THE EFFECT OF ALTERATIONS OF PULMONARY ARTERIAL AND ALVEOLAR GAS TENSIONS ON THE PRESSURE-VOLUME CURVE AND SURFACE TENSION OF DOG LUNGS.

Part I. A review of the literature.

Part II. An experimental study.

by

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Acute cessation of pulmonary blood flow in ventilated lobes and perfusion of lobes with hypoxic, hypercapnic blood increased the retractive forces of the lobes. These changes were reversed by perfusion with hypoxic, eucapnic blood.

Reduction in pulmonary blood flow, hypoxia-eucapnia, hypercapnia in the presence of high oxygen tension and non-respiratory acidosis did not alter the mechanical properties of the lobes.

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Part I

The current literature on the effects of alteration in pulmonary arterial blood flow and in alveolar gas tensions on the mechanical properties of the lung is reviewed. Pulmonary artery ligation, cardio-pulmonary bypass, oxygen at high pressures, and high concentrations of CO_2 in the inspired gas cause changes in the lung mechanics associated with a decrease in the activity of the lung lining material. Little is known regarding the mechanisms by which inactivation of surfactant takes place under these conditions. However, there is some evidence that on exposure of lung tissue to high pressures of oxygen, the inactivation of surfactant is secondary to inhibitors originating from plasma that has exuded onto the alveolar surface. Whether CO_2 exerts its effect directly or indirectly by altering the hydrogen ion concentrations of the cells is not yet clearly defined.

Part II

To further understand the roles of oxygen, CO_2, and pH on surface forces of the lung these studies were undertaken. The effects of acute alterations of pulmonary blood flow, pulmonary arterial blood gas tensions and alveolar gas tensions on the mechanical properties of the lungs were studied on the left lower lobes of open-chest dogs. The pressure-volume characteristics (expressed as percentage of the maximum lobe air volume) of ventilated lobes deprived of pulmonary

PART I. A REVIEW OF THE LITERATURE

A. Introduction

Over the past decade an appreciation of the role of the alveolar lining layer has provided new insight into the metabolism and function of the lung in health and disease. Many studies have been carried on in order to find evidence for the existence of a surface active substance in the lung, its source, its chemical identification, its relation to the lung metabolism, the effects of its alterations on lung function, and factors affecting the activity of the substance in the lung. One approach to understanding the conditions essential for the integrity of pulmonary surfactant is to examine the conditions in which it may be altered.

The present thesis is the study of the effects of alterations in the pulmonary circulation, the pulmonary blood gas tensions, and the alveolar gas tensions on the activity of the lung lining material (surfactant).

B. Pulmonary Artery Ligation

The effect of occlusion of the pulmonary artery on the lung has been studied by several workers. Schlaepfer (1) ligated one pulmonary artery in dogs and rabbits and noted atelectasis and some alveolar hemorrhage 10 hours after the ligation in rabbits and 8 days after the procedure in dogs. Several months after pulmonary artery occlusion, all of the lungs appeared normal. Lindskog and Gilman (2) ligated the pulmonary artery to the right lower lobe in 10 dogs and subsequently found no atelectasis. However, they sacrificed their animals from 15 - 256 days after ligation and had no information on the condition of the lungs during the first two post-operative weeks. Liebow and associates (3) ligated the left pulmonary artery in 11 dogs. They examined the lungs from these dogs 9 weeks to 25.6 months later and found no difference in the sizes of the left and right alveoli. Davis and co-workers (4) found hemorrhagic infarctions of the lungs in monkeys which were sacrificed 50 hours after pulmonary arterial occlusion. Catena and associates (5) noted that pulmonary edema and atelectasis had occurred in dogs sacrificed 7 days after pulmonary artery occlusion. However, the alveolar structure was normal in animals sacrificed several weeks after the surgical procedure.

The anatomical changes of unilateral pulmonary artery ligation consists of focal atelectasis, bronchial degeneration, and hemorrhage several days after ligation with an estimated reduction

of the resting lung volume by 1/3 to 1/2. As bronchial collateral circulation increases, these changes regress; within several months the lungs are grossly normal. Bloomer and colleagues (6) measured the collateral blood flow following ligation of the left pulmonary artery in dogs. In their studies the collateral blood flow averaged 300 ml./minute/sq. meter body surface area one month after pulmonary artery ligation and 450 ml. after six months.

Van Allan et al (7) suggested that atelectasis was secondary to obstruction of bronchioles due to degeneration and desquamation of the lining epithelium. Clements et al (8) demonstrated that stability of alveoli is dependent upon a highly surface-active alveolar lining layer, the pulmonary surfactant. This substance is produced by the lung tissue and its integrity is presumably dependent upon an adequate pulmonary capillary blood flow (9, 10). Therefore a reduction or cessation in pulmonary artery blood flow could cause an alteration in lung cellular function affecting production of surfactant which could lead to atelectasis. Thus it could be postulated that atelectasis observed following pulmonary artery ligation has been the result of a deficiency of surfactant as well as secondary to obstruction of airways.

In recent years a number of investigators have carried out studies in order to find a relationship between atelectasis following unilateral pulmonary artery ligation and a deficiency in surface activity of the lung lining material. Finley et al (11) measured minimal surface tension of dog lung extracts in chronic pulmonary

artery ligation and found elevated values 15 hours after ligation which returned to normal after six months. Giammona et al (12) showed that 4 hours after ligation of one pulmonary artery, the surface tension measured on the lung extracts and the deflation pressure-volume curves were unaltered. However, two weeks after ligation the lung had a smaller air volume at each pressure during deflation and its extracts had a higher minimal surface tension. Their data suggests a close relationship between altered surface forces and the pressure-volume characteristics of the lung after pulmonary artery ligation. Chernick et al (13) carried out similar studies on 17 dogs for a period of 2 - 98 days. They showed that the total lung volume was reduced by 40 - 50% of control lungs for the first 35 days following ligation. Lobes ligated from 2 - 14 days had a significant decrease in proportional volume deflation, indicating increased retractive forces of inflatable alveoli. Minimal surface tension was significantly elevated for this group. By 25 - 35 days, both minimum surface tension and increased retractive forces had returned to normal values. The decrease in total lung volume was converted to normal after 50 days of pulmonary artery ligation. Edmunds et al (14) in their extensive studies, have demonstrated that unilateral pulmonary artery occlusion reduces inflatable lung volume, causes an increase in lung weight and produces focal areas of hemorrhagic atelectasis which cannot be inflated by either gas or saline. In the hemorrhagic areas alveoli are filled with red cells and alveolar septa are thickened and infiltrated with macrophages, lymphocytes, plasma cells, and a few

polymorphonuclear cells. Other areas of lung remain entirely normal and show no ultrastructural alterations. These areas contain surface-active alveolar lining material and show no change in pulmonary mechanical relationships. These areas are unchanged biochemically from opposite non-ligated lung but the hemorrhagic atelectatic areas of the ligated lung show a decrease in phospholipid content, dipalmitoyl phosphatidyl choline, and alterations in esterified fatty acid composition and enzyme activity. Their data indicates that pulmonary arterial occlusion does not cause a generalized increase in alveolar surface forces, but that focal rather than general changes occur in lung following pulmonary artery ligation.

Alteration in surface activity of lung lining material in conjunction with the changes following pulmonary arterial occlusion has been demonstrated. It is not yet shown whether the deficiency in the activity of surfactant is primary or secondary to the structural changes in the lung such as hemorrhage or edema.

Howatt et al (15) have shown that occlusion of the left pulmonary artery, for 2 - 7 hours, in the fetal lambs affected the surface properties of the upper part of the lung at 124 - 128 days gestation (when the alveolar surface properties are undergoing active development), but had no effect on more developed lung at 135 days gestation. They have suggested that the actively developing lung is particularly susceptible to the effects of diminished blood flow. This difference could have been a result of different experimental conditions designed

for the two groups of lambs under study. The smaller lambs were not ventilated but the larger lambs were ventilated with room air. However, in a similar study, Adams et al (16) demonstrated that unilateral pulmonary arterial occlusion had no effect on surface activity and phospholipid content in the non-ventilated fetal lambs at gestational age of 128 - 140 days.

C. Cardio-pulmonary Bypass

One of the most frequent pulmonary complications following open-heart surgery and circulatory bypass procedures is pulmonary congestion and edema, which could be a potentially lethal event in the post-operative period (17, 18, 19, 20). In the most severe form of "pulmonary congestion syndrome" the patient usually leaves the operating room, following intracardiac surgery in reasonably good condition, but, within the first few hours he develops fever, increasing cyanosis, dyspnea and hypotension. Physical examination may reveal increasing signs of pulmonary edema, and radiologic examination may reveal diffuse clouding of the pulmonary fields.

