human medicine supports this assumption. The change in size of the heart during the cardiac cycle has been shown not to be clinically relevant, with cardio-thoracic ratio changes being <2% between systole and diastole.[18,19] Exclusion of the cardiac silhouette would seem to represent a more physiologic assessment of changes in lung expansion and would obviate any potential influence of systole and diastole on measurements obtained. Consequently, considering the current data, we conclude that the hemithorax measurement provides a more appropriate method for estimating thoracic area change during tidal respiration.

A significant relationship was observed between weight and percent change in lung area. This relationship appeared to be independent of BCS because there was minimal variation in BCS within the study sample (SD = 0.62). However, as weight increased, percent change in thoracic area decreased. This observation is in agreement with a study that found that body weight could predict absolute expiratory and inspiratory static lung compliance in healthy dogs under anesthesia as measured using forced pulmonary inflation.[20]

We found no correlation between percent lung area change and age. Aging in people is associated with degeneration in the elasticity and distensibility of the lung parenchyma, which alters lung compliance.[21] Additional age-related changes to chest wall compliance, such as costal cartilage calcification and degeneration of the spine, augment the decline in thoracic compliance.[22] Although our study population included a wide range

Table 3. Allometric modeling of percent change in thoracic and lung area

Y Variable	X Variable	Fitted Equation	*P*-Value
% Change in Area Total Method	Weight	% Change = 21.9 $\text{Weight}^{-0.20}$	<0.001
% Change in Area Hemithorax Method	Weight	% Change = 38.0 $\text{Weight}^{-0.22}$	<0.001
% Change in Area Total Method	Height-Width Ratio	% Change = 12.6 $\text{Ratio}^{-0.57}$	0.157
% Change in Area Hemithorax Method	Height-Width Ratio	% Change = 20.8 $\text{Ratio}^{-0.49}$	0.200

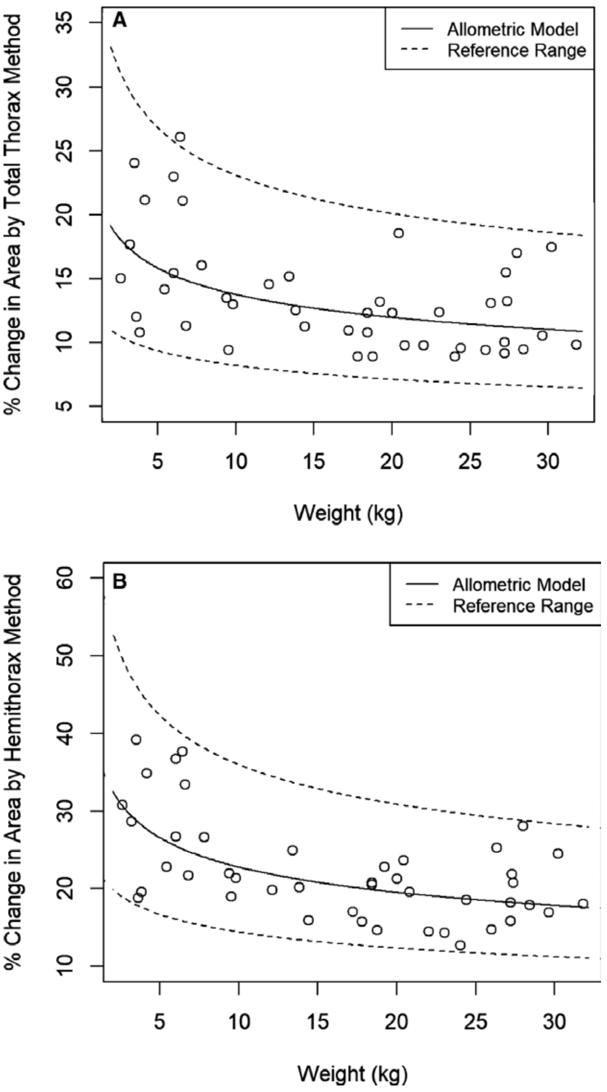

Fig 8. Percent change in thoracic area calculated by the total measurement method **(A)** and in lung area calculated by the hemithorax measurement method **(B)** was correlated with body weight. $P < 0.001$.

of ages, the dogs were relatively young (median age, 3.5 years), and any effect of age could have been missed as a consequence of low numbers of geriatric participants. Further studies with larger age variation are warranted to investigate the role of age on fluoroscopic changes in lung area in dogs.

