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Pulmonary Blood Flow Redistribution with Low Levels of Positive End-expiratory Pressure

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Background: **Recent studies have questioned the importance of the gravitational model of pulmonary perfusion. Because low levels of positive end-expiratory pressure (PEEP) are commonly used during anesthesia, the authors studied the distribution of pulmonary blood flow with low levels of PEEP using a high spatial resolution technique. They hypothesized that if hydrostatic factors were important in the distribution of pulmonary blood flow, PEEP would redistribute flow to more dependent lung regions.**

Methods: **The effects of zero cm H_2O PEEP and 5 cm H_2O PEEP on pulmonary gas exchange were studied using the multiple inert gas elimination technique; the distribution of pulmonary blood flow, using fluorescent-labeled microspheres, was also investigated in mechanically ventilated, pentobarbital-anesthetized dogs. The lungs were removed, cleared of blood, dried at total lung capacity, and then cubed to obtain approximately 1,000 small pieces of lung (~1.7 cm^3).**

Results: **Positive end-expiratory pressure increased the partial pressure of oxygen by 6 ± 2 mmHg ($P < 0.05$) and reduced all measures of ventilation and perfusion heterogeneity ($P < 0.05$). By reducing flow to nondependent ventral lung regions and increasing flow to dependent dorsal lung regions, PEEP increased ($P < 0.05$) the dorsal-to-ventral gradient. Redistribution of blood flow with PEEP accounted for 7 ± 3%, whereas structural factors accounted for 93 ± 3% of the total variance in blood flow.**

Conclusions: **The increase in dependent-to-nondependent gradient with PEEP is partially consistent with the gravitationally based lung zone model. However, the results emphasize the greater importance of anatomic factors in determining the distribution of pulmonary blood flow. (Key words: Fluorescent microspheres; gravitational gradient; multiple inert gas elimination technique; pulmonary gas exchange.)**

POSITIVE end-expiratory pressure (PEEP) is often used to improve arterial oxygenation in patients. This technique improves pulmonary gas exchange by decreasing intrapulmonary shunt and perfusion of low ventilation-perfusion ($\dot{V}_A$-$\dot{Q}$) by improving the $\dot{V}_A$ of these regions.[1] High levels of PEEP (20 cm H_2O) markedly decrease cardiac output and redistribute blood flow from the core to the periphery and from the nondependent ventral lung to the dependent dorsal lung.[2,3] These changes in pulmonary blood flow traditionally have been interpreted within the context of the gravitational model, where pulmonary blood flow is thought to be distributed passively by the hydrostatic gradient due to gravity.[4-7] However, recent studies using high-resolution techniques have shown that gravity is a minor determinant of the distribution of pulmonary blood flow in animals, including dogs,[8-12] horses,[13-15] and sheep.[16] Newer high-resolution studies in humans have also described gravity-independent[17] and gravity-dependent[18] distributions of blood flow.

The overall dominance of the structure of the pulmonary arterial system in the distribution of blood flow[9,10,12,14,16,19] suggests that redistribution in the face of increased alveolar pressure with PEEP may also play a lesser role than was thought previously. This study determined the effects of low levels of PEEP (5 cm H_2O), an amount that has a minimal effect on cardiac output, on the distribution of pulmonary blood flow. We hypothesized that if hydrostatic factors were important in the distribution of pulmonary blood flow, PEEP would enhance preexisting dorsal (dependent)-

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to-ventral (nondependent) blood flow gradients. Pulmonary gas exchange was also studied, with the multiple inert gas elimination technique, to determine whether the redistribution of pulmonary blood flow was associated with an increase in high $\dot{V}_A$-$\dot{Q}$ units and dead space, as would be predicted by the gravitational model.

Methods

Anesthesia and Surgical Preparation

The study was approved by the University of Washington Animal Care Committee. Sixteen mongrel dogs of either sex (weight, 14-21 kg) were anesthetized with pentobarbital sodium (30 mg/kg given intravenously, supplemented with 30-90 mg every 20-30 min). Neuromuscular blockade was maintained during experimental measurements (0.1 mg/kg pancuronium given 15 min before measurements). The trachea was intubated, and the lungs were ventilated with the dogs in the supine position with $F_IO_2 = 0.21$ at a tidal volume of 15 ml/kg (Harvard piston ventilator; Harvard Apparatus, Holliston, MA). The respiratory rate was adjusted to yield a partial pressure of carbon dioxide (Pa_{CO_2}) of 37 ± 2 mmHg (mean ± SD). The lungs were hyperinflated (30-40 cm H_2O) every 30 min during the experimental preparation period and 30 min before experimental measurements.

