Pulmonary Parenchymal Tissue Volume Measurements in Graded Degrees of Pulmonary Edema in Dogs

By Frederick L. Glauser, Archie F. Wilson, Linn Carothers, Judy Higi, David White, and James Davis

ABSTRACT

We investigated the accuracy and the sensitivity of a modification of the acetylene inhalation technique for the determination of lung tissue volume (Vt) during various grades of hemodynamic pulmonary edema in 23 dogs. After base-line acetylene measurements were obtained, intravascular driving force (pulmonary wedge pressure minus intravascular colloid osmotic pressure) was varied between -8 and +17 mm Hg by the inflation of an intra-aortic balloon and the infusion of isotonic saline. After 30 minutes at this new driving force, four timed acetylene samples were again collected. Vt (when factored by alveolar volume, V A) increased from base line to 0.23 ± 0.07 ml/ml between a driving force of 0 and +17 mm Hg. This same change in Vt/V A was accompanied by an increase in the lung wet weight-dry weight ratio from 3.84 ± 0.31 to 5.2 ± 0.25. Vt was 271 ± 57 ml compared with an actual lung wet weight of 288 ± 57 g; Vt tended to overestimate lung wet weight in severe pulmonary edema. Alloxan-induced pulmonary edema (6 dogs) tended to parallel these data. We conclude that the acetylene method may be a relatively accurate noninvasive method for the determination of increasing lung water in pulmonary edema.

KEY WORDS acetylene inhalation pulmonary wedge pressure colloidal osmotic pressure lung wet weight-dry weight ratio lung water

A multitude of pathophysiological events can occur simultaneously or sequentially during the course of acute hemodynamic pulmonary edema. These events include increased pulmonary blood volume (1), extra-alveolar vessel fluid accumulation particularly in the dependent portions of the lung (2), reversal of pulmonary blood flow (3), interstitial edema accumulation affecting the alveolar-capillary membrane (4), fluid adherence at the angles of the alveolar walls, and, finally, alveolar flooding (2, 4). Objective quantification of these changes can be made by observing decreased pulmonary compliance (static or frequency dependent) (5), abnormal regional ventilation-perfusion imaging (6), increased small airway resistance (7, 8), other abnormal pulmonary function studies (5), chest x-ray changes (3), abnormalities of arterial blood gases (9, 10), abnormal double indicator-dilution studies employing either tritiated water and radioactive albumin (11) or heat and saline conductivity (12). All of these methods except the last two are nonspecific, since many pathological conditions other than pulmonary edema can also produce similar abnormalities. Although the latter two methods measure lung water, they are both invasive. Moreover, the technique utilizing tritiated water is technically difficult, employs radioisotopes, and requires blood withdrawal (13). The heat conductivity method obviates many of these difficulties but still requires extensive instrumentation.

We have previously investigated the feasibility of employing a modification of the acetylene (C 2H 2) method of Cander and Forster (14) for determination of pulmonary parenchymal tissue volume during the development of pulmonary edema. In that study we noted that fluid accumulation could be detected by the acetylene method when pulmonary wedge pressure (PWP) exceeded intravascular colloid osmotic pressure (COP) by approximately 5 mm Hg.

Measurement of lung water over a wide range of intravascular driving forces (driving force = PWP - COP) would be of great interest, since in recent years it has become apparent that earlier concepts regarding pulmonary fluid accumulation in hemodynamic pulmonary edema are oversimplified. Application of Starling's law of water movement to the lung suggests that fluid should transudate from pulmonary capillaries when intravascular hydrostatic pressure becomes larger than intravascular COP (normally 25 mm Hg), i.e., not until left atrial pressure or PWP (as a reflection of pulmo-
nary capillary pressure) exceeds 25 mm Hg should fluid movement occur (15). Recent studies now seem to indicate that, because of the complex interplay between interstitial pressures and lymphatic factors, fluid movement can occur at much lower values of left atrial pressure (16, 17). Indeed, in the experimental animal, peribronchial interstitial fluid accumulation has been reported to occur at left atrial pressures as low as 15 mm Hg (7). Once the critical pressure is obtained, parenchymal fluid accumulation occurs at an ever-increasing rate. Because of the potential utility of the acetylene method as a noninvasive technique for quantifying pulmonary edema, we examined lung water accumulation over a wide range of intravascular driving forces to further evaluate both the validity of the method and the details of pulmonary water accumulation under these conditions.

