From the Department of Molecular Medicine and Surgery,
Section for Radiology, Karolinska University Hospital and
Karolinska Institute, Stockholm, Sweden.

The effect of posture, airway pressure and anesthesia on regulation of the regional ventilation and perfusion distribution in healthy humans

Sven Nyrén

M.D.

Karolinska Institutet

Stockholm 2010

Published and printed by Karolinska University Press Box 200, SE-171 77 Stockholm, Sweden

Layout Ringvor Hägglöf

ISBN 978-91-7457-060-1

If you think this is clear
- just wait until I explain it to you.

My mother's school teacher

To my beloved wife and my two wonderful sons

Abstract

Gas exchange has been observed to vary with posture in adult respiratory distress syndrome (ARDS) patients. In this thesis, the effect of posture on the regional distribution of ventilation (V) and perfusion (Q) in the lungs under normal breathing with and without continuous positive airway pressure (CPAP) and during general anesthesia with mechanical ventilation was studied. Additionally, endogenously produced nitric oxide (NO) may influence the effect of posture on the Q distribution and this was also studied. Further, since inhalation anesthesia known to impair pulmonary gas exchange, we investigated the effect of inhaled sevoflurane on the V and Q regional distribution. All studies were performed on healthy volunteers with Single Photon Emission Tomography (SPECT). Isotope tagged albumin macro aggregates, trapped in the pulmonary capillaries were used to measure regional Q and isotope labeled aerosol or labeled ultrafine carbon particle aerosol to measure regional V.

Prone posture, compared to supine, favors a more uniform Q distribution in the anterior to posterior direction both awake and during mechanical ventilation. CPAP, compared to normal breathing, redistributes Q to more dependent parts of the lungs, resulting in a less beneficial V/Q matching. We found indications of a higher release of endothelial NO in dorsal, compared to ventral, parts of the lungs. This may contribute to the more uniform Q distribution seen in prone compared to supine posture. NO from paranasal sinuses contributes to a more homogenous Q distribution in upright position. Inhalation anesthesia with sevoflurane decreases Q distribution heterogeneity but increases V/Q matching heterogeneity. No significant influence on the V distribution was observed in any of the studies. In conclusion, all studied factors in healthy volunteers have an effect on the Q regional distribution but not on V, suggesting that variations in V/Q ratio distribution are a consequence of changes in the intrapulmonary Q distribution.

Keywords: Anesthesia, ALI, ARDS, CPAP, Dual isotope, Mechanical ventilation, Nitric oxide, Nuclear medicine, Prone position, Regional ventilation, Regional perfusion, SPECT.

List of Publications

I Nyrén S, Mure M, Jacobsson H, Larsson SA, Lindahl SG.
Pulmonary perfusion is more uniform in the prone than in the supine position: scintigraphy in healthy humans.
J Appl Physiol. 1999 Apr;86(4):1135-41.

II Mure M, Nyrén S, Jacobsson H, Larsson SA, Lindahl SG.
High continuous positive airway pressure level induces ventilation/ perfusion mismatch in the prone position.
Crit Care Med. 2001 May;29(5):959-64.

III Rimeika D, Nyrén S, Wiklund NP, Koskela LR, Torring A, Gustafsson LE, Larsson SA, Jacobsson H, Lindahl SG, Wiklund CU.
Regulation of regional lung perfusion by nitric oxide.
Am J Respir Crit Care Med. 2004 Aug 15;170(4):450-5.

IV Nyrén S, Radell P, Lindahl SG, Mure M, Petersson J, Larsson SA Jacobsson H, Sánchez-Crespo A.
Lung Ventilation and Perfusion in Prone and Supine Postures with Reference to Anesthetized and Mechanically Ventilated Healthy Volunteers.
Anesthesiology. 2010 Mar;112(3):682-7.

V Nyrén S, Radell P, Mure M, Petersson J, Larsson SA Jacobsson H, Lindahl SG, Sánchez-Crespo A.
Inhalation anesthesia increases V/Q regional heterogeneity during spontaneous breathing in healthy subjects.
Accepted for publication in Anesthesiology.

VI Sanchez-Crespo A, Hallberg J, Lundberg JO, Lindahl SG, Jacobsson H, Weitzberg E, Nyrén S.
Nasal nitric oxide and regulation of human pulmonary blood flow in the upright position.
J Appl Physiol. 2010 Jan;108(1):181-8.

CONTENTS

List of abbreviations..... 11
Introduction..... 13
Background..... 13
The history of pulmonary physiology..... 13
The development of nuclear medicine and its use in respiratory physiology..... 14
Methods determing V,Q and V/Q matching..... 15
Current knowledge on regional V, Q and V/Q distribution in the lung..... 18
General definitions..... 18
Factors influencing regional V distribution..... 19
Factors influencing regional Q distribution..... 19
Factors influencing V/Q matching..... 23
ALI/ARDS..... 24
Aims of the Thesis..... 27
Material and Methods..... 29
Subjects and patients..... 29
Radiopharmaceuticals..... 29
Imaging technique..... 30
Anesthesia..... 30
Experiment setups..... 30
Data analysis..... 34
Results..... 37
Effects on V,Q AND V/Q matching from posture, during normal breathing (Studies I,II,V,VI)..... 37
V,Q and V/Q matching, effects from CPAP (Study I,II)..... 39
Effects on V,Q,V/Q distribution from posture during intravenous anesthesia with muscle relaxtion and mechanical ventilatin (Study IV)..... 40
The effect from endothelial NO on regional Q distribution (Study III)..... 40
Distribution of Q as a function of inhaled nasal NO (Study VI)..... 41
V,Q,V/Q distribution as a function of inhalation anesthesia (Study V)..... 41
Discussion..... 43
General limitations of the SPECT technique..... 43
Specific limitations in each study..... 44
Comparison with previous knowledge..... 45
Clinical implications..... 49
Future perspectives..... 50
Conclusions..... 51
Acknowledgements..... 53
References..... 55
Papers

LIST OF ABBREVIATIONS

ALI	Acute lung injury
ARDS	Acute respiratory distress syndrome
ATP	Adenosine tri phosphate
cAMP	Cyclic Adenosine mono phosphate
cGMP	Cyclic Guanosine mono phosphate
CMV	Controlled mechanical ventilation
CPAP	Continuous positive airway pressure (cmH_2O)
CT	Computed tomography
DTPA	Diethylenetriamine pentaacetic acid
eNOS	Enothelial nitric oxide synthase
$EtCO_2$	End tidal concentration of exhaled carbon dioxide
FBP	Filtered back projection
FiO_2	Fraction of inhaled oxygen
FRC	Functional residual capacity
FWHM	Full Width Half Maximum
GTP	Guanosine tri phosphate
HPV	Hypoxic Pulmonary Vasoconstriction
^{113m}In	Metastable indium
keV	Kiloelecron-Volt
L-NMMA	N^0-monomethyl-L-arginine
LyoMAA	Labeled macro aggregates of human albumin
MAP	Mean arterial pressure (cmH_2O)
MBq	Megabecquerel
MIGET	Multiple Inert Gas Elimination Technique
NO	Nitric Oxide
NOS	Nitric Oxide Synthase
PEEP	Positive end expiratory pressure (cmH_2O)
Q	Lung perfusion
SPECT	Single photon emission tomography
^{99m}Tc	Metastable technetium
V	Lung ventilation
VOI	Volume of interest
V/Q	Ventilation-perfusion ratio

INTRODUCTION

BACKGROUND

For oxygenation of blood and elimination of carbon dioxide a close contact between blood and air in the lungs is necessary (1,2). As neither perfusion nor ventilation of the lungs is uniform (3) it is important for gas exchange that well ventilated parts receive more blood flow than less ventilated parts and vice versa. At normal conditions this is effective and the blood levels of oxygen and carbon dioxide, as well as blood pH, are kept within close limits (4). During disease this regulation is often disturbed which sometimes results in critically low levels of blood oxygen. Conditions that disturb this coordination of regional ventilation (V) and lung perfusion (Q) include pulmonary embolism and lung atelectasis. In other conditions, such as Acute Respiratory Distress Syndrome (ARDS) and Acute Lung Injury (ALI), the anatomical location of this disturbance is not so obvious. Many of these conditions induce increased venous admixture and shunting (mixed venous blood passing to the systemic arteries without being oxygenated in the lungs). Shunt is also induced by general anesthesia for unclear reasons (5). It has been shown that body posture can influence blood oxygenation (6-12) but the underlying explanation remains unclear.

The overall aim of this thesis is to investigate factors related to posture or intensive care that could influence regional V, Q and V/Q distributions and if these factors could explain the improvement in blood oxygenation in patients with acute lung injury (ALI) seen during prone posture. We studied if posture, both awake and during anesthesia with mechanical ventilation, continuous positive airway pressure (CPAP) or inhalation anesthesia could influence regional V,Q and V/Q distributions in healthy volunteers. Moreover, we investigated if endothelial and autoinhaled nitric oxide (NO) modified regional Q distribution or if an inhalational anesthestetic agent could influence V/Q matching.

The history of pulmonary physiology

Since ancient times it has been obvious that breathing is essential for life. An Egyptian papyrus from 1820 BC stated that "As to the air that penetrates the nose. It enters into the heart and the lungs. They are those which give air to the entire body." A Roman physician, Galen (129-199AD), made a number of studies into anatomy and physiology. He, unfortunately, misunderstood some basic mechanisms. As he remained an authority for more than 1000 years, his dogmas hindered further development of understanding. After the decline of the Roman Empire much of the knowledge in anatomy and physiology from Rome and Ancient Greek disappeared from Europe but some survived in the Byzantine and Arabic empires (13).

With the Renaissance, through human dissections and questioning of old dogmas, the understanding of the human anatomy increased. Through the microscope, the structure of the lung was studied from the 17th century. During this period, the similarity between combustion and metabolism gradually became evident and in the 18th century it was proven that the metabolism is taking place in the tissue and not in the heart and lungs and that excess of carbon oxide stimulates breathing (13).

In the 19th century, improved laboratory equipment made accurate measurements possible. The mechanics of breathing and lung volumes were studied by Hutchingson, who constructed the first spirometer and produced the first lung compliance curve in humans. It was also shown that the partial pressure rather than the concentration of a gas affects biological systems (14). The nature of blood oxyhaemoglobin dissociation was also reveled during late 19th and early 20th century (13) and quantitative measurements of the chemical control of breathing and the interactions between carbon dioxide and exercise were published by Haldane and Priestly in 1905 (13).

Von Neergard found in 1929 that the tissue elasticity alone could not explain the recoiling properties of the lung and that the surface tension of the alveoli is lower than expected from Laplace's law (15). He therefore proposed the existence of another, unknown, factor and in 1955 Pattle demonstrated that lung tissue contains an insoluble protein layer and thus discovered the surfactant (13).

Major contributions for the understanding of the pulmonary function were made by von Euler and Liljestrand in the mid 1940:ies. They found, among other things, that lung vessels, opposed to vessels in other tissues, reacts to low oxygen concentrations with vasoconstriction (16-18).

From the late 1950:ies studies on regional distribution of Ventilation, Perfusion and V/Q matching were initiated (19). Furchgott and Zawadzki reported, in 1980, that acetylcholine had a constricting effect on pulmonary vessels if endothelial cells were damaged, but a relaxing effect if the endothelial lining was intact (20). This led to the suspicion of a labile, intermediate, endothelial derived factor which was named "endothelial-derived relaxing factor, EDRF" (21). Later, this substance was proven to be identical with nitric oxide, NO (22). This opened a new field of research (23,24).

The development of nuclear medicine and its use in respiratory physiology

In 1896, Becquerel described natural radioactive substances. In 1913 de Hevesy proposed the "tracer principle", the idea that radioactive substances could be used to follow biological processes without interfering with them. Geiger-Müller counters were first used to locate radioisotopes but from the 1940:ies imaging equipments were gradually developed and refined. In the 1960: ies the vertical distribution of blood flow (Q) in the erect man was studied. Inert, radioactive gases (e.g. ^{133}Xe, ^{81m}Kr) were either inhaled or injected and detected by external scintillation counters. The spacial resolution was poor, amd it was not possible to depict inhomogeneities in the isogravitational plane. However, all these studies showed a higher perfusion in basal, dependent parts of the lungs compared to apical (25,26). Investigations on the distribution of ventilation, with radioactive inert gases, (27) showed a similar pattern, with basal dominance. These findings of a basal dominance of Q eventually led to the so-called "West model" of a gravity dependent Q distribution in the lungs (28).

With the arrival of the gamma camera technique in the 1960:ies there were tools for a better localization of ventilation and perfusion in the lungs. This technique made it possible for two-dimensional images of the distribution of radiotracers in the lungs (29). In the 1960:ies it was also found that radioisotope labeled albumin aggregates could be injected intravenously and emission images, reflecting the lung perfusion, could be produced. Pulmonary scintigraphy with radioisotope labeled macroaggregates (microspheres) for diagnosis of lung embolism (30) were established in the 1970:ies, but has in recent years partly been replaced by Computer Tomography. It soon became evident that other conditions than pulmonary embolism could result in perfusion

defects and that this could lead to diagnostic errors. One cause of perfusion defects is, for instance, local vasoconstriction secondary to regional hypoxia. By combining perfusion images with images representing ventilation the diagnostic precision could be improved (30). Many of these techniques were used in the early phases of basic research in pulmonary physiology (31,32). Technical advances including increased spatial resolution (3), 3D acquisitions with SPECT (33), improved scatter and attenuation correction (34, 35) and simultaneous registration of isotopes with different energy levels (35) have in recent years improved the possibility of quantitative measurements. SPECT has the capability of quantifying regional V,Q and V/Q and by subtraction or dual-isotope techniques this can also be made simultaneous (35,36). An alternative approach, the PET technology, which also produces 3D images (37), was developed during the 1960:ies and 70:ies. These later, high precision imaging techniques have led to new knowledge and the questioning of previous results (38-41).

Methods determing V,Q and V/Q matching

Global measuresment

The respired volumes can be measured with a spirometer, either directly or indirectly through integrating gas flow. An alternative is a pletysmograph that indirectly measures lung volumes from the cross sectional area of the chest and abdomen. The residual capacity has to be determined by other means, either by gas dilution or using a body pletysmograph (42,43).

By repeated measurement of tracer concentration in the blood after it has passed the lungs and comparing it with the known amount injected before it reached the lungs, pulmonary blood flow and blood volume can be calculated. This is known as the tracer dilution technique. The pulmonary blood flow can also be calculated by dividing the amount of oxygen extracted from the lungs with the difference in oxygen content between arterial and venous blood, known as the "Ficks's principle". Both these methods require invasive procedures (1,44). An overview of the techniques is presented in table 1.

Table 1. Overview of methods used for measurements of global V,Q and V/Q, without regional or anatomical information.

	Measure			Advantages	Disadvantages
	V	Q	V/Q		
Spirometry	+	-	-	Cheep, harmless	Limited information
Single breath	-	+	+	Simple, portable equipment	Inexact
Tracer dilution	-	+	-	Exact	Invasive

The multiple inert gas technique (MIGET)

The matching of V and Q can be studied in the lung as a whole with physiologic methods. The arterial blood gases give an indirect measurement of the effectiveness of V/Q matching. The gas exchange in the lungs can also be studied via measurements of the exchange of probe gases, a principle that has been elaborated in the MIGET technique (1,45). Tracer amounts of six inert gases are dissolved in saline and infused i.v. The concentrations of the gases are thereafter measured in exhaled air and in blood. From this data V/Q ratios can be calculated and presented in graphic

form. This gives excellent information on the relative occurrence of different V/Q ratios. The method is complicated to perform and has mainly been used for research but has served as a reference for other studies (36).

Regional and anatomical measurements

A general overview of methods depicting regional V,Q and V/Q distributions that yield both regional and anatomical information is presented in table 2.

Table 2. Overview of imaging and other techniques that yield both regional and anatomic used in lung physiology.

	Measure			Advantages	Disadvantages
	V	Q	V/Q		
Microsphere	+	+	+	Spatial resolution. Simultaneous multiple tracers. "Freezes" conditions.	Animals have to be scarified, not physiologic conditions, expensive
MRI	+	+	+	No ionizing radiations. Multiple measurements.	V difficult to monitor. Non-linear quantification. Images represent only recumbent conditions.
CT	-	+	-	Spatial resolution.	Images represent only recumbent conditions. Ionizing radiation.
PET	+	+	+	Sensitivity and spatial resolution.	Images represent only recumbent conditions. Ionizing radiation.
Impedance	+	-	-	Continuous measurement possible, harmless	Very low special resolution. Only semiquantative.
SPECT	+	+	+	Quantification. "Freezes" conditions.	Partial volume effects. Ionizing radiation

MICROSPHERES

The microsphere technique is based on the principle that microspheres that are injected i.v. and/or inhaled are trapped in the lungs in direct proportion to blood flow or ventilation (38,46,47). The microspheres are labeled with fluorescent dye or radioisotopes and administered at the condition that is in focus for the investigation. After the microspheres are trapped, the animal is sacrificed and the lungs fixated and dissected into small pieces. This gives an exact measurement and multiple conditions can be estimated simultaneous as multiple dyes or radioisotopes can be used. Disadvantages with this method are that the lungs are fixated at full expansion and that only animals can be used. Full expansion does not reflect the distribution of lung parenchyma in a living organism (48), but on the other hand the measurements are believed to reflect the distribution in relation to alveoli (47).