Extra-corporeal circulation produces changes in lung function. Studies on human subjects and experimental dogs have shown that carbon monoxide and oxygen diffusion are diminished, alveolar-arterial gradients for oxygen is increased and an average right to left shunt of 24% of the cardiac output is noted when breathing air (21, 22). Gas volumes in all functional compartments except residual volume are reduced and lung compliance is decreased (23, 24). If death occurs it is with the symptoms of acute pulmonary failure and this usually occurs within the first two days. Post-mortem studies show that the lungs are dark red and congested, with focal zones of collapse and parenchymal hemorrhages. Microscopically there are usually many small zones of hemorrhage with blood cells and clear proteinacious

edema fluid filling the alveoli and bronchi, in association with focal collapse, pulmonary edema and engorgement of small blood vessels.

There are so many events during and after the operation which could predispose to pulmonary hemorrhage that it is difficult to assign primary significance to any one. Littlefield et al (25) have concluded from their experiments on dogs that the pulmonary hemorrhages and congestion seem to be caused by a positive surge of pulmonary capillary pressure during or at the end of perfusion. This damaging transient or prolonged increase in pulmonary capillary pressure was the result of overfilling of the left atrium. Baer et al (20) and Kottmeier et al (26) have shown that "pulmonary congestion syndrome" does develop in experiments where the left auricle is drained, concluding that the left auricular distension and pulmonary back flow could not be considered to be responsible for the pulmonary changes. No correlation between perfusion volume, blood flow, arterial blood pressure, oxygen saturation, and pathologic changes was found. However, they (26) were able to find embolic material grossly and microscopically in the lungs of several animals sacrificed within the first 24 hours. Material found in the oxygenator resembled that which was found in the lung. Hepps et al (27) showed that the addition of low molecular weight dextran to the blood and hemodilution perfusion where the hematocrit was reduced to less than 15% lessened the adverse effects of cardio-pulmonary bypass upon the lung, as evidenced by lowered minimal surface tension and improved microscopic

appearance. Many advantages have been ascribed to low molecular weight dextran, including the prevention of intravascular aggregation and sludging. It has been suggested by Bernstein and associates (28) that the reduction in red blood cell aggregation by the addition of low molecular weight dextran is due to an increase in red blood cell electronegativity which produces repulsion between red blood cells and therefore prevents aggregation. It is possible then that micro-embolization of the pulmonary vascular bed occurs during cardio-pulmonary bypass and leads to an ischemic injury, resulting in the release of circulating inhibitors of the surfactant. Gardener et al (29) measured the surface tension of lung extracts prepared from lungs of patients or experimental dogs undergoing cardio-pulmonary bypass for a period of between 36 - 96 minutes. The minimum surface tension of these extracts were much higher than the control values. They also demonstrated that lung extracts mixed with blood, which had been re-circulated in the bubble oxygenator from 4 - 12 hours, had a high minimum surface tension. When the same extracts were mixed with un-perfused blood, minimal surface tensions were within normal limits. They have suggested that the denaturation of the plasma proteins in the oxygenators, where the blood was in direct contact with air or oxygen (30), could be a factor in the alteration of surface-active material. However, Mandelbaum et al (31) have failed to show an inhibitory substance of pulmonary surfactant in the pump oxygenator blood. In their study blood inhibited pulmonary surface activity of normal lung extracts before and after circulation through the disc oxygenator.

Although the etiology of lung pathology following cardiopulmonary bypass is not clear, the changes have been shown to be associated with alterations in the surface forces of the lung. To what extent alterations in the surfactant are primary or secondary to edema and hemorrhage is not clear.

D. Experimental Atelectasis

Avery and Mead in their study of lungs of infants with hyaline membrane disease, have considered the possibility that atelectasis alone could be deleterious to the surfactant (32). Since then a number of investigators have studied the effect of induced atelectasis in experimental animals and pathologic atelectasis in human subjects on the surface forces of the lungs. Avery and Chernick (33) found that after 24 hours atelectasis induced by a large pneumothorax in rabbits, some diminution in surfactant was present. They postulated that the occasional recollapse of lungs following re-expansion after prolonged atelectasis (34) could be on the basis of the loss of surfactant. Finley et al (11) studied the effect of absorption atelectasis (following bronchial ligation) in dogs on surface forces of the lungs. They found an increase in minimum surface tension after 48 hours of bronchial ligation which returned to normal by 50 days. They postulated that airlessness per se was not detrimental since surface tension was normal in extracts prepared from lungs that were still collapsed 8 and 10 weeks after ligation of the bronchus; and that the increase in surface tension following bronchial ligation was most probably secondary to a loss of pulmonary blood flow. Their conclusion is based on the alterations of surface forces observed following pulmonary artery ligation and the fact that following bronchial ligation blood flow to the lung is reduced proportionately to the degree

of pulmonary collapse (35, 36, 37, 38). The blood flow through an atelectatic area is continuously decreasing during the first month, until practically no blood is passing through the non-ventilated part of the lung. According to Peters and associates (37), 10 - 15% of the cardiac output returns to the lung 1 - 2 months after ligation of the bronchus. On the basis of this observation, Finley et al (11) have suggested that the return to normal surface tension 8 - 10 weeks after bronchial ligation in their experiments may be related to the return of blood flow through alveolar capillaries. Sutnick and Soloff (39) demonstrated a decrease in surface activity of the lungs as evidenced by an increase in minimum surface tension as early as 45 - 90 minutes following ligation of the bronchus in dogs. They further demonstrated that the inflated portions of the atelectatic lungs had normal surface activity as compared to that of the controls.

Up to this time studies dealing with the effect of atelectasis upon the surface active properties of saline lung extracts were not in agreement. Using different extraction techniques, the investigators had reported early (39), time related (33, 11), or no (40) alteration in extract activity in experimental atelectasis. Levine and Johnson (41) studied the effect of the extraction method on apparent activity of lung extract using three extraction techniques - chopping, mincing, and pestle homogenization. They noted that all extraction methods gave highly active extracts when aerated lungs were used; but extracts from airless lungs prepared by chopping or mincing, invariably showed higher minimum surface tensions, lower stability indices and took

more time to reach minimum tension than did extracts of the same lungs when aerated. However, lung extracts made by homogenization from airless lungs showed normal surface activity. The authors indicated that the differences could be due to a decreased area of saline-alveolar contact during the extraction and suggested that the lungs should be inflated prior to saline extraction for measurements of surface activity. Yeh et al (42) confirmed the findings of Levine and Johnson. They showed that the collapsed lungs from 1 - 194 days did show higher surface tensions when studied in the collapsed state, but if they were re-inflated immediately before extraction of the surfactant, no abnormalities were found. This indicates that atelectasis per se is not deleterious to the surfactant.

Levine and Johnson (43) extended their experiments on atelectatic lungs to include a study of the pressure-volume characteristics of the lungs. They produced left lung collapse by pneumothorax in rabbits for a period of 90 minutes to 8 days. The atelectatic lungs showed a progressive decrease in "inflatability" with duration of collapse. An increase in airway pressure at any inflation volume as well as a decrease in total volume at maximum inflation pressure were seen. The alteration in opening pressure with duration of atelectasis was also seen during saline inflation of atelectatic lungs as compared to the controls. However, the deflation portion of the air pressure-volume diagram when compared on the basis of percent were not different from those of the controls. Since a highly surface active material was

demonstrated in the collapsed lung from the tracheal foam, the alterations in the saline and air inflation pressure-volume curves were considered to be due to altered tissue characteristics as demonstrated previously (38, 44) by histologic studies of the lung after collapse. These changes begin in the elastic fibers 30 minutes after bronchial ligation, and consist of alteration in position and form of the fibers, and, to a lesser degree, in disturbance of their structure (thickening, swelling). The changes are progressive and by the 5th. to 9th. months thin, tightly stretched elastic fibers arranged in bundles are found in the bronchi, the visceral pleura and the intra-alveolar septa. These changes could be secondary to the reduced pulmonary arterial blood flow which accompanies atelectasis. In spite of these structural changes, the experimental observations reveal that the lung easily and completely re-expands when bronchial ligation is released and the endobronchial mucus is aspirated (45). Furthermore, the lung does not seem to be functionally handicapped, since the blood that flows through gets normally oxygenated after re-expansion of the lung.