**Methods**

Thirty mongrel dogs were lightly anesthetized with sodium pentobarbital (30 mg/kg, iv) and placed in the supine position. The dogs were divided into the following groups. Group 1 consisted of one 25-kg dog without pulmonary edema. Eleven individual lung tissue volume determinations were made on five separate occasions to assess the accuracy and the reproducibility of the acetylene technique for normal lungs. At the end of the experimental period the dog was killed, and the lung wet weight–dry weight ratio was obtained.

In group 2, 23 mongrel dogs were subjected to hemodynamic pulmonary edema. A polyethylene endotracheal tube was inserted in each dog. The dogs were allowed to breathe spontaneously except when their lungs were periodically hyperinflated with air at 20 cm H₂O via a Bird Mark VII ventilator. Several intravascular catheters were placed through skin incisions: a no. 7 Fogarty balloon catheter was advanced through one femoral artery into the descending aorta to the level of the diaphragm, a no. 5 Swan-Ganz flow-directed balloon catheter was inserted under pressure monitoring into the pulmonary wedge position, and no. 260 PE tubing was pushed through the common carotid artery into the arch of the aorta.

Base-line acetylene lung tissue volume measurements were obtained, and then PWP was elevated by simultaneously inflating the Fogarty balloon catheter and infusing 0.9% saline into the carotid artery catheter. COP was determined every 10–15 minutes. A spectrum of intravascular driving force values from approximately −8 to +17 mm Hg was obtained in this manner, although individual dogs were exposed to only one driving force. When the desired driving force was reached, 30 minutes was allowed before four timed acetylene samples were obtained. Following this last sample, the lungs were removed and the wet weight–dry weight ratio obtained.

In group 3, six dogs were subjected to alloxan-induced nonhemodynamic pulmonary edema. The dogs had 65–200 mg/kg of alloxan injected rapidly through a no. 260 PE jugular venous line into their right atrium following a base-line determination of lung tissue volume. Then, 15–30 minutes after the alloxan injection, lung tissue volume was again determined. The dogs were killed, and the lung wet weight–dry weight ratio was obtained.

**SPECIFIC PROCEDURES**

**Determination of Pulmonary Parenchymal Tissue Volume.**—Two 1-liter plastic syringes were connected to the endotracheal tube through a large-bore three-way vinyl adapter with vinyl tubing. Maximal expiration was obtained with 15–20 mm Hg of negative pressure with one syringe. The lungs were then rapidly inflated with 20–40 mm Hg of positive pressure with the second syringe, which had been previously flushed and filled with a mixture of 0.80% acetylene, 0.93% neon, 19.5% oxygen, and the balance nitrogen. At the end of approximately six seconds of breath holding, 200 ml of dead-space gas was withdrawn into one syringe, and immediately thereafter (about 8 seconds of elapsed time) the remainder of the expire was obtained with negative pressure employing the other syringe. The collection time of the alveolar sample was approximately 0.5 seconds. The exact timing was obtained as follows. A side opening on the three-way vinyl adapter was connected through no. 260 PE tubing to a Statham P23db pressure transducer connected in turn to an Electronics-for-Medicine VR6 physiological recorder. At the time of lung inflation and deflation, pressure changes were recorded on time-calibrated photosensitive paper. Time of breath holding was taken from beginning of inspiration to end of collection of the alveolar sample.