MAGNETIC RESONANCE IMAGING (MRI)

Magnetic resonance imaging (MRI) has been used to depict local lung perfusion and lung density. As the method is not based on ionizing radiation it is harmless and very suitable for repeated measurements in humans. Vascular anatomy can be imaged and flow in the larger vessels measured with "phase-contrast imaging" (49,50). Imaging of the parenchyma has some inherent problems, especially the "susceptibility artifact" which result in signal loss produced by the proximity between areas with different magnetic properties, here vessels and air (51). There is also a non-

linear relation between the concentration of contrast agents and signal, which can cause problems in measurements of local perfusion (52). This problem can be solved with the "spin labeling" or "arterial spin labeling" technique (53-57).

The ventilation is not visible with conventional MRI but can be demonstrated with direct imaging of hyperpolarized noble gases (^{129}Xe and ^{3}He) (58,59). Attempts have been made to combine technologies into simultaneous or almost simultaneous ventilation and perfusion imaging with MRI (60). A cheaper method is indirect imaging of ventilation through the effects on the local magnetic properties from the inhalation of pure oxygen (61). This, however, requires repeated measurements and effects from the oxygen on the distribution of ventilation cannot be excluded. Attempts to study the distribution of lung water and to estimate local pleural pressure with MRI has also been made (62). Common to all MRI studies of the lungs are that they are limited to the conditions in the recumbent position in the MRI camera.

POSITRON EMISSION TOMOGRAPHY (PET)

This technology, like single photon emission tomography (SPECT), is based on 3D depiction of radioisotopes within the lungs. In PET, isotopes emitting positrons (positively charged electrons) are used. When a positron is emitted from the decaying radioisotope, it soon encounters a negatively charged electron. When this happens, both are annihilated and the mass is transformed into energy in the form of two photons (37). The energy of these two photons is 511 keV and they are emitted at an angle of almost exactly 180^{O}. A ring of detectors registers these photons and as they are detected simultaneously the location of the annihilation event can be determined. The positron travels only a short distance before it encounters an electron and gets annihilated and, thus, the position of the annihilation serves as a good estimation of the location of original decay of the isotope (37). As the energy of the photons are always 511 keV independent of what radioisotope that originally produced the positron, simultaneous V and Q imaging cannot be made. By dynamic studies of the turnover of ^{13}N (63,64) in the lungs a number of parameters, including local ventilation, perfusion and V/Q matching can be calculated but only in recumbent position. ^{15}O labeled water can be used for estimation of regional Q, local blood volume and water content (65).

COMPUTED TOMOGRAPHY (CT)

From axial reconstructions of repeated measurement of the radio density a 2D image of a slice in the thorax can be calculated by this technology. If a bolus of radio dense contrast medium is injected in the bloodstream, followed by repeated 2D images, regional perfusion in that slice can be calculated with accuracy (66). Ventilation cannot be measured with CT but regional lung aeration could be used as an alternative.

ELECTRIC IMPEDANCE TOMOGRAPHY (EIT)

This new imaging method is based on measurements of the resistances between several measuring sites that are placed round the chest (67). From this, a coarse map of the local electric impedance in a section of the chest can be made and air content, pleural fluid, atelektasis can be monitored. The images exhibit a low special resolution and measurements are only semiquantitative. The method is harmless and the equipment can be attached to a patient for a long period without causing any inconvenience, which makes it suitable for continuous measurements in intensive care patients.

SINGLE PHOTON EMISSION COMPUTED TOMOGRAPHY (SPECT)

An overview over radioisotopes commonly used for SPECT, their characteristics and usual applications are presented in table 3.

Table 3. Example of radioisotopes used as tracers in SPECT imaging of the lungs.

Radio-nuclide	Half-life	Energy	Imaging application
^{99m}Tc	6 h	140 keV	Labeling of albumin microspheres for regional perfusion ^{99m}Tc-DTPA aerosol or Technegas for regional of ventilation
^{113}Xe	5.3 days	81 keV	Steady state for alveolar volume, washout for regional ventilation
^{81m}Kr	13 sec	190 keV	Inhalation for regional ventilation, i.v. infusion for regional perfusion
^{113m}In	100 min	392 keV	Labeling of albumin macro aggregates or technegas for perfusion

SPECT is based on transforming multiple planar images into a three dimensional reconstruction. The photons from gamma ray emitting radioisotopes are detected with one or a number of planar detectors, which are rotated round the individual examined. The radioisotopes (Table 3) are usually attached to a carrier (ligand) and the compound is called a radiopharmaceutical (68). A number of radioisotopes can be utilized but few are practical and cost effective. The most commonly used isotope is ^{99m}Tc (69), which is produced in a ^{99}Mo/^{99m}Tc-generator. This technique makes the tracer always available at the laboratory. The tracers are, hereby, comparably inexpensive and the half-life of 6 hours makes them easy to handle while a disadvantage is the exposure to ionizing radiation. Positive is the fact that the injected microparticles and inhaled particles or aerosol remains fairly stable in the lungs after being fixed (30,70,71). Hereby, the SPECT images reflect the conditions at administration. The distribution of V,Q and V/Q during conditions other than the ones in the SPECT camera can thus be imaged, e.g. Q distribution during hypergravity (72) and V,Q and V/Q distribution in upright (73). Another possibility is subtraction or dual isotope techniques that enable direct visualization of the changes in V or Q distribution between two conditions in the same individual.

CURRENT KNOWLEDGE ON REGIONAL V, Q AND V/Q DISTRIBUTION IN THE LUNG

General definitions

Local ventilation (V) refers to the amount of air that enters a volume of lung during a limited time frame and perfusion (Q) to the amount of pulmonary blood flow entering a volume during a time period. In this thesis, every image voxel represents a fraction of the total V or Q in the lung. V/Q can be calculated as the quotient of the relative V and Q in a larger volume of interest (VOI) as in **study II**, or local V/Q in a VOI can be defined as the mean of pixel wise calculated V/Q values as in **Study IV** and **V**. There is some confusion in the literature as local V,Q and V/Q some times have been defined as distribution per unit lung tissue and sometimes related to a volume in the chest. With the latter definition, the posture at registration will influence the results.

Dead space is described ventilation not contributeting to gas exchange. This can be divided into anatomical dead space from gas in the airways and "alveolar dead space" which is caused by variations in alveolar V/Q matching (1). **Shunt, or venous admixture** is defined as the fraction of venous blood passing to the arterial side without being oxygenated. It consists of "true shunt", blood that have not at all been in contact with the inhaled air, but also of blood that have passed alveoli with low V/Q values. The two causes of impaired gas exchange can be separated by studying the how arterial blood oxygenation reacts to increase the fraction of inhaled oxygen (1). **Heterogeneity** for V,Q or V/Q can be defined as the sum of squares of the deviation from the mean pixel value in the entire lungs or in each isogravitational plane.

Factors influencing regional V distribution

The posture is considered to influences distribution of ventilation. In awake humans, at ordinary respiration rates, dependent parts of the lungs are more ventilated than the non-dependent part. This is true in lateral, supine and erect positions where lowest parts are most ventilated (1). In prone, there is no consensus on the distribution of ventilation in awake humans. Some authors claim a more ventral, dependent (31,74-76) and some claim a more uniform or dorsal, non-dependent dominance of the lung ventilation (77,78). Rapid inhalation makes the ventilation more uniform (1). Anesthesia, both with spontaneous breathing and during mechanical ventilation are by some believed to reverse the distribution of regional ventilation so that non dependent parts are more ventilated both supine and in lateral position (1). A more dependent distribution of ventilation has been explained by less expanded alveoli in dependent parts at functional residual capacity, believed to be due to a vertical gradient in pleural pressure (79) and compression from the heart (80). Alveoli are more compliant when less expanded and thus receive a larger fraction of inhaled gas. The reason for the gradient in pleural pressure is not clear, but the weight of the lungs seems to be of importance (80). The effect is most obvious in erect posture, were apical alveoli tends to be more expanded, but the concept has been applied to other postures (3,80). In prone, the vertical gradient in pleural pressure seems to be smaller (62), and this might be explained by the anatomy, e.g. shape and compliance of the chest wall (80) or compression from the heart in supine (81). The regional distribution of V is also influenced by changes in end expiratory lung volumes, tidal volumes and mechanical ventilation. The relative effect may also be changed by posture (31,39,82-84). There are reports of heterogeneity of V that can not be entirely explained by the factors mentioned above (85). Factors influencing regional ventilation seem to be mainly of a "passive, mechanical" nature and the author has not been able to find any reports of active mechanisms altering the relative distribution of V in the lungs.

Factors influencing regional Q distribution

Gravity

It has for a long time been an accepted truth that gravity is the dominating factor determining the regional distribution of Q. This originates from West and collaborators who proposed in the 60:ies a model of up to 4 zones of pulmonary blood flow along the gravitational axis (3,4,28,38). An outline of this theory is presented in table 4.

Table 4. The West model of the influence from gravity on lung perfusion.

	Pressure difference driving the blood flow	Resulting blood flow
Zone 1	Alveolar > Arterial	No flow in the pulmonary arteries
Zone 2	Arterial > Alveolar	Gradually increasing flow along the direction of gravity
Zone 3	Arterial > Venous	
Zone 4	Arterial > Tissue	Decreasing flow in most dependent parts

The **Zone 1** is the highest, most non-dependent and **Zone 4** the lowest, most dependent. The underlying idea is that regional blood flow is governed by the balance between pressure in the lung arteries, pressure in alveolar air and pressure in the lung veins. **Zone 1** is seldom present at normal conditions in recumbent. In this zone, the intra alveolar air pressure is higher than the pressure in the pulmonary artery which inhibits local perfusion. These conditions may appear in the uppermost parts of the lungs in erect position. **Zone 2** covers most of the lungs and here the flow is governed by the difference between the pressure in the pulmonary arteries and the pressure of the gas in the alveoli. As the gas pressure is the same in all parts of the lungs and arterial pressure gradually increases, down this zone, the net result is a gradual increase in Q towards lower parts of this zone. In **Zone 3** the flow is determined by the differences in pressure between pulmonary arteries and veins, this difference also increases down the direction of gravity and hereby Q. Sometimes, a decreased blood flow is seen in the most dependent parts of the lungs and this is called **Zone 4**. Here, from the weight of the lung, the pressure of the surrounding lung tissue is larger than the pressure in the veins. The force driving the blood flow is the arterial pressure minus the tissue pressure which results in a decreased Q in the most dependent parts (38).

Hypoxic pulmonary vasoconstriction

In the systemic circulation vessels dilate in response to tissue hypoxia. The opposite occurs in the pulmonary vessels, which constrict in response to both low PO_2 in the pulmonary artery and to low PO_2 in the alveoli, the latter stimuli being the most potent (17). The response is non linear and strongest if the oxygen levels are very low. The mechanism is beneficial as it diverts pulmonary blood flow away from lung portions with low ventilation and thus helps to optimize V/Q relationships (17). It is doubtful if this mechanism is active during normoxia (86) but it is important in the fetus because it diverts blood flow from the unventilated lungs. This reflex mainly affects arterioles of 30-200 μm in diameter. An onset of constriction within 5 minutes of local hypoxia is seen. In vitro and in animal studies have shown that this is followed by relaxation and thereafter a second vasoconstriction after 40 minutes (17).

Cardiac output and lung inflation

In the absence of pathologic conditions the blood flow through the pulmonary circulation is approximately equal to that in the systemic circulation. Pulmonary circulation can adapt to a large increases in blood flow with only a small increases in pulmonary arterial pressure. The reason is a simultaneous decrease in vascular resistance which is believed to be an effect from passive distention, and recruitment of lung vessels (17). It is suggested that this effect is most pronounced in **Zone 1** which may imply a less dependent Q distribution with higher cardiac output but this has, to my knowledge, not been proven. Lung inflation influences resistance in the pulmonary

vessels. At FRC the resistance is minimal and increases at both higher and lower lung volumes, this being most pronounced at high lung volumes (17). During CPAP, both lung volumes and the intra alveolar pressure increase which might increase resistance in non-dependent lung vessels more than in dependent vessels. This may result in a redistribution of Q to dependent parts of the lungs during CPAP (17).

Neural control of pulmonary perfusion

Transplanted lungs retain hypoxic pulmonary vasoconstriction which implies that this reflex is not nerve mediated. Adrenergic – sympathetic nerves, originating from the first five thoracic nerves, follows the pulmonary vessel and act on smooth muscles in arteries and arterioles down to 60 μm in diameter. Both α_1 receptors, mediating constriction and α_2 and β_2 receptors mediating relaxation and dilatation are present in the vessel walls. The net effect from sympathetic stimulation seems to be a slight constriction and increase in resistance in the pulmonary vascular circulation, less pronounced than in the systemic circulation. Cholinergic – parasympathetic nerves from the vagus nerve release acetylcholine (ACh). Via effects on the endothelium and subsequent release of NO this leads to vasodilatation, but the significance of cholinergic nerves in the lungs is unclear (17).

Humural control and cellular mechanisms controlling pulmonary vascular tone

The most studied substances affecting the vascular resistance of the lungs are listed in Table 5. There are several types of receptors, with sometimes opposing effects on both endothelial and smooth muscle cells of the pulmonary vasculature why the effect on vascular resistance from a substance is difficult to predict. Some of the effects are dependent on a preserved endothelial lining and it seems that NO is a common pathway for producing vascular relaxation from a variety of stimuli. Many have only been studied *in vitro* or in animals why there role in humans is unclear (17). There are no reports, to my knowledge, on local difference implying that any of these substances could cause regional redistribution of pulmonary blood flow.

Table 5. Effect of substances on flow in the pulmonary circulation. ↑=increased flow, ↓=decreased flow. Receptor in parenthesis. *Modified from reference 17.*

Substance	Effect
Noradrenalin	↓ (α_1), ↑ (α_2)
Adrenaline	↑
Acetylcholine	↑
Histamine	Variable (H_1), ↑ (H_2)
5-Hydroxytryptamine (5HT)	Variable
Adenosine triphosphate (ATP)	↓ (P_{2x}), ↑ (P_{2y})
Adenosine	↓ (A1), ↑ (A2)
Tromboxane A2	↓
Prostacycklin (PGI2)	↑
Substans P	↑
Neurokinin A	↓
Vasoactive intestinal peptide (VIP)	↑
Angiotensin	↓
Atrial natriuretic peptide (ANP)	↑
Bradykinin	↑
Endothelin	↓ (ET_A), ↑ (ET_B)
Adrenomedullin	↑
Vasopressin	↑

Nitric oxide (NO)

Nitric oxide, also called nitric monoxide, is a small molecule the readily passes through membranes. It has a molecular weight of 30 Dalton (87). NO is produced through the oxidation of L-arginine to citrullune, catalyzed by NO-synthase (NOS) (88, 89). There are three known forms of NOS: nNOS only found in neural tissue, eNOS found in vascular endothelium and iNOS (inducible NOS in macrophages, neutrophiles and smooth muscle cells). The two former enzymes depend on the presence of Ca^{2+} ions and calcium modulated protein and the last enzyme is sometimes called Ca^{2+} independent NOS (90). NO can also be produced non-enzymatically during ischemia but the pathway is not clear (91).

The mechanism of induced vascular relaxation in the pulmonary circulation through the NO pathway is initiated by endothelial cell stimulation via a receptor which leads to an increase in intracellular Ca^{2+}. This activates NOS which promotes the conversion of L-arginine to one molecule of L-citrulline and one NO molecule. NO is mainly produced locally in the endothelium and its effect is pre capillary dilatation (92). As NO rapidly binds to hemoglobin it is inactivated before reaching the systemic circulation (93). The NO molecule diffuses freely over to the neighboring smooth muscle cell where NO activates guanelate cyclase.
This enzyme converts GTP to cGMP which decrease intracellular Ca^{2+} levels causing relaxation of the smooth muscles via enzyme activation (17). CGMP also directly causes smooth relaxation through Ca^{2+}–desensitization of myofilaments (94). Increased levels of Ca^{2+} in the smooth muscles, on the contrary, induce contraction by Ca^{2+} activating myosin light chain kinase by forming complex enabling interaction with actine and by binding with caldesmon which is detached from muscle filament and makes them available for sliding (94).

Humans and other higher primates release NO from the nasal cavity and paranasal sinuses which is conveyed to the lungs with the inhaled air (95-97). This autoinhalation of endogenous NO from the nasal airways has been shown to improve arterial oxygenation and reduce pulmonary vascular resistance (95), but it has no direct influence on the systemic circulation (93). The importance of NO in the respiratory system is well studied (98) and exogenous NO added to the inhaled air has been extensively used in clinical practice to reduce pulmonary hypertension and improves gas exchange in intensive care patients (99).