E. Oxygen Toxicity

Shortly after the discovery of oxygen (Priestley 1775), Lavoisier (46) (1785) described pulmonary damage and right heart failure in guinea pigs which had died after prolonged exposure to an oxygen rich environment. In 1878, Bert (47) described that a high oxygen pressure can kill all forms of living beings. The lung pathology resulting from high oxygen concentrations was first described by Smith in 1899 (48). Since then many conflicting reports have appeared regarding the importance of and mechanisms of oxygen toxicity in man and experimental animals. There are several reviews on the subject of oxygen poisoning by Stadie et al (49), Bean (50), Ohlsson (51) and Dickens (52).

The lung is the first organ in higher vertebrates which responds to oxygen poisoning. When the ambient pressure is maintained low enough so that the alveolar PO_2 is not greater than normal, inspiration of 100% oxygen produces no toxicity. The toxicity is intensified rather rapidly with increasing oxygen pressure even at ordinary atmospheric pressure. The type of damage also depends on the partial pressure prevailing. At a PO_2 around one atmosphere, the respiratory system appears to react first, while at pressures above one atmosphere the primary disturbances are noted in the central nervous system. The pulmonary lesion in animals after exposure to pure oxygen for 3 - 4 days is atelectasis, edema, hemorrhage and

occasionally hyaline membrane formation. The lung is heavy, large and dark in color, usually described as "liver-like".

The interest on the effect of oxygen at high pressure on the activity of surfactant was aroused partly because of the following observations: 1) The similarity of the histological findings of oxygen toxicity and hyaline membrane disease (53). 2) The fact that hyaline membrane disease (respiratory distress syndrome) was reported to be associated with alterations in surfactant (32). 3) The statement by Pattle (40) that increased alveolar surface tension could cause exudation of fluid into the alveoli. 4) Finally, because of the therapeutic use of high concentrations of oxygen in respiratory distress syndrome. Therefore, a number of investigators attempted to relate the pathological changes observed in the lung following prolonged inspiration of oxygen under high pressure to possible alterations in the surface forces of the lung. Rabbits, cats, dogs, rats, mice and guinea pigs were exposed to high concentrations of oxygen at one atmosphere or more for different periods. The state of lung lining material was then assessed by means of the surface tension balance or by chemical analysis. The results reported by different investigators, using the same or different species of animals, are conflicting. Pattle and Burgess (54), Fujiwara et al (55) and Giammona et al (56) found no alterations in the activity of the lung lining material in mice, guinea pigs and rats after exposure to oxygen at one atmosphere. Other investigators (57, 58, 59, 60, 61, 62) have demonstrated a reduction in the activity of

surfactant in dogs, rabbits, guinea pigs, rats and cats following oxygen intoxication. These findings do not prove nor disprove the fact that alterations in surfactant are primary to the effect of high pressure oxygen or secondary to pulmonary edema and hemorrhage, since surface-activity of the lung lining material was measured when pathological changes including edema and hemorrhage were already present in most of the animals.

It has been shown that the high activity of the crude powder of surfactant is maintained for several days when exposed to oxygen (63). Faridy et al (64) have shown that the pressure-volume characteristics of excised lobes exposed to pure oxygen for 3 hours are unaltered. The histological examination, pressure-volume characteristics and surface-activity of lung extracts of dogs exposed to oxygen for 48 hours were found to be normal (65). Morgan et al (61) were able to demonstrate an increase in minimal surface tension of lung extracts of dogs exposed to oxygen for periods of 44 - 52 hours only in those which had developed pulmonary edema. Fujiwara et al (55) who measured the lipid composition and the surface tension of lungs of guinea pigs treated with oxygen, found them not to be different from the control animals. These findings suggest that the lung lining material is not inactivated by direct action of pure oxygen but rather that its inactivation is secondary to pulmonary edema, hemorrhage and inhibitors of surfactant which come to the alveolar surface.

Electron microscopic examinations of the lung substantiate

this possibility (66, 67, 68, 69). By morphometric methods and by electron microscopy, Kistler et al (68) elucidated the nature and time course of damage occurring in the lungs of rats breathing pure oxygen at atmospheric pressure. The first changes were detected on the second day, consisting of edematous imbibition of the interstitial space. Since the fine structure of endothelial cells was unaltered, it was assumed that the edema formation was due to an increase permeability of the capillary lining. During the third day a structural damage to endothelial cells was noted with a decrease in capillary volume and capillary surface area. Later on a profuse exudation of a plasma-like fluid containing fibrin and numerous free cells oblitrated up to two-thirds of the alveoli. This was accompanied by remarkably little damage to alveolar epithelium. In contrast to the massive destruction of capillary endothelium, the vast majority of epithelial cells remained normal. The authors believe the primary injury resulting from breathing pure oxygen is localized in endothelial cells of alveolar capillaries and the other changes are its consequence. The question why epithelial cells, which are directly exposed to the high oxygen pressure, are not damaged, whereas endothelial cells undergo such early and drastic changes, is not yet understood.

F. CO_2 Poisoning

One of the techniques by which investigators have been able to produce hyaline-like membranes in experimental animals (rats, guinea pigs) is the exposure of the animal to high concentrations of CO_2 for periods of a few hours to a few days (70, 71). This pathological condition was also shown to be associated with alterations in surface activity of lung lining material (72). Guinea pigs exposed to 1.5% CO_2 in 21% oxygen for periods of from one day to six months showed a 50% incidence of atelectasis with no other pathological findings in their lungs (73). As the concentration of CO_2 in inspired air was increased to 3 and 15%, the pathological changes observed in the lungs of animals (hyaline membranes, atelectasis, perivascular edema, and alveolar edema) were increased proportionately. Kloos and Malrony (71) observed that guinea pigs exposed to 14 - 17% CO_2 in 21% oxygen for 8 - 22 hours developed pulmonary hyaline membranes while guinea pigs with chronic metabolic acidosis produced by the ingestion of ammonium chloride for periods of 3 - 24 days did not develop lung pathology and hyaline membranes. On the basis of these findings they concluded that hyaline membrane formation was caused specifically by CO_2 rather than by acidosis.

Neimoeller and Schaefer (73) and Schaefer et al (72) in their elegant studies, demonstrated four phases in the pulmonary pathology caused by exposure to 15% CO_2: 1) Up to 6 hours there is severe respiratory acidosis associated with pulmonary effusion and

an increase in lung weight and a decrease in the lamellar bodies of the granular pneumocytes. The surface tension is not affected by these early changes, nor is there any evidence of hyaline membranes during this period. 2) From 6 - 24 hours, hyaline membranes begin to form and are mainly of inspissated proteinacious material containing cell debris and occasional strand of fiber. All normal lamellar bodies disappear and there is an elevation in the minimum surface tension. 3) From the second to the seventh day the respiratory acidosis gradually becomes compensated, lung weight gradually decreases to normal values, surface tension measurements return to normal and there is an increased number of normal lamellar bodies. The pulmonary edema is absorbed and the hyaline membrane disappears. 4) In the final phase the pH is compensated although the PCO_2 remains elevated, and a normal cytological pattern is re-established.

Since alterations in the activity of lung lining material was associated with a decrease in the number of normal lamellar bodies, the authors have concluded that the granular pneumocytes have lost their ability of producing surfactant as a result of CO_2 in the inspired gas. They also believe that the pathological changes are secondary to a change in hydrogen ion concentration rather than a direct effect of CO_2. Their conclusion is based on the recovery of the pathological findings when the respiratory acidosis is compensated. One can argue that had they prevented the compensation of respiratory acidosis from the effects of high concentrations of CO_2

in inspired air, it is possible that the animals would still have recovered.

G. Asphyxia

One of the factors commonly associated with the respiratory distress syndrome of newborn is intrauterine asphyxia and complications of pregnancy leading to fetal hypoxemia (74, 75). This statistical observation is confirmed by James who found severe degrees of hypoxia, acidosis, and hypercapnia in the cord blood of prematures who went on to have respiratory distress, compared with non-distressed prematures (76).

A number of investigators have designed experiments to assess the influence of pre-natal asphyxia on the respiratory adaptation at birth of newborn in experimental animals and in particular, to observe whether changes similar to those of the respiratory distress syndrome could be demonstrated. Dawes et al (77, 78, 79) and Adamson et al (80) were able to induce experimental respiratory distress in the fetal lamb and fetal monkey following a period of up to 15 minutes asphyxia by clamping the umbilical cord and by preventing the fetus from breathing air.