Samples were also obtained with breath-holding times of approximately 10, 12, and 16 seconds. A rest period of at least 10 minutes was allowed between samples; the total time required to obtained one series of samples was 30 minutes. Alveolar samples were collected in vinyl bags. Since rubber absorbs acetylene, rubber was replaced with vinyl in all parts of the system that had contact with the gas.

**Determination of Lung Wet Weight–Dry Weight Ratios.**—Following collection of the last acetylene sample, the dogs were mechanically ventilated and the chest entered through a sternum-splitting incision. All of the blood vessels supplying the lungs were simultaneously clamped and the lungs were removed within 10 minutes. Excess tissue was removed, blood drained, and the lungs weighed. The lungs were then inflated to full size with a continuous flow of compressed air through the trachea. One week later the dried lungs were removed and reweighed, and the wet weight–dry weight ratio was calculated. Normal wet weight–dry weight ratios for this laboratory are 3.8 ± 0.25.

**Specific Measurements.**—Acetylene and neon concentrations were measured with a Loenco AD2000 gas chromatograph. Pulmonary tissue volume was calculated from the intercept of the fractional alveolar concentration of acetylene vs. time plot utilizing the method of Cander and Forster (14). Alveolar volume (Vₐ) was determined by the single-breath neon method (18). Plasma colloid osmotic pressures were calculated from plasma protein refractometer readings taken before and after saline infusions and referred to a standard total protein–osmotic pressure curve (19). Data were analyzed for significant changes by standard linear regression and tests of significance (20).
TABLE 1

Reproducibility Measurements (Group 1)

<table>
<thead>
<tr>
<th>Exp. no.</th>
<th>Vt (ml)</th>
<th>Vt/VA (ml/ml)</th>
<th>Intercept</th>
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<td>3</td>
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<tr>
<td></td>
<td>283</td>
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<td>0.875</td>
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Vt = pulmonary tissue volume, and Vt/VA = pulmonary tissue volume divided by alveolar volume.

Results

GROUP 1

Table 1 gives the individual data on the eleven experiments in this dog. The mean pulmonary parenchymal tissue volume (Vt) was 230 ± 35 ml, and Vt/VA was 0.14 ± 0.02 ml/ml. The actual lung wet weight was 241 g (not significantly different compared with Vt), and the wet weight/dry weight ratio was 4.1.

GROUP 2

Mean data for the 23 experimental dogs in group 2 are presented in Table 2. The dogs in this group were divided into two subgroups: those with an intravascular driving force of 0 mm Hg or less (group 2A) and those with an intravascular driving force of more than 0 mm Hg (group 2B). Intravascular driving force varied from -8 to +17 mm Hg because of changes in both PWP and intravascular COP. Individual intravascular driving force values were compared with associated changes in Vt and lung weight.

Base-line normal prepulmonary edema data for all dogs in group 2 revealed a mean dog weight of 19.5 ± 3.5 kg with an actual Vt of 207 ± 64 ml and a mean Vt/VA of 0.147 ± 0.047 ml/ml. Base-line PWP was 4.6 ± 0.1 mm Hg with an intravascular COP of 26.2 ± 2.7 mm Hg, giving a net intravascular base-line driving force of -21.6 ± 1.75 mm Hg.

After pulmonary edema had been induced in group 2 dogs, Vt averaged 271 ± 57 ml and actual lung weight averaged 228 ± 57 g (P > 0.05). It is apparent that, although the overlap of values was great, and the acetylene method tended to overestimate the actual lung weights in pulmonary edema, Vt measured by the acetylene method gave a base-line normal pulmonary edema data for all dogs in group 2 revealed a mean dog weight of 19.5 ± 3.5 kg with an actual Vt of 207 ± 64 ml and a mean Vt/VA of 0.147 ± 0.047 ml/ml. Base-line PWP was 4.6 ± 0.1 mm Hg with an intravascular COP of 26.2 ± 2.7 mm Hg, giving a net intravascular base-line driving force of -21.6 ± 1.75 mm Hg.