Femoral and carotid artery catheters, a pulmonary artery catheter inserted into an external jugular vein, and a femoral venous catheter were placed *via* peripheral cutdown. Animals were mechanically ventilated for at least 120 min before the experimental protocol to allow for stabilization of gas exchange.[20] Systemic arterial, pulmonary arterial, pulmonary arterial wedge pressure, and airway pressures were measured continuously and recorded on a Western Graphtec Mach 12 data management system DMS 1000 with Validyne amplifiers (Irvine, CA). Blood temperature was maintained at 38 ± 1°C using heating lamps and pads. Thermodilution cardiac outputs ($\dot{Q}_T$) were obtained in triplicate (Edwards SAT-2 Cardiac Output Computer, Santa Ana, CA). Arterial and mixed venous blood gases were measured and corrected for temperature. Inspired, mixed expired, and end-tidal F_{CO_2} and F_IO_2 were measured using a mass-spectrometer (Perkin Elmer, Plumsteadville, PA).

Experimental Protocol

Each animal received both 0 cm H_2O PEEP (ZEEP) and 5 cm H_2O PEEP in random order determined by the flip of a coin. After 30 min in each phase, experimental measurements were obtained. Seven animals were studied in the microsphere study, and 12 animals were studied in the inert gas study. In three animals, microsphere and inert gas measurements were obtained. This experimental design occurred because inert gas measurements were added to the protocol after the first few microsphere animals were studied.

Measurement of Functional Residual Capacity

Functional residual capacity (FRC) was measured twice during each experimental condition by rebreathing helium (He) dilution.[21] The FRC was calculated as: $FRC = V_i (He_i/He_f) - V_i$, where V_i is the initial volume of the gas syringe, and He_i and He_f are the initial and final concentrations of He, respectively.

Inert Gas Measurements

The multiple inert gas elimination technique, as previously described, was used to assess pulmonary gas exchange in 12 animals.[22-24]

Gas exchange was assessed by changes in the $\dot{V}_A/\dot{Q}$ distributions predicted by the 50-compartment model of Evans and Wagner[22] and Wagner *et al.*[23] Inert gas shunt (V_D/V_T), mean $\dot{V}_A/\dot{Q}$ ratios of the $\dot{V}_A$ and $\dot{Q}$ distributions (mean $\dot{V}_A/\dot{Q}$ of $\dot{V}$ and mean $\dot{V}_A/\dot{Q}$ of $\dot{Q}$, respectively), log standard deviations of the $\dot{Q}$ (log SD_Q) and $\dot{V}_A$ (log SD_V) distributions, percentage of $\dot{Q}$ to low $\dot{V}_A/\dot{Q}$ units ($\dot{V}_A/\dot{Q}$ ratio < 0.1), and percentage of $\dot{V}_A$ to high $\dot{V}_A/\dot{Q}$ units ($\dot{V}_A/\dot{Q}$ ratio of 10-100) were calculated from the 50-compartment model. The $\dot{V}_A/\dot{Q}$ inequality was also assessed by the arterioalveolar difference ([a−A]D area), a model-independent measure.[25]

Fluorescent-labeled Microsphere Techniques

Pulmonary blood flow was measured using fluorescence-labeled microspheres.[26] The technique and the statistical analysis have been described in detail.[13,14,16] Briefly, one of five colors (orange, scarlet, blue-green, red, or crimson) of 15 μM (16.5 ± 0.10 μM; coefficient of variation, 14%) fluorescent latex microspheres (FluoSpheres 0.2% solids; Molecular Probes, Eugene, OR) was randomly selected, sonicated for 5 min, and vortexed immediately before slow injection (for 60 s) of $2-3 \times 10^6$ microspheres through the femoral venous cannula. After the injection, the catheter was flushed with saline.

After the final microsphere injection, the animals were treated with heparin (20,000 U), given papaverine (60 mg) to facilitate flushing of the lung, and were

exsanguinated through the arterial cannula. A median sternotomy was performed, and the pulmonary artery and left atrium were cannulated and perfused with a 2% dextran solution at low pressure (10–20 cm H_2O). The lungs were excised *en bloc*, the trachea was connected to a pressure source (approximately 25 cm H_2O) to inflate the lungs at total capacity, and the lungs were suspended to dry. The anatomic configuration of the lungs was approximately preserved by gluing together the apical and most ventral rims of the left and right lungs with a small amount of cyanoacrylate glue (Duro Superglue, Loctite Corp., Cleveland, OH).

The lungs were allowed to dry completely for 6–8 days and then coated with a 1-cm-thick layer of polyurethane foam (Kwik Foam, DAP Inc., Dayton, OH). The foam-covered lungs were suspended in a plastic-lined square box so that isogravitational planes were parallel to the caudal–cranial axis. The box was filled with a rapidly setting foam (polyol and isocyanate; International Sales, Seattle, WA). The foam block was sliced into 1.2-cm-thick slices using a band saw with a blade designed to eliminate tearing and tissue loss, and the slices were cut into squares (1.2 × 1.2 cm) to yield cubes approximately 1.7 cm^3 in volume, in a miter box. Samples that weighed less than 0.008 g were discarded to avoid increased experimental error associated with measurement of weight-normalized flow in small samples. The remaining pieces were assigned unique, three-dimensional spatial coordinates. The percentage of airway present in the sample was estimated by visual inspection.

Fluorescent dye was extracted from the lung tissue samples by soaking in 1.5 ml 2-ethoxy ethyl acetate (Cellosolve, Aldrich Chemical, Milwaukee, WI) for 48 h. The supernatant was placed into cuvettes using a pipette and read in a fluorescence spectrophotometer (Perkin Elmer LS 50B, Perkin Elmer Corp., Norwalk, CT) at the dye-specific excitation and emission wave lengths.

To evaluate the presence of microsphere shunting across the pulmonary vasculature, a sample of each animal's kidney was harvested and digested for 24–48 h in 4N KOH and filtered through a 10-μm-pore polycarbonate filter (Poretics, Livermore, CA). The filter containing the microspheres was soaked in Cellosolve for 4 h and the fluorescence from the supernatant was measured.

Statistical Analysis

Tissue samples with an airway content of 25% or more based on visual inspection were not included in the final analysis.[9,10] The effect of excluding these pieces on the study results was quantified by additional analysis with these pieces present. Fluorescence was corrected for the weight of each piece. Relative pulmonary blood flow to each piece of lung was calculated by dividing the fluorescence of each piece by the mean fluorescence of all pieces.

The coefficient of variation (standard deviation/mean) was used to describe the heterogeneity of blood flow. Normalized flow was characterized as the linear function of the three-dimensional coordinates or radial distance using least-squares regression analysis. A slope of −4.0%/cm, for example, means that flow decreased 0.04 normalized flow units per centimeter across the lung's full span in the dimension evaluated. The slope was expressed in terms of the percentage per centimeter because the mean normalized flow for the entire lung for each animal was 100%. Because of the interdependence of blood flow in the coronal and transverse planes in the supine position, the dorsal–ventral gradient was recalculated after correction for trends in the caudocranial direction.[16] The Pearson's correlation coefficients (r) between relative pulmonary blood flow and the linear vectors were determined, and the percentage of variance in flow in each study phase accounted for by each of the linear vectors was estimated by the square of the Pearson's correlation coefficients (r^2).

The variation in relative flow for the entire experiment was partitioned into (1) a component due to spatial position of the piece within the lung (*i.e.*, structure) and (2) a component representing the experimental manipulation (ZEEP *versus* PEEP), time, and methodologic noise, using previously described techniques.[13] The component of variation due to piece location represents variation due to structural factors common to ZEEP and PEEP. The second component is variation which primarily represents the portion of flow that is changed with administration of PEEP. Methodologic noise was previously reported as being small.[26] Bias from the variation of flow over time was controlled through randomization.

Linear slopes were compared with zero using a single-sample two-tailed *t* test. Hemodynamic, blood gas, inert gas, and pulmonary blood flow distribution measurements during ZEEP and PEEP were compared by paired-comparison *t* tests. The coefficient of variation of isogravitational flow during ZEEP and PEEP was tested using a two-factor repeated-measures analysis of variance and the Scheffé *post hoc* test. Probability values <0.05

Table 1. Cardiopulmonary Variables

	ZEEP	PEEP
FRC (ml)	593 ± 105	818 ± 144*
RR (breaths/min)	14 ± 2	16 ± 3*
V_T (ml)	270 ± 30	270 ± 30
P_{aw} (cmH_2O)	9.4 ± 1.5	13.9 ± 2.1†
P_{sa} (mmHg)	126 ± 20	125 ± 15
P_{pa} (cmH_2O)	22.7 ± 3.9	24.1 ± 4.4‡
P_w (cmH_2O)	8.7 ± 2.5	11.7 ± 2.8*
HR (beats/min)	154 ± 21	155 ± 20
$\dot{Q}_T$ (L/min)	3.66 ± 0.60	3.30 ± 0.52‡
Pa_{O_2} (mmHg)	97 ± 6	103 ± 4†
Pa_{CO_2} (mmHg)	37 ± 2	37 ± 2
*p*Ha	7.34 ± 0.03	7.33 ± 0.02
$P\bar{v}_{O_2}$ (mmHg)	51 ± 5	49 ± 3
Hct (%)	35 ± 4	34 ± 3
Temp (°C)	38.4 ± 0.8	38.5 ± 0.8

Values are mean ± SD (n = 12).

ZEEP = zero end-expiratory pressure; PEEP = 5 cmH_2O end-expiratory pressure; FRC = functional residual capacity; RR = respiratory rate; V_T = tidal volume; P_{aw} = peak airway pressure; P_{sa} = mean systemic arterial pressure; P_{pa} = mean pulmonary arterial pressure; P_w = pulmonary capillary wedge pressure; HR = heart rate; $\dot{Q}_T$ = cardiac output; Pa_{O_2} = arterial O_2 tension; Pa_{CO_2} = arterial C_{O_2} tension; *p*Ha = arterial *p*H; $P\bar{v}_{O_2}$ = mixed venous O_2 tension; Hct = hematocrit; Temp = blood temperature.

* $P < 0.01$, *versus* ZEEP.

† $P < 0.001$, *versus* ZEEP.

‡ $P < 0.05$, *versus* ZEEP.

Table 2. Pulmonary Gas Exchange

	ZEEP	PEEP
Mean $\dot{V}_A/\dot{Q}$ of $\dot{Q}$	0.49 ± 0.10	0.63 ± 0.15*
Log $SD_{\dot{Q}}$	0.76 ± 0.11	0.68 ± 0.14*
Mean $\dot{V}_A/\dot{Q}$ of $\dot{V}$	0.97 ± 0.25	1.03 ± 0.28
Log $SD_{\dot{V}}$	0.87 ± 0.30	0.70 ± 0.28†
$\dot{Q}_S/\dot{Q}_T$ (%)	0.6 ± 1.0	0.3 ± 0.4
$\dot{Q}$ of low $\dot{V}_A/\dot{Q}$ (%)	0.6 ± 1.1	0.3 ± 0.8
V_D/V_T (%)	32.3 ± 6.1	37.6 ± 6.1‡
$\dot{V}$ of high $\dot{V}_A/\dot{Q}$ (%)	2.4 ± 3.7	1.4 ± 2.7†
(a-A)D	0.28 ± 0.12	0.20 ± 0.09†

Values are means ± SD (n = 12).

Mean $\dot{V}_A/\dot{Q}$ of $\dot{Q}$ and mean $\dot{V}_A/\dot{Q}$ of $\dot{V}$ = mean ventilation-perfusion ($\dot{V}_A/\dot{Q}$) ratios of perfusion ($\dot{Q}$) and ventilation ($\dot{V}_A$) distributions, respectively; log $SD_{\dot{Q}}$ and log $SD_{\dot{V}}$ = log standard deviations of $\dot{Q}$ and $\dot{V}$ distributions, respectively; $\dot{Q}_S/\dot{Q}_T$ = inert gas shunt; $\dot{Q}$ of low $\dot{V}_A/\dot{Q}$ = $\dot{Q}$ to low $\dot{V}_A/\dot{Q}$ units ($\dot{V}_A/\dot{Q}$ ratio <0.1); V_D/V_T = dead space; $\dot{V}$ of high $\dot{V}_A/\dot{Q}$ = $\dot{V}$ to high $\dot{V}_A/\dot{Q}$ units ($\dot{V}_A/\dot{Q}$ ratio 10–100); (a-A)D = arterial–alveolar difference area.

* $P < 0.01$ *versus* ZEEP.

† $P < 0.05$ *versus* ZEEP.

‡ $P < 0.001$ *versus* ZEEP.

were deemed significant. Mean values ± SD are presented.

Results

Hemodynamics and Respiratory Measurements

Five cm H_2O PEEP increased FRC by 38% ($P < 0.01$; table 1). The general experimental conditions, including blood temperature, hematocrit concentration, partial pressure of carbon dioxide (Pa_{CO_2}), *p*H, $P\bar{v}O_2$, systemic arterial pressure, and heart rate were constant throughout the study (table 1). The respiratory rate was increased during PEEP to maintain constant Pa_{CO_2}. The partial pressure of oxygen (Pa_{O_2}), pulmonary arterial pressure, and pulmonary wedge pressure were increased minimally ($P < 0.05$) by PEEP (table 1). The $\dot{Q}_T$ was decreased by 10 ± 4% ($P < 0.05$) by PEEP.

Gas Exchange Measurements

Positive end-expiratory pressure reduced all measures of $\dot{V}_A/\dot{Q}$ heterogeneity, including log $SD_{\dot{Q}}$, log $SD_{\dot{V}}$, and the (a − A)D area ($P < 0.05$; table 2). PEEP reduced ventilation of high $\dot{V}_A/\dot{Q}$ units ($P < 0.05$) and increased V_D/V_T ($P < 0.001$, table 2). $\dot{Q}_S/\dot{Q}_T$ and perfusion of low $\dot{V}_A/\dot{Q}$ units were not affected by PEEP in these normal lungs (table 2).