The next step has been an attempt to demonstrate an alteration in surface activity of fetal lungs following a period of asphyxia as a cause of induced experimental respiratory distress in the newborn animal. Reynolds et al (81) studied the effect of pre-natal asphyxia on 12 lambs with gestational ages ranging from 131-143 days (full term is approximately 147 - 150 days). Asphyxia was produced by ventilating the ewe with low concentrations of oxygen and decreasing the minute

volume. After 1 1/2 to 4 hours of asphyxia, the umbilical cord was ligated and respiration established. After 1 - 5 1/2 hours of breathing, the lambs were sacrificed and the surface tension of lung extracts was measured. Four of six lambs (gestational age 130 - 136 days) subjected to severe pre-natal asphyxia developed respiratory distress with some hyaline membrane formation and in three, abnormal surface tensions were demonstrated. Only one of six lambs above 136 days gestation showed signs of respiratory distress following prolonged pre-natal asphyxia. Though the numbers of animals studied in their experiments were small, they concluded that the influence of pre-maturity for respiratory distress is more important than that of asphyxia. In the experiment where the newborn animals (guinea pigs, rabbits, rats and mice) were exposed to 3-4% inspired oxygen for periods of 15 - 25 hours, results indicated that hypoxia could not be a predisposing factor for the formation of hyaline membrane disease in term animals (82). The lungs of these animals were histologically normal without any evidence of atelectasis or of hyaline membrane formation. Adams et al (16) studied the effect of acute maternal hypoxemia, hypercapnia, and acidosis on the surfactant of fetal lamb lung (gestational age 114 - 140 days). They found no difference in surface activity of lung extracts from these lambs subjected to different experimental conditions in comparison to the controls. In addition the lipid composition of lung saline extracts were comparable to those of the controls. However, their findings cannot rule out hypoxemia, hypercapnia and acidosis as predisposing factors of

respiratory distress, since they did not allow the lambs to breath. Hyaline membranes are not demonstrable in the lungs of stillborns and respiratory distress develops mostly a few hours after the newborn has started breathing. Is it possible that the failure to allow the fetuses to breath (16) have masked changes that would have occurred following respiration? There is some evidence that surfactant is depleted by ventilation and that if the metabolic activity of the lung tissue is reduced, surfactant is not replenished (64, 83).

The pulmonary vascular bed in fetal and neonatal lambs is capable of strong vasoconstriction particularly in response to asphyxia (84, 85). Since the fetal pulmonary arterial flow is about 10% of the cardiac output and the fetal lung consumes about 8% of the oxygen supplied to the fetus (86), shunting of blood flow away from the lung tissue, induced by fetal asphyxia, may reduce the supply of oxygen and metabolites to levels that result in damage to the alveolar cells which are not fully developed. There is evidence that alveolar epithelial cells and alveolar surface-active material are not fully developed until late in gestation (87, 88, 89, 90).

H. Mechanical Stress

Brown et al (91) have shown that the surface film from a lung extract can be reversibly compressed to 50% of its initial area. After further compression, the film apparently ruptures on re-expansion. On expansion of the surface after film collapse, the insoluble portions of the condensed film may persist as islands, and if there is excess surfactant in the hypophase, more molecules come to the new surface (92). On the basis of studies of the kinetics of surface films, Clements et al (93) stated that long-term stability of the lungs requires periodic replenishment of surfactant.

Faridy et al (64) attempted to alter the surface forces of the lung by ventilation, that is, by the expansion and compression of the alveolar surface area. They found that following ventilation the excised lobes of lungs of dogs retained less air volume at a given transpulmonary pressure. This correlated with an increase in the minimum surface tension of the lung extract. When ventilation was stopped and the lobe was kept at a constant volume for several hours, the effects of ventilation were reversed, if the temperature of the environment was appropriate and the lobe was not deprived of oxygen at any stage of the experiment. This dependability of the recovery phase on the temperature of the environment and the concentration of oxygen led to the following hypothesis; that ventilation depletes the surfactant present at the air fluid interface of the alveoli. If however, the alveolar lining cells are functioning normally, new

surfactant is formed and replaces that which is depleted by ventilation.

MacLennahan and Urtnowski (83) after carrying out similar studies came to the same conclusion. In addition they showed that when rat lungs were ventilated at 37°C no alterations were found in the surface activity of the lung lining material, and that lungs perfused with potassium cyanide showed no signs of recovery from the effects of ventilation even though these were incubated at 37°C.

Another method of altering the activity of surfactant is over-inflation of the lung. Greenfield et al studied the effects of over-inflation on dog lungs using a respirator (94). After 2 - 6 hours of cyclic inflation and deflation at a rate of 20 per minute with a peak pressure of 26 - 32 cm. H_2O, no immediate changes were noted. However, 23 hours later, atelectasis associated with high surface tension was present in the over-inflated lungs, which recovered by 48 hours. No alteration in surface tension of lung extracts was observed in dogs ventilated with normal volume and pressure for periods up to 6 hours. Schulz (95) showed in the electron microscopic studies of over-inflated lungs of dogs, granular cytoplasmic degeneration and disruption of normal architecture. The capillary endothelium was the first structure within the blood-air-pathway to be destroyed, followed by the epithelium. It is possible that inactivation of surfactant is secondary to disruption of the capillaries, pulmonary edema and hemorrhage which in fact has been observed by Greenfield et al (94) in some of their experimental animals whose lungs were over-inflated.

I. Summary

During the past decade, interest has been focused on factors affecting the activity of the lung lining material. The effects of pulmonary artery ligation, cardio-pulmonary bypass, high pressures of oxygen, high concentrations of CO_2 in the inspired gas, asphyxia, atelectasis and over-inflation on the mechanical properties of the lung are reviewed from the literature. All of these conditions, except for experimental atelectasis, were found to be associated with a decrease in the activity of surfactant. However, common pathological findings under these circumstances are pulmonary edema and hemorrhage which are known to be inhibitors of surfactant. Since the measurements of surface activity of the lung lining material were made at the final stage of the lung pathology, it remains to be elucidated whether alterations of surface forces are primary or secondary to edema and hemorrhage. Electron microscopic studies of lungs exposed to high pressures of oxygen have presented some suggestive evidence that the inactivation of surfactant is secondary to inhibitors from plasma exudate. In CO_2 poisoning it is not known whether CO_2 exerts its effect directly or indirectly by changing the hydrogen ion concentration.

J. Bibliography

1. Schlaepfer, K.: Ligation of the pulmonary artery of one lung with and without resection of the phrenic nerve. Arch. Surg. 9: 25-94, 1924.

2. Lindskog, G.E. and Gilman, A.: Effect of pulmonary artery ligation on the histamine content of lung, with observations on concomitant structural changes. Am. J. Physiol. 152: 417-422, 1948.

3. Liebow, A.A., Hales, M.R., Bloomer, W.E., Harrison, W. and Lindskog, G.E.: Studies on the lung after ligation of the pulmonary artery. Am. J. Pathol. 26: 177-195, 1950.

4. Davis, H.A., Gordon, W.B., Hayes, E.W. Jr. and Wasley, M.T.: Effects upon lung of varying periods of temporary occlusion of pulmonary artery. A.M.A. Arch. Surg. 64: 464-474, 1952.

5. Catena, E., D'Alfonso, G., Gioretti, G.P. and Fiorelli, E.: The lung after ligation of the pulmonary artery (anatomohistological and anatomo-angiographic studies). Angio Tisiol. 13: 861, 1958.

6. Bloomer, W.E., Harrison, W., Lindskog, G.E. and Liebow, A.A.: Respiratory function and blood flow in the bronchial artery after ligation of the pulmonary artery. Am. J. Physiol. 157: 317-328, 1949.

7. Van Allen, C.M., Nocoll, G.L. and Tuttle, W.M.: Lung changes after occlusion of pulmonary artery branches by embolus and by ligature. Yale J. Biol. Med. 2: 363-389, 1929-1930.

8. Clements, J.A., Hustead, R.F., Johnson, R.P. and Gribetz, I.: Pulmonary surface tension and alveolar stability. J. Appl. Physiol. 16: 444-450, 1961.

9. Bozic, C.: Pulmonary hyaline membranes and vascular anomalies of the lung. Pediatrics 32: 1094-1097, 1963.

10. Chu, J., Clements, J.A., Cotton, E., Klaus, M.H., Sweet, A.Y., Thomas, M.A. and Tooley, W.H.: The pulmonary hypoperfusion syndrome. Pediatrics 35: 733-742, 1965.