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reasonable approximation of actual lung weights. Vt (ml) correlated significantly ($r = 0.44, P < 0.05$) though poorly with the wet weights (g) of the edematous lungs (Fig. 1). Although there was a statistically significant correlation, Vt tended to underestimate slightly the actual wet weights of edematous lungs at low weight levels and to overestimate the wet weights at higher weight levels. Since the lower lung wet weights were associated with wet weight–dry weight ratios of 4.3 or less and intravascular driving force values of less than 0 mm Hg, pulmonary edema was either not present or minimal at these values.

$V_t/V_A$ increased linearly with increasing driving force (Fig. 2) once PWP equaled or exceeded intravascular COP; Vt was divided by alveolar volume, $V_A$, to allow comparison between lungs of different sizes. Factoring by $V_A$ also helped to improve the statistical variation of the data. Since $V_t/V_A$ varied linearly with the lung wet weight–dry weight ratio (Fig. 3), it is not surprising that Vt compared favorably with the actual lung weights. As noted in Table 2, when dogs with an intravascular driving force of more than 0 mm Hg were compared with those with an intravascular driving force of 0 mm Hg or less, there was a statistically significant difference ($P < 0.001$) in all of the following categories: pulmonary edema PWP, driving force, pulmonary edema Vt, $\Delta V_t/V_A$, lung weight, and lung wet weight–dry weight ratio.

**Discussion**

A rapid noninvasive technique is needed for the quantification and detection of pulmonary edema at the clinical level. On a theoretical basis, the acetylene method of Cander and Forster (14) would seem ideally suited to meet this need. Because of the rapid solubility of acetylene in lung tissues, including supporting structures, bronchi, and fine interstitial tissue, a certain amount of acetylene is rapidly removed from the inspired gas. The remainder of the gas is more slowly but continuously absorbed into pulmonary capillary blood; the gas disappears at a rate dependent on the rapidity of blood flow. Therefore, as Cander and Forster (14) have pointed out, the slope of the plot of fractional alveolar concentration of acetylene vs. time is proportional to pulmonary capillary blood flow, and the intercept is proportional to lung tissue volume (Vt). Depression of the intercept point indicates greater absorption of acetylene by lung tissue and thus an increase in Vt. Since acetylene is absorbed almost equally by both tissue and...
LUNG TISSUE VOLUME IN PULMONARY EDEMA

increases in Vt/Va with increases in the lung wet weight-dry weight ratio. A statistically significant relationship exists. The broken lines represent the standard error of the estimate. See Figure 2 for definitions of Vt and Va.

Increases in fluid accumulation of fluid during development of pulmonary edema should be associated with greater absorption of acetylene.

In the present study, we attempted to determine the sensitivity and the accuracy of the acetylene method by studying hemodynamic pulmonary edema at various values of intravascular driving force. In this way, we intended to investigate several degrees of pulmonary edema varying from mild to severe. We found that once intravascular driving force exceeded zero, Vt/Va increased in a highly predictable manner with increasing driving force (Fig. 2). Vt/Va increased in a similar fashion compared with the lung wet weight-dry weight ratio (Fig. 3). This observation is consistent with the results of other investigators who have utilized different methods and emphasizes the validity of the acetylene method for monitoring changes in pulmonary fluid accumulation during pulmonary edema (21-23).

We were somewhat surprised that the acetylene method was unable to detect pulmonary edema below an intravascular driving force of zero. Since we calculated our intravascular driving force as PWP (mm Hg) - intravascular COP (mm Hg), lung interstitial pressure was neglected. Because interstitial fluid pressure may be negative (in these dogs in the supine position, it is probably about -5 mm Hg), this force acting in concert with PWP (hydrostatic pressure) tends to draw fluid from the intravascular compartment. Therefore, interstitial water accumulation should occur to some degree before PWP exceeds intravascular COP. Our inability to detect this early accumulation of lung water may be explained by some combination of the following: the inherent accuracy (or inaccuracy) of the acetylene method, the technique as employed in dogs, the fact that lung interstitial pressure is so minimal in the supine position that its effect was overlooked, or the possibility that we did not produce minimal edema with our procedure. In defense of the acetylene method, it should be noted that lung wet weight-dry weight ratios also increased only at driving forces greater than 0 mm Hg.

