Regional CO₂ tension quantitatively mediates homeostatic redistribution of ventilation following acute pulmonary thromboembolism in pigs

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Tsang JY, Lamm WJ, Swenson ER. Regional CO₂ tension quantitatively mediates homeostatic redistribution of ventilation following acute pulmonary thromboembolism in pigs. J Appl Physiol 107: 755–762, 2009. First published July 16, 2009; doi:10.1152/japplphysiol.00245.2009.—Previous studies reported that regional CO₂ tension might affect regional ventilation (V˙) following acute pulmonary thromboembolism (APTE). We investigated the pathophysiology and magnitude of these changes. Eight anesthetized and ventilated piglets received autologous clots at time 0 min until mean pulmonary artery pressure was 2.5 times baseline. The fluorescent microsphere (FMS) technique allows separation of regional CO₂ tension and ventilation (V˙) and perfusion (Q˙) at four different times (5 min, 30, 60, 120 min) was mapped by fluorescent microspheres. Regional V˙ and Q˙ were examined postmortem by sectioning the air-dried lung into 900–1,000 samples of ~2 cm³ each. After the redistribution of regional Q˙ by APTE, but in the scenario assuming that no V˙ shift had yet occurred, CO₂ tension in different lung regions at 30 min post-APTE (PₓCO₂) was estimated from the V/Q data and divided into four distinct clusters: i.e., PₓCO₂ < 10 Torr; 10 < PₓCO₂ < 25 Torr; 25 < PₓCO₂ < 50 Torr; PₓCO₂ > 50 Torr. Our data showed that the clusters in higher V/Q regions (with a PₓCO₂ < 25 Torr) received ~35% less V˙ when measured within 30 min of APTE, whereas, in contrast, the lower V/Q regions showed no statistically significant increases in their V˙. However, after 30 min, there was minimal further redistribution of V˙. We conclude that there are significant compensatory V˙ shifts out of regions of low CO₂ tension soon following APTE, and that these variations in regional CO₂ tension, which initiate CO₂-dependent changes in airway resistance and lung parenchymal compliance, can lead to improved gas exchange.

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Previous investigators reported that temporary unilateral pulmonary artery occlusion by inflation of a balloon-tipped pulmonary artery catheter resulted in partial redistribution of ventilation (V) to the unobstructed side (16, 33), and that such changes were prevented by inhalation of CO₂ into the vascularly obstructed lung (23, 26). The proposed mechanism was presumably hypocapnic bronchoconstriction and reduction in parenchymal compliance (pneumocostriction) in the obstructed lung in which local airway narrowing and increased tissue resistance were mediated by the lowered regional CO₂ tension (6, 7, 24). In the contralateral lung, elevation in CO₂ led to bronchodilation and increased parenchymal compliance (pneumodilation). The subsequent rise in tissue pH in these hypocapnic regions was mainly due to the decrease in blood flow (Q) and CO₂ delivery. Evidence that carbonic anhydrase inhibition by acetazolamide, given to slow the change in tissue pH during vascular occlusion, also slowed the rate of V˙ shift further supported the plausible concept of local regulation of V˙ by CO₂ (27). As a result of such regional regulation, the magnitude of ventilation-perfusion (V/Q) mismatch and gas exchange impairment arising from vascular obstruction became partially reduced because of these intrinsic homeostatic mechanisms.

Using a two-compartment model, the estimation for the redistribution of regional V˙ was based on the idea that the entirety of nonperfused lung had become dead space. Therefore, if and when the actual decrease in alveolar V˙ (V˙A), estimated by arterial and exhaled CO₂, was less than that predicted by the mass of hypoperfused lung following the vascular obstruction, any difference in effective V˙A was attributed to the local ventilatory shifts away from the hypoperfused side toward the better perfused side (15). These findings were also confirmed by Allgood et al. (1) and Swenson et al. (27) using radioactive isotope labeling techniques of V˙, showing that complete vascular balloon occlusion could rapidly reduce regional V˙ by roughly one-third with a half time of 75 s.

Leyv and Simmons (14) also reported that there were shifts in V˙ between different regions of the lung after acute pulmonary thromboembolism (APTE), in which large blood clots were scattered within the embolized lung, creating many hypoperfused areas. They found that regions of higher V/Q and thus lower regional CO₂ tension would redistribute V˙ away to the lower V/Q regions, and that these changes could be attenuated by a pretreatment with heparin or simultaneous inhalation of CO₂. However, their method in labeling regional Q˙ was based on relatively gross approximation postmortem, and their estimation on regional V˙ was not obtained by direct measurements. Consequently, proper quantification of these changes at different times following APTE was limited. However, other investigators showed that acute pulmonary microembolism induced by polystyrene beads of different size and load did not result in significant global change in V heterogeneity, using the multiple-breath helium washout technique (31). It should be noted that changes in regional V˙ following APTE were unlikely to be caused by other mechanical factors, such as de novo obstruction of the airway by edema fluid or other secretions.