Pulmonary Blood Flow Distribution

Lung pieces (n = 969 ± 140) were analyzed for each animal. Because of the presence of more than 25% air-

Table 3. Pulmonary Blood Flow Distribution: Heterogeneity and Gradients as a Linear Function of Spatial Vectors

	ZEEP	PEEP
Coefficient of variation (%)	53 ± 7	56 ± 10
Left-to-right gradient (%/cm)	1.6 ± 2.6	1.1 ± 2.3
Dorsal-to-ventral gradient (%/cm)	−5.4 ± 4.8†	−8.3 ± 5.7*‡
Caudal-to-cranial gradient (%/cm)	−0.7 ± 2.5	−1.9 ± 3.4
Hilar-to-peripheral gradient (%/cm)	−9.7 ± 5.3‡	−9.4 ± 4.2§

Values are mean ± SD (n = 7).

* $P < 0.05$, versus ZEEP.

† $P < 0.05$, *versus* 0.

‡ $P < 0.01$, *versus* 0.

§ $P < 0.001$, *versus* 0.

Table 4. Components of Variation of Pulmonary Blood Flow

	Variance (relative flow units)			Variance (% of total)	
Dog No.	Structure	PEEP	Total	Structure	PEEP
1	0.412	0.024	0.436	95	5
2	0.231	0.034	0.265	87	13
3	0.262	0.033	0.295	89	11
4	0.251	0.014	0.265	95	5
5	0.198	0.007	0.204	97	3
6	0.350	0.030	0.38	92	8
7	0.267	0.019	0.286	93	7
Mean ± SD				93 ± 3	7 ± 3

ways (in 98% of deleted specimens) or sample loss or lack of identification of spatial coordinates (2% of deleted specimens), 114 ± 37 lung pieces, representing 10.5 ± 3% of the total samples, had been deleted before analysis. Kidney samples did not have any fluorescence, indicating that all of the microspheres were trapped in the lungs. Left-to-right and caudal-to-cranial gradients over the whole lung were not different from zero and were not significantly affected by PEEP (table 3). Inclusion or exclusion of lung samples with more than 25% airways did not significantly affect any of the gradients, the coefficient of variation, or the effect of PEEP.

The heterogeneity of the distribution of pulmonary blood flow estimated by the coefficient of variation was not affected by PEEP (table 3). The heterogeneity of pulmonary blood flow within horizontal isogravitational slices in the dorsal (dependent)-ventral direction also were not affected by PEEP. The coefficient of variation of blood flow was greater in the ventral (nondependent) lung regions than in the dorsal (dependent) lung regions ($P < 0.001$) with ZEEP and PEEP. During ZEEP, the coefficient of variation of flow was 39 ± 14% in the dependent-most vertical slice and 57 ± 14% in the upper-most vertical slice. Similar values were observed during PEEP. More than 93 ± 3% of the variance in the distribution of pulmonary blood flow was attributed to structural factors (table 4). Redistribution of blood flow with PEEP accounted for <7 ± 3% of the total variance in pulmonary blood flow (table 4).

Pulmonary blood flow decreased with increasing distance from the ipsilateral hilus (table 3). Pulmonary blood flow decreased linearly by 9.7 ± 5.3% per centimeter diagonally away from the hilus during ZEEP (95% CI of the slope = [−14.5%/cm to −4.7%/cm]; $P < 0.01$ compared with zero). The distance from the ipsilateral hilus accounted for 18 ± 4% of the variance in the whole lung during ZEEP. The hilar-to-peripheral gradient was not altered by PEEP.

The pulmonary blood flow was also distributed vertically, with a decrease in pulmonary blood flow of 5.4

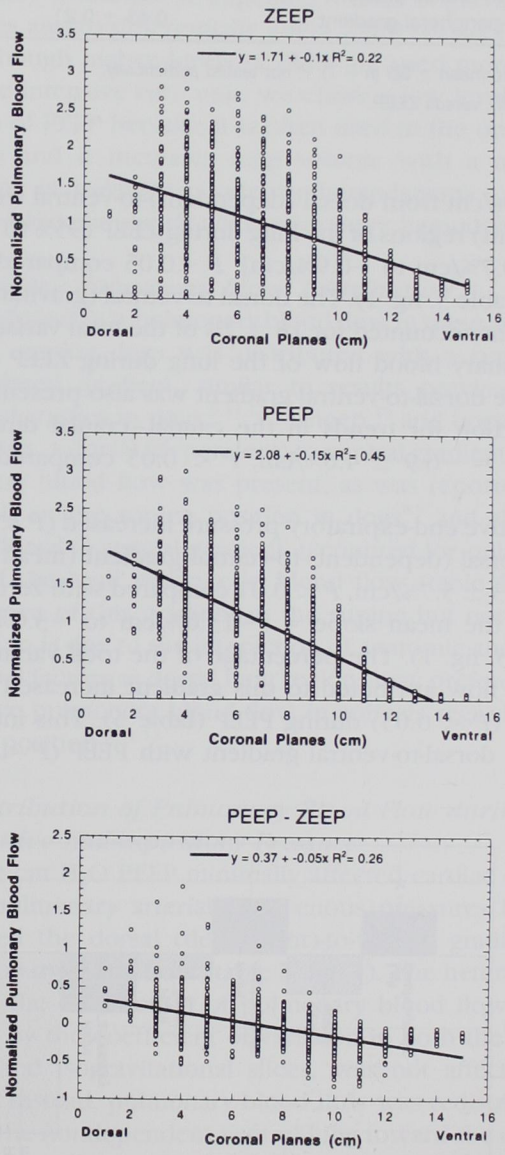

Fig. 1. Dorsal (dependent)-to-ventral pulmonary blood flow gradients in one representative animal during ventilation with zero cm H_2O positive end-expiratory pressure (PEEP) (ZEEP, *top*), 5 cm H_2O PEEP (PEEP, *middle*), and change in pulmonary blood flow between PEEP and ZEEP (PEEP-ZEEP, *bottom*). The dorsal-to-ventral gradient (slope = −10%/cm, *top*) became steeper with PEEP (slope = −15%/cm, *middle*), which was associated with an increase in relative pulmonary blood flow to dependent dorsal areas and a decrease to nondependent ventral areas (*bottom*).

Table 5. Linear Association (Pearson's Correlation Coefficients [r]) between Pulmonary Blood Flow and Spatial Vectors

Gradient	ZEEP		PEEP	
	r	r^2	r	r^2
Left-to-right gradient	0.15 ± 0.17	0.02 ± 0.03	0.16 ± 0.08	0.03 ± 0.01
Dorsal-to-ventral gradient	0.42 ± 0.16	0.18 ± 0.02	0.57 ± 0.16*	0.33 ± 0.02
Caudal-to-cranial gradient	0.20 ± 0.12	0.04 ± 0.01	0.28 ± 0.18*	0.08 ± 0.03
Hilar-to-peripheral gradient	0.43 ± 0.21	0.18 ± 0.04	0.41 ± 0.19	0.17 ± 0.04

Values are mean ± SD (n = 7). r^2 not tested statistically.

* $P < 0.05$, *versus* ZEEP.

± 4.8%/cm from dorsal (dependent)-to-ventral (nondependent) regions of the lung during ZEEP (95% CI slope = [−9.7%/cm to −0.9%/cm], $P < 0.05$ compared with zero; table 3, fig. 1). The dorsal-to-ventral (gravitational) gradient accounted for 18 ± 2% of the total variance in pulmonary blood flow of the lung during ZEEP (table 5). The dorsal-to-ventral gradient was also present after correction for trends in the caudal-cranial direction (slope = −6.9 ± 4.6%/cm, $P < 0.05$ compared with zero).

Positive end-expiratory pressure increased ($P < 0.05$) the dorsal (dependent)-to-ventral gradient (mean slope = −8.3 ± 5.7%/cm, $P < 0.01$ compared with zero; 95% CI of the mean slope = [−13.6%/cm to −3.0%/cm]; table 3, fig. 1). The percentage of the total variance in blood flow attributed to this gradient increased to 33 ± 2% ($P < 0.05$) during PEEP (table 5). This increase in the dorsal-to-ventral gradient with PEEP ($P < 0.05$) persisted after correction for trends in the caudal-cranial direction (slope = −9.4 ± 5%/cm; $P < 0.01$ compared with zero).

The steeper dorsal-to-ventral gradient with PEEP was associated with an increase in relative flow to dependent, dorsal lung regions and a decrease in relative flow to nondependent, ventral lung regions (fig. 1). Identification of these segments by lung lobe (fig. 2) revealed significant decreases in relative flow to the right middle lobe ($P < 0.01$) and increases in relative flow to the left diaphragmatic lobe ($P < 0.05$, fig. 2). There was a nonsignificant trend toward decreased relative flow in other upper lung lobes (left apical, right apical, and right accessory) and toward increased relative flow in the dependent right diaphragmatic lobe.

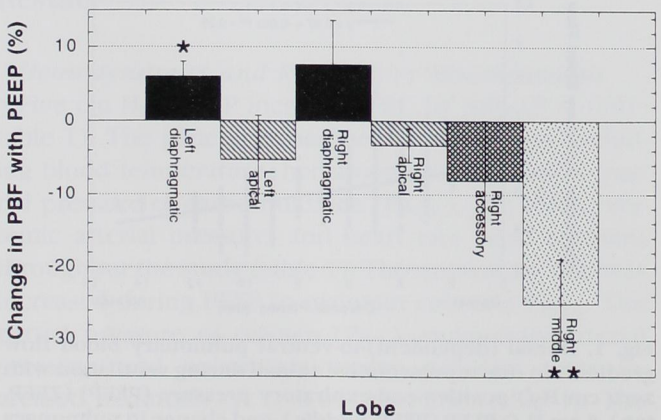

Fig. 2. Percentage change in mean relative pulmonary blood flow with positive end-expiratory pressure (PEEP) by lung lobe location (n = 7). Pulmonary blood flow decreased significantly in the right middle lobe ($P < 0.01$) and increased significantly in the left diaphragmatic lobe ($P < 0.05$). *$P < 0.05$; **$P < 0.01$ comparing zero cm H_2O PEEP and PEEP.

Discussion

This is the first study to describe the redistribution of pulmonary blood flow with low levels of PEEP using a high-resolution technique. Five cm H_2O PEEP increased the dorsal (dependent)-to-ventral gradient of pulmonary blood flow by increasing flow relatively to dependent lung regions and decreasing flow relatively to nondependent lung regions in anesthetized, supine-positioned dogs. Although these results are consistent with the traditional, gravitationally based lung zone model, the redistribution of flow with PEEP accounted for only a small amount of the total variance in the pulmonary blood flow distribution.

Methodologic Issues

Use of 15-μm fluorescent-labeled microspheres was recently validated to measure regional pulmonary blood flow.[26] Simultaneous injection of fluorescent-labeled and radioactivity-labeled microspheres yielded a Pearson's correlation coefficient of 0.98 ± 0.01, with a slope

of 0.95 and intercept of 0.05.[26] To estimate the distribution of regional pulmonary blood flow accurately, the spheres must be extracted completely by the pulmonary microcirculation. Spheres measuring 15 μm are almost completely entrapped by the pulmonary circulation,[27] with <2% shunting reported.[28] We found this in the current experiment by the absence of fluorescence in the kidney samples. We injected $2-3 \times 10^6$ microspheres, which was calculated to ensure a sufficient number (approximately 400) per tissue sample to limit the effect of methodologic error.

We removed 114 ± 37 lung pieces that contained more than 25% airways, representing 10% of the samples, before analysis because their inclusion would result in an erroneously low weight-corrected pulmonary blood flow. Flow in samples from central areas with large vessels and airways may have been underrepresented because of the exclusion criteria. Central lung pieces that were included in the analysis (<25% airways) may be slightly more likely to have a disproportionate additional weight because airway cartilage caused the weight-normalized blood flow to be less than expected. Reanalysis of all data, including pieces with >25% airways, revealed that exclusion of these specimens did not alter the results of the study.

As our focus was to examine gradients in pulmonary blood flow and the relative flow distribution, flows in each lung piece were normalized to the mean flow of all pieces per animal. The flow signals were corrected for piece weight because many tissue pieces from peripheral parts of the lungs had a volume <1.7 cm^3. Because the lung was inflated to total lung capacity before drying, all alveoli were of uniform size and the density was the same. Thus weight is a surrogate of volume. In contrast to many older studies, flow was normalized to total lung capacity and not to other lung volumes. We previously normalized flow to FRC and still observed large isogravitational heterogeneity in flow.[29] However, we have not evaluated the effect of normalization of flow at TLC compared with FRC on the magnitude of the gravitational gradient.

The lungs were dried in anatomic reapproximation when inflated to a pressure of 25 cm H_2O. The configuration of the lung after drying may be slightly different than that *in vivo*. The experimental lung volume may have been slightly greater than in the intact lung, which would increase the linear dimensions slightly. The orientation of gravity is slightly different when the lung is suspended by the trachea. The lung may be slightly distorted by this and the lack of physical constraints of the chest wall and diaphragm. In addition, the weight of the heart *in vivo* may result in some compression of the lung below it. Expansion of the lung from FRC to total lung capacity may move adjacent parenchymal units 1-2 cm but has little effect on the overall orientation of the lung.[30] These factors are difficult to quantify, but they would not be expected to alter isogravitational planes and to differentially affect ZEEP *versus* PEEP.

Although higher levels of PEEP are used more often in the intensive care unit, we chose a low level (5 cm H_2O) of PEEP because it is often used in the operating room and it increases lung volume with a minimal change in pulmonary vascular hemodynamics, which would independently alter pulmonary hemodynamics.