11. Finley, T.N., Tooley, W.H., Swenson, E.W., Gardner, R.E. and Clements, J.A.: Pulmonary surface tension in experimental atelectasis. Am. Rev. Respirat. Diseases 89: 372-378, 1964.

12. Giammona, S.T., Mandelbaum, I., Foy, J. and Bondurant, S.: Effects of pulmonary artery ligation on pulmonary surfactant and pressure-volume characteristics of dog lung. Circulation Research 18: 683-691, 1966.

13. Chernick, V., Hodson, W.A. and Greenfield, L.J.: Effect of chronic pulmonary artery ligation on pulmonary mechanics and surfactant. J. Appl. Physiol. 21: 1315-1320, 1966.

14. Edmunds, L.H. Jr. and Huber, G.L.: Pulmonary artery occlusion. J. Appl. Physiol. 22: 990-1016, 1967.

15. Howatt, W.F., Avery, M.E., Humphrey, P.W., Normand, I.C.S., Reid, L. and Strang, L.B.: Factors affecting pulmonary surface properties in the foetal lamb. Clin. Sci. 29: 239-248, 1965.

16. Adams, F.H., Nozaki, M., Chida, N., El Salawy, A. and Norman, A.: Effects of hypoxemia, hypercarbia, acidosis and reduced pulmonary blood flow on the surfactant of fetal lamb lung. J. Pediat. 71: 396-403, 1967.

17. Dodrill, F.D.: The effects of total body perfusion upon the lungs. in Allen, J.G. Extracorporeal circulation. Springfield, Illinois. Charles C. Thomas. 1958, pp 327-335.

18. Kolff, W.J., Effler, D.B., Groves, L.K., Hughes, C.R. and McCormack, L.J.: Pulmonary complications of open-heart operations, their pathogenesis and avoidance. Cleveland Clin. Quart. 25: 65-83, 1958.

19. Muller, W.H., Littlefield, J.B. and Dammann, J.F.: Pulmonary parenchymal changes associated with cardiopulmonary by-pass. In Allen, J.G. Extracorporeal Circulation. Springfield, Illinois. Charles C. Thomas. 1958. pp 336-341.

20. Baer, D.M., Osborn, J.J.: The post perfusion pulmonary congestion syndrome. Am. J. Clin. Pathol. 34: 442-445, 1960.

21. McClenahan, J.B., Sykes, M.K. and Young, W.: Changes in pulmonary physiology following extracorporeal circulation. (abstract) Circulation 28: 767, 1963.

22. Schramel, R.J., Cameron, R., Ziskind, M.M., Adam, M. and Creech, O. Jr.: Studies of pulmonary diffusion after open heart surgery. J. Thoracic Cardiovascular Surg. 38: 281-291, 1959.

23. Howatt, W.F., Talner, N.S., Sloan, H. and DeMuth, G.R.: Pulmonary function changes following repair of heart lesions with the aid of extracorporeal circulation. J. Thoracic Cardiovascular Surg. 43: 649-657, 1962.

24. Shimizu, T. and Lewis, F.J.: An experimental study of pulmonary function following cardiopulmonary bypass. J. Thoracic Cardiovascular Surg. 52: 565-570, 1966.

25. Littlefield, J.B., Dammann, J.F., Ingram, P.R. and Muller, W.H.: Changes in pulmonary artery pressure during cardiopulmonary bypass. J. Thorac. Surg. 36: 604-614, 1958.

26. Kottmier, P.K., Adamsons, J., Stuckey, J.H., Newman, M.M. and Dennis, C.: Pathological changes after partial and total cardiopulmonary bypass in humans and animals. Surgical Forum 9: 184-189, 1958.

27. Hepps, S.A., Roe, B.B., Wright, R.R. and Gardner, R.E.: Amelioration of the pulmonary postperfusion syndrome with hemodilution and low molecular weight dextran. Surgery 54: 232-241, 1963.

28. Bernstein, E.F., Castaneda, A., Evans, R.L. and Varco, R.L.: Alterations in red blood cell charge with extracorporeal circulation. Surg. Forum 13: 193-195, 1962.

29. Gardner, R.E., Finley, T.N. and Tooley, W.H.: The effect of cardiopulmonary bypass on surfact activity of lung extracts. Bulletin de la Societe Internationale de Chirurgie 21: 542-551, 1962.

30. Clements, J.A.: Surface tension of lung extracts. Proc. Soc. Exp. Biol. (N.Y.) 95: 170-172, 1957.

31. Mandelbaum, I. and Giammona, S.T.: Extracorporeal circulation, pulmonary compliance, and pulmonary surfactant. J. Thorac. Cardiovascular Surg. 48: 881-889, 1964.

32. Avery, M.E. and Mead, J.: Surface properties in relation to atelectasis and hyaline membrane disease. A.M.A.J. Dis. Child. 97: 517-523, 1959.

33. Avery, M.E. and Chernick, V.: Alterations of the alveolar lining layer in living rabbits (abstract). J. Pediat. 63: 762, 1963.

34. Carlson, R.I., Classen, K.L., Grollan, F., Grobbel, W.G., Sherman, D.E. and Christensen, R.O.: Pulmonary edema following the rapid re-expansion of a totally collapsed lung due to a pneumothorax. A clinical and experimental study. Surg. Forum 9: 367-371, 1958.

35. Coryllos, P.M. and Birnbaum, G.L.: Circulation in compressed atelectatic pneumonic lung (pneumothorax-apneumatosis-pneumonia). Arch. Surg. 19: 1346-1423, 1929.

36. Björk, V.O. and Salen, E.F.: The blood flow through an atelectatic lung. J. Thorac. Surg. 20: 933-942, 1950.

37. Peters, R.M. and Roos, A.: The effects of atelectasis on pulmonary blood flow in the dog. J. Thoracic Surg. 24: 389-397, 1952.

38. Ahmed, F.S. and Harrison, C.V.: The effect of prolonged pulmonary collapse on the pulmonary arteries. J. Pathol. Bact. 85: 357-360, 1963.

39. Sutnick, A.I. and Soloff, L.A.: Surface tension reducing activity in the normal and atelectatic human lung. Am. J. Med. 35: 31-36, 1963.

40. Pattle, R.E.: Properties, function and origin of the alveolar lining layer. Proc. Roy. Soc. London. Ser. B. 148: 217-240, 1958.

41. Levine, B.E. and Johnson, R.P.: Surface activity of saline extracts from inflated and degassed normal lungs. J. Appl. Physiol. 19: 333-335, 1964.

42. Yah, T.J., Manning, H., Ellison, L.T. and Ellison, R.G.: Alveolar surfactant in chronic experimental atelectasis. Am. Review Resp. Dis. 93: 953-956, 1966.

43. Levine, B.E. and Johnson, R.P.: Effects of atelectasis on pulmonary surfactant and quasi-static lung mechanics. J. Appl. Physiol. 20: 859-864, 1965.

44. Neminushchaya, L.I.: Changes in the elastic tissue of the lung in experimental lobar atelectasis. Fed. Proc. 22: T657-T660, 1963.

45. Webb, W.R. and Burford, T.H.: Studies of the re-expanded lung after prolonged atelectasis. A.M.A. Arch. Surg. 66: 801-809, 1953.

46. Lavoisier, A.L.: L'alteration qu'éprouve l'air respiré (1785). Memoires sur la respiration et la transpiration des animaux (Gauthiers-Villans et cie Paris (1929).

47. Bert, P.: Pression barometrique (Masson, Paris 1878).

48. Smith, J.L.: The pathological effects due to increase of oxygen tension in the air breathed. J. Physiol. 24: 19-35, 1899.

49. Stadie, W.C., Riggs, B.C. and Haugaard, N.: Oxygen poisoning. Am. J. Med. Sci. 207: 84-114, 1944.

50. Bean, J.W.: Effects of oxygen at increased pressure. Physiol. Review 25: 1-147, 1945.

51. Ohlsson, W.T.L.: A study on oxygen toxicity at atmospheric pressure. Acta Med. Scand. Suppl. 190: 1-93, 1947.

52. Dickens, F.: The toxic effect of oxygen on nervous tissue. In Neurochemistry edited by K.A.C. Elliot, I.H. Page and J.H. Quastel. Springfield, Illinois. Thomas, 1962. pp 851-869.

53. Berfenstam, R., Edlund, T. and Zettergren, L.: The Hyaline Membrane Disease. Acta Paediat. 47: 82-100, 1958.

54. Pattle, R.E. and Burgess, F.: The lung lining film in some pathological conditions. J. Path. Bact. 82: 315-331, 1961.

55. Fujiwara, T., Adams, F.H. and Seto, K.: Lipids and surface tension of extracts of normal and oxygen-treated guinea pig lungs. J. Pediat. 65: 45-52, 1964.

56. Giammona, S.T., Kemer, D. and Bondurant, S.: Effect of oxygen breathing at atmospheric pressure on pulmonary surfactant. J.A.P. 20: 855-858, 1965.

57. Caldwell, P.R.B., Giammona, S.T., Lee, W.L. and Bondurant, S.: Effect of oxygen breathing at one atmosphere on pulmonary surfactant in dogs (abstract). Clin. Res. 11: 301, 1963.

58. Hackney, J.D., Collier, C.R. and Rounds, D.E.: Mechanism of pulmonary damage in oxygen poisoning. Proc. 2nd. International Pharmacological Meeting. Vol. 11. Drugs and Respiration. Macmillan Co. N.Y. 1964. pp 93-103.

59. Collier, C.R., Hackney, J.D. and Rounds, D.E.: Alterations of surfactant in oxygen poisoning. Dis. Chest 48: 233-238, 1965.

60. Webb, W.R., Lanius, J.W., Aslami, A. and Reynolds, R.C.: The effects of hyperbaric-oxygen tensions on pulmonary surfactant in guinea pigs and rats. J.A.M.A. 195: 279-280, 1966.

61. Morgan, T.E., Finley, T.N., Huber, G.L. and Fialkow, H.: Alterations in pulmonary surface active lipids during exposure to increased oxygen tension. J. Clin. Invest. 44: 1737-1744, 1965.

62. Kennedy, J.H.: Hyperbaric oxygenation and pulmonary damage. The effect of exposure at two atmospheres upon surface activity of lung extracts in the rat. Med. Thorac. 23: 27-35, 1966.

63. Clements, J.A.: Surface phenomena in relation to pulmonary function. Physiologist 5: 11-28, 1962.

64. Faridy, E.E., Permutt, S. and Riley, R.L.: Effect of ventilation on surface forces in excised dogs' lungs. J. Appl. Physiol. 21: 1453-1462, 1966.

65. Faridy, E.E. and Chernick, V.: (unpublished data)

66. Cedergren, B., Gyllensten, L. and Wersäll, J.: Pulmonary damage caused by oxygen poisoning. An electron microscopic study in mice. Acta Paediatrica 48: 477-494, 1959.

67. Schaffner, F., Felig, P. and Trachtenberg, E.: Structure of rat lung after protracted oxygen breathing. Arch. Path. 83: 99-107, 1967.

68. Kistler, G.S., Caldwell, P.R.B. and Weibel, E.R.: Development of fine structural damage to alveolar and capillary lining cells in oxygen-poisoned rat lungs. J. Cell Biol. 32: 605-628, 1967.

69. Bowden, D.H.: Personal communication.

70. De, T.D. and Anderson, G.W.: The experimental production of pulmonary hyaline-like membranes with atelectasis. Amer. J. Obst. & Gynec. 68: 1557-1567, 1954.

71. Kloos, K., Malorney, G. and Wolf, H.: Experimentelle pulmonale hyaline membranen. Verh. dtsch. Ges. Path. 180: 1957.

72. Schaefer, K.E., Avery, M.E. and Bensch, K.: Time course of changes in surface tension and morphology of alveolar epithelial cells in CO_2 induced hyaline membrane disease. J. Clin. Invest. 43: 2080-2093, 1964.

73. Niemoeller, H. and Schaefer, K.E.: Development of hyaline membranes and atelectasis in experimental chronic respiratory acidosis. Proc. Soc. Exp. Biol. Med. 110: 804-808, 1962.

74. Glystad, W., Landing, B.H. and Smith, C.A.: Pulmonary hyaline membranes in newborn infants. Pediatrics 8: 5-21, 1951.

75. Cohen, M.M., Weintraub, D.H. and Lilienfeld, A.M.: The relationship of pulmonary hyaline membrane to certain factors in pregnancy and delivery. Pediat. 26: 42-50, 1960.

76. James, L.S.: Physiology of respiration in newborn infants and in the respiratory distress syndrome. Pediat. 24: 1069-1101, 1959.

77. Dawes, G.S., Mott, J.C. and Stafford, A.: Prolongation of survival in the anoxic foetal lamb. J. Physiol. (London) 153: 16P, 1960.

78. Dawes, G.S., Mott, J.C., Shelley, H.J. and Stafford, A.: The prolongation of survival time in asphyxiated immature foetal lambs. J. Physiol. 168: 43-64, 1963.

79. Dawes, G.S., Jacobson, H.N., Mott, J.C., Shelley, H.J. and Stafford, A.: The treatment of asphyxiated, mature foetal lambs and Rhesus monkeys with intravenous glucose and sodium carbonate. J. Physiol. 169: 167-184, 1963.

80. Adamsons, K., Behrman, R., Dawes, G.S., James, L.S. and Koford, C.: Resuscitation by positive pressure ventilation and tris-hydroxymethylaminomethane of Rhesus monkeys asphyxiated at birth. J. Pediat. 65: 807-818, 1964.

81. Reynolds, E.O.R., Jacobson, H.N., Motoyama, E.K., Kikkawa, Y., Craig, J.M., Orzalesi, M.M. and Cook, C.D.: The effect of immaturity and prenatal asphyxia on the lungs and pulmonary function of newborn lambs: The experimental production of respiratory distress. Pediat. 35: 382-392, 1965.

82. Avery, G.R.: The effect of hypoxia on newborn animals, with reference to hyaline membrane disease. Pediat. 32: 801-807, 1963.

83. McClenahan, J.B. and Urtnowski, A.: Effect of ventilation on surfactant, and its turnover rate. J.A.P. 23: 215-220, 1967.

84. Dawes, G.S. and Mott, J.C.: The vascular tone of the foetal lung. J. Physiol. 164: 465-477, 1962.

85. Cook, C.D., Drinker, P.A., Jacobson, H.N., Levison, H. and Strang, L.B.: Control of pulmonary blood flow in the foetal and newly born lamb. J. Physiol. 169: 10-29, 1963.

86. Dawes, G.S., Mott, J.C. and Widdicombe, J.G.: The foetal circulation in the lamb. J. Physiol. (London) 126: 563-587, 1954.

87. Buckingham, S. and Avery, M.E.: Time of appearance of lung surfactant in the foetal mouse. Nature 193: 688-689, 1962.

88. Campiche, M., Jaccottet, M. and Juilland, E.: La Pneumonose a membranes hyalines. Ann. Paediat. 199: 74-88, 1962.

89. Pattle, R.E.: The formation of a lining film by foetal lung. J. Pathol. Bact. 82: 333-343, 1961.

90. Woodside, G.L. and Dalton, A.J.: The ultrastructure of lung tissue from newborn and embryo mice. J. Ultrastruct. Res. 2: 28-54, 1958.

91. Brown, E.S., Johnson, R.P. and Clements, J.A.: Pulmonary surface tension. J. Appl. Physiol. 14: 717-720, 1959.

92. Avery, M.E. and Said, S.: Surface phenomena in lungs in health and disease. Medicine 44: 503-526, 1965.

93. Clements, J.A., Tierney, D.F. and Trahan, H.J.: Influence of kinetic behaviour of surface films on pulmonary elasticity. Physiologist 6: 159, 1963.

94. Greenfield, L.J., Ebert, P.A. and Benson, D.W.: Effect of positive pressure ventilation on surface tension properties of lung extracts. Anesthesiol. 25: 312-316, 1964.

95. Schulz, H.: The submicroscopic anatomy and pathology of the lung. Berlin, Springer-Verlag, 1959, p. 128.

PART II. THE EFFECT OF ALTERATIONS OF PULMONARY ARTERIAL AND ALVEOLAR GAS TENSIONS ON THE PRESSURE-VOLUME CURVE AND SURFACE TENSION OF DOG LUNGS.

A. Introduction

Changes in the mechanical properties of the lung are known to occur where the lung is subjected to a variety of abnormal conditions including exposure to abnormal gas tensions (28, 29, 30), pulmonary artery occlusion (31, 32) and abnormal patterns of ventilation (7, 12). Since several of these conditions may co-exist it is often difficult to distinguish with certainty the effect of a specific factor.

The present experiments were undertaken to evaluate the effects of ventilation, blood flow, CO_2 tension, O_2 tension and hydrogen ion concentration singly and in combinations on mechanical and surface properties of the lung in vivo. A systematic study of these variables was done on the left lower lobe of living anesthetized dogs.

B. Methods

Eighty-three mongrel dogs of both sexes, free of respiratory disease, weighing between 9 and 21.5 Kg., were used for this study. The animals were anesthetized with 30 mg. of sodium pentobarbital/Kg. of body weight. Additional 20 mgm. doses were administered as necessary during the experiment. Tracheostomy was then performed and a metallic tube was placed into the trachea. The end of the tracheal tube was connected to a thick rubber tube, 3 inches long, which had an opening of 1/4 of an inch in diameter on one side. Through this opening a Tygon tube, bearing a plastic cannula at the tip, was passed half way into the trachea. The Tygon tube was then clamped and the thick rubber rube was connected to a Harvard Respiration pump. The dog was placed on the right side. The chest was opened on the left between the fifth and sixth ribs. The branch of the pulmonary artery to the left lower lobe and the left lower lobe bronchus were dissected. An umbilical tape was passed around each. The Tygon tube was then pushed further down into the trachea till the plastic cannula was felt to be in the left lower lobe bronchus. The umbilical tape was then firmly tied around the plastic cannula to prevent air leakage around the tube and also to cut the bronchial circulation to the left lower lobe. The chest incision was covered with a few layers of gauze wet with normal saline. At this point the clamp was removed from the Tygon tube and the tube was connected to a second respiration pump. Thus the right lung and the left lower lobe could be ventilated separately. The left upper lobe was not functioning since the Tygon

tube was obstructing its bronchus and throughout the experiments this lobe remained almost atelectatic. In some experiments the left lower lobe bronchus was firmly ligated so that it was not ventilated. When the experimental condition required, the pulmonary artery to the left lower lobe was also ligated.

Four different conditions were tested on the left lower lobe: 1) ventilation, with intact pulmonary arterial circulation, 2) ventilation with no blood circulation, 3) no ventilation with intact circulation and 4) no ventilation and no circulation.

The tidal volume, ventilation rate and gases used for ventilation of the right lung were changed appropriately to control the blood gas tensions of the pulmonary artery, that is, the blood supply to the left lower lobe. The left lower lobe was ventilated at a rate of 12/minute, with a tidal volume of 10 ml./Kg. of body weight. This is approximately equal to 30% of the maximum lobe air volume (MLV) at 40 cm H_2O pressure. The blood samples for gas analysis were directly drawn from the left pulmonary artery by needle aspiration at intervals of 20 minutes. The blood gas tensions and blood pH were measured on a Radiometer microelectrode system using CO_2, O_2 and pH electrodes.

At the end of four hours, the dog was exsanguinated. The left lower lobe was then excised and weighed.

Deflation Pressure-Volume Curve:

Following bronchial cannulation the lobe was degassed in a vacuum jar. The jar was evacuated until water vaporization was noted

to occur. The vacuum was then interrupted and upon recompression of the lobe to atmospheric pressure, the water vapor condensed leaving the lobes gas free. The cannulated degassed lobe was then attached to the T tube of a pressure-volume apparatus (Fig. 1) similar to that previously described by others (1). One limb of the T tube was connected to a water manometer and the other limb was connected to an air reservoir in series with a water filled burette. By raising and lowering the burette, water was displaced to and from the reservoir which in turn displaced air to and from the attached lobe at pressures simultaneously registered by the manometer. Changes in lobe volume were considered equal to changes in water volume in the burette after corrections were made for volume changes of the air within the apparatus.

The degassed lobe was inflated with air to 40 cm H_2O pressure. Inflation from the degassed state tends to be segmental, nonuniform, and irregular (2). Deflation volume changes are more uniform and following maximum inflation are representative of a maximum number of alveoli. For these reasons, the initial volume for maximum inflation and the subsequent deflation volumes were used to relate pressure-volume properties.

Maximum inflation pressure was maintained until the lobe volume remained constant for 10 seconds. The air volume observed at this transpulmonary pressure, considered as maximum lobe air volume (MLV), was designated as 100% and each volume subsequently observed

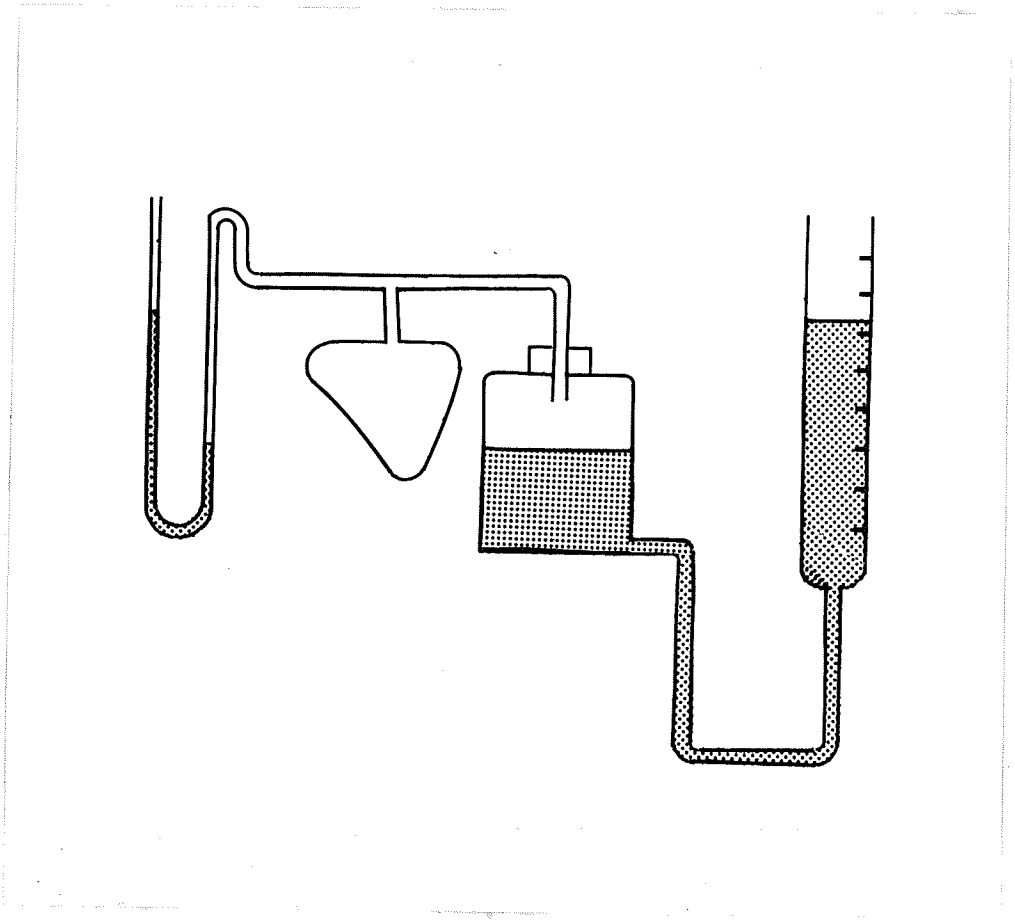

Figure 1. Apparatus for inflation and deflation of the lung.

following deflation to a predetermined transpulmonary pressure (30, 20, 15, 10, 5 and 0 cm. H_2O) was expressed as a percentage of the maximum lung air volume. During deflation the pressures were maintained to allow volumes to stabilize. If volumes did not stabilize at high pressures, air leaks were assumed to be present and such lungs were excluded from the study. The relative volume remaining at transpulmonary pressure of 10 cm H_2O ($V\%_{10}$) was used for comparing different experimental conditions.

Stability Ratio:

In some experiments, the activity of the lung lining material was measured by the technique of Pattle (3). A small piece of the inflated portion of the lung (inflated with air using a hypodermic needle) was squeezed into a drop of water hanging under a microscope slide. This slide was placed over another slide with a large hollow (hanging drop preparation). The bubbles were then watched through a microscope fitted with an eye piece graticule. Ten bubbles not smaller than 20 micra and not larger than 60 micra were chosen at random. A sketch was made giving the diameters of the bubbles. After 20 minutes their diameters were measured again. The ratio of the final to the original surface area of each bubble (the stability ratio) was then calculated. A total of 40 bubbles were examined from from each lobe, that is, 4 groups of 10 bubbles taken from four different areas of the lung tissue. The mean stability ratio of 40 bubbles was used as a measure of the state of the lining film.

Dry Weight:

Dry weights were obtained in some lobes through the following procedure. The lobe was fully inflated with air. Then the surface was punctured with a No. 25 hypodermic needle at numerous places. A continuous air flow through the bronchus into the lung and out from these punctured holes dried the lung within 24 - 48 hours. When the weight of the lobe remained constant for 12 hours the lobe was considered dry.

A t test of unpaired variates was used to compare the lung average volumes expressed as percentage of MLV of the different groups of lobes (4).

C. Results

Control:

In a previous study, the static deflation pressure-volume curves of 104 canine lower lobes (right and left) were determined within one hour after exsanguination (5) and were considered the control pressure-volume curves for the present study. In Figure 2 and Table I the mean and the standard deviation of these curves are indicated.

Pulmonary Artery Ligation:

A) Not Ventilated: In six dogs the pulmonary artery to the left lower lobe and the left lower lobe bronchus were ligated. After 4 hours, the pressure-volume characteristics of these lobes were within normal limits (Fig. 3).

B) Ventilated: In 8 dogs, following the ligation of the pulmonary artery to the left lower lobe, the left lower lobe was ventilated for 4 hours with a gas containing 5% oxygen in nitrogen (6 lobes) or with room air (2 lobes). The pressure-volume curve was depressed in all and their $V\%_{10}$ were significantly different ($p < .001$) from the controls (Fig. 3).

Hypoxia and Eucapnia:

C) Not Ventilated: In 5 dogs, the left lower lobe bronchus was ligated and the pulmonary circulation was left intact. The pulmonary arterial blood gas tensions were: $PO_2 < 42$ mm Hg, $PCO_2 < 44$ mm Hg and $pH > 7.30$.

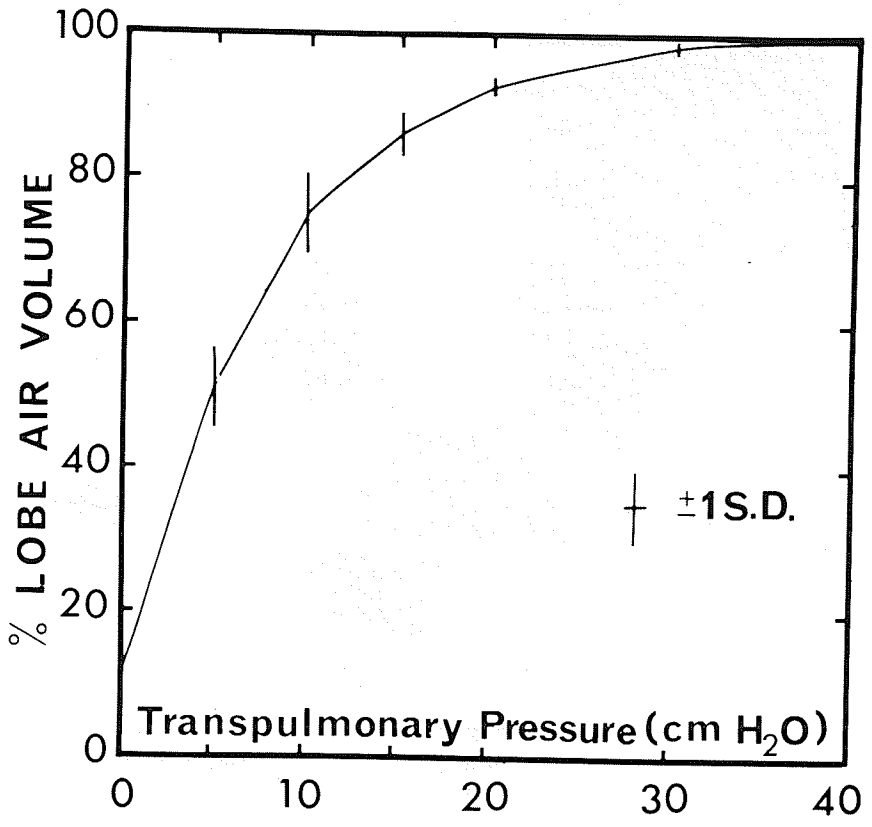

Figure 2. Mean deflation pressure-volume curve of 104 lower lobes of control dogs, expressed as percentage of lobe air volume at 40 cm H_2O transpulmonary pressure. The vertical lines represent one standard deviation to either side of the mean.

TABLE I

VOLUME OF 104 EXCISED LOWER (RIGHT AND LEFT) LOBES OF DOG LUNGS AT VARIOUS STATIC DEFLATING PRESSURES, EXPRESSED AS PERCENT LOBE AIR VOLUME AT 40 cm H_2O TRANSPULMONARY PRESSURE.

P_{TP}	MEAN	S.D.	S.E.
30	98.17	± 0.84	± .08
20	92.62	± 1.48	± 0.15
15	86.03	± 3.10	± 0.30
10	75.00	± 5.50	± 0.54
5	50.89	± 5.56	± 0.55
0	11.11	± 3.15	± 0.31

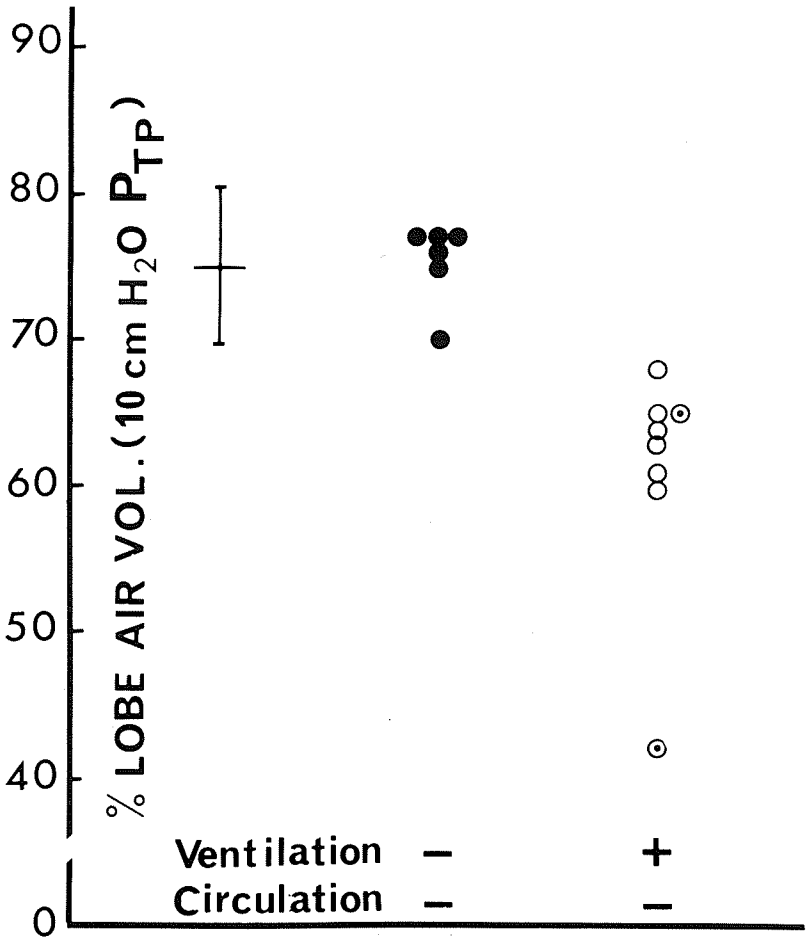

Figure 3. The effect of pulmonary artery ligation on $V\%_{10}$ of non-ventilated and ventilated lobes.

● = Lobes not ventilated (atelectatic).
○ = Lobes ventilated with 5% oxygen in nitrogen.
⊙ = Lobes ventilated with room air.

The $V\%_{10}$ of the controls is indicated with one standard deviation to either side of the mean for comparison.

The lobes became atelectatic within 1/2 to 1 hour after the bronchial ligation. The pressure-volume characteristics of these lobes were not different from those of the controls (Fig. 4).

D) Ventilated: In 11 dogs, the left lower lobes were ventilated with pure nitrogen while being perfused with pulmonary arterial blood containing $PO_2 < 48$ mm Hg, $PCO_2 < 46$ mm Hg and $pH > 7.30$. The pressure-volume curves of these lobes were also within the normal range (Fig. 4).

E) Reduced Pulmonary Blood Flow: In 4 dogs, the pulmonary blood flow to the left lower lobe was reduced to 15% of the control value, that is, a blood flow of about 50 - 70 ml./min. The pulmonary arterial blood gas tensions were kept at $PO_2 < 32$ mm Hg, $PCO_2 < 46$ mm Hg and $pH > 7.25$. Following 4 hours of ventilation with pure nitrogen the lobes retained their normal pressure-volