Regional lung blood flow and ventilation in upright humans studied with quantitative SPECT

Johan Petersson a,b,*, Malin Rohdin c, Alejandro Sánchez-Crespo d,e, Sven Nyrén f, Hans Jacobsson d,f, Stig A. Larsson d,e, Sten G.E. Lindahl a,b, Dag Linnerström c, Blazej Neradilek g, Nayak L. Polissar g, Robb W. Glennya

a Department of Anesthesiology and Intensive Care, Karolinska University Hospital Solna, 171 76 Stockholm, Sweden
b Department of Physiology and Pharmacology, Section of Anesthesiology and Intensive Care Medicine, Karolinska Institutet, 171 77 Stockholm, Sweden
c Department of Physiology and Pharmacology, Section of Environmental Physiology, Karolinska Institutet, 171 77 Stockholm, Sweden
d Section of Nuclear Medicine, Department of Hospital Physics, Karolinska University Hospital Solna, 171 76 Stockholm, Sweden
e Medical Radiation Physics, Department of Oncology-Pathology, Stockholm University and Karolinska Institutet, 171 77 Stockholm, Sweden
f Department of Radiology, Karolinska University Hospital Solna, 171 76 Stockholm, Sweden
g Mountain Whisper Light Statistical Consulting, Seattle, WA, USA
h Departments of Medicine and Physiology and Biophysics, University of Washington, Seattle, WA 98195, USA

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Abstract
We used quantitative Single Photon Emission Computed Tomography (SPECT) to study the effect of the upright posture on regional lung blood flow and ventilation. Nine (upright) plus seven (prone and supine) healthy volunteers were studied awake, breathing spontaneously. Regional blood flow and ventilation were marked in sitting upright, supine and prone postures using 113mIn-labeled macroaggregates and inhaled Technegas (99mTc); both remain fixed in the lung after administration. All images were obtained while supine. In comparison with horizontal postures, both blood flow and ventilation were greater in caudal regions when upright. The redistribution was greater for blood flow than for ventilation, resulting in decreasing ventilation-to-perfusion ratios down the lung when upright. We conclude that gravity redistributes regional blood flow and ventilation in the upright posture, while the influence is much less in the supine and prone postures.

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1. Introduction

The influence of posture and gravity, on the regional distributions of lung blood flow and ventilation has been of longstanding interest to the physiologic community (Martin et al., 1953; Rahn et al., 1956). Early human studies, using methods with a spatial resolution limited to one or two directions, demonstrated greater blood flow and ventilation in dependent regions independent of posture (West and Dollery, 1960; Bryan et al., 1964; Anthonisen and Milic-Emili, 1966; Glazier and DeNardo, 1966; Kaneko et al., 1966; Milic-Emili et al., 1966; Hughes et al., 1968). The non-uniform, but well-matched, distributions of regional lung blood flow and ventilation were therefore explained by the shared influence of gravity. Later animal and human studies demonstrated heterogeneity of blood flow within isogravitational planes (Reed and Wood, 1970; Greenleaf et al., 1974; Beck and Rehder, 1986; Hakim et al., 1987; Lisbona et al., 1987; Hakim et al., 1988a,b), which led to the conclusion that blood flow heterogeneity must be determined by other factors in addition to gravity. Further animal studies have demonstrated that gravity (posture) has an important, but secondary, influence on the distribution of blood flow (Glenny et al., 1991a,b, 1999). No study has previously employed high-resolution imaging methods to simultaneously quantify the effect of gravity on the distribution of both regional lung blood flow and ventilation in upright humans.

In animal studies, regional distributions have been described as per unit lung tissue, i.e. per alveolus. In contrast, most human studies employing high-resolution methods have described the distributions as per unit lung volume. Interpretation of results from these studies is complicated by the non-uniform and posture-dependent distribution of lung tissue. In each posture the amount of lung tissue per unit lung volume varies between regions. Similarly, the amount of lung tissue in a unit volume of the lung, defined by its relationship to anatomical landmarks, varies between postures. Recently we used a novel application of the Single Photon Emission Computed Tomography (SPECT) technique to demonstrate that...
these phenomena greatly contribute to the differences in blood flow and ventilation distributions imaged in the supine and prone postures (Petersson et al., 2007). With SPECT, regional blood flow and ventilation can be marked using radiotracers that remain fixed in the lung parenchyma after administration. In this study of spontaneously breathing healthy volunteers, we used these radiotracers to mark regional lung blood flow and ventilation in the sitting upright, supine and prone postures. All images of the radiotracer distributions were acquired in the supine posture; regional blood flow and ventilation were therefore observed as per unit volume supine lung. These distributions were further characterized as cranial-to-caudal gradients of relative blood flow and ventilation per unit volume supine lung. The gradients were compared, looking for the effect of subjects having been supine, prone or upright at the time of radiotracer administration, which allowed us to quantify the effect of posture/gravity on the distribution of blood flow and ventilation within the lung parenchyma. We believe that the current work adds to the ongoing debate on the importance of gravity to lung blood flow and ventilation distributions by: (1) studying upright humans, a posture that is not amenable to other modern imaging methods, (2) confirming that gravity is an important determinant of regional lung blood flow and ventilation in upright humans, (3) obtaining simultaneous measurements of regional blood flow and ventilation allowing assessment of regional ventilation-to-perfusion ratios, (4) acquiring measurements during normal breathing as oppose to special breathing manoeuvres, and (5) using a novel approach to exclude the confounding effect of lung tissue distribution when comparing blood flow and ventilation distributions in different postures.

2. Methods

This study consists of new data from previously reported experiments (Petersson et al., 2004, 2007). The current study, however, includes no previously published data on the regional distributions of lung blood flow or ventilation.

2.1. Subjects

Eighteen healthy volunteers (9 men and 9 women), age 20–40 years were studied. All subjects were of normal weight (range 57–81 kg) and height (range 160–179 cm). We recruited subjects less than 180 cm in height to ensure that all lung regions would fit into the scanning field of the SPECT-camera. None of the subjects had a history of pulmonary disease, all were non-smokers and all had normal spirometry and lung volumes. The subjects received written information about the procedure and informed verbal consent was obtained. Results from two of the subjects had to be excluded from the study. One due to technical problems with the transmission source causing artifacts in the SPECT results and the other because the subject was unable to maintain a fixed position during image acquisition. The local committees for research ethics and radiation safety approved the study.

2.2. SPECT imaging

The dual isotope SPECT technique used in this work has previously been described in detail (Sánchez-Crespo et al., 2002; Petersson et al., 2004).

2.2.1. Radiopharmaceuticals

Regional distribution of ventilation was marked using inhaled Technegas, microscopic graphite particles labeled with radioactive Technetium (99mTc) (Burch et al., 1986). Regional lung blood flow was marked using macroaggregates of albumin labeled with radioactive Indium (113mIn). Technegas was inhaled during quiet tidal breathing from a Technegas generator. This was done through a box that mixed Technegas (initially 100% Argon) with air. Pulse oximetry was monitored during the Technegas inhalation to document normal hemoglobin oxygen saturation. After the inhalation of Technegas 100 or 75 MBq 113mIn-LyoMAA was administered intravenously via a peripheral venous catheter for the subjects studied in the sitting upright or horizontal postures, respectively. The subjects were estimated to receive a total effective dose of about 5mSv.

2.2.2. SPECT

SPECT images were obtained with a three-headed gamma camera equipped with medium energy general-purpose parallel-hole collimators. SPECT scans were performed in 72 projections, 62 s per projection using a four-energy window acquisition protocol. Image acquisition required 25 min. Thus, four images (128 × 128 pixels with a pixel size of 3.56 mm × 3.56 mm) were obtained at each data acquisition angle. An additional transmission tomography with a moving 99mTc line source was performed in order to obtain data for the attenuation correction routine. The two sets of projected images, one for each principal photon energy (140 and 392 keV), were corrected for photon scattering and attenuation as well as for the contribution of high-energy photons in the lower photon energy window and the radioactive decay before image reconstruction. The lung areas were delineated in the reconstructed transverse transmission images using a previously described edge detection algorithm (Sánchez-Crespo et al., 2002). To verify that only lung tissue was included in the images they were reviewed by a radiologist. In a few of the images, a small number of peripheral pixels were considered non-lung tissue and therefore removed.