The fluorescent microsphere (FMS) technique allows separate mapping of regional V˙ and Q˙ in high resolution (2, 3) by direct aerosolization and intravenous infusion of these beads into very small lung regions, down to the size of 2 cm³, and with amounts insufficient to cause measurable changes in airway or vascular resistance (8, 18). Using this method, we were able to characterize the value of V/Q in all lung regions.

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and their anatomic relationship with the thromboemboli, as well as their regional PCO2 concentration (2). Our laboratory’s earlier study showed that V/Q mismatch after acute pulmonary embolism was mostly due to the divergent regional Q created by the mechanical obstruction of the thromboemboli in the pulmonary vasculature (30). The higher V/Q regions were located in the periphery of the lung, where there was reduced Q distal to the thromboemboli, while the lower V/Q regions were located in the less or unembolized regions. Along with the others, we observed that there were dynamic changes in V/Q distribution after the initial embolic injury (5).

In the present study, we were interested to test the hypothesis that there was indeed significant redistribution of V following APTE between the lung regions of varying CO2 tension and whether there was a physiological threshold in regional CO2 tension that quantified these changes.

METHODS

Surgical Preparations and Physiological Measurements

The experimental protocol was approved by the University of Washington Animal Care Committee. Eight piglets (23 ± 3 kg) were premedicated with ketamine (20 mg/kg im) and xylazine (2 mg/kg im) and a bolus of thiopental sodium (20 mg/kg iv). They were maintained under general anesthetic for the entire experiment using intravenous pentothal, initially set at 100 mg/h, and the dose was only occasionally titrated afterwards. These animals were ventilated with room air in the supine posture, while the tracheostomy and vascular line insertions were completed.

One femoral arterial line and two femoral venous lines were inserted for the purposes of monitoring systemic mean blood pressure (MBP), fluid infusion, and FMS injection, respectively. A Swan-Ganz catheter (Edwards Laboratory) was inserted in the right external jugular vein for the measurement of pulmonary arterial pressure (Ppa), wedge pressure, and cardiac output (Qt) using the thermodilution technique, while a large-bore catheter (0.5-cm internal diameter) was inserted in the left external jugular vein for the rapid infusion of preformed blood clots (see below). Generally, these animals received normal saline at 100 ml/h during the experiments. They were kept warm using a warming blanket so that the body temperature was maintained at ~38°C. No heparin was used.

After the insertion of the femoral arterial line, 80 ml of blood were withdrawn and mixed with 2,500 units of thrombin-JMI at room temperature, so that clots were allowed to form and fibrinized over the next 2 h.

Upon completion of the surgical procedures, the animals were placed in the prone posture and received stacking of at least three consecutive breaths to remove residual atelectasis. Their control ventilatory settings were determined according to the arterial blood-gas results, such that arterial CO2 tension (PaCO2) was set at 35–40 mmHg, tidal volume was 12–15 ml/kg, respiratory rate was 18–20/min, and inspired oxygen was room air. End-tidal CO2 (PetCO2) was also continuously monitored during the experiment. Once these V parameters were established, there was no further adjustment of these settings for the remaining part of the experiment. At each of the subsequent data collection time points, hemodynamic parameters, such as MBP, Ppa, wedge pressure, heart rate, and Qt were measured, along with hemoglobin, arterial and venous blood gases, Fowler dead space (MacLab at 100 mm/s), tidal volume, airway pressure, and respiratory rate.

The time for the induction of APTE was defined as time = 0 min, and all of the events before and following that point would be recorded in relation to that time. Two control runs were performed at −30 and −5 min before APTE to establish the consistency of the animal’s physiological condition at baseline.

In each experiment, eight different FMS of distinct colors (Molecular Probes, Eugene, OR) were chosen and used in random orders: four for V (1 μm) and four for Q (15 μm), i.e., at −5, 30, 60, and 120 min. Due to the overlapping spectra of different FMS colors, an optimal number of eight colors was allowed per experiment in order that the most reliable data could be obtained (22). The details of the FMS aerosolization and injection techniques have been well described (8, 18).

After the control runs were performed and all of the physiological measurements were recorded, APTE was induced at time = 0 min. Approximately 12–16 pieces of preformed fibrinized clots (roughly 1.5 × 0.5 × 0.5 cm3 per piece) were suspended in normal saline in a large catheter tip syringe and injected into the left external jugular vein over the next 10–15 min, until Ppa was 2.5 times the baseline value. On completion, there were no further injections of clots. At time = 30, 60, and 120 min after the induction of APTE, physiological measurements and blood samplings were similarly done as in the control runs, followed by FMS aerosolization and injection using FMS of different colors. This would mark the regional V and Q in the first 2 h following APTE.

Postmortem Lung Preparations

At the end of the experiment, the animals were deeply anesthetized with intravenous pentothal, heparinized with 5,000 units, and exsanguinated. The lungs were extracted after gentle saline flush, suspended vertically, and inflated to no more than 25 cmH2O. The lobes were kept in their resting anatomical position by a small amount of cyanocrylate glue and blown dry with warm air through the lungs for 72 h. Small puncture holes were made to allow good airflow through the lungs during the drying process. The injected thromboemboli were not macerated and were readily seen in the major pulmonary arteries.

After the thorough drying of the harvested lungs, they were sectioned into 2-cm3-sized cubes, with each sample carefully assigned to three-dimensional coordinate, according to a preestablished grid pattern. Approximately 900–1,000 lung pieces were analyzed per animal (see RESULTS). For each lung sample, its spatial location, weight, amount of airway tissue, and the presence or absence of blood clots in arteries >1 mm were recorded.

The fluorescent intensities of all eight FMS embedded in each sample, which marked the regional V and Q at 4 different time points (i.e., time = −5, 30, 60, and 120 min), were measured as previously described (2). Briefly, the fluorescent signal, which reflected the number of a given microsphere trapped in a lung piece, was determined by measuring its intensity in a spectrofluorimeter (Perkin-Elmer LS-50B), following four days of soaking in 2 ml of organic solvent (Cellosolve, Sigma-Aldrich, MO). Overlaps from adjacent colors were then corrected using a matrix inversion method (22).

Data Analysis

Physiological data. Regional V was chosen as the primary variable, since it was the key factor in our hypothesis. Other physiological parameters listed below, such as Ppa, MBP, Qt, arterial PO2, PaCO2, venous Po2, venous PCO2, mean peak airway pressure, and mean dynamic compliance were considered as secondary variables.

Results were expressed as means ± SE, unless indicated otherwise. P < 0.05 was used to designate statistical significance.

Microsphere data. From the fluorescent intensities in each sample, the minute V, and the Fowler dead space, we calculated its regional VA in milliliters per minute and its percentage of the total minute VA. From these data and the sample weight, we could also follow their weight-normalized V in milliliters per minute per gram and their changes in different experimental conditions.

Since large airway and blood vessels added substantially to the weight, pieces designated as containing >20% large airways or vessels, which, on the average, accounted for <10% of the original total number of lung pieces, were omitted from the data set. A value
of 50 was assigned to V/Q for lung pieces with V/Q > 50 (usually due to very low flow, resulting in extremely high V/Q) to include them in our V/Q analysis.

Estimation of regional CO₂ tension. The corresponding regional CO₂ tension of each lung sample can be estimated using the validated method of Altemeier et al. (2) in both the normal and diseased lung. Briefly, the method uses the microsphere V/.Q weight-normalized data for each region to determine its V/Q and effect on respiratory gas exchange. Alveolar tensions of O₂ and CO₂ (Paco₂) and end-capillary O₂ and CO₂ contents for each lung piece are determined by solving mass balance equations for each gas, given that piece’s unique V/Q ratio and the relevant inspired and mixed venous blood gas tensions, temperature, barometric pressure, and hemoglobin concentration. Altemeier et al. (2) have shown that this microsphere-based V/Q analysis predicts global pulmonary gas exchange with high reliability compared with that predicted by the 50 compartmental analyses of the multiple-inert gas exchange elimination technique.

Clustering of lung samples. We allocated the lung samples into four clusters, according to their projected regional Paco₂ (Paco₂) at time = 30 min after APTE, assuming that there was not yet redistribution of regional V occurring and that the extent of vascular obstruction did not change in those first 30 min after APTE (see below). This Paco₂ was determined by the V/Q from the FMS data, taking the measurement of V at −5 min and Q at 30 min. This theoretical Paco₂ (or regional Pco₂ just after embolization but before any V readjustment occurs) establishes the putative regional Pco₂ hypothesized to evoke regional V shifts. Paco₂, using V values at −5 min, is to be contrasted with the real-time Paco₂, which uses the actual real-time regional V values at 30 min. Differences observed between Paco₂ and Pco₂ thus represent the extent to which regional V is altered from its pre-APTE value in response to the decreases in Q in embolized regions and to increases in Q in nonemobilized regions.

The four clusters represented were as follows: cluster 1, which included lung pieces that had Paco₂ < 10 Torr; cluster 2, which included lung pieces that had a Paco₂ between 10 and 25 Torr; cluster 3, which included lung pieces that had a Paco₂ between 25 and 50 Torr; and cluster 4, which included lung pieces that had a Paco₂ > 50 Torr, all at 30 min after APTE. These ranges were chosen to encompass values of Paco₂ in the truly hypocapnic and hypercapnic ranges, below and above which homeostatic mechanisms might begin to be activated, as well as those in the midranges.

Our first measurement post-APTE was feasible only at 30 min, in order that a steady state in hemodynamics and blood gases could be obtained. Swenson et al. (27) found that, with acute nonthrombotic balloon occlusion, V diversion after the cessation of Q, as assessed by regional nuclide (krypton) scanning, was completed in a matter of minutes, which was too fast for us to make any accurate real-time FMS measurements. Thus a more practical way to estimate this projected regional CO₂ tension immediately after APTE but before the completion of V shift was to estimate Paco₂ using V values at −5 min and Q at 30 min from the FMS data.

Once the clusters were defined as stated above, their corresponding Paco₂ at −5, 60, and 120 min, as well as their regional CO₂ tension in real-time (Pco₂), which used the FMS data of V and Q obtained simultaneously, at −5, 30, 60, and 120 min was calculated. Finally, for both the projected and real-time data sets and for each of these four clusters defined above, their median V/Q and their composite Pco₂ (Pco₂, CO₂), which represented the CO₂ tension of blood exiting the lung from that cluster at different time points, were also determined. Median V/Q was used instead of mean V/Q due to a small number of samples with extremely high V/Q.

In testing our hypothesis, we were interested to see if and how this theoretical Paco₂ would differ in all clusters from the real-time Paco₂, when the actual data of V and Q were used simultaneously at 30, 60, and 120 min. In other words, if the real-time Paco₂, calculated by the same mass equation, was higher than that theoretical Paco₂ described above, we would conclude that shifts in regional V had redistributed away from that particular cluster, because Q was held constant at that same time point and was thus eliminated as a confounding variable. On the other hand, if Paco₂ was lower than Paco₂, regional V would have distributed toward it. If they were the same, there would have been no V shift at that same time point. We chose to examine regional CO₂ tension instead of V/Q here, because the former was more central to our hypothesis and had a narrower physiological range.

To quantify the amount of V shifts during the experiments, we summed up all of the regional Va from the same cluster in either data set already defined above, at −5, 30, 60, and 120 min and compared them within the same cluster over time, as well as among the corresponding clusters at the same time. For example, if a given cluster received less V over time, e.g., it had received less V at time = 30 min after APTE than at time = −5 min, regional V in that cluster would have shifted away from it, and vice versa.

Finally, to better explore a possible physiological threshold or relationship between the calculated Paco₂ immediately after APTE and the magnitude and direction of V shift, we refined our stratification of Paco₂ into six groups instead of four, ranging from 0–10, 10–20, 20–30, 30–40, 40–50, and >50 Torr.

RESULTS

Animal Physiological Data

Table 1 shows the hemodynamic and blood-gas changes during all phases of the experiments. The data show that, after APTE, there was a significant increase in Ppa, as one would expect from our experimental protocol, while MBP and Qr remained comparable. After APTE, arterial Pao₂ dropped sig-

Table 1. Animal physiological data

<table>
<thead>
<tr>
<th>Time</th>
<th>30 min</th>
<th>60 min</th>
<th>120 min</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ppa, cmH₂O</td>
<td>22.8±4.7</td>
<td>22.6±4.1</td>
<td>54.8±2.5†</td>
</tr>
<tr>
<td>BP, mmHg</td>
<td>93.5±6.1</td>
<td>96.4±5.4</td>
<td>105.0±12.6</td>
</tr>
<tr>
<td>Qt, l/min</td>
<td>2.82±0.49</td>
<td>2.74±0.46</td>
<td>2.69±0.56</td>
</tr>
<tr>
<td>PaO₂, Torr</td>
<td>108.8±6.0</td>
<td>111.0±5.6</td>
<td>58.6±12.3†</td>
</tr>
<tr>
<td>PacO₂, Torr</td>
<td>36.3±2.6</td>
<td>35.4±2.8</td>
<td>49.0±8.1†</td>
</tr>
<tr>
<td>PacO₂, Torr</td>
<td>36.0±1.6</td>
<td>35.6±2.8</td>
<td>32.4±3.6†</td>
</tr>
<tr>
<td>PacO₂, Torr</td>
<td>40.4±4.2</td>
<td>40.0±4.0</td>
<td>33.1±6.5†</td>
</tr>
<tr>
<td>PacO₂, Torr</td>
<td>43.0±1.9</td>
<td>42.3±3.2</td>
<td>58.9±5.6†</td>
</tr>
<tr>
<td>PacO₂-Paco₂, Torr</td>
<td>0.3±0.7</td>
<td>−0.3±0.4</td>
<td>16.6±9.9†</td>
</tr>
</tbody>
</table>

Values are means ± SD. Acute pulmonary thromboembolism (APTE) is at 0 min. Ppa, pulmonary arterial pressure; BP, systemic blood pressure; Qt, cardiac output; PaO₂ and PacO₂, arterial (a) and mixed venous (v) oxygen tension, respectively; PaCO₂ and Pco₂, arterial (a), and mixed venous (v) carbon dioxide tension, respectively; PacO₂, end-tidal PacO₂; PacO₂-Paco₂, arterial and end-tidal CO₂ gradient. †P < 0.01 at time = −5 min vs. 30 min, based on paired t-test.

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nificantly from ~111.0 to 58.6 Torr, while PaCO₂ increased from 35.4 to 49.0 Torr, as minute V was held constant. PertCO₂ was also reduced from 35.6 to 32.4 Torr, resulting in increased gradient between PaCO₂ and PertCO₂, as previously described (9, 11). In terms of pulmonary mechanics (Table 2), there was also a reduction of the dynamic respiratory system compliance after APTE, and some mild recovery was noted afterwards.

Cluster Analyses

Using cluster analyses described in Methods, four subpopulations of lung regions were identified, according to their theoretical P_{xCO₂} at 30 min, as if no V shift had occurred, and Q at 30 min was measured directly by FMS at that time. Cluster 1 represented regions in which P_{xCO₂} was <10 Torr; cluster 2 represented regions in which P_{xCO₂} was between 10 and 25 Torr; cluster 3 represented regions in which P_{xCO₂} was between 25 and 50 Torr; and cluster 4 represented regions in which P_{xCO₂} was >50 Torr. After the clusters were selected, their corresponding real-time P_{xCO₂} was also determined. For both the real-time and projected data set, their median V/Q, total V, and percentages of total minute V_A, as well as the estimated P_{comCO₂} of the blood exiting the lung from each of these four clusters, were calculated for −5, 30, 60, and 120 min (see Methods).

Table 3 shows the number of pieces in each cluster, as defined a priori by its P_{xCO₂}, as well as its percentage of the total. These four clusters, although not containing an equal number of samples, had sufficient numbers to permit sound statistical analysis.

Table 4 shows the percentage of total V_A and percentage of total Q in each cluster over time. Before APTE at time = −5 min, all clusters had well-matched fractions of total Q and V. After APTE, cluster 1 showed the greatest V shifts immediately, falling almost 50% from 23.3 to 13.4% at 30 min, indicating a considerable early shift of V away from this cluster, which remained relatively stable afterwards. This cluster also had the greatest decrease in Q (from 23.3 to 0.3% of total Q) at 30 min. Over the next 90 min, there was a modest return of Q (from 0.3 to 4.3%), indicative of some modest clot breakdown or movement. Cluster 2 behaved in a similar fashion, but with only about a 25% fall in V at 30 min accompanying a lesser degree of Q interruption compared with cluster 1. Cluster 3, whose P_{xCO₂} spanned the range of 25–50 Torr, had only <10% increase in V at 30 min accompanying its 25% reduction in Q. Cluster 4 demonstrated a 25% increase in V at 30 min, which, thereafter, remained relatively stable, accompanying its over 50% increase in Q after APTE. In addition to the V shifts, the recovery of Q to the low-flow areas (clusters 1 and 2) and reduced Q to cluster 4 certainly also contributed to the gradual normalization in their V/Q over time. The overall recovery of V/Q heterogeneity was due to both V and Q independently, as they were also measured separately.

The percentage of total minute V_A from clusters 1 and 2 was ~36.5% before APTE, compared with 23.7% at 30 min, a reduction of 35%. However, after 30 min, there was minimal further redistribution of V.

Figure 1 shows the changes in median V/Q over time for the four clusters. The dotted lines represent that scenario in which no V shift was projected to have occurred since time = −5 min (using V in the pre-APTE state in the calculation and Q in real time), whereas the solid line represented the observed situation in which both V shifts and repercussion to these clusters occurred simultaneously (using real-time data for both V and Q from the FMS data). The dotted line allowed for the independent assessment of the physiological impact from V shifts alone, since the value of Q was the same, thus eliminating it as a confounding variable. The results show that, in the early phase of APTE, clusters 1 and 2 were the most affected by shifts in V away from them, since even a little change in V would make a substantial difference here because of the low value of Q in the denominator of their V/Q. The median V/Q of clusters 1 and 2 was much higher in the dotted line for P_{xCO₂} compared with the solid line for P_{xCO₂}, illustrating the difference due to the shifts. While there was statistical significant difference in the median V/Q in cluster 2, as one would expect, such difference was not seen in cluster 1. This result could be explained by the artificial capping of maximal V/Q at 50 (see Methods) and the large standard errors in cluster 1 because all samples were included. Because of this, the difference in V/Q matching with or without shift in V in clusters 1 and 2 might be better demonstrated by P_{comCO₂} (Fig. 2), where the standard errors were smaller. In cluster 3, V/Q was unaffected by any V shifts, as it occupied the more normal physiological range. Cluster 4, which represented the unembolized units that received more diverted Q after APTE, was...
Table 4. Percentages of total minute alveolar ventilation and total pulmonary blood flow in clusters 1–4 over time

<table>
<thead>
<tr>
<th>%Total alveolar ventilation</th>
<th>5 min</th>
<th>30 min</th>
<th>60 min</th>
<th>120 min</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cluster 1 P_{\text{a}}CO_2 &lt; 10</td>
<td>23.3±5.6</td>
<td>13.4±6.8</td>
<td>13.2±5.0</td>
<td>13.9±5.1</td>
</tr>
<tr>
<td>Cluster 2 10 &lt; P_{\text{a}}CO_2 &lt; 25</td>
<td>13.2±5.2</td>
<td>10.3±4.2</td>
<td>10.6±4.2</td>
<td>11.4±4.7</td>
</tr>
<tr>
<td>Cluster 3 25 &lt; P_{\text{a}}CO_2 &lt; 50</td>
<td>29.7±7.4</td>
<td>32.3±8.1</td>
<td>32.7±8.1</td>
<td>32.4±8.1</td>
</tr>
<tr>
<td>Cluster 4 P_{\text{a}}CO_2 &gt; 50</td>
<td>33.8±11.5</td>
<td>43.9±12.1</td>
<td>43.5±11.4</td>
<td>42.3±12.1</td>
</tr>
</tbody>
</table>

%Total perfusion

<table>
<thead>
<tr>
<th>%Total perfusion</th>
<th>5 min</th>
<th>30 min</th>
<th>60 min</th>
<th>120 min</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cluster 1 P_{\text{a}}CO_2 &lt; 10</td>
<td>23.3±5.8</td>
<td>0.3±0.1</td>
<td>2.6±1.8</td>
<td>4.3±2.7</td>
</tr>
<tr>
<td>Cluster 2 10 &lt; P_{\text{a}}CO_2 &lt; 25</td>
<td>12.9±5.5</td>
<td>1.5±0.7</td>
<td>3.8±1.9</td>
<td>5.5±3.0</td>
</tr>
<tr>
<td>Cluster 3 25 &lt; P_{\text{a}}CO_2 &lt; 50</td>
<td>28.8±7.7</td>
<td>21.0±12.2</td>
<td>23.4±9.3</td>
<td>27.1±10.1</td>
</tr>
<tr>
<td>Cluster 4 P_{\text{a}}CO_2 &gt; 50</td>
<td>35.0±11.7</td>
<td>77.2±12.6</td>
<td>70.2±8.7</td>
<td>63.1±10.5</td>
</tr>
</tbody>
</table>

Values are means ± SD in percent.

also affected modestly by shifts in $V$ toward them as a result of the decreases in $V$ to clusters 1 and 2. But this additional $V$ had less impact on its overall V/Q because of the higher value of $Q$ in the V/Q denominator.

In analyzing the median V/Q from 30 to 120 min (in both dotted and solid lines), they all changed toward their pre-APTE value as clusters 1 and 2 gradually received more Q over time and cluster 4 gave up some of the diverted Q (Table 4), presumably when some clot in situ might have moved or lysed.

Figure 2 shows the $P_{\text{com}}CO_2$ from each cluster plotted over time. Again, the dotted lines represented the projected scenario in which no V shift had occurred since time = −5 min, using the $P_{\text{com}}CO_2$ data, whereas the solid line represented the real-time data (P_{\text{com}}CO_2), when V shifts had already occurred. The $P_{\text{com}}CO_2$ from cluster 1 was estimated to be only ~3 Torr, if no shift in V took place, but was significantly increased to 13 Torr after the shifts. Similarly, cluster 2 behaved like cluster 1, but to a lesser degree. Cluster 3 remained relatively unaffected due to its $P_{\text{com}}CO_2$ in the relatively normal range. Cluster 4 showed a slightly lower $P_{\text{com}}CO_2$ derived from $P_{\text{com}}CO_2$, as one would expect due to the increase in V, resulting from the diversion of V. All clusters recovered toward their pre-APTE value as reperefusion took place. Therefore, without the concomitant V shifts (dotted lines), the median V/Q in the hypocapnic clusters (clusters 1 and 2) would have been much higher (Fig. 1), and the corresponding regional CO2 tension much lower (Fig. 2), especially in the first 30 min.

In short, the data of $P_{\text{com}}CO_2$ vs. $P_{\text{com}}CO_2$ at 30 min in Fig. 2 showed the impact of V shifts that took place early, while the same data of $P_{\text{com}}CO_2$ and $P_{\text{com}}CO_2$ over time showed the impact of both V and Q, with the latter becoming much more significant after the initial phase.

Figure 3 plots the change in weight-normalized regional $V_A$ at 30 min post-APTE against six subgroups defined by their calculated $P_{\text{com}}CO_2$ at time of embolization, spanning a range from 0 to 65 Torr. Each range was compared statistically with the one representing $P_{\text{com}}CO_2$ at 30–40 Torr. It was evident that there was a progressive redistribution of $V_A$ away from regions of low $P_{\text{com}}CO_2$. However, the increase in V toward those with higher $P_{\text{com}}CO_2$ was not significant. On statistical analysis of the data with a highly conservative Bonferroni correction at $P < 0.01$, the threshold for the redistribution of V appeared to be <20 Torr.

DISCUSSION

We used a clinically relevant experimental model of APTE and a high-resolution V/Q mapping method to study how changes in regional CO2 tension might affect regional V. Our results showed that the marked dispersion of regional V/Q that occurred soon after APTE was partially restored toward normal values within 30 min by compensatory regional V shifts and that the extent of these V shifts correlated well with changes in the regional CO2 estimated by the V/Q data. Our observations
Further supported the mechanisms of hypocapnic bronchoconstriction and/or pneumoconstriction in the higher V/Q regions, acting to reduce V in the more embolized areas (Fig. 3).

Our model of APTE used to study V/Q relationships and gas exchange is a well-characterized model that mimics many features of clinical pulmonary thrombembolism, including features such as significant hypoxemia, increases in V/Q heterogeneity, pulmonary hypertension, reduction in dynamic respiratory system compliance, and, in the case of controlled V, the development of hypercapnia (Table 1). Furthermore, in the course of the first 2 h after APTE, there is a dynamic evolution of physiological changes that do not similarly occur with other models of vascular obstruction, such as intravascular balloon occlusion or embolization by inert microspheres. Mapping of regional V and Q by FMS yields both regional and global information over time and permits accurate calculation of regional gas tensions to estimate how closely changes in V are linked to the changes in regional Pco2.

Our study is the first to show directly that, after APTE (as opposed to large-vessel balloon occlusion or inert beads), alteration in regional Pco2 correlates quantitatively with the changes in regional V. We expanded on the earlier work by Levy et al. (15), who used a contrast dye postmortem in low resolution to measure changes in regional Q after thrombembolism and found that the calculated PaCO2-PetCO2 difference predicted on the basis of volumetric loss of perfused lung tissue was lower than expected. They subsequently deduced that there had to be some changes in regional V to account for the lower estimated physiological dead space. This was later confirmed by Vidal-Melo et al. (34), who used positron emission tomography to mark changes in regional V in APTE at 1 h and demonstrated that V decreased by an average of 25% in embolized regions and increased by ~10% in perfused areas. They did not, however, have any means to estimate regional Pco2 in relation to the magnitude or direction of V shifts.

We reported that essentially all of the V redistribution and changes in regional CO2 tension occurred within 30 min after APTE (Table 4, Figs. 1 and 2). Thereafter there was normalization toward baseline V/Q values, which was largely due to gradual reperfusion in embolized areas and reduction of Q to the unembolized areas. The lack of subsequent further shifts in V in the next 90 min (Table 4) was likely due to the fact that regional CO2 tension in most of these clusters was no longer below the threshold at ~20 Torr, according to the data we have obtained in Fig. 3.

Our results and those of others with vascular thrombotic obstruction, which showed measurable regional V shifts, differed from those of Altemeier et al. (3), who found no V diversion when 780-µm-sized inert microspheres were injected to obstruct Q, or those of Tsang et al. (30), who found no significant change in V heterogeneity using the multiple-breath helium washout technique when dogs were given 250-µm polystyrene beads. However, the latter data were more global than regional in nature and had lower resolutions in which regional Pco2 was not measured. Their embolization materials, experimental methods, and physiological end points were also different. Finally, the role of vasoactive or bronchoconstrictive mediators under the various experimental conditions could not be readily ruled out.

We find evidence for a bronchoconstrictive and/or pneumoconstrictive effect of lower Pco2. Figure 3 showed the change in V at 30 min post-APTE in relation to Pco2. These results suggested a physiological threshold of ~20 Torr in regional CO2 tension, below which regional V is shifted away significantly, while the rest of the data showed no statistically significant V shifts toward regions with CO2 tension > 50 Torr.

CO2 has potent effects on airway, blood vessel, and lung parenchyma properties that can confer significant V/Q regulation (28). With vascular obstruction, local alveolar, distal airway, and parenchymal Pco2 falls. Parenchymal tissue Pco2 will be dominated by Pco2 changes, which may affect a variety of contractile interstitial cells containing smooth muscle actin-myosin elements in alveolar tissue. These cells, including interstitial myofibroblasts and interstitial cells around pre- and postcapillary vessels (13, 19), may be responsible for the phenomenon of reversible parenchymal mechanical changes induced either by CO2/pH changes (7), methacholine (38), noradrenaline, and angiotensin (20), through their attachments to the interstitial fiber network (36, 38). Changes in Pco2 will also act on distal airway tone via diffusion of CO2 across the airway epithelium to the surrounding bronchial smooth muscle and accompanying distal pulmonary arteries in the bronchovascular bundle (25, 29).

Our data with regard to the potent effect of hypocapnia to decrease V are compatible with those reported by Coon et al. (4), Ingram (10), and Kaise et al. (12), who found that, in dog lungs, airway resistance and static compliance did not change until Pco2 fell very low, e.g., <15–20 Torr. The failure of these studies, along with ours, to observe any CO2-mediated bronchodilation or pneumolysis is at odds with the work of others in either lungs or airway smooth muscle cells (7, 17, 21, 24, 32, 37) who studied higher Pco2 concentrations (i.e., >40 Torr) and found evidence for a continuous dose response of CO2-mediated changes in V, from decreases with low Pco2 to increases with high Pco2. The lack of unanimity with respect to the continuous dose-response characteristics of CO2-mediated...
changes in regional \( V \), bronchomotor tone, and parenchymal compliance remains to be clarified.

It is interesting that, in all experimental models examining the phenomenon of \( Q \)-mediated changes in regional \( V \), it appears that the maximum \( V \) diversion is roughly 30%, even with total loss of \( Q \) into that lung region. This may represent either the true intrinsic maximum effect or the presence of other factors, which, particularly in the live animal with thromboembolism, can oppose or limit these \( V \) shifts. Reinspiration of \( CO_2 \) derived from other lung regions contained within the common dead space and \( CO_2 \) delivery via the bronchial arterial circulation can also potentially act to buffer the full impact of change in \( PaCO_2 \) arising from changes in local pulmonary \( Q \). Additionally, there is the consequence of diffuse bronchoconstriction arising either from systemic hypercapnia (as seen in our experiments, Table 1) and/or release of circulating bronchoactive mediators from platelets and red cells as endogenous lytic mechanisms are activated. The impact of generalized bronchoconstriction will likely be greater on those units attempting to bronchodilate than on those units where the airways are already constricted in the high \( V/Q \) areas. The net result will be that maximal local bronchodilation and increased \( V \) in unembolized areas will be partially restrained, making them less able to receive more \( V \) from embolized regions and, in effect, limiting the ability of embolized regions to redivert their \( V \).

Nonetheless, a few limitations of our methods deserve mention. The marking of regional \( V \) by particulate FMS and not by gases may underestimate the diffusive component of \( V \) in very distal alveolar regions, including \( V \) via bronchiolo-alveolar and interbronchioloalveolar collateral pathways (35). Our first practical time point post-APTE was only at 30 min, in order that a more established steady state in hemodynamics and blood gases could be obtained. Consequently, we were unable to ascertain if the \( V \) shifts had occurred sooner. However, Swenson et al. (27) found that, with vascular balloon occlusion, \( V \) shifts after the cessation of \( Q \), as measured by nuclide scanning, was completed in a matter of minutes. This time frame was too short for us to make any accurate real-time FMS mapping measurements. Thus, to estimate regional \( P_aCO_2 \) immediately after APTE but before the completion of \( V \) shift, we had to use \( V \) values at \(-5 \) min and \( Q \) at 30 min.

Another potential weakness is the assumption that, within the first 30 min after APTE, there was no change in the magnitude of clot obstruction, either by lysis or distal movement. If any relief of vascular obstruction had taken place in the first 30 min, our method would have led to an underestimation of the initial total \( V/Q \) heterogeneity. However, the percentage of total flow in cluster 1, which was the cluster with the most significant \( V \) shifts, remained very low at 0.3% at 30 min (Table 4); thus the extent of change in \( Q \) was likely to remain small during that period of time.

Finally, we did not test directly the influence of changes in regional \( CO_2 \) by adding it into the inspired gas, because we considered it very likely that this maneuver would have introduced more acid base disturbance, hemodynamic or even neurohumoral changes to obscure the analysis. For that matter, we could not exclude other factors related to local neural reflex or release of mediators concomitant with clot lysis that might have additionally contributed to or opposed local \( V \) redistribution. However, we think the key mediator in affecting regional \( V \) remains to be the regional \( CO_2 \) tension. Along with our data, it also makes the best sense from the evolutionary standpoint, as \( CO_2 \) can act both rapidly and locally to reestablish \( V/Q \) homeostasis for improved survival.

As mentioned above, there are potentially other factors that may affect the regional \( CO_2 \) tension and thus the validity of our conclusions, such as collateral \( V \) pathways, reinspiration of dead space gas, \( CO_2 \) delivery via bronchial arterial \( Q \), high solubility of \( CO_2 \) in blood, and diffusive component of \( V \) in the distal airway. However, the main determinant of regional \( PCO_2 \) remains the \( V/Q \).

In conclusion, APTE is a dynamic disease. It causes marked initial regional \( V/Q \) dispersion, which is quickly restored toward normal by compensatory regional \( V \) shifts of roughly 35% from hypoxic regions within the first 30 min and later by gradual reperfusion to the heavily embolized area. The magnitude and direction of these changes in regional \( V \) correlate well with changes in the regional \( CO_2 \) tension estimated near the time of embolization. These findings support the homeostatic roles of hypoxic bronchoconstriction and pulmonary \( O_2 \) diffusion, which facilitate redistribution of \( V \) after APTE to optimize \( V/Q \) matching.

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REFERENCES