In people, differences in hormones between males and females have been linked to disparities in the occurrence of lung disease. Asthma, lung cancer, idiopathic pulmonary hypertension, and pulmonary fibrosis occur more commonly in either men or women.[23–26] Despite characteristic differences in lung size and airway diameters between the sexes, healthy men and women share the same degree of lung elasticity.[27] Similarly, we found that sex did not affect lung area changes in the examined healthy dogs.

Dogs were categorized as typical, deep, and barrel chested based on thoracic height-width ratio, but this had no effect on percent area change obtained. Interestingly, a previous study found significant differences in static respiratory compliance between narrow and broad chested breeds.[1] The narrow chested Border Collies and German Shepherds had higher absolute compliance than did the broad chested Labrador and Rottweiler breeds, presumably because of larger lung volume. Although the study recommended further characterization of the thoracic cavity by a height-width ratio and predicted that a relationship would be observed, these values were not calculated. Further studies including a larger number of dogs with greater variation in thoracic conformation would be necessary to ensure that there is no relationship of body conformation to percent change in lung area.

We anticipated that BCS could affect change in lung area by altering the ability of the thoracic musculature to change dimension. However, no effect was noted and this observation is likely a consequence of the relatively uniform lean BCS in the study population. In people, it is well known that obesity predisposes individuals to lung function complications.[28] Further studies are needed to evaluate the effect of BCS on inspiratory to expiratory volume change because obesity affects the ability of the thorax to expand and could thus impact measurable values.

We were unable to determine whether breed influenced percent change in pulmonary area, but a diverse group of healthy dogs was analyzed. Certain breeds are predisposed to ILDs and specific reference intervals might need to be established for a breed such as the West Highland Terrier, which frequently develops pulmonary fibrosis.[29] In people, ethnicity is considered a predictor of lung function. Reference values for spirometric measurements are calculated by ethnic correction factors, and recent recommendations have suggested basing lung function measures on genetic ancestry for greater accuracy.[30] Additional studies focusing upon percent change in pulmonary area in specific breeds would be necessary to determine if this factor must be considered an influential variable.

Additional limitations of this study need to be addressed. First, we were able to recruit only 50 dogs to this study in order to complete analyses within the confines of equipment availability and the time frame of the study. More importantly, we used a 2-dimensional assessment of area change to evaluate a 3-dimensional organ, and the use of these measurements to approximate compliance, which is the change in volume for a given change in pressure, is impossible to predict. Studies in experimental subjects that directly compare fluoroscopic assessment with pulmonary function measurements in anesthetized animals are required to validate the precision of this method. Correlation with less invasive methodology, such as whole-body plethysmography and tidal breathing flow volume loops, also should be considered.

Conclusion

We propose a reference range for percent change in thoracic and pulmonary area in healthy dogs and recommend further evaluation of the hemithorax measurement to identify potential abnormal values that might suggest decreased lung compliance. Both single frame and summated methods are valid, permitting assessment of small and large dogs, but reference ranges are influenced by body size. Future investigations in dogs with respiratory disease are required to determine the degree and clinical applicability of the percent change in pulmonary area for diagnosis, prognosis, and therapeutic monitoring.

Footnotes

[a] EasyDiagnostEleva, Philips Medical Systems, N.A., Bothell, WA

[b] QuickTime, Apple Inc., Cupertino, CA

[c] NIH ImageJ, Bethesda, MD

[d] GraphPad Prism Version 5.0f, San Diego, CA

[e] R, Version 3.4.0, RStudio Inc., Boston, MA

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Conflict of Interest Declaration: Authors declare no conflict of interest.

Off-label Antimicrobial Declaration: Authors declare no off-label use of antimicrobials.

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