Spatial coordinates and number of events per voxel within the total delineated lung region were extracted from original reconstructed transverse SPECT 113mIn-LyoMAA and 99mTc-technegas image data. The data sets thus consist of a number of voxels, each with a number of counts representing either regional blood flow or ventilation and coordinates for each voxel in the left–right, ventral–dorsal and cranial–caudal directions. The size of each voxel was 3.56 mm × 3.56 mm × 3.56 mm. Number of events per voxel was normalized to the mean number of events for all voxels. The voxel data thus represent the amount of blood flow or ventilation for each voxel relative to the mean for all voxels. The normalized voxel data for blood flow and ventilation were also used to calculate ventilation-to-perfusion ratios for each voxel.

2.3. Experimental protocol

Seven subjects were studied in the horizontal postures. In these subjects the radiotracers were once administered in the supine posture and at another occasion in the prone posture. A time interval of at least 48 h was required between the two occasions. For four of the subjects the first study was done with radiotracer administration in the supine posture, the remaining subjects were initially studied prone. In nine subjects the radiotracers were administered in the sitting upright posture. All images were obtained in the supine posture. The subjects breathed air at all times, radiotracer administration and image acquisitions were performed during continuous spontaneous breathing. During imaging, an expiratory pressure of 2.5 cmH2O was used for the subjects to whom the radiotracers were administered in the upright posture.

2.4. Data analysis

For each data set the lung was divided into 10 segments each representing 10% of the total cranial-to-caudal distance. The distributions of blood flow and ventilation were visualized using plots of
mean flow and ventilation per voxel for all voxels within each segment. That is, the plots show the mean flow or ventilation per voxel within 10 transverse planar slices of the lung. Cranial-to-caudal gradients were estimated as the regression coefficients from linear regressions of regional blood flow, ventilation and ventilation-to-perfusion ratio on the cranial-to-caudal distance. Gradients are thus quantified as the change in normalized flow, ventilation and ventilation-to-perfusion ratio per cm. To adjust for potential confounding of gradients, other spatial variables were added into the linear regression model. Variables tested were distance from the edge, ventral-to-dorsal distance, cranial-to-caudal distance, right-to-left distance and an indicator variable for the edge vs. the interior of the lung. All these variables were considered in linear and quadratic form. Gradients were calculated using linear regression models with and without adjusting for the additional variables that provided a non-trivial increase in the coefficient of determination (at least 0.02 average increase in $R^2$ across the images).

2.5. Statistics

Values are means and standard deviations. One sample $t$-test was used to test whether gradients were different from zero. Unpaired $t$-tests were used for statistical comparisons of gradients after radiotracer administration in the upright vs. horizontal postures. Paired $t$-tests were used for statistical comparisons of gradients after radiotracer administration in the prone vs. the supine postures. We used statistical software R Version 1.9.0 for all the calculations.

3. Results

3.1. SPECT

Subjects received 62–105 MBq of $^{113m}$In, corresponding to 270,000–700,000 Lyo-MAA particles. Technegas breathing required 120–387 s. The lowest pulse oximetry reading recorded during Technegas administration was 95%.

3.2. Plots of blood flow and ventilation distributions

In the following, dependent, non-dependent and posture always refers to conditions at the time of radiotracer administration. The profiles for the distribution of blood flow and ventilation in the cranial-to-caudal direction are presented in Fig. 1. In upright regional blood flow and ventilation are greater in caudal than apical regions. In contrast, the plots suggest even distributions of blood flow and ventilation between apical and caudal regions in the supine and prone postures, with little difference between the postures. Thus, the plots clearly demonstrate that a change from either the supine or the prone posture is associated with a redistribution of blood flow and ventilation from apical to caudal regions. While in upright the ventilation-to-perfusion ratio decreases in the cranial-to-caudal direction, there is less change in this direction in the horizontal postures. In other words, a change from the horizontal postures to upright is associated with an increasing ventilation-to-perfusion ratio in apical regions and a decreasing ratio in caudal regions.

averaged within each section. Error bars illustrate standard deviations for the individual values for each section. Values at the extremes of the cranial-to-caudal distances are influenced by the edge effect (underestimation of the radiotracer concentration at the lung periphery). The extremes also correspond to very few voxels, which might explain deviations at the ends of the profiles. The linear regression gradients (Fig. 2) are much less sensitive to these phenomena.
### Table 1
Cranial-to-caudal and ventral-to-dorsal gradients.

<table>
<thead>
<tr>
<th>Gradient</th>
<th>Supine</th>
<th>Upright</th>
<th>Prone</th>
<th>$P$ value upright vs. supine</th>
<th>$P$ value upright vs. prone</th>
<th>$P$ value supine vs. prone</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cranial-to-caudal gradients</td>
<td>−0.003 ± 0.009</td>
<td>0.051 ± 0.013†</td>
<td>−0.007 ± 0.006†</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>0.24</td>
</tr>
<tr>
<td>Blood flow</td>
<td>0.005 ± 0.008</td>
<td>0.024 ± 0.008†</td>
<td>−0.002 ± 0.008†</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>0.11</td>
</tr>
<tr>
<td>Ventilation-to-perfusion ratio</td>
<td>0.008 ± 0.017</td>
<td>−0.043 ± 0.025†</td>
<td>0.007 ± 0.009</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>0.91</td>
</tr>
</tbody>
</table>

Estimates of cranial-to-caudal gradients obtained from least squares linear regression. Values are mean ± standard deviation; units are normalized flow, ventilation or ventilation-to-perfusion ratio per cm.

1 $P < 0.05$ vs. zero.

### 3.3. Linear regression model

Regional blood flow and ventilation correlated with several spatial variables. Adjusting for other spatial variables did not change statistical conclusions about the difference in gradients between the postures. Estimates of the gradients for regional blood flow, ventilation and ventilation-to-perfusion ratios are summarized in Table 1 and illustrated in Fig. 2. The conclusions from the analysis of the linear regression models agree with the results of the visual inspection of the plots in Fig. 1.

#### 3.3.1. Cranial-to-caudal blood flow gradients

In the horizontal postures, the mean cranial-to-caudal gradients were similar ($P = 0.24$ prone vs. supine), with a small decrease in blood flow in the caudal direction that was significant only in the prone posture ($P = 0.02$ vs. zero). In contrast, in the sitting upright posture there was a much greater gradient with increasing blood flow in the caudal direction ($P < 0.001$ vs. zero and vs. the prone and supine postures). Thus, a change from the horizontal postures to the sitting upright posture is associated with a redistribution of blood from cranial to caudal regions.

#### 3.3.2. Cranial-to-caudal ventilation gradients

The cranial-to-caudal gradients for regional ventilation were small in both the prone and supine postures ($P = 0.63$ and 0.18 vs. zero) and not significantly different between the postures ($P = 0.11$). In the sitting upright posture, there was a greater mean gradient, with regional ventilation increasing in the caudal direction ($P < 0.001$ vs. zero and vs. the prone and supine postures). Thus, similar to the effect on regional blood flow, a change from the prone or supine postures to sitting upright causes a redistribution of ventilation from cranial to caudal regions.

#### 3.3.3. Regional ventilation-to-perfusion ratios

In the horizontal postures, the mean ventilation-to-perfusion ratio gradients in the cranial-to-caudal direction were small in and statistically not different from zero. In contrast, in the upright posture the ventilation-to-perfusion ratio decreased in the cranial-to-caudal direction ($P < 0.001$ vs. zero and vs. the prone and supine postures).

### 4. Discussion

The relative effect of gravity on regional lung blood flow and ventilation continues to be debated (Glenny et al., 2007). At least some of this controversy stems from differing results between studies of upright humans and horizontal animals. These studies also differ in the spatial resolution of the employed methodologies. In addition, prior works have also differed in their consideration of lung compression as a confounder of observed blood flow and
ventilation distributions (Hopkins et al., 2007; Petersson et al., 2007). Most modern, high-resolution, imaging methods do not allow direct imaging of upright humans. However, radiotracers that remain fixed within the lung after administration, allow upright humans to be studied with SPECT and elimination of the confounding effect of lung compression when comparing the distribution of blood flow and ventilation in different postures. To our knowledge, this is the first study that used such radiotracers and quantitative SPECT, corrected for attenuation and scatter, to report the regional distributions of both lung blood flow and ventilation in upright humans. Using the same method, we have previously reported the small effect of gravity on regional lung blood flow and ventilation in humans in the horizontal postures (Petersson et al., 2007).

In this paper we postulated that gravity might play a greater role in upright humans, which is suggested by earlier work using methods with a low spatial resolution. In the current study of healthy spontaneously breathing volunteers, we have confirmed a greater effect of gravity in the upright posture. The main results are that a change from the prone and supine postures to sitting upright causes a shift in the distribution of regional blood flow and ventilation within the lung parenchyma towards caudal regions. When taken together with our previous study (Petersson et al., 2007), which similarly to prior animal studies demonstrated little effect of gravity on regional blood flow and ventilation in supine and prone postures, we believe our results support consistent observations between prior human and animal studies.

4.1. Methodological issues

We have discussed the characteristics and limitations of the SPECT method in previous publications (Sánchez-Crespo et al., 2002; Petersson et al., 2004). The present discussion is therefore limited to methodological issues important for the interpretation of the current study. The SPECT method uses radionuclide-labeled particles that are deposited in the lung in proportion to regional blood flow and ventilation, which has been confirmed with other methods (Melsom et al., 1995; Johansson et al., 2004). Quantitative SPECT measurements are hampered by attenuation and scatter of the radiation emitted from the radiotracer within the body. In this study we applied new methods of correcting for attenuation and scatter. The methods include transmission tomography to obtain data for the attenuation correction routine. Phantom studies have shown that these methods produce corrected values with a mean deviation from the true value of −0.8% for 111In and −1.5% for 99mTc (Sánchez-Crespo et al., 2002).

Linear regression coefficients are clearly not perfect descriptors of the profiles presented in Fig. 1. However, linear regression is easy to comprehend and it identifies any general trend for blood flow or ventilation to change in a certain direction. The deviations at the extremes of the profiles are greatly influenced by the edge effect and also by the motion of the lungs during imaging. It is not possible to differentiate these effects from any true reduction of blood flow or ventilation at the lung edges. Although the edge effect might influence estimates of gradients in the different postures, it is less likely to influence differences between gradients in the three postures. We therefore believe that the effect of gravity is better estimated from the comparison of gradients after administration in different postures than from the study of only one posture.

At lung volumes below total lung capacity there is a gravitational gradient with deceasing alveolar size down the lung (Milic-Emili et al., 1966; Glazier et al., 1967; Brudin et al., 1987; Mayo et al., 1995). Dependent lung regions therefore contain more lung tissue per unit volume and the distribution of lung tissue is dependent on posture. Recently we demonstrated that the parenchymal shift with a change in posture is one important factor contributing to the depicted distributions when regional lung blood flow and ventilation are imaged in different postures (Petersson et al., 2007). However, the effect of the shift in tissue distribution with a change in postures is excluded if radiotracer administration in different postures is followed by imaging in one posture. In the current study all images were obtained in a single posture (supine). Lung tissue distribution was therefore equal for all images. Differences between images can thus only be explained by different distribution of blood flow or ventilation within the lung parenchyma (per alveolus) at the time of radiotracer administration. Thus, our study design allowed us to capture the effect of posture on the distribution of blood flow and ventilation within the lung vasculature and airways, respectively.

The non-uniform distribution of lung parenchyma is usually attributed to a gravitational gradient in pleural pressure (Milic-Emili et al., 1966; Glazier et al., 1967; Rehder et al., 1977; Hoffman, 1985; Hubmayr et al., 1987). In supine, cranial-to-caudal gradients represent gradients within an isogravitational plane, i.e. tissue distribution at the time of imaging can be assumed to be uniform in this direction (Glazier et al., 1967; Rhodes et al., 1981). Thus any gradient in blood flow or ventilation in this direction can be assumed to represent a gradient per alveolus.

A limitation of the present study is that different subjects were studied in the horizontal and upright postures. The main result of the study is the significant redistribution of blood flow and regional ventilation to caudal regions in the upright posture. There was no overlap between the cranial-to-caudal blood flow gradients observed in subjects prone or supine and the gradients observed in upright subjects (Fig. 2). For regional ventilation, only one subject had a cranial-to-caudal gradient that was greater than any gradient observed in the upright subjects (Fig. 2). We therefore consider it unlikely that differences between the subjects studied upright vs. supine and prone are responsible for the main findings of our study. Breathing against a PEEP of 2.5 cmH2O during image acquisition was employed only in the upright subjects. This was done to maintain a similar lung volume during imaging as during radiotracer administration (Petersson et al., 2004). It is unlikely that this influenced the major results of our study, which is supported by two experiments where we repeated imaging with and without PEEP and found no effect of PEEP on the imaged profiles (data not shown).

4.2. Regional distribution of blood flow

Animal studies, using high-resolution methods, have found posture to have only a relatively minor influence on the regional distribution of blood flow (Glenny et al., 1991a, 1999). In contrast, prior studies of upright humans have found a greater blood flow to dependent regions (West and Dollery, 1960; Bryan et al., 1964; Anthonisen and Milic-Emili, 1966; Glazier and DeNardo, 1966; Hughes et al., 1968; Arborelius and Lilja, 1972; Landmark et al., 1977), suggesting a major effect of gravity on regional blood flow. In a primate study, Glenny et al. (1999) found a significant redistribution of blood flow in the upright posture, although the influence of gravity was small in relation to the overall blood flow variability. In the current study we found a similar effect of gravity in healthy human subjects. In the prone and supine postures we observed a more uniform distribution of blood flow between cranial and caudal regions, which is in agreement with previous studies demonstrating either no or small gradients in either direction (Bryan et al., 1964; Reed and Wood, 1970; Arborelius and Lilja, 1972; Engel and Prefaut, 1981; Almqist et al., 1997).

Our previous study suggests that gravity does not have a major influence on the distribution of blood flow in the ventral-to-dorsal direction. In contrast, in the current study we found a clear effect of posture (gravity) on the distribution of blood flow along the cranial-
to-caudal direction. According to the zonal (West et al., 1964) model the greatest gradient is located within zone 2.

The comparison of the ventral-to-dorsal gradients is a comparison of zone 3 gradients (Kaneko et al., 1966; Maeda et al., 1983). In contrast, the comparison of the cranial-to-caudal gradients in the horizontal postures vs. the upright postures represents a comparison of the gradients within an isogravitational plane with the gradient within zones 1–3. Any effect of gravity is therefore more likely to be observed in the comparison of the cranial-to-caudal gradients than in the comparison of the ventral-to-dorsal gradients. Accordingly, Glenny et al. demonstrated that gravity explains 25% of the blood flow heterogeneity in upright baboons, but only 7% and 5% in the supine and prone postures, respectively (Glenny et al., 1999).

4.3. Regional distribution of ventilation

We found an even distribution of ventilation between cranial and caudal regions in the supine and prone postures, but greater ventilation in caudal regions when sitting upright (Figs. 1 and 2). Previous studies have obtained comparable results (West and Dollery, 1960; Bryan et al., 1964; Anthonisen and Milic-Emili, 1966; Glazier and DeNardo, 1966; Milic-Emili et al., 1966; Bake et al., 1967; Hughes et al., 1968; Newhouse et al., 1968; Secker-Walker et al., 1975; Rehder et al., 1977; Amis et al., 1984; Orphanidou et al., 1986). In our prior study we could not demonstrate any significant effect of gravity on the ventral-to-dorsal distribution of ventilation in the supine and prone postures. Thus, as it is for regional blood flow, the effect of gravity on regional ventilation is most apparent in the cranial-to-caudal direction.

4.4. Regional ventilation-to-perfusion ratios

The uniformity of ventilation-to-perfusion ratios in both the supine and prone postures is in agreement with recent studies (Mure et al., 2001; Musch et al., 2002) and with similar arterial oxygenation in these postures at normal gravity (Rohdio et al., 2003). Although a change from the supine or prone posture to sitting upright was associated with a redistribution of both blood flow and ventilation to caudal regions, the redistribution was greater for blood flow than for ventilation. In the upright posture the ventilation-to-perfusion ratio therefore decreased in the cranial-to-caudal direction, which has been demonstrated in earlier studies (West and Dollery, 1960; Bryan et al., 1964; Glazier and DeNardo, 1966; Newhouse et al., 1968; Harf et al., 1978).

4.5. Conclusions

Using a novel application of the SPECT method, we have employed a modern imaging method to study regional lung blood flow and ventilation, in spontaneously breathing normal subjects in both upright and horizontal postures. We have shown that a change from the supine or prone posture to sitting upright, redistributes regional blood flow and ventilation within the lung parenchyma towards caudal lung regions, the redistribution being greater for blood flow than for ventilation. In contrast, the redistributions caused by a change between the supine and prone postures are much less. Hence, gravity plays a role for the distribution of regional lung blood flow and ventilation in the upright posture, but much less so in the prone and supine postures.

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References