ACCURACY OF THE ACETYLENE METHOD

The accuracy of the acetylene method in detecting increasing fluid accumulation secondary to pulmonary edema is dependent on many factors. Cander and Forster (14) have discussed in detail the difficulties inherent in this method when it is applied to normal Vt and capillary flow. In the present study, in the base-line normal dog (no pulmonary edema present) the variability for the acetylene method was ±14% (SD). In the 6 dogs with alloxan-induced pulmonary edema, mean Vt was 391 ± 191.5 ml and actual lung wet weight was 298 ± 101 g; in the 23 dogs with hemodynamic pulmonary edema, the respective values were 271 ± 57 ml and 228 ± 57 g; i.e., there was no statistically significant difference between Vt and wet weight in either group.

However, when Figure 1 is examined, it is apparent that at low lung wet weights, the acetylene method underestimates and at high lung weights (above 225 g) overestimates true lung weight (this situation was also found in dogs with alloxan-induced pulmonary edema). Review of our previous experiments reveals the same findings: in eight normal dogs, Vt underestimated true lung weight by 33% and during pulmonary edema (intravascular driving force approximating +5 mm Hg) overestimated true lung weight by 30%. Underestimation under normal conditions is not surprising considering the fact that several structures would not be measured by acetylene; these include large blood vessels, blood retained in the drained lung, and the nonsuperficial tissues of the airways. As pulmonary edema develops, the following causes of overestimation may be operative. (1) Pressures in the range of 80 mm Hg are needed to inflate the lungs as pulmonary edema develops and the lungs become less compliant. Since absorption of acetylene is dependent on alveolar pressure, an increased absorption of approximately 5% relative to normal would be expected. (2) As pulmo-
nary edema becomes more severe, both pulmonary blood volume and lung water increase. During the process of lung removal from the chest, most of the pulmonary blood volume and some of the lung water is lost. This loss tends to decrease the actual lung weights. (3) The coefficient of acetylene partition may change with development of pulmonary edema. An increase in this parameter would increase apparent Vt.

ADVANTAGES OF THE ACETYLENE METHOD

Lung water can also be determined quantitatively by indicator-dilution techniques utilizing either isotopes (13) or heat and salt conductivity (12). Both procedures necessitate sophisticated equipment and invasive techniques, and the first procedure also has the disadvantage of blood withdrawal, multiple samples, and radioisotopes. In addition, these techniques also measure pulmonary blood volume; thus, an increase in this compartment can be misinterpreted as an actual increase in true lung water.

The acetylene method obviates these disadvantages by being somewhat less sophisticated (thus employable in more hospitals), noninvasive, and relatively easy to perform. Meticulous attention to detail is needed, and the patient must be able to hold his breath for 5-20 seconds with an open glottis and be able to tolerate a procedure that lasts 30-40 minutes. The acetylene method does not depend on actual pulmonary capillary blood flow as do indicator-dilution methods but does depend on distribution of inspired ventilation. In pulmonary edema, this distribution will be abnormal and the acetylene method thus may suffer from the same problem as the indicator-dilution methods. Factoring Vt by V, may partially obviate this problem.

As noted earlier in this paper, the indicator-dilution methods will detect any increases in pulmonary blood flow. Our data in the six dogs with allooxan-induced pulmonary edema (in which pulmonary blood volume should not have increased) indicate that the acetylene method may not have this problem, since the mean values for Vt/V, compared with the lung wet weight: dry weight ratio and for Vt compared with actual lung wet weight in grams were essentially identical to those obtained in dogs with hemodynamic pulmonary edema.

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References

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