Baseline Pulmonary Blood Distribution

We found that pulmonary blood flow in these anesthetized, supine dogs was distributed with a central-to-peripheral gradient, similar to results previously reported studies in dogs,[9,11,12,19] sheep,[16] and horses.[15] In addition, a dorsal (dependent)-to-ventral gradient in pulmonary blood flow was present, as was reported previously in the supine position in dogs[8,9] and sheep.[16] The dorsal-to-ventral gradient accounted for only 18 ± 2% of the total variance in blood flow (table 5). The presence of this gradient in the supine but not prone position is due to the interaction of anatomic and gravitational factors as dorsal lung regions may preferentially receive pulmonary blood flow in animals regardless of their position.[8,11,12]

Distribution of Pulmonary Blood Flow during Positive End-expiratory Pressure

Five cm H_2O PEEP minimally affected cardiac output and pulmonary arterial and venous pressures but increased the dorsal (dependent)-to-ventral gradient of pulmonary blood flow (table 3, fig. 1). The heterogeneity of the distribution of pulmonary blood flow, measured by the coefficient of variation in both the entire lung and isogravitational slices, was not affected by PEEP. Instead, pulmonary blood flow was redistributed from the nondependent ventral lung toward the dependent dorsal lung. The increase in the dorsal-to-ventral gradient with PEEP persisted after correction for blood flow in the caudocranial direction. Therefore, PEEP independently altered the pulmonary blood flow distribution in the gravitational distribution, independent of caudocranial anatomic factors. PEEP decreased pulmonary blood flow most in the right middle lobe and less in the apical lobes, perhaps due to the middle lobe's

more ventral orientation, with fewer dorsal regions, than in the apical lobes in the dog. Previous studies using high levels of PEEP (20 cm H_2O) profoundly depressed cardiac output and redistributed blood flow to dependent lung regions.[2,3]

The increased dorsal (dependent)-to-ventral gradient with PEEP is consistent with the gravitationally based lung zone model.[4-7] The increased slope could represent the shifting of the predominant zonal conditions in upper lung regions from zone 2 to zone 1, thereby decreasing blood flow to nondependent ventral areas. The increased dorsal-to-ventral gradient also could result from the shift of the predominant zonal conditions from zone 4 to zone 3 in the most dependent lung regions, as has been postulated to occur with increased lung volumes.[7] However, it is unlikely that the improved relative perfusion of the dorsal lung was due to improved ventilation and release of hypoxic pulmonary vasoconstriction, in dorsal lung regions, because shunt and low $\dot{V}_A/\dot{Q}$ were absent in this study.

The $\dot{V}_A/\dot{Q}$ heterogeneity was reduced by PEEP because of a reduction in high $\dot{V}_A/\dot{Q}$ regions and an increase in dead space (table 2). This effect is in contrast to the gas-exchange effect in normal lungs observed with larger amounts ($\geq$10 cm H_2O) of PEEP, which increased $\dot{V}_A/\dot{Q}$ heterogeneity by depressing cardiac output and creating high $\dot{V}_A/\dot{Q}$ peaks.[1,2] Low levels of PEEP had minimal effects on gas exchange in healthy dogs but dramatically reduced pulmonary shunt and low $\dot{V}_A/\dot{Q}$ regions in injured lungs.[1,2]

In the current study, 5 cm H_2O PEEP did not affect the hilar-to-peripheral gradient of pulmonary blood flow, in contrast to earlier findings with higher levels of PEEP.[3] With 20 cm H_2O PEEP, blood flow was redistributed from the core to the periphery of the lung.[3] As the magnitude of this central-to-peripheral gradient may be related to cardiac output,[19] little change may occur when cardiac output is minimally changed, as in the current study.

Although our results can be interpreted within the context of the classical model in which zonal conditions are vertically stacked, the marked heterogeneity of pulmonary blood flow in isogravitational planes emphasize that the mechanisms that determine the distribution are far more complex. Regional pulmonary blood flow to each lung piece was observed to vary by as much as 100 times within isogravitational planes, a degree of variability that is similar to that seen between different isogravitational planes. In addition, redistribution of flow with PEEP only accounted for $<7 \pm 3\%$ of the total variance in pulmonary blood flow. In contrast, $93 \pm 3\%$ of the variance in pulmonary blood flow was attributed to structural factors (table 4). The finding of marked isogravitational plane heterogeneity that is unchanged with PEEP suggests that vascular branching patterns and regional variation in conductance to blood flow are more important determinants of pulmonary perfusion.

Clinical Implications

These results cannot be applied directly to the clinical treatment of persons with lung disease who require mechanical ventilation. There may be important species differences in the role of gravity in the distribution of blood flow in upright humans compared with quadruped animals.[17,18,31-34] Thus it is likely that PEEP would enhance the gravitational gradient in pulmonary blood flow to a greater degree in humans than in dogs.

In conclusion, low levels of PEEP increased the dependent-to-nondependent gradient of pulmonary blood flow in supine-positioned dogs, which is partially consistent with the traditional gravitationally based zone model of pulmonary blood flow. However, the mechanisms that determine the overall distribution of pulmonary blood flow are more complex and most likely involve anatomic factors.

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