The function of NO in the pulmonary circulation can be studied indirectly by blocking NOS and thereby the endogenous production. Synthesis of NO can be partially blocked by competitive inhibitors to the substrate L-Arginine. L-N^G-monomethyl-L-arginine (L-NMMA), a non selective inhibitor, can be used for this purpose (88).

Branching architecture of the vessels

With the introduction of high resolution techniques like fluorescent microspheres a heterogeneity of blood flow, not explained by gravity, has been observed (40,41,100). A considerable heterogeneity of pulmonary blood flow remains during weightless conditions (101,102). A similar heterogeneity in regional ventilation distribution (103) has been seen, but matching between perfusion and ventilation seem to remain tight (104,105). This has led to a theory that the vascular structure is the most important determinant of regional blood flow distribution (106).

Drug effects on the pulmonary circulation

Pulmonary hypertension is a common complication to chronic hypoxia from a variety of lung diseases. This often leads to right sided heart failure, which can be fatal. Despite a wide range of receptor-agonist systems in the pulmonary vascular systems, there are few effective drugs regulating the pulmonary circulation (17). One reason for the limited therapeutic arsenal is the

unspecific nature of many receptors and that the drugs often have adversary effects on the systemic circulation. Another reason is that pulmonary vasodilatators reduce the hypoxic pulmonary vasoconstriction which is the main mechanism for compensating for poor respiratory function at respiratory disease. This can sometimes be avoided by delivering drugs by inhalation (17).

INHALED DRUGS

Inhaled NO has been used as a selective pulmonary vasodilatator and no systemic effects are seen as it is rapidly inactivated. It also improves V/Q matching since it stimulates blood flow to well ventilated areas of the lungs. In combination with oxygen NO is, however, rapidly oxidized to NO_2 which reacts with water to injurious nitric and nitrous acids which can result in pneumoitis and pulmonary edema, limiting the concentrations of NO that can be used. In addition, rebound phenomenon, with increased pulmonary arterial pressure, can occur if NO treatment is withdrawn rapidly (17). Prostacyclin i.v. has been used to reduce pulmonary pressure but causes significant adverse effects from the systemic circulation, which can be avoided by inhalation administration (17).

SYSTEMIC DRUGS

Angiotensin-converting enzyme inhibitors reduce pulmonary vascular resistance during long time treatment. Angiotensin II receptor antagonists can be used for more rapid reduction in pulmonary artery pressure without detriment in oxygen saturation (17). Phosphodiesterase inhibitors can inhibit breakdown of cAMP and cGMP and, thus, reduce intracellular Ca^{2+} with consequent relaxation of smooth muscles and reduced pulmonary hypertension (17). Calcium antagonists can reduce secondary pulmonary hypertension but may produce right heart failure due to its negative inotropic effect. Oxygen saturation may also drop as a result of negative effect on the hypoxic pulmonary vasoconstriction reflex. These two side effects limit the dose that can be administered (17). Endothelin receptor antagonists compete with endothelin on the A and B receptors, thus counteracting the constrictive effect and reduce pressure in the vascular circulation but the drugs are quite new and not fully studied (17). Anesthesia, especially with inhalation anesthetics, increases the alveolar-arterial oxygen tension difference, and the underlying mechanism is believed to be an increase in V/Q variability. The effect may be related to atelectasis formation (107). Formation of atelectasis are considered to secondary to a decrease in FRC (108,109,110) and the use of high concentrations of oxygen in inhaled air (39). Inhalation anesthetic agents decrease the force of the respiratory musculature (110), but the direct effect on the pulmonary circulation is more unclear. There are studies suggesting a vasodilatory effect through an inhibition of the hypoxic pulmonary vasoconstriction reflex (111) but also a direct vasoconstriction (112).

Factors influencing V/Q matching

Ideally, all inhaled air would meet the blood flowing through the lungs in each alveolus in equal proportions, but this is not the case. Part of the air does not get in contact with the bloodstream (dead space) and part of the blood does not get in contact with the air (venous admixture or shunt) (1). During normal conditions, in young people, dead space is around 30% of the inhaled air and venous admixture 1-2% but these fractions can increase significantly during disease, e.g. ALI/ARDS (1). Both ventilation and perfusion show a dependent dominance in upright, prone and supine postures, why V/Q matching is less affected by posture than each factor separately. Increase in dead space, increased venous admixture or shunt and increased heterogeneity in alveolar V/Q matching contribute to impaired gas exchange. As there is a high correlation

between alveolar ventilation and perfusion (3) there must be mechanisms coordinating the two. There is one known mechanism, the hypoxic pulmonary vasoconstriction (17), but as it is doubtful if this reflex is active during normal levels of oxygen in the inhaled air (87) also other mechanisms must be expected.

ALI/ARDS

In modern intensive care, pulmonary complications are common and show a high mortality rate. Severe complications such as ALI and ARDS cause death in over 40% of patients (113-117) and mortality has not decreased significantly in recent years (118). Treatment with high concentration of O_2 and high ventilator pressures has other adverse effects (119,120). In 1976 and 1977 two articles reported positive effects from treating patients with acute respiratory failure in prone position (6,7), but the method did not became widespread. In the late 1980:ies and in the 1990:ies the interest for this method was renewed, and the effects were confirmed by other studies (8-12). The physiologic background to this effect was, however, unclear. During the same period, the established truth, that gravity is the dominating factor influencing the local distribution of pulmonary blood flow (25,28,74,121,122) was coming under debate (40,41). The observation that vascular conductance is higher in the dorso-caudal lung regions irrespective of body posture (46) strengthened the concept that factors other than gravity are important for the understanding of the local distribution of lung perfusion. There are now several studies supporting that perfusion is more uniform (1,123) and that V/Q matching is better (124,125) in prone. The basic mechanism is still, however, unknown.

The syndrome of ARDS was described in 1967 (126). As it shows features similar to Infant Respiratory Distress Syndrome it was named Adult Respiratory Distress Syndrome (ARDS). Current recommendations are however that "Adult" should be replaced with "Acute". Synonym names for the syndromes that have been used are: acute respiratory failure, shock lung, pump lung and Da Nang lung. Acute Lung Injury (ALI) is used for milder forms of same condition (127). ALI/ARDS describes a characteristic form of lung disease but the severity covers a wide range, from mild dyspnoea to terminal lung failure. There has previously been some confusion on the definitions of ALI and ARDS but a consensus has now been reached that is widely accepted (114).

The diagnosis of ALI requires the following criteria:

1. Acute onset of symptoms.
2. Alveolar (Pao_2) to inhaled (FIo_2) O_2 concentration ratio $\leq$ 300 mmHg.
3. Bilateral diffuse infiltrations on chest X-ray.
4. Pulmonary artery wedge pressure $\leq$ 18mmHg (to exclude other causes).

The definition of ARDS is the same except that hypoxia is worse, Pao_2/FIo_2 ratio $\leq$ 200 mmHg. The incidence of ARDS reported differs greatly between studies and has been estimated to be between 1.5 and 88.6 per year per 100,000 population (128). Most studies show an overall mortality over 40% (113-117,129).

A division of the patients with ALI/ARDS syndrome in two groups based on the cause has been proposed (130). As the two groups caused by "direct lung injury" and "indirect lung injury" differ in pathological mechanisms, appearance on radiological examinations, respiratory

mechanical abnormalities and response to ventilatory strategies they might be considered separately. The most common "direct" causes are pneumonia and aspiration of gastric content and the most common "indirect" causes are sepsis, severe trauma and multiple blood transfusions.

Computed tomography (CT) of the lungs during ARDS show opacities representing collapsed areas distributed throughout the lungs with predominance for dependent regions. In supine they are dorsal but in prone these consolidations moves to the ventral, now dependent regions, within minutes. Often a shunt of >40% is seen, why increased FIO_2 cannot produce normal PO_2. Sometimes, regions of overdistended lung tissue contribute to increased dead space, and dead space in ARDS patients may exceed 70%. An other characteristic of ALI/ARDS is increase resistance to inspired air, measured as a reduction in compliance. The compliance of the lung itself is reduced in patients with intrinsic lung disease while reduced chest wall compliance is the main cause of reduced compliance seen in patients with extrapulmonary induced ALI/ARDS. The resistance to air flow in ARDS is about three times as high as in anesthetized patients with normal lungs. Functional residual capacity is reduced from the collapsed lung areas and increased elastic recoil and alveolar-capillary permeability is substantially increased (127).

Aims of the Thesis

General aim: To investigate factors that might influence V,Q and V/Q distributions and blood oxygenation.

Specific aims were to find out if:

- regional Q distribution in normal breathing healthy volunteers differ between supine and prone posture (**Study I**)
- CPAP influences regional V and Q distributions in prone and supine posture in healthy volunteers (**Study I, II**)
- V/Q matching is better in prone than supine position during general anesthesia and mechanical ventilation in healthy volunteers (**Study IV**)
- endothelial NO contributes to the observed differences in Q distribution between ventral and dorsal parts of the lungs (**Study III**)
- endogenous, paranasal NO release in humans effect Q distribution (**Study VI**)
- inhalation anesthesia, in spontaneous breathing humans, increase V/Q mismatch (**Study V**)

MATERIAL AND METHODS

SUBJECTS AND PATIENTS

Altogether, sixty-seven healthy volunteers and twenty-one patients were included in this thesis. The healthy volunteers were all non-smokers and without any history of pulmonary or cardiovascular disease. The mean age and weight at group level for each study, are presented in table 6. Biopsy samples from ventral and dorsal regions of the lungs in 21 patients (9m,12f) scheduled for lung surgery were analyzed in **Study III**. Average age at the time for biopsy was 69 years (range 41-92). All subjects and patients were informed about the procedure and risk associated with the investigation and informed consent was obtained. The local ethical committee approved all studies.

Table 6. Overview of the healthy volunteers participating in the studies included in this thesis.

Study	Number	Male-female	Mean age years (range)	Mean weight kg (range)
I	10	5 m 5f	39 (24-57)	68 (55-85)
II	16	7m 9f	29 (22-54)	67 (61-80)
III	9	5m 4f	31 (25-48)	70 (59-80)
IV	7	3m 4f	31 (26-39)	70 (57-91)
V	10	5m 5f	25 (20-34)	68 (58-81)
VI	15	8m 7k	29 (20-55)	66 (52-88)
Total:	67	33m 34f	30 (20-57)	68 (52-91)

RADIOPHARMACEUTICALS

Macroaggregates of human albumin (LyoMAA) (TechneScan LyoMAA, Mallickrodt Medica, Petten, The Netherlands) were used to depict regional Q in all studies. In **Studies I**, **II**, **III** and **VI** LyoMAA was labeled with technetium-99m (^{99m}Tc) and in **Studies IV**,**V** and **VI** with indium-113m (^{113m}In). In **Study VI** both radionuclides were used to depict the regional Q. As tracer for V an aerosol of water and ^{99m}Tc-labeled diethylenetriamine pentaacetic acid (DTPA) was used in **Study II** and ^{99m}Tc-Technegas aerosol (Tetley Manufacturing Ltd., Sydney, Australia) was used in **Studies V**, **IV** and **V** (Table 8). The maximum allowed administered activities by the local radiation protection committee for each study are presented in table 7. The subjects in all studies were estimated to receive an effective radiation dose less than 5 mSv.

Table 7. Radionuclides, activity levels and SPECT techniques used in this thesis.
^{99m}Tc = meta stable technetium, ^{113m}In = meta stable indium, MBq = megabecquerel.

Study	Q (1st injection)	Q (2nd injection)	V	SPECT Technique
I	50MBq ^{99m}Tc	100MBq ^{99m} Tc	-	Subtraction
II	70MBq ^{99m} Tc	-	30 MBq ^{99m} Tc	Subtraction
III	50MBq ^{99m}Tc	100MBq ^{99m}Tc	-	Subtraction
IV	50MBq ^{113m}In	-	50MBq ^{99m} Tc	Dual energy
V	50MBq ^{113m}In	-	50MBq ^{99m} Tc	Dual energy
VI	50MBq ^{113m}In	50MBq ^{99m}Tc	-	Dual energy

IMAGING TECHNIQUE

SPECT was used in all imaging studies and were made with a three-headed Triad XLT gamma camera (Trionix, Twinsburg, OH, USA). A matrix size of 128x128 giving a voxel size of 3.6 mm^3 was used through out the studies. Filtered back projection was used for image reconstruction. In **Studies I** and **III** the same radionuclide (^{99m}Tc) was used to trace two different conditions and in **Study II** to trace V and Q. Hence, two consecutive SPECT scan were acquired using low energy high-resolution collimators. The first SPECT scan corresponds to the Q distribution during the first condition (**Studies I**, **III**), or V distribution (**Study II**) and the second representing the combined effect of the Q distributions during the first and second condition, (**Studies I**, **III**) or V and Q distributions (**Study II**). The first SPECT scan was then pixel-wise subtracted from the second SPECT in order to obtain a new set of images without the influence from the activity in the first SPECT. In **Studies IV**, **V** and **VI** the photon emission data was simultaneously registered for ^{99m} Tc and ^{113m} In using medium energy general-purpose collimators and a four-energy windows technique. Two windows, centered at 140 keV and 392 keV, were used to register the primary emission photons from ^{99m}Tc and ^{113m}In, respectively. Two secondary windows, placed just below each of the primary window, were used to registered scattered photons. After registration of emission images, a 15 min transmission scan was made with a ^{99m}Tc filled line source. These images were used for attenuation correction of the emission images and as anatomical reference (51). Table 7 summarizes the different radionuclides and SPECT techniques used in this thesis.

ANESTHESIA

In **study IV** an intravenous catheter was first inserted into a peripheral vein. Monitoring equipment was attached and anesthesia was induced by i.v. injection of 200 mg propofol, followed by an infusion of propofol, at a rate of 8 mg $\cdot kg^{-1} \cdot h^{-1}$. Endotracheal intubation was carried out in supine position after establishing muscle relaxation by i.v. injection of Rocuronium Bromide 0.6 mg $\cdot$ kg body $weight^{-1}$. Alfentanil was used for analgesia. The subjects were then connected to a Servo 900C ventilator (Siemens-Elema, Stockholm, Sweden) set in a volume controlled mode. A PEEP of 3-4 cm H_2O was applied. Thereafter muscle relaxation was reversed and anesthesia terminated.

In **study V** anesthesia was induced by inhalation of sevoflurane at a concentration of 2.3% until a minimum alveolar concentration of sevoflurane of approximately 1 was reached and spontaneous breathing was preserved though out the experiment. The subject was returned to consciousness before the SPECT. In both studies (**IV,V**), standard monitoring equipment was used during the anesthesia in both studies.

EXPERIMENT SETUPS

All imaging studies were performed with SPECT. If repeated experiments with radiotracers were performed, at least 2 days elapsed between the experiments to avoid interference from remaining activity and the order was randomized. When examined in prone, supports were put under the upper thorax and the pelvis to allow for free movement of the chest. The subjects fasted for 6 h before the experiment in **Studies IV** and **V**. An overview of breathing patterns, postures and interventions are presented in table 8.

Table 8. Overview of the experiments in this thesis.

Study	Anesthesia	Breathing pattern	Position at administration	Position at registration	Factors studied
I	None	All with and without CPAP	Prone/supine	Supine	Posture
II	None	Half with and half without CPAP	Prone/supine	Prone/Supine	Posture and CPAP
III	None	Spontaneous	Prone/supine	Supine	L-NMMA
IV	Propofol iv + infusion	Mechanical ventilation with PEEP	Prone/supine	Supine	Posture
V	Inhalation Sevoflurane	Spontaneous	Supine	Supine	Inhalation anesthesia
VI	None	Nasal or oral breathing	Upright	Supine	Breathing pattern or exogenous NO

Study I, comparing Q distribution between prone and supine

Two sessions were used, one with CPAP breathing (n=9) and one with normal breathing (n=8). The subject was first positioned in prone on the gamma camera couch, with or without a mask with 10 cmH_20 CPAP. In that position ^{99m}Tc-labeled LyoMAA was injected through a vein catheter in the right arm. After the injection was completed the subject turned supine and a SPECT acquisition was made. In unaltered supine position a second injection of ^{99m}Tc-labeled LyoMAA was injected through the same catheter and a second SPECT acquisition was made (Fig. 1).

Figure 1.

Study II, comparing V,Q and V/Q distribution between CPAP and normal breathing in prone and supine posture

Half of the subjects (n=8) were examined during normal breathing in prone and supine at two different occasions in a randomized order. An inhalation of a ^{99m}Tc aerosol was given through a tight fitting mask and a first SPECT acquisition was made. Thereafter, ^{99m}Tc-labeled LyoMAA was injected intravenously and a second SPECT acquisition was made. The subject remained in a fixed position though out the experiment. The other half of the subjects (n=8) were also studied in prone and supine but at both occasions with 10 cmH_2O CPAP through a tight fitting mask during the experiment (Fig. 2).

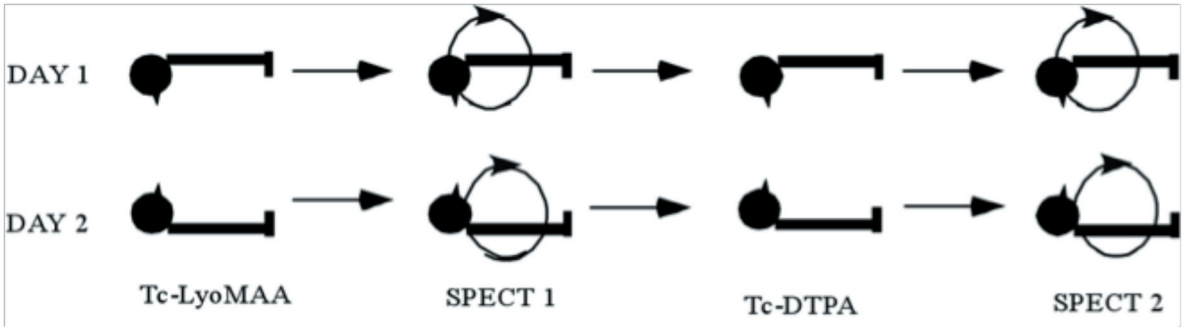


Figure 2

Study III, comparing the activity of NO between ventral and dorsal parts of the lungs and studying the effect from endothelial NO on Q distribution

Patients: Biopsy samples were taken *in situ* in the operating theatre from the ventral and the dorsal regions of the lower lob (n=21). Sample was taken from areas judged as normal and were frozen within 1 minute in liquid nitrogen. In 13 of the 21 patients total RNA was isolated using the RNeasy Mini Kit (Qiagen, Hilden, Germany) and quantified by spectrophotometry. 2 mg of total RNA was used for cDNA synthesis and approximately 50 ng of cDNA real time polymerase chain reaction using primers probes. The cDNA was analyzed in duplicate and polymerase chain reaction amplification. The eNOS mRNA expression was quantified in relation to the expression of mRNA of β-actin. Further, the activity of NOS was assed in all the 21 samples by the conversion of L-(U-C^{14}) arginine to L-(U-C^{14}) citrulline. To differentiate between Ca^{2+} dependent and independent activity the analysis was performed both with and without the presence of a calcium chelator.

Volunteers: The subjects (n=9) were examined at two different occasions, prone or supine. When studied prone, the subject was positioned on the gamma camera couch and remained in a fixed position throughout the experiment. First ^{99m}Tc-labeled LyoMAA was injected intravenously to label Q, and a first SPECT acquisition was made. Thereafter endogenous NO production of was blocked by an infusion of N^0-monomethyl-L-arginine (L-NMMA, ClinAlfa, Laufelfingen, Switzerland). 2.4 mg kg^{-1} body weight min^{-1} was infused during 5 minutes followed by 0.1 mg kg^{-1} body weight min^{-1} during 60 minutes. Thereafter a second SPECT acquisition was performed. After a second injection of ^{99m}Tc-labeled LyoMAA a third, final, SPECT acquisition was done (Fig.3).

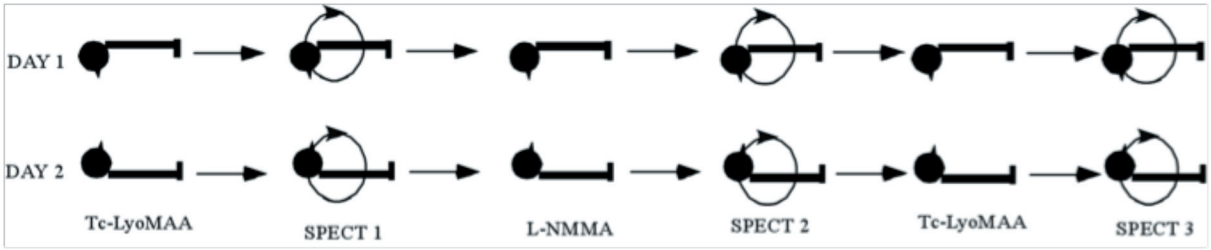


Figure 3

Study IV, comparing V,Q and V/Q distributions between prone and supine during general anesthesia and mechanical ventilation

The volunteers (n=7) were examined during general anesthesia and mechanical ventilation at two different occasions, one aimed at V,Q and V/Q distributions in prone and the other at distributions in supine. The radiopharmaceuticals were administered in prone or supine but SPECT was always performed in supine. After anesthesia the subject was placed either in prone or supine on the gamma camera couch and a recruitment maneuver with 30 cm H_2O, sustained for 30 s was performed. After 10 minutes, ^{99m}Tc-labeled Technegas was added to the inhaled gas. Simultaneously, ^{113m}In-labelled LyoMAA was injected intravenously. If the radiopharmaceuticals were administered in prone the subject was turned supine, otherwise the volunteer remained in the same position. After a second recruitment maneuver, similar to the previous, a SPECT acquisition was made (Fig 4).

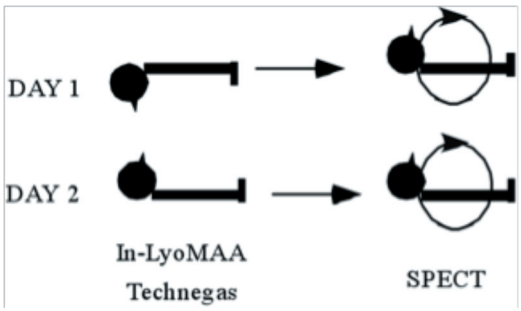


Figure 4

Study V, comparing V,Q and V/Q distributins between awake and during inhalationanesthesia in supine

Volunteers (n=10) were studied at two occasions, once awake and once during inhalation anesthesia. After the anesthesia was established ^{113m}In-labeled LyoMAA was injected and simultaneously ^{99m}Tc-labeled Technegas was added to the inhaled gas. After administration of radiopharmaceuticals the subject was returned to consciousness and a SPECT acquisition was performed (Fig 5).

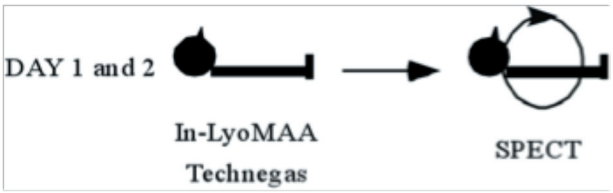


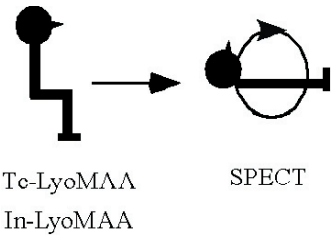
Figure 5.

Study VI, studying the effect of NO from the nasal airways on Q distribution in upright position

The radiopharmaceuticals were administered in sitting position, but SPECT acquisition was made in supine. The subjects were divided into three groups. In the experimental group (n=7) the subjects began, in the sitting upright position, with a controlled 20-min oral breathing of humidified NO-free air (AGA Gas AB, Stockholm, Sweden), which was followed by intravenous administration of LyoMAA labeled with one of the tracers (^{99m}Tc or ^{113m}In). Ten minutes later the subjects switched to controlled nasal breathing of humidified NO-free air, inhaling through

the nose and exhaling through the mouth during 10 min. At the end of this period the second tracer was administered and SPECT was performed. The order of the tracers was randomized.

In one of the control groups (n=3) ^{99m}Tc and ^{113m}In labeled LyoMAA were injected simultaneously during normal, regular, breathing followed by a SPECT. In the other control group (n=4) the subjects began, in the sitting upright position, with a controlled 20-min oral breathing of humidified NO-free air, which was followed by intravenous administration of LyoMAA labeled with one of the tracers. Ten minutes later the subjects switched to oral breathing of a mixture of humidified NO free air mixed with exogenous NO (AGA Gas AB, Stockholm, Sweden) to a final concentration of approximately 150 ppb. The subjects inhaled the exogenous NO from a 25



L Douglas bag, through the mouth via a mouthpiece and exhaled through the nose. Thereafter LyoMAA labeled with the other tracer was injected and the SPECT was performed (Fig 6).

Figure 6

DATA ANALYSIS

Image quantification

In **Study I, II** and **III** the activity distribution in volumes of interest (VOI's), from the anterior to posterior border, in the right lung were evaluated. The activity in these was normalized so that the total activity in the VOI was considered to be 100%. The VOI were subsequently divided in to two (**Study III**) or three (**Study I**, **II**) equal volumes, in the anterior to posterior direction. The activity in each part was considered to represent the relative Q or V and presented as percent of the activity in the entire VOI. The anterior and posterior borders of the VOI's, equal to the outer borders of the lungs, were defined as the intersection between the mean values in a 128 pixel long profile with the actual activity profile in the same direction. In **Study I,** VOI's at 1/5, 2/5 and 3/5, in **Study II** a VOI at 2/3 and in **Study III** at 1/3 and 2/3 from the apex to the base were selected. The VOI's, from the ventral to the dorsal border of the lung, were 40 mm from side to side and 10 mm in the apical to basal direction in **Study I** and **II**. In **Study III** the size was 18 x 18 mm and the length from the ventral to the dorsal border of the lung. Furthermore, in **Study II**, the activity profiles in the anterior to posterior direction, from the entire lungs, were fitted to a linear function representing the increase or decrease of V and Q in the direction of gravity. In **Study IV**,**V** and **VI** the SPECT data was pixel-wise normalized to the total activity in the lungs. Hereby, each pixel activity represents the relative V and Q in that position (in percent of total lung V and Q). The lungs in each individual were divided into 21 (**Study IV**) or 20 (**Study V**) equal volumes in the anterior to posterior direction and also 3 volumes of equal size (**Study IV** and **V**). The V/Q ratio in every pixel was calculated by pixel-wise dividing the relative V and Q. The heterogeneity of in the entire lungs for V,Q and V/Q was calculated as the sum of squares of the pixel-wise deviations from the mean (SS_{total}). The heterogeneity in each isogravitational plane was calculated as the sum of squares of the pixel-wise deviation from the mean pixel value in

that plane and all these deviations were added ($SS_{residual}$). Finally, the heterogeneity in the anterior to posterior, vertical, direction was calculated as SS_{total} - $SS_{residual}$ and the vertical contribution to heterogeneity in percent as 100 x (SS_{total} -$SS_{residual}$)/ SS_{total}.

Statistics

A double tailed paired Student's t-test was used to compare Q distribution between VOI's in prone and supine positions (**Study I,IV**), V and Q (**Study II**), with and without the influence from NO (**Study III and VI**) and differences between anesthesia and awake (**Study V**) (Excel, Microsoft Corporation, Redmond, USA, SAS institute, Cary, NC, USA in **Study VI**). Bonferroni corrections were used in **Study IV** and **V**. A single sample Students t-test and a two factor analysis of variance with Fisher's least significance test was used to compare the linear gradients of V and Q with a zero gradient in **Study II**. In **Study III** a Wilcoxon sum rank test was used to compare the NOS activity in ventral and dorsal parts of the lungs and repeated measurement of variance to assess the effect of L-NMMA on exhaled NO.

RESULTS

EFFECTS ON V,Q AND V/Q MATCHING FROM POSTURE, DURING NORMAL BREATHING (STUDIES I,II,V,VI)

An overview of the results are presented in table 9 (**Study I,II**), table 10 (**Study V**), table 11 (**Study VI**) and table 12 (**Study IV**).

Table 9. Results from study I and II. From study I the relative Q in the dependent (dorsal in supine and ventral in prone) and non-dependent thirds of a VOI in the basal part of the right lung are presented as % of total Q in the VOI. In study II, the gradients of V,Q and V/Q are presented as percentage of change per centimeter in the anterior-posterior direction**.**

	Supine			Prone			P-value Supine-Prone	
Study I	**Without CPAP**	**With CPAP**	**P - value**	**Without CPAP**	**With CPAP**	**P - value**	**Without CPAP**	**With CPAP**
Dependent	38 ±3%	49 ±7%	<0.001	32 ±4%*	39 ±4%	< 0.01	< 0.01	< 0.01
Non dependent	25 ±3%	16 ±4%	<0.001	31 ±5%*	24 ±5%	< 0.01	< 0.05	< 0.01
Study II								
V	2.7 ±1.5	3.4±0.8	ns	2.6±0.9	2.9±1.0	ns	ns	ns
Q	3.7±1.5	5.5±1.0	<0.05	1.6±1.9	-2.2±0.8	<0.05	ns	<0.05
V/Q	-1.5±3.5	-3.4±2.4	ns	1.5±3.5	8.3±1.1	<0.05	ns	<0.05

ns = non significant. *In the published article (**Study II**) findings were presented in a graph.

Table 10. Results from **study V**. The relative distributions of V and Q in ventral and dorsal thirds, of equal size, of the entire lungs were compared between awake and during sevoflurane inhalation anesthesia with spontaneous breathing. The mean of pixel-wise calculated V/Q values in the ventral and dorsal thirds are also presented as well as the total heterogeneity for V,Q and V/Q distributions. Full width half maximum (FWHM) is the width of the distribution of the frequency of V/Q values in the lungs, at half of the maximum value of frequency. This is an alternative estimation of V/Q heterogeneity.

	1/3 of lungs	AWAKE	ANESTHESIA	P value
V	**Ventral**	24.7 ±2.5%	23.5 ±4.6%	ns
	Dorsal	41.5 ±3.4%	44.5 ±4.9%	ns
	Total heterogeneity	0.57 ±0.15	0.48 ±0.29	ns
Q	**Ventral**	26.4 ±2.3%	25.3 ±2.0%	ns
	Dorsal	39.3 ±3.2%	40.9 ±3.9%	ns
	Total heterogeneity	0.37 ±0.09	0.24 ±0.14	0.002
V/Q matching	**Total heterogeneity**	0.08 ±0.02	0.13 ±0.02	0.002
	Vertical heterogeneity	0.008 ±0.003	0.03±0.03	0.028
	FWHM of V/Q ratios	0.57 ±0.12	0.77 ±0.12	0.009

ns = non significant (p >0.05)

Table 11. Results from **study VI**. The relative distribution of Q between thirds, of equal size is studied. Comparison between Q distribution while breathing NO free air and while breathing air with NO added endogenous by breathing through the nose or exogenously by adding ~150 parts of NO to the inhaled NO free air was made.

Third of lungs	Oral breathing (n=7)	Breathing NO (n=11)	P -value
Cranial	8.3 ±3.3	12.0 ±3.6	<0.001
Mid	47.2 ±4.9	47.2 ±5.4	ns
Basal	40.7 ±7.2	44.5 ±7.5	<0.01
Ventral	13.5 ±3.3	12.7 ±3.2	ns
Mid	38.5 ±3.1	35.8 ±3.4	<0.001
Dorsal	47.9 ±4.9	51.4 ±5.4	<0.001

ns = non significant (p >0.05).

Table 12. Results in **study IV**. The relative distributions of V and Q in ventral and dorsal thirds, of equal size, of the entire lungs were compared between supine and prone postures. The mean of pixel-wise calculated V/Q values in the ventral and dorsal thirds are also presented as well as the contribution to the total heterogeneity explained by the vertical direction for V, Q and V/Q, expressed in %.

	Third of lungs	Supine	Prone	P-value
V	**Ventral**	25 ±3%	24 ±2%	0.28
	Dorsal	41 ±2%	42 ±3%	0.62
	$SS_{vertical}$	18.0 ±6.7%	19.6 ±9.8%	0.589
Q	**Ventral**	17 ±4%	31 ±4%	0.0007
	Dorsal	49 ±4%	33 ±5%	0.0008
	$SS_{vertical}$	45.8 ±7.9%	20.0 ±10.3%	0.0006
V/Q	**Ventral**	1.8 ±0.5%	0.8 ±0.2%	0.003
	Dorsal	0.8 ±0.1%	1.4 ±0.3%	0.002
	$SS_{vertical}$	31.4 ±14.1%	16.4 ±14.2%	0.0639

$SS_{vertical}$ = Vertical component of heterogeneity

Q distribution in prone, supine and upright positions

In **Study I** the dependent of three equal sized thirds, in the anterior to posterior direction, of a VOI in the basal part of the right lung received a higher percentage of total Q in supine than in prone (p <0.01) (table 9). The differences of Q distribution, between prone and supine posture, did not reach significant levels in VOI's in the apical and mid parts of the right lung. Q distribution between three equal sized parts, in the anterior to posterior direction, of the entire lungs were examined during normal breathing, awake and in supine posture in **Study V**. The dependent (dorsal) third of the lungs received a higher percentage of Q than the non-dependent (ventral) third (table 10). In **Study VI**, Q distribution was mainly basal in upright position (table 11).

V distribution, during normal breating, in supine

Study V showed a dorsal dominance of V during normal breathing in supine (table 10).

V/Q distribution during normal breathing, in prone and supine

V/Q ratios, assessed as a linear function of spatial vector in the ventral to dorsal direction, (**Study II**) was uniform (i.e. not significantly different from a zero gradient) in both postures (table 9).

V,Q AND V/Q MATCHING, EFFECTS FROM CPAP (STUDY I,II)

During CPAP, Q distribution between equally sized thirds of a VOI in a basal part of the right lung, was more dependent in supine than in prone posture (**Study I)** (table 9).

CPAP breathing, compared to breathing without CPAP, resulted in a shift of blood flow from non-dependent to dependent parts of the lungs in both prone and supine (**study I**). In **study II** CPAP increased the ventral to dorsal gradient of Q in supine ($p<0.05$) and decreased the gradient in prone ($p<0.05$). The gradient of V was not altered significantly from CPAP in either of the positions. CPAP did not change the ventral to dorsal gradient of local V/Q matching significantly in supine but increased the gradient in prone ($p <0.05$, table 9). This indicates a more uneven V/Q matching in the ventral to dorsal direction in prone than in supine during CPAP breathing. Mean V/Q values in three equal sized thirds of a VOI in the basal part of the right lung were significantly different from values during normal breathing in the ventral an dorsal third in prone but only in the ventral third in supine. This indicates that CPAP induces a V/Q mismatch in both postures, this being most pronounced in prone (Fig. 7).

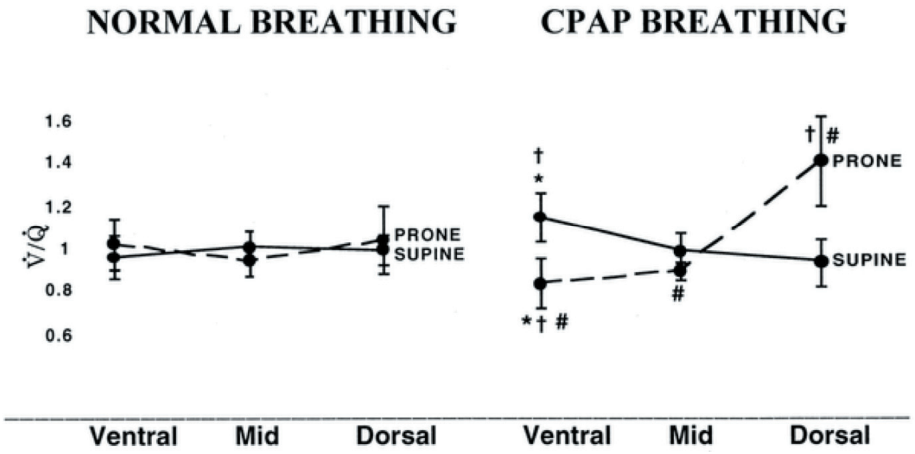


Figure 7. *Results from study II presenting the quotient of activity, representing V and Q, in three VOI's, in a basal section of the right lung. The quotient is considered to reflect mean V/Q ratios and the effect from posture and CPAP on the distribution of V/Q. The left graph presents the V/Q distribution during normal breathing and the right V/Q distribution during CPAP. *p < 0.05 for the difference between ventral and dorsal third, prone or supine; #p < 0.05 for difference between supine and prone posture; †p < 0.05 for differences between normal and CPAP breathing.*

EFFECTS ON V,Q,V/Q DISTRIBUTION FROM POSTURE DURING INTRAVENOUS ANESTHESIA WITH MUSCLE RELAXATION AND MECHANICAL VENTILATION (STUDY IV)

In **study IV,** Q was highest in the dorsal and lowest in the ventral third of the lungs in supine. In prone, Q was more evenly distributed between the ventral and dorsal thirds (table 12). The vertical component of total Q heterogeneity was larger in supine than in prone (P = 0.0006). V was highest in the dorsal third of the lungs in both prone and supine and did not differ between postures. The vertical component of total V heterogeneity was also unaltered between postures. This resulted in high mean V/Q values in the ventral third of the lungs while in supine position and in the dorsal third of the lungs when in prone posture. The vertical component of V/Q heterogeneity showed a tendency to be higher in supine than in prone (p =0.0639).

THE EFFECT FROM ENDOTHELIAL NO ON REGIONAL Q DISTRIBUTION (STUDY III)

Blocking endogenous NO production (**study III**) resulted in a decreased Q in the dorsal half of a VOI in the basal part of the right lung ($p < 0.05$). Q distribution did not change in prone (Fig. 8). The influence from endogenous NO on regional Q distribution is further supported from a higher eNOS mRNA expression and higher Ca^{2+} dependent eNOS activity in biopsies from dorsal than from ventral parts of the lungs ($p< 0.05$) (figure 9).

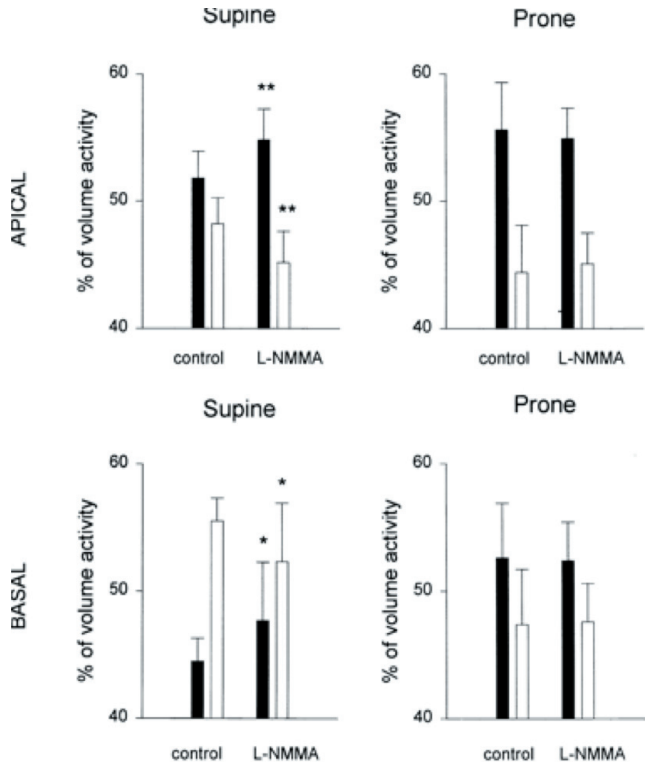


Figure 8. *Regional Q distribution in supine and prone positions between VOI's representing the ventral (filled bars) and dorsal (open bars) half of the lungs, 1/3 (apical) and 2/3 (basal) from apex. Before (control) and after (L-NMMA) infusion of L-NMMA is shown. *p < 0.05, **p <0.01.*

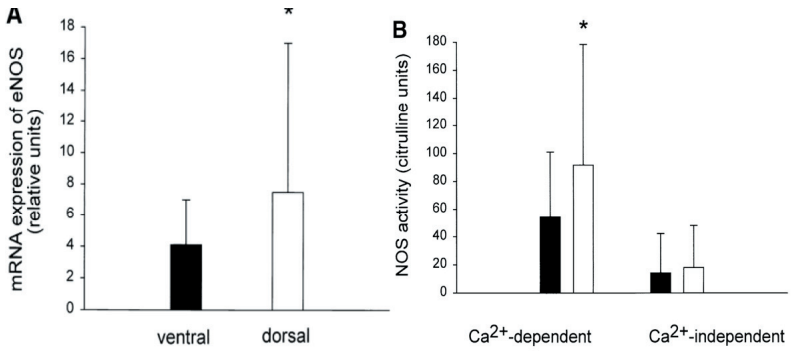


Figure 9. A. *Expression of mRNA for endothelial NOS in biopsies ventral (filled bar) and dorsal (open bar) lung tissue samples from basal lower lob in patients (n=13), expressed as relative units.*
B. *NOS activity in ventral (filled bar) and dorsal (open bar) lung tissue samples from the basal lower lob in patients (n=21). Left side describe activity with and the right side without added Ca^{2+}. Wilcoxon rank sum test was used for analysis. *p < 0.05.*

DISTRIBUTION OF Q AS A FUNCTION OF INHALED NASAL NO (STUDY VI)

Endogenous NO from paranasal sinuses by nose breathing or adding small concentrations of NO to inhaled air (**study VI**) showed a redistribution of Q from basal to apical parts of the lungs, compared to breathing NO free air though the mouth ($p < 0.001$). The shift of Q from basal to apical third of the lungs was in mean 3.8%. The apical third of the lungs, in upright position under regular spontaneous breathing, receives 16% of Q (table 11). Thus, a shift of 3.8% accounts for a net increase of ~24% in the apical third.

V,Q,V/Q DISTRIBUTION AS A FUNCTION OF INHALATION ANESTHESIA (STUDY V)

Inhalation anesthesia with maintained spontaneous breathing did not change the regional distribution of V, Q or V/Q between the three equal parts of the lungs, in the ventral to dorsal direction (**Study V,** table 10). The total heterogeneity of Q was lower during anesthesia than awake ($P = 0.002$) while the heterogeneity of V did not change, neither the total nor the vertical component. The resulting heterogeneity of local V/Q was higher during anesthesia than awake. This indicates a less beneficial V/Q matching during inhalation anesthesia.

DISCUSSION

In this thesis, factors related to posture and intensive care influencing the regional distribution of V and Q, and how these may influence local V/Q matching, has been studied. The background is the observation that some patients with ALI/ARDS improve their gas exchange when turned prone (6-12). The thesis is mainly based on measurements obtained from SPECT images in healthy volunteers. The basic concept has been to study every factor in isolation and in a fixed position to avoid interference from differences in anatomical conditions. To study changes induced by each factor, either a subtraction or a dual energy technique has been used.

GENARAL LIMITATIONS OF THE SPECT TECHNIQUE

The findings are based on the assumption that the radiotracers distributions truly reflect V and Q distributions and that the acquired images, in their turn, truly represent the tracer distribution. The deposition of LyoMAA, diethyleneetriamamine acid and Technegas has been shown to be proportional to the distribution of blood flow and ventilation, respectively (30,70,71,131). Phantom and physiologic studies using the same imaging technique and correction algorithms as in this work have shown that the corrected images are good estimates of the real distribution of the radiotracers in the lungs (35,36).

The subtraction technique used in **Study I**,**II** and **III**, requires fixed positions throughout the entire experiment. As some experiments lasted for over one hour this was difficult to achieve. We made efforts to position the volunteer as comfortably as possible by padding with cushions and by pelvic and upper thorax supports when in the prone. No obvious movements were observed between the acquired images during the experiments. Additionally, in these studies, no scatter correction was performed which could influence the quantification of the regional distributions of V and Q. However, when comparing the effect on the distribution of V or Q between two conditions, we only compared differences between distributions registered in the same position. Consequently, the impact of scatter should be the same.

To obtain images with good quality the experiments had to be performed with the arms over the head. The anatomy and physiology of the lungs may then differ slightly from conditions with the arms along the sides of the body. The aim was, however, to examine differences between two conditions and the position was identical between the acquisitions.

Partial volume effects in SPECT could, when two different isotopes were used simultaneously, lead to an incorrect quantification if no proper correction algorithms for scatter, attenuation and organ outline are carried out. In **Studies IV**, **V** and **VI** we use a correction scheme to minimize the effect of partial volume in image quantification (35). In **Study VI** we simultaneously administer intravenously LyoMAA labeled with ^{99m}Tc and with ^{113m}In to check the accuracy of the corrections. This resulted in no differences in the regional distribution of Q as mapped with both isotopes after proper corrections for scatter, attenuation and organ outline

were performed. Further, the spatial resolution of SPECT precludes measurements of V,Q and V/Q ratios in volumes smaller than approximately 3 cm3 even though gas exchange takes place in acini with a volume of approximately 0.18 cm3 (132) and measurements from single pixels are influenced by the activity in adjacent pixels. This results in an underestimation of V,Q and V/Q heterogeneity (36,100). The SPECT method, however, has other advantages that made it the best available technique for in vivo measurements of V and Q in the studies included made.

The SPECT methods used measure activity reflecting regional V and Q, per unit lung volume. As posture effects the distribution of lung tissue in the thorax (133) this is not the same as V and Q distribution per alveolus. Therefore, the posture at registration will affect the results and the same V and Q distribution and the same distribution per alveolus would yield different results if examined prone or supine. This makes comparisons with previous publications difficult. On the other hand, comparing SPECT registration of V and Q distributions in supine, registered in supine, with V and Q distributions in prone registered in prone will measure the sum of two effects. Consequently, studying the effect from posture on V and Q distributions but both registered in supine could be a strategy of showing the true magnitude of changes.

SPECIFIC LIMITATIONS IN EACH STUDY

In **Study I, II and III** we lack anatomical information from transmission or CT scans. This information is needed for both attenuation correction and organ outline of the SPECT images. This has a direct effect on our estimates of the regional distributions of V and Q. Hence, the shape of the profiles in the ventral to dorsal direction used to quantify the effect of posture on V and Q regional distribution have some uncertainty. The magnitude of this uncertainty is difficult to quantify. In **Study I**, we compared the differences in Q distribution only from the SPECT scans performed in supine position, but with the tracers administered in supine and in prone. Hence, the obtained difference is less affected by these limitations, but makes it difficult to draw conclusions on Q distribution per alveolus (133). However, the study demonstrates a true change in Q distribution from posture and a more uniform distribution of Q in prone, in the ventral to dorsal direction, in relation to lung tissue as distributed in supine.

We cannot draw any firm conclusions about V and Q distributions in the ventral to dorsal direction from the profiles presented in **Study II** as they were acquired in different anatomical positions. Additionally, these profiles were not normalized to the area of each coronal plane why they represent the total activity in each plane for V and Q. Therefore, with this quantification even with a homogenous distribution of the total Q and V in the entire lungs, we would have obtained a gradient for Q and V, respectively, in the ventral to dorsal direction, because of the anatomy. In **Study II**, the V/Q ratios were derived by simply dividing the sum of the total activity representing V and Q obtained at each position in the ventral to dorsal direction. This is not the same as performing the V/Q ratios pixel-wise and calculating the average of all obtained V/Q ratios in a volume as in **Study IV** and **V**. However, as previously presented (35,36) the frequency distribution of V/Q ratios in the lung of a healthy volunteer follows a normal distribution around V/Q=1 with a standard deviation of about ±0.2. Hence, about 60% of the entire lung volume is represented by V/Q ratios in the narrow interval between [0.8, 1.2]. Therefore the simplification used in the quantification of the V/Q made in **Study II** is a good approximation of the real distribution of average V/Q ratios along the ventral to dorsal direction. Furthermore, the major focus of this paper was to measure the magnitude of the

effect of CPAP in prone and in supine separately. In this case the described limitations are of less importance as the comparisons with and without CPAP were performed under the same anatomical conditions.

In **Study III** we separately compared the effect of LNMMA in the Q distribution in supine and prone posture. The difference induced by L-NMMA at each position is thus less affected by anatomical differences between postures.

In **Study IV, V** and **VI**, we perform scatter, attenuation and organ outline correction (35) which avoids many of the limitations described for **Study I**,**II** and **III**.
The number of observations in **Study IV**,**V** and **VI** is low (n=7 for **Study IV**, n=10 for **Study V**, n=14 for **Study VI**). Throughout these studies the Student t-test was used for statistical analysis of the data. This assumes that the data is normally distributed which is questionable for some small groups. However biological processes are generally normally distributed. Furthermore, in complex physiological studies that involve some hazardous components like anesthesia or mechanical ventilation, large series cannot be expected.

The distribution of V and Q per lung volume, and not per alveolus, is measured in **Study IV**, with the same limitations as discussed above.

COMPARISON WITH PREVIOUS KNOWLEDGE

Q distribution during normal breathing, in prone, supine and upright positions

Most prior studies agree on a dependent dominance of Q in upright position (25,26,32.77,134) which is in line with our findings in **Study VI**. Most also agree on a dependent dominance of Q in supine (74,77,78,81,134-137) in line with findings in **Study I**,**II** and **V**. Q distribution in prone position is more controversial. Most previous human studies report a ventral dominance of Q distribution in prone but they compare Q distribution in prone examined in prone with Q distribution in supine examined in supine, thus measuring the added effect of Q and tissue distribution. These include early human studies (74) using external detectors and more recent using SPECT (77,134,135). Two of these studies, however, show a tendency towards a more even distribution in prone (77,134). Animal studies with SPECT (136) and with microsphere technique (104,137,138) has shown a ventral Q distribution in prone. As these studies are performed by studying fixated, excised lungs, they do not suffer from the problem with the position at registration but as the lungs are dried at total lung capacity they do not exactly reflect the anatomic conditions in vivo and there may also be differences from human physiology. Human studies with PET (139), CT (140) and MRI (141) have also shown a dominant ventral Q distribution in prone. Even though efforts have been taken to compensate for the distribution of lung parenchyma, these studies still suffer from the examinations being performed in different postures. There are also reports that SPECT studies without attenuation correction might underestimate the gradient of regional Q in the direction of gravity (142). These are in conflict with the more uniform Q distribution in prone found in **Study I**. However, microsphere, animal studies in sheep (143) and horses (144) demonstrate a uniform Q distribution in prone, in agreement with the findings in **Study I**. **Study II** shows a mainly dorsal distribution of regional Q in prone but these findings are influenced by the form of the lungs, with larger lung volumes in the dorsal parts of the lungs and thus not comparable with studies measuring Q per lung volume or per alveoli.

In conclusion, most previous studies have shown a ventral dominance of Q in prone in conflict with our findings in **Study I** even if some studies are in agreement. However, definitions of to what Q should be related differs. The basic question if Q distribution, in the anterior to

posterior direction, is somewhat more uniform in prone than in supine posture has not been finally solved.

V distribution during normal breathing, in supine

There seems to be complete consensus on regional V distribution being mainly dorsal in supine (31,74,77,78,145). This is in agreement with the findings in **Study II** even if our results are influenced by anatomy and not comparable to previous studies.

V/Q distribution during normal breathing, in prone and supine

As both V and Q show a dorsal dominance in supine there are prerequisites for a good regional V/Q matching in both ventral and dorsal parts of the lungs as was found in **Study II** and **V**. This is supported by previous studies (146) and by the narrow V/Q distribution in supine seen with MIDGET (147). There also seems to be a good regional V/Q matching in prone (78,123,138) in agreement with the findings in **Study II**.

Q distribution during CPAP, in prone and supine

CPAP is extensively used in intensive care settings and during the postoperative period as a means to improve blood oxygenation and to prevent atelectasis formation (148,149). Few, if any, studies exist on regional Q distribution in supine during spontaneous breathing with CPAP. Increased lung volumes in humans (122,150), a situation that might be comparable with CPAP breathing, show a dependent dominance of regional Q in supine in agreement with the results in **Study I**.

To the best of my knowledge no prior specific studies on the effect of CPAP on regional V,Q and V/Q distributions have been carried out. One study on the effect from different levels of PEEP, comparable to CPAP, during general anesthesia in sheep (151) showed an increasing dorsal dominance of regional Q distribution from increased PEEP. In prone, however, Q distribution was uniformly distributed and not influenced by PEEP up to 20 cm H_2O. This is in contradiction with the findings in **Study II**, where a redistribution of regional Q towards dependent parts of the lungs from CPAP was seen in prone. However, there may be differences between sheep and humans as sheep probably are completely adapted to standing on four legs.

Effects on V,Q and V/Q distributions from posture during intravenous anesthesia with muscle relaxtion and mechanical ventilation

During general anesthesia with mechanical ventilation, studies show a preferentially dorsal Q distribution in supine (39-41,152-155) in agreement with the findings in **Study IV**. In prone most studies show, in agreement with **Study IV**, a uniform regional Q distribution in the anterior to posterior direction in animals (152-156). Two studies in dogs, however, show a dorsal (40,46) and one study in man (32) show a ventral regional Q distribution. As the studies showing a basically uniform distribution of Q are more recent there is evidence that Q distribution is at least more uniform in the anterior to posterior direction in prone than in supine posture during mechanical ventilation. Regional V distribution is more controversial. Two studies, in man (31) and dogs (157), mainly show a dorsal regional V distribution in supine in agreement with our findings in **Study IV**. However, one study in man (39), two in dogs (48,146) and one in pigs (153) show mainly ventral regional V distribution. In prone posture most previous studies show a uniform regional V distribution in dogs (48,146) and pigs (155) while two show a ventral V distribution in man (31) and pigs (153) and only one, in dogs (157), a dorsal V distribution in agreement with our findings in **Study IV**. The question

of posture at registration is however crucial. If region V distribution in prone would have been registered in prone in **Study IV**, this could have shown a uniform distribution. Some of the studies (153,156) were done with microsphere technique which reflects V distribution per alveolus and not in relation to lung volume as in **Study IV**. Ventilation per alveolus might be a better way of describing V distribution but **Study IV** demonstrates that V distribution does not change between prone and supine.

Effects from nitric oxide on Q distribution, related to posture

There is now some evidence that regional Q distribution in the lungs, in the anterior to posterior direction, is more uniform in prone than in supine posture during spontaneous breathing and not simply a "reversal" of the distribution in supine (140,144). One possible explanation is differences in vessel conductance between ventral and dorsal parts of the lungs (24,41,46). NO is a known strong vasodilator and as endogenous NO is rapidly bound to hemoglobin and inactivated (158) there are prerequisites for a local action. There are indications suggesting that endogenous NO has a role in the regulation of regional Q (23,24) as the results in **Study III**. NO may be a mediating factor explaining the difference in vascular resistance between ventral and dorsal parts of the lungs. In quadruped animals lower vascular resistance in dorsal parts of the lungs (24) may be beneficial as it could make Q distribution more uniform by counteracting gravity. In upright position it is generally accepted that there is a basal dominance of regional Q distribution (25,26,32,77,81,134) and that V is less basally distributed (159-161). As humans are bipedal, in contrast to almost all animals, the distance between the highest and lowest parts, in the gravitational plane, of the lungs is larger than in most species. Gravity may therefore have greater influence on the distribution of regional Q distribution in upright humans (26) than in quadruped animals and this may have a negative effect on gas exchange. Furthermore, poor perfusion to some lung areas may make them more susceptible to some infections, most notably tuberculosis. This is supported by the fact that tuberculosis generally affects the highest parts of the lungs in animals (162). The production of high concentration of NO in the paranasal sinuses is also exclusive to humans and higher primates (163). Therefore, NO production in the paranasal sinuses may reflect an adoption to the bipedal state. It has not been possible to find any comparable studies following the same line of thought as in **Study VI**.

The effect of inhalation aneshesia on V,Q and V/Q distributions

It is well known that general anesthesia, including inhalation anesthesia causes a deteriorated gas exchange (164) and direct effects from inhalation anesthesia on the pulmonary circulation (111, 112,165) has been shown. We, therefore hypothesized, that the anesthetic gas (sevoflurane) could it self cause a deterioration of V/Q matching. V/Q matching during inhalation anesthesia with spontaneous breathing has previously been studied with MIDGET (109) and a wider distribution of V/Q matching during anesthesia was found in agreement with the findings in **Study V**. The previous study (109) found, however, a slightly bimodal V/Q distribution with a small peak of high V/Q values, representing shunts, which increased during inhalation anesthesia. No such second peak could be demonstrated in our study. One explanation could be the younger age group (range 20-34 years) in **Study V** compared to the age in the previous study (range 37-64 years) as shunt is known to increase with age (166). Another explanation might be the poor spatial resolution in **Study V**, compared to MIDGET as shunt could be located in small areas. A third explanation may be the measures we took to avoid atelectasis formation. No previous study on the anatomical location of disturbance of regional V/Q matching during inhalation anesthesia has to my knowledge been performed. MIDGET has long been considered

a golden standard for describing V,Q and V/Q distributions in the lungs. It does not suffer from limitations in spatial resolution but gives no information on the anatomical location of V,Q and V/Q. A limitation with MIDGET is an interdependence of V and Q as they are presented as a function of V/Q which can give a false impression of an increased variation of both V and Q even if only one of them is altered. This is illustrated by a simple example with 13 compartments. If all compartments have V/Q =1, a MIDGET examination will of cause show one single peak centered around 1 for both V and Q (Table 13, Fig. 10).

Table 13. A number of compartments (alveoli) with homogenous distribution of V and Q

	No alveoli	% of alveoli	V	% of total V	Q	% of total Q	V/Q
	1	7.7	1	7.7	1	7.7	1
	2	23.1	1	23.1	1	23.1	1
	5	38.5	1	38.5	1	38.5	1
	3	23.1	1	23.1	1	23.1	1
	1	7.7	1	7.7	1	7.7	1
Sum:	13	100 %		100%		100%	

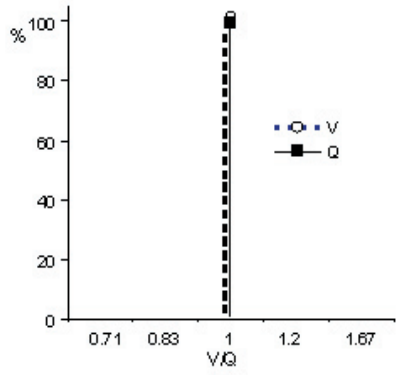


***Figure 10**. V and Q distribution as expected from MIDGET if V/Q is 1 in all compartments.*

With a redistribution of Q but with the same, even, distribution of V between the compartments the resulting MIDGET graph would show a broadening of the distributions for both V and Q (Table 14, figure 11).

Table 14. A number of compartments (alveoli) with homogenous distribution of V but inhomogeneous of Q

	No alveoli	% of alveoli	V	% of total V	Q	% of total Q	V/Q
	1	7.7	1	7.7	1	4.6	1.67
	2	23.1	1	23.1	1	18.5	1.25
	5	38.5	1	38.5	1	38.5	1
	3	23.1	1	23.1	1	27.7	0.83
	1	7.7	1	7.7	1	10.8	0.71
Sum:	13	100 %		100%		100%	

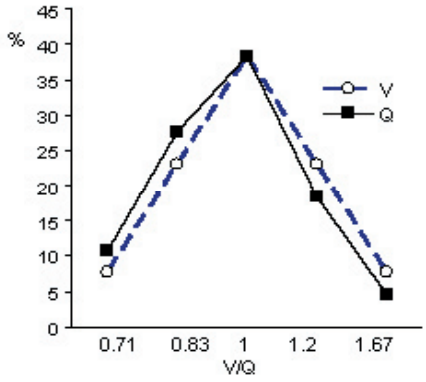


Figure 11. *V and Q distribution as expected from MIDGET if V has the same distribution between compartments but Q is reattributed*.

CLINICAL IMPLICATIONS

The fundamental clinical problem in ALI/ARDS patients is a deteriorated gas exchange caused by mismatch between V and Q. Even small improvements in gas exchange could have a large impact in these patients. Therefore, every factor influencing V/Q matching is of importance. That Q distribution in prone differs from that in supine is shown in **Study I** and this fundamental knowledge in physiology could add to the understanding of the positive effects from treatment of ALI/ARDS patients in the prone position. Understanding how Q distribution is influenced by posture in combination with CPAP could lead to modifications in the treatment of ALI/ARDS patients in prone position, e.g. the value of CPAP in prone might be smaller than in patients treated in supine posture. It could even be suggested that CPAP should perhaps be avoided when treating patients in their prone position (**Study II**). How the endogenous NO system is interacting with posture could also have an impact on treatment in ALI/ARDS patients, e.g. modifications of the relative magnitude of NO from endothelium and airways may alter V/Q matching. After an oral or endotracheal intubation NO from paranasal sinuses are by-passed. Small concentrations of NO in the inhaled air might be needed to counteract a possible negative effect of endothelial NO. That inhaled NO does influence Q distribution is shown in **Study III** and **VI**, and this may indicate that small amounts of NO should be added to inspired gas in marginal, intubated, patients with severe ALI. During general intravenous anesthesia with mechanical ventilation, Q distribution is mainly dependent in both postures while V is not affected by posture (**Study IV**). A tendency towards a more even vertical V/Q distribution in prone could, however, indicate an advantage for gas exchange in prone. Less well matched V/Q during inhalation anesthesia than awake is seen in **Study V** which may implicate that this form of anesthesia have negative effects in patients with already deranged V/Q matching.

FUTURE PERSPECTIVES

The methods used in this thesis make it possible to image V,Q and V/Q ratio distributions in various situations and to compare distributions between different conditions. As a good matching of V and Q in the lungs is fundamental for efficient gas exchange and maldistribution of Q is the most common cause of impaired oxygenation of the arterial blood (1), studies of this is of interest. Arterial blood oxygenation is impaired in many types of lung pathology but the exact mechanisms are often not known. The advantages with the methods employed in this thesis are:

1. The radiotracers used stay fixed in relation to lung parenchyma after the administration. This makes it possible to study the regional distribution of V,Q and V/Q ratios in any posture even though the registration is done in supine.

2. SPECT is currently the superior method for independent imaging of the regional distribution of V,Q and V/Q ratios in vivo.

3. The methods can be used to study changes in the regional distribution of V,Q and V/Q ratios registered under identical anatomical conditions which excludes many confounding factors.

4. The possibility to independently study the regional V,Q and V/Q ratio heterogeneity in vivo is so far unique.

Studies of V,Q and V/Q ratio distributions during diseases affecting gas-exchange could give insights into fundamental patho-physiological mechanisms. Conditions that could be studied are pulmonary hypertension, chronic obstructive lung disease and different types of interstitial lung disease. More detailed insights could also be gained by repeating some of the older studies in this thesis with the more modern dual energy technique, e.g. study the influence from NO on regional V/Q matching. Studying patients with ALI/ARDS could yield important information but performing such studies would pose large logistic and safety problems.

CONCLUSIONS

Based on the results from healthy volunteers in this thesis we observed that:

- In awake healthy volunteers Q distribution is more uniform in prone than in supine (**Study I**).
- Regional V distribution was uninfluenced by CPAP, both in supine and prone posture (**Study II**). Regional Q distribution is more dependent (**Study I,II**) and V/Q matching less uniform at CPAP breathing in both postures, and this is most pronounced in prone (**Study II**)
- In anaesthetized and mechanically ventilated healthy volunteers, regional V does not differ between prone and supine position whereas Q is more uniform in the ventral to dorsal direction in prone (**Study IV**).
- There is larger expression of nitric oxide synthase (NOS) and production of NO is higher in dorsal compared to ventral parts of the lungs. As NO is a powerful vaso-dilatator this might be a factor contributing to a more uniform Q distribution in prone compared to supine position (**Study III**).
- Paranasal, endogenous, NO induces a shift of regional Q from basal to apical parts of the lungs (**Study VI**).
- Inhalation anesthesia with sevoflurane does not alter regional V,Q and V/Q distributions in ventral to dorsal direction. The heterogeneity of regional V was un-affected while Q heterogeneity decreased and V/Q heterogeneity increased during inhalation anesthesia.

In general, it was concluded, that V distribution was un-affected by the tested variables whereas observed changes occurred in regional Q distribution.

ACKNOWLEDGEMENTS

I would like to express my gratitude to:

Professor Hans Jacobsson, my principal tutor for his enthusiasm and loyalty. Thank you for never giving up on me!! You have given me lots of wise advices that I've learned to appreciate.

Professor Sten Lindahl, my co-tutor, a great scientist who refuses to become a pure bureaucrat and always finds time for research despite a tight schedule. Thank you for your help at the end stage and thank you for all your ideas that has largely been the basis for this thesis.

Professor Stig Larsson, a true scientist and one of the smartest people I have ever known. Thank you for stimulating discussions and for your never-ending curiosity. I really admire you and miss you at work.

Alejandro Sanchez-Crespo, my co-tutor and friend. A rely brilliant man who will make a remarkable career as a scientist. Thank you for all the work you've put in to help me. Without you I would not have succeeded. It's been a pleasure working with you and I really hope our cooperation can continue. And I would like to apologize to **Jenny Hallberg** and **Santiago** for spoiling their holyday with Alex.

All my co-authors, especially **Margareta Mure**, **Johan Petersson**, **Eddy Weitzberg** and **Peter Radell** for help, support and stimulating discussions. Without your knowledge in anesthesiology and physiology this thesis had not been possible.

My colleagues at the department of thoracic radiology, especially **Åke Moritz**, **Jacek Pavlowski**, **Kerstin Klinge** and **Vendla Riesenfeldt** for support and for tolerating my mental absence during the last months.

My colleagues at the department of radiology who have taught me all I know about radiology, including my excellent head of department, **Lott Bergstrand** who have supported me.

All the brave **healthy volunteers**, who are the most important persons in this thesis.

Annette Ebberyd, who have organized the experiments and recruited the volunteers. Without you this would not have been possible.

Ringvor Hägglöf, for highly professional help with the Lay Out.

All the **staff at the department of nuclear medicine** that have helped and supported me and never complained.

My wife, **Miruna Nyrén**, who have been my best coach and my sons **Paul** and **Peter Nyrén** who I have not seen enough of me during the final stage of the thesis.

My **friends** and other **family** that I have neglected.

REFERENCES

1. Lumb AB (2005) Nunn's applied respiratory physiology. 6th edn. ELSEVIER Butterworth-Heinemann. Oxford, UK. Page 110-133.

2. Gal TJ. Anatomy and physiology of the respiratory system and the pulmonary circulation. Kaplan JA, Slinger PD, eds. Thoracic anesthesia. 3rd edition, Philadelphia, Elsevier Science 2003. Page 57-70.

3. Glenny RW. Determinants of regional ventilation and blood flow in the lung. Intensive Care Med. 2009 Nov;35(11):1833-42. Review.

4. West, JB. Respiratory Physiology – the essentials, Eighth edition, Williams & Wilkins 2008. Page 55-89.

5. Hedenstierna G, Edmark L. The effects of anesthesia and muscle paralysis on the respiratory system. Intensive Care Med. 2005 Oct;31(10):1327-35. Review.

6. Piehl MA, Brown RS. Use of extreme position changes in acute respiratory failure. Crit Care Med. 1976 Jan-Feb;4(1):13-4.

7. Douglas WW, Rehder K, Beynen FM, Sessler AD, Marsh HM. Improved oxygenation in patients with acute respiratory failure: the prone position. Am Rev Respir Dis. 1977 Apr;115(4):559-66.

8. Albert RK, Leasa D, Sanderson M, Robertson HT, Hlastala MP. The prone position improves arterial oxygenation and reduces shunt in oleic-acid-induced acute lung injury. Am Rev Respir Dis. 1987 Mar;135(3):628-33.

9. Lamm WJ, Graham MM, Albert RK. Mechanism by which the prone position improves oxygenation in acute lung injury. Am J Respir Crit Care Med. 1994 Jul;150(1):184-93.

10. Langer M, Mascheroni D, Marcolin R, Gattinoni L. The prone position in ARDS patients. A clinical study. Chest. 1988 Jul;94(1):103-7.

11. Murdoch IA, Storman MO. Improved arterial oxygenation in children with the adult respiratory distress syndrome: the prone position. Acta Paediatr. 1994 Oct;83(10):1043-6.

12. Mure M, Martling CR, Lindahl SG. Dramatic effect on oxygenation in patients with severe acute lung insufficiency treated in the prone position. Crit Care Med. 1997 Sep;25(9):1539-44.

13. Lumb AB (2005) Nunn's applied respiratory physiology. 6th edn. ELSEVIER Butterworth-Heinemann. Oxford, UK. Page 209-25.

14. Hitchcock MA, Hitchcock FA. Paul Bert. Barometric pressure. Research in experimental physiology. (1943) Collage Book Company. Columbus, OH, USA.

15. Lumb AB (2005) Nunn's applied respiratory physiology. 6th edn. ELSEVIER Butterworth-Heinemann. Oxford, UK. Page 25-28.

16. Euler, von US, Liljestrand G. Observations on the pulmonary arterial blood pressure in the cat. Acta Phys. Scandinav. 1946;12:301-19.

17. Lumb AB (2005) Nunn's applied respiratory physiology. 6th edn. ELSEVIER Butterworth-Heinemann. Oxford, UK. Page 92-109.

18. Peacock AJ (1996) Pulmonary Circulation A handbook for clinicians. 1st edn. Chapman & Hall Medical. London, UK. Page 71-85.

19. Svanberg L. Influence of posture on the lung volumes, ventilation and circulation in normals; a spirometric-bronchospirometric investigation. Scand J Clin Lab Invest. 1957;9(Suppl 25):1-195.

20. Furchgott RF, Zawadzki JV. The obligatory role of endothelial cells in the relaxation of arterial smooth muscle by acetylcholine. Nature. 1980 Nov 27;288(5789):373-6.

21. Ignarro LJ. Endothelium-derived nitric oxide: actions and properties. FASEB J. 1989 Jan;3(1):31-6. Review.

22. Ignarro LJ, Buga GM, Wood KS, Byrns RE, Chaudhuri G. Endothelium-derived relaxing factor produced and released from artery and vein is nitric oxide. Proc Natl Acad Sci U S A. 1987 Dec;84(24):9265-9.

23. Mitchell JA, Ali F, Bailey L, Moreno L, Harrington LS. Role of nitric oxide and prostacyclin as vasoactive hormones released by the endothelium. Exp Physiol. 2008 Jan;93(1):141-7.

24. Pelletier N, Robinson NE, Kaiser L, Derksen FJ. Regional differences in endothelial function in horse lungs: possible role in blood flow distribution? J Appl Physiol. 1998 Aug;85(2):537-42.

25. Bryan AC, Bentivoglio LG, Beerel F, MacLeish H, Zidulka A, Bates DV. Factors affecting regional distribution of ventilation and perfusion in the lung. J Appl Physiol. 1964 May;19:395-402.

26. Anthonisen NR, Milic-Emili J. Distribution of pulmonary perfusion in erect man. J Appl Physiol. 1966 May;21(3):760-6.

27. Bryan AC, Milic-Emili J, Pengelly D. Effect of gravity on the distribution of pulmonary ventilation. J Appl Physiol. 1966 May;21(3):778-84.

28. West JB, Dollery CT. Distribution of blood flow and the pressure-flow relations of the whole lung. J Appl Physiol. 1965 March;20(2):175-83.

29. Newhouse MT, Wright FJ, Ingham GK, Archer NP, Hughes LB, Hopkins OL. Use of scintillation camera and 135-xenon for study of topographic pulmonary function. Respir Physiol. 1968 Mar;4(2):141-53.

30. Wagner HN Jr.: Regional ventilation and perfusion. In: Principles of Nuclear Medicine, edited by Wagner HN, Jr, Szabo Z and Buchanan JW. Philadelphia, PA: Saunders, 1995, pp 881-95

31. Rehder K, Knopp TJ, Sessler AD. Regional intrapulmonary gas distribution in awake and anesthetized-paralyzed prone man. *J Appl Physiol* 1978;45:528–535.

32. Reed JH Jr, Wood EH. Effect of body position on vertical distribution of pulmonary blood flow. J Appl Physiol. 1970 Mar;28(3):303-11.

33. Larsson SA. Gamma camera emission tomography. Development and properties of a multi-sectional emission computed tomography system. Acta Radiol Suppl. 1980;363:1-75.

34. Msaki P, Axelsson B, Dahl CM, Larsson SA. Generalized scatter correction method in SPECT using point scatter distribution functions. J Nucl Med. 1987 Dec;28(12):1861-9.

35. Sanchez-Crespo A, Petersson J, Nyrén S, Mure M, Glenny RW, Thorell JO, Jacobsson H, Lindahl SG, Larsson SA. A novel quantitative dual-isotope method for simultaneous ventilation and perfusion lung SPET. Eur J Nucl Med Mol Imaging. 2002;29:863-75.

36. Petersson J, Sanchez-Crespo A, Rohdin M, Montmerle S, Nyrén S, Jacobsson H, Larsson SA, Lindahl SG, Linnarsson D, Glenny RW, Mure M. Physiological evaluation of a new quantitative SPECT method measuring regional ventilation and perfusion. J Appl Physiol. 2004 Mar;96(3):1127-36.

37. Ter-Pogossian MM, Phelps ME, Hoffman EJ, Mullani NA. A positron-emission transaxial tomograph for nuclear imaging (PETT). Radiology. 1975 Jan;114(1):89-98.

38. West JB, Dollery CT, Naimark A. Distribution of blood flow in isolated lung; relation to vascular and alveolar pressure. J Appl Physiol. 1964 Jul; 19:713-24.

39. Tokics L, Hedenstierna G, Svensson L, Brismar B, Cederlund T, Lundquist H, Strandberg A. V/Q distribution and correlation to atelectasis in anesthetized paralyzed humans. J Appl Physiol. 1996 Oct;81(4):1822-33.

40. Glenny RW, Lamm WJ, Albert RK, Robertson HT. Gravity is a minor determinant of pulmonary blood flow distribution. J Appl Physiol. 1991;71:620-9.

41. Glenny RW, Bernard S, Robertson HT, Hlastala MP. Gravity is an important but secondary determinant of regional pulmonary blood flow in upright primates. J Appl Physiol. 1999 Feb;86(2):623-32.

42. Jonson B, Westling H, White T, Wollmer P. Klinisk Fysiologi, Liber, Stocholm. 1st Edn 1998, Page 33-100.

43. Kendrick AH. Comparison of methods of measuring static lung volumes. Monaldi Arch Chest Dis. 1996 Oct;51(5):431-9. Review.

44. Riley RL, Cournand A. Ideal alveolar air and the analysis of ventilation-perfusion relationships in the lungs. J Appl Physiol. 1949 Jun;1(12):825-47.

45. Wagner PD, Saltzman HA, West JB. Measurement of continuous distributions of ventilation-perfusion ratios: theory. J Appl Physiol. 1974 May;36(5):588-99.

46. Beck KC, Rehder K. Differences in regional vascular conductances in isolated dog lungs. J Appl Physiol. 1986 Aug;61(2):530-8.

47. Robertson HT, Hlastala MP. Microsphere maps of regional blood flow and regional ventilation. J Appl Physiol. 2007 Mar;102(3):1265-72. Review.

48. Hoffman EA, Ritman EL. Effect of body orientation on regional lung expansion in dog and sloth. J Appl Physiol. 1985 Aug;59(2):481-91.

49. Gefter WB, Hatabu H. Evaluation of pulmonary vascular anatomy and blood flow by magnetic resonance. J Thorac Imaging. 1993 Spring;8(2):122-36. Review.

50. Hecht EM, Rosenkrantz A. Pulmonary MR angiography techniques and applications. Magn Reson Imaging Clin N Am. 2009 Feb;17(1):101-31. Review

51. Kauczor HU, Kreitner KF. MRI of the pulmonary parenchyma. Eur Radiol. 1999;9(9):1755-64. Review.

52. Kauczor HU, Kreitner KF. Contrast-enhanced MRI of the lung. Eur J Radiol. 2000 Jun;34(3):196-207. Review.

53. Prisk GK, Yamada K, Henderson AC, Arai TJ, Levin DL, Buxton RB, Hopkins SR: Pulmonary perfusion in the prone and supine postures in the normal human lung. J Appl Physiol 2007; 103:883-94.

54. Mai VM, Berr SS. MR perfusion imaging of pulmonary parenchyma using pulsed arterial spin labeling techniques: FAIRER and FAIR. J Magn Reson Imaging. 1999 Mar;9(3):483-7.

55. Uematsu H, Levin DL, Hatabu H. Quantification of pulmonary perfusion with MR imaging: recent advances. Eur J Radiol. 2001 Mar;37(3):155-63. Review.

56. Burnham KJ, Arai TJ, Dubowitz DJ, Henderson AC, Holverda S, Buxton RB, Prisk GK, Hopkins SR. Pulmonary perfusion heterogeneity is increased by sustained, heavy exercise in humans. J Appl Physiol. 2009 Nov;107(5):1559-68.

57. Hopkins SR, Henderson AC, Levin DL, Yamada K, Arai T, Buxton RB, Prisk GK. Vertical gradients in regional lung density and perfusion in the supine human lung: the Slinky effect. J Appl Physiol. 2007 Jul;103(1):240-8.

58. Kauczor H, Surkau R, Roberts T. MRI using hyperpolarized noble gases. Eur Radiol. 1998;8(5):820-7. Review.

59. Emami K, Stephen M, Kadlecek S, Cadman RV, Ishii M, Rizi RR. Quantitative assessmen of lung using hyperpolarized magnetic resonance imaging. Proc Am Thorac Soc. 2009 Aug 15;6(5):431-8. Review.

60. Hatabu H, Chen Q, Levin DL, Tadamura E, Edelman RR. Ventilation-perfusion MR imaging of the lung. Magn Reson Imaging Clin N Am. 1999 May;7(2):379-92,.Review.

61. Ohno Y, Sugimura K, Hatabu H. Clinical oxygen-enhanced magnetic resonance imaging of the lung. Top Magn Reson Imaging. 2003 Jun;14(3):237-43. Review.

62. Mayo JR, MacKay AL, Whittall KP, Baile EM, Paré PD. Measurement of lung water content and pleural pressure gradient with magnetic resonance imaging. J Thorac Imaging. 1995 Winter;10(1):73-81.

63. Rhodes CG, Valind SO, Brudin LH, Wollmer PE, Jones T, Buckingham PD, Hughes JM. Quantification of regional V/Q ratios in humans by use of PET. II. Procedure and normal values. J Appl Physiol. 1989 Apr;66(4):1905-13.

64. Rhodes CG, Valind SO, Brudin LH, Wollmer PE, Jones T, Hughes JM. Quantification of regional V/Q ratios in humans by use of PET. I. Theory. J Appl Physiol. 1989 Apr;66(4):1896-904.

65. Neeb D, Kunz RP, Ley S, Sz√°bo G, Strauss LG, Kauczor HU, Kreitner KF, Schreiber LM. Quantification of pulmonary blood flow (PBF): validation of perfusion MRI and nonlinear contrast agent (CA) dose correction with H(2)15O positron emission tomography (PET). Magn Reson Med. 2009 Aug;62(2):476-87.

66. Wolfkiel CJ, Rich S. Analysis of regional pulmonary enhancement in dogs by ultrafast computed tomography. Invest Radiol. 1992 Mar;27(3):211-6.

67. Costa EL, Lima RG, Amato MB. Electrical impedance tomography. Curr Opin Crit Care. 2009 Feb;15(1):18-24. Review.

68. Pettersson H. edit. (1998) The encyclopedia of medical imaging, volume 1, The NICER institute, Oslo, Norway.

69. Petersson J, Sanchez-Crespo A, Larsson SA, Mure M. Physiological imaging of the lung: single-photon-emission computed tomography (SPECT). J Appl Physiol. 2007 Jan;102(1):468-76. Review.

70. Amis TC, Crawford AB, Davison A, Engel LA. Distribution of inhaled 99mtechnetium labelled ultrafine carbon particleaerosol (Technegas) in human lungs. Eur Respir J 1990; 3:679–85.

71. Tapling GV, MacDonald NS. Radiochemistry of macroaggregated albumin and newer lung scanning agents. Semin Nucl Med 1971; 1:132–52.

72. Petersson J, Rohdin M, Sánchez-Crespo A, Nyrén S, Jacobsson H, Larsson SA, Lindahl SG, Linnarsson D, Glenny RW, Mure M. Paradoxical redistribution of pulmonary blood flow in prone and supine humans exposed to hypergravity. J Appl Physiol. 2006 Jan;100(1):240-8.

73. Petersson J, Rohdin M, Sánchez-Crespo A, Nyrén S, Jacobsson H, Larsson SA, Lindahl SG, Linnarsson D, Neradilek B, Polissar NL, Glenny RW, Mure M. Regional lung blood flow and ventilation in upright humans studied with quantitative SPECT. Respir Physiol Neurobiol. 2009 Mar 31;166(1):54-60.

74. Kaneko K, Milic-Emili J, Dolovich MB, Dawson A, Bates DV. Regional distribution of ventilation and perfusion as a function of body position. J Appl Physiol. 1966 May;21(3):767-77.

75. Bake B, Bjure J, Grimby G, Milic-Emili J, Nilsson NJ. Regional distribution of inspired gas in supine man. Scand J Respir Dis. 1967;48(3):189-96.))

76. Milic-Emili J, Henderson JA, Dolovich MB, Trop D, Kaneko K. Regional distribution of inspired gas in the lung. J Appl Physiol. 1966 May;21(3):749-59.

77. Amis TC, Jones HA, Hughes JM. Effect of posture on inter-regional distribution of pulmonary perfusion and VA/Q ratios in man. Respir Physiol. 1984 May;56(2):169-82.

78. Orphanidou D, Hughes JM, Myers MJ, Al-Suhali AR, Henderson B. Tomography of regional ventilation and perfusion using krypton 81m in normal subjects and asthmatic patients. Thorax. 1986 Jul;41(7):542-51.

79. Lai-Fook SJ. Pleural mechanics and fluid exchange. Physiol Rev. 2004 Apr;84(2):385-410. Review.

80. Albert RK, Hubmayr RD. The prone position eliminates compression of the lungs by the heart. Am J Respir Crit Care Med. 2000 May;161(5):1660-5.

81. Landmark SJ, Knopp TJ, Rehder K, Sessler AD. Regional pulmonary perfusion and V/Q in awake and anesthetized-paralyzed man. J Appl Physiol. 1977 Dec;43(6):993-1000.

82. Neumann P, Wrigge H, Zinserling J, Hinz J, Maripuu E, Andersson LG, Putensen C, Hedenstierna G. Spontaneous breathing affects the spatial ventilation and perfusion distribution during mechanical ventilatory support. Crit Care Med. 2005 May;33(5):1090-5.

83. Reber A, Engberg G, Sporre B, Kviele L, Rothen HU, Wegenius G, Nylund U, Hedenstierna G. Volumetric analysis of aeration in the lungs during general anaesthesia. Br J Anaesth. 1996 Jun;76(6):760-6.

84. Olson LE, Rodarte JR. Regional differences in expansion in excised dog lung lobes. J Appl Physiol. 1984 Dec;57(6):1710-4.

85. Hubmayr RD, Rodarte JR, Walters BJ, Tonelli FM. Regional ventilation during spontaneous breathing and mechanical ventilation in dogs. J Appl Physiol. 1987 Dec;63(6):2467-75.

86. Arai TJ, Henderson AC, Dubowitz DJ, Levin DL, Friedman PJ, Buxton RB, Prisk GK, Hopkins SR. Hypoxic pulmonary vasoconstriction does not contribute to pulmonary blood flow heterogeneity in normoxia in normal supine humans. J Appl Physiol. 2009 Apr;106(4):1057-64.

87. Culotta E, Koshland DE Jr. NO news is good news. Science. 1992 Dec 18;258(5090):1862-5. Review.

88. Vallance P, Chan N. Endothelial function and nitric oxide: clinical relevance. Heart. 2001 Mar;85(3):342-50. Review.

89. Palmer RM, Moncada S. A novel citrulline-forming enzyme implicated in the formation of nitric oxide by vascular endothelial cells. Biochem Biophys Res Commun. 1989 Jan 16;158(1):348-52.

90. Wang Y, Marsden PA. Nitric oxide synthases: biochemical and molecular regulation. Curr Opin Nephrol Hypertens. 1995 Jan;4(1):12-22. Review.

91. Zweier JL, Samouilov A, Kuppusamy P. Non-enzymatic nitric oxide synthesis in biological systems. Biochim Biophys Acta. 1999 May 5;1411(2-3):250-62. Review.

92. Ferrario L, Amin HM, Sugimori K, Camporesi EM, Hakim TS. Site of action of endogenous nitric oxide on pulmonary vasculature in rats Pflugers Arch. 1996 Jul;432(3):523-7.

93. Frostell C, Fratacci MD, Wain JC, Jones R, Zapol WM. Inhaled nitric oxide. A selective pulmonary vasodilator reversing hypoxic pulmonary vasoconstriction. Circulation. 1991 Jun;83(6):2038-47.

94. Barnes PJ, Liu SF. Regulation of pulmonary vascular tone. Pharmacol Rev. 1995 Mar;47(1):87-131. Review.

95. Lundberg JO, Settergren G, Gelinder S, Lundberg JM, Alving K, Weitzberg E. Inhalation of nasally derived nitric oxide modulates pulmonary function in humans. Acta Physiol Scand. 1996 Dec;158(4):343-7.

96. Schedin U, Röken BO, Nyman G, Frostell C, Gustafsson LE. Endogenous nitric oxide in the airways of different animal species. Acta Anaesthesiol Scand. 1997 Oct;41(9):1133-41.

97. Törnberg DC, Marteus H, Schedin U, Alving K, Lundberg JO, Weitzberg E. Nasal and oral contribution to inhaled and exhaled nitric oxide: a study in tracheotomized patients. Eur Respir J. 2002 May;19(5):859-64.

98. Ricciardolo FL, Sterk PJ, Gaston B, Folkerts G. Nitric oxide in health and disease of the respiratory system. Physiol Rev. 2004 Jul;84(3):731-65. Review.

99. Hobbs AJ, Higgs A, Moncada S. Inhibition of nitric oxide synthase as a potential therapeutic target. Annu Rev Pharmacol Toxicol. 1999;39:191-220. Review.

100. Glenny RW, Bernard SL, Robertson HT. Pulmonary blood flow remains fractal down to the level of gas exchange. J Appl Physiol. 2000 Aug;89(2):742-8.

101. Glenny RW, Lamm WJ, Bernard SL, An D, Chornuk M, Pool SL, Wagner WW Jr, Hlastala MP, Robertson HT. Selected contribution: redistribution of pulmonary perfusion during weightlessness and increased gravity. J Appl Physiol. 2000 Sep;89(3):1239-48.

102. Prisk GK, Guy HJ, Elliott AR, West JB. Inhomogeneity of pulmonary perfusion during sustained microgravity on SLS-1. J Appl Physiol. 1994 Apr;76(4):1730-8.

103. Altemeier WA, McKinney S, Glenny RW. Fractal nature of regional ventilation distribution. J Appl Physiol. 2000 May;88(5):1551-7.

104. Melsom MN, Kramer-Johansen J, Flatebø T, Müller C, Nicolaysen G. Distribution of pulmonary ventilation and perfusion measured simultaneously in awake goats. Acta Physiol Scand. 1997 Mar;159(3):199-208. Erratum in: Acta Physiol Scand 1997 Jul;160(3):297.

105. Altemeier WA, Robertson HT, Glenny RW. Pulmonary gas-exchange analysis by using simultaneous deposition of aerosolized and injected microspheres. J Appl Physiol. 1998 Dec;85(6):2344-51.

106. Hlastala MP, Glenny RW. Vascular structure determines pulmonary blood flow distribution. News Physiol Sci. 1999 Oct;14:182-6. Review.

107. Strandberg A, Tokics L, Brismar B, Lundquist H, Hedenstierna G. Atelectasis during anaesthesia and in the postoperative period. Acta Anaesthesiol Scand. 1986 Feb;30(2):154-8.

108. Rothen HU, Sporre B, Engberg G, Wegenius G, Reber A, Hedenstierna G. Atelectasis and pulmonary shunting during induction of general anaesthesia—can they be avoided? Acta Anaesthesiol Scand. 1996 May;40(5):524-9.

109. Bindslev L, Hedenstierna G, Santesson J, Gottlieb I, Carvallhas A. Ventilation-perfusion distribution during inhalation anaesthesia. Effects of spontaneous breathing, mechanical ventilation and positive end-expiratory pressure. Acta Anaesthesiol Scand. 1981 Aug;25(4):360-71.

110. Westbrook PR, Stubbs SE, Sessler AD, Rehder K, Hyatt RE. Effects of anesthesia and muscle paralysis on respiratory mechanics in normal man. J Appl Physiol. 1973 Jan;34(1):81-6.

111. Marshall BE. Effects of anesthetics on pulmonary gas exchange. In: Stanley TH. Sperry RJ eds. *Anesthesia and the lung*. London: Kluwer Academic Publishers, 1989; 117–25.

112. Takemura M, Shiokawa Y, Okamoto S, Uno H, Futagawa K, Koga Y. Volatile anesthetics constrict pulmonary artery in rabbit lung perfusion model. *J Anesth* 2005; 19:343–6.

113. Ware LB, Matthay MA. The acute respiratory distress syndrome, N Engl J Med. 2000 May 4;342(18):1334-49. Review

114. Bernard GR, Artigas A, Brigham KL, Carlet J, Falke K, Hudson L, Lamy M, Legall JR, Morris A, Spragg R. The American-European Consensus Conference on ARDS. Definitions, mechanisms, relevant outcomes, and clinical trial coordination. Am J Respir Crit Care Med. 1994 Mar;149(3 Pt 1):818-24. Review.

115. Kallet RH, Jasmer RM, Pittet JF, Tang JF, Campbell AR, Dicker R, Hemphill C, Luce JM. Clinical implementation of the ARDS network protocol is associated with reduced hospital mortality compared with historical controls. Crit Care Med. 2005 May;33(5):925-9.

116. Luhr OR, Antonsen K, Karlsson M, Aardal S, Thorsteinsson A, Frostell CG, Bonde J. Incidence and mortality after acute respiratory failure and acute respiratory distress syndrome in Sweden, Denmark, and Iceland. The ARF Study Group. Am J Respir Crit Care Med. 1999 Jun;159(6):1849-61.

117. Martin M, Salim A, Murray J, Demetriades D, Belzberg H, Rhee P. The decreasing incidence and mortality of acute respiratory distress syndrome after injury: a 5-year observational study. J Trauma. 2005 Nov;59(5):1107-13.

118. Phua J, Badia JR, Adhikari NK, Friedrich JO, Fowler RA, Singh JM, Scales DC, Stather DR, Li A, Jones A, Gattas DJ, Hallett D, Tomlinson G, Stewart TE, Ferguson ND. Has mortality from acute respiratory distress syndrome decreased over time?: A systematic review. Am J Respir Crit Care Med. 2009 Feb 1;179(3):220-7.

119. Winter PM, Smith G. The toxicity of oxygen. Anesthesiology. 1972 Aug;37(2):210-41. Review.

120. Donahoe M. Basic ventilator management: lung protective strategies. Surg Clin North Am. 2006 Dec;86(6):1389-408. Review.

121. Ball WC Jr, Stewart PB, Newsham LG, Bates DV. Regional pulmonary function studied with xenon 133. J Clin Invest. 1962 Mar;41:519-31.

122. Hughes JM, Glazier JB, Maloney JE, West JB. Effect of lung volume on the distribution of pulmonary blood flow in man. Respir Physiol. 1968 Jan;4(1):58-72.

123. Musch G, Layfield JD, Harris RS, Melo MF, Winkler T, Callahan RJ, Fischman AJ, Venegas JG: Topographical distribution of pulmonary perfusion and ventilation, assessed by PET in supine and prone humans. J Appl Physiol 2002; 93:1841-51.

124. Richter T, Bellani G, Scott Harris R, Vidal Melo MF, Winkler T, Venegas JG, Musch G. Effect of prone position on regional shunt, aeration, and perfusion in experimental acute lung injury. Am J Respir Crit Care Med. 2005 Aug 15;172(4):480-7.

125. Altemeier WA, McKinney S, Krueger M, Glenny RW. Effect of posture on regional gas exchange in pigs. J Appl Physiol. 2004 Dec;97(6):2104-11.

126. Ashbaugh DG, Bigelow DB, Petty TL, Levine BE. Acute respiratory distress in adults. Lancet. 1967 Aug 12;2(7511):319-23.

127. Lumb AB (2005) Nunn's applied respiratory physiology. 6th edn. ELSEVIER Butterworth-Heinemann. Oxford, UK. Page 408-418.

128. Rubenfeld GD. Epidemiology of acute lung injury. Crit Care Med. 2003 Apr;31(4 Suppl): S276-84. Review.

129. Luce JM. Acute lung injury and the acute respiratory distress syndrome. Crit Care Med. 1998 Feb;26(2):369-76. Review.

130. Gattinoni L, Pelosi P, Suter PM, Pedoto A, Vercesi P, Lissoni A. Acute respiratory distress syndrome caused by pulmonary and extrapulmonary disease. Different syndromes? Am J Respir Crit Care Med. 1998 Jul;158(1):3-11.

131. O'Doherty MJ, Miller RF. Aerosols for therapy and diagnosis. Eur J Nucl Med. 1993 Dec;20(12):1201-13. Review

132. Weibel ER, Sapoval B, Filoche M. Design of peripheral airways for efficient gas exchange. Respir Physiol Neurobiol. 2005 Aug 25;148(1-2):3-21

133. Petersson J, Rohdin M, Sánchez-Crespo A, Nyrén S, Jacobsson H, Larsson SA, Lindahl SG, Linnarsson D, Neradilek B, Polissar NL, Glenny RW, Mure M. Posture primarily affects lung tissue distribution with minor effect on blood flow and ventilation. Respir Physiol Neurobiol. 2007 Jun 15;156(3):293-303.

134. Maeda H, Itoh H, Ishii Y, Todo G, Mukai T, Fujita M, Kambara H, Kawai C, Torizuka K. Pulmonary blood flow distribution measured by radionuclide-computed tomography. J Appl Physiol. 1983 Jan;54(1):225-33.

135. Ross DJ, Wu P, Mohsenifar Z. Assessment of postural differences in regional pulmonary perfusion in man by single-photon emission computerized tomography. Clin Sci (Lond). 1997 Jan;92(1):81-5.

136. Hakim TS, Dean GW, Lisbona R. Effect of body posture on spatial distribution of pulmonary blood flow. J Appl Physiol. 1988 Mar;64(3):1160-70.

137. Melsom MN, Flatebø T, Kramer-Johansen J, Aulie A, Sjaastad OV, Iversen PO, Nicolaysen G. Both gravity and non-gravity dependent factors determine regional blood flow within the goat lung. Acta Physiol Scand. 1995 Apr;153(4):343-53.

138. Melsom MN, Flatebø T, Nicolaysen G. No apparent effect of nitric oxide on the local matching of pulmonary perfusion and ventilation in awake sheep. Acta Physiol Scand. 2000 Mar;168(3):361-70.

139. Brudin LH, Rhodes CG, Valind SO, Jones T, Hughes JM. Interrelationships between regional blood flow, blood volume, and ventilation in supine humans. J Appl Physiol. 1994 Mar;76(3):1205-10.

140. Jones AT, Hansell DM, Evans TW. Pulmonary perfusion in supine and prone positions: an electron-beam computed tomography study. J Appl Physiol. 2001 Apr;90(4):1342-8.

141. Stock KW, Chen Q, Levin D, Hatabu H, Edelman RR. Demonstration of gravity-dependent lung perfusion with contrast-enhanced magnetic resonance imaging. J Magn Reson Imaging. 1999 Apr;9(4):557-61.

142. Almquist HM, Palmer J, Jonson B, Wollmer P. Pulmonary perfusion and density gradients in healthy volunteers. J Nucl Med. 1997 Jun;38(6):962-6.

143. Walther SM, Domino KB, Glenny RW, Polissar NL, Hlastala MP: Pulmonary blood flow distribution has a hilar-to-peripheral gradient in awake, prone sheep. J Appl Physiol 1997; 82:678-85.

144. Hlastala MP, Bernard SL, Erickson HH, Fedde MR, Gaughan EM, McMurphy R, Emery MJ, Polissar N, Glenny RW. Pulmonary blood flow distribution in standing horses is not dominated by gravity. J Appl Physiol. 1996 Sep;81(3):1051-61.

145. Suga K, Nishigauchi K, Kume N, Koike S, Takano K, Matsunaga N. Regional ventilatory evaluation using dynamic SPET imaging of xenon-133 washout in obstructive lung disease: an initial study. Eur J Nucl Med. 1995 Mar;22(3):220-6.

146. Treppo S, Mijailovich SM, Venegas JG: Contributions of pulmonary perfusion and ventilation to heterogeneity in V(A)/Q measured by PET. J Appl Physiol 1997; 82:1163-76.

147. Wagner PD, Laravuso RB, Uhl RR, West JB. Continuous distributions of ventilation-perfusion ratios in normal subjects breathing air and 100 per cent O2. J Clin Invest. 1974 Jul;54(1):54-68.

148. Stock MC, Downs JB, Gauer PK, Alster JM, Imrey PB. Prevention of postoperative pulmonary complications with CPAP, incentive spirometry, and conservative therapy. Chest. 1985 Feb;87(2):151-7.

149. Ferreyra GP, Baussano I, Squadrone V, Richiardi L, Marchiaro G, Del Sorbo L, Mascia L, Merletti F, Ranieri VM. Continuous positive airway pressure for treatment of respiratory complications after abdominal surgery: a systematic review and meta-analysis. Ann Surg. 2008 Apr;247(4):617-26. Review.

150. Kosuda S, Kobayashi H, Kusano S. Change in regional pulmonary perfusion as a result of posture and lung volume assessed using technetium-99m macroaggregated albumin SPET. Eur J Nucl Med. 2000 May;27(5):529-35.

151. Walther SM, Johansson MJ, Flatebø T, Nicolaysen A, Nicolaysen G. Marked differences between prone and supine sheep in effect of PEEP on perfusion distribution in zone II lung. J Appl Physiol. 2005 Sep;99(3):909-14.

152. Wiener CM, Kirk W, Albert RK: Prone position reverses gravitational distribution of perfusion in dog lungs with oleic acid-induced injury. J Appl Physiol 1990; 68:1386-92.

153. Mure M, Domino KB, Lindahl SG, Hlastala MP, Altneier WA, Glenny RW: Regional ventilation-perfusion distribution is more uniform in the prone position: J Appl Physiol 2000; 88:1076-83.

154. Walther SM, Domino KB, Glenny RW, Hlastala MP. Pulmonary blood flow distribution in sheep: effects of anesthesia, mechanical ventilation, and change in posture. Anesthesiology. 1997 Aug;87(2):335-42.

155. Beck KC, Vettermann J, Rehder K: Gas exchange in dogs in the prone and supine positions. J Appl Physiol 1992; 72:2292-7.

156. Robertson HT, Glenny RW, Stanford D, McInnes LM, Luchtel DL, Covert D. High-resolution maps of regional ventilation utilizing inhaled fluorescent microspheres. J Appl Physiol. 1997 Mar;82(3):943-53.

157. Venegas JG, Yamada Y, Burnham C, Hales CA. Local gas transport in eucapnic ventilation: effects of gravity and breathing frequency. J Appl Physiol. 1990 Jun;68(6):2287-95.

158. Jia L, Bonaventura C, Bonaventura J, Stamler JS. S-nitrosohaemoglobin: a dynamic activity of blood involved in vascular control. Nature. 1996 Mar 21;380(6571):221-6.

159. West JB, Dollery CT. Distribution of blood flow and ventilation-perfusion ratio in the lung, measured with radioactive carbon dioxide. J Appl Physiol. 1960 May;15:405-10.

160. West JB. Distribution of gas and blood in the normal lungs. Br Med Bull. 1963 Jan;19:53-8.

161. West JB. Regional differences in gas exchange in the lung of erect man. J Appl Physiol. 1962 Nov;17:893-8.

162. Goodwin RA, Des Prez RM. Apical localization of pulmonary tuberculosis, chronic pulmonary histoplasmosis, and progressive massive fibrosis of the lung. Chest. 1983 May;83(5):801-5.

163. Schedin U, Röken BO, Nyman G, Frostell C, Gustafsson LE. Endogenous nitric oxide in the airways of different animal species. Acta Anaesthesiol Scand. 1997 Oct;41(9):1133-41.

164. Lumb AB (2005) Nunn's applied respiratory physiology. 6th edn. ELSEVIER Butterworth-Heinemann. Oxford, UK. Page 297-326.

165. Hedenstierna G. Contribution of multiple inert gas elimination technique to pulmonary medicine. 6. Ventilation-perfusion relationships during anaesthesia. Thorax. 1995 Jan;50(1):85-91. Review.

166. Gunnarsson L, Tokics L, Gustavsson H, Hedenstierna G. Influence of age on atelectasis formation and gas exchange impairment during general anaesthesia. Br J Anaesth. 1991 Apr;66(4):423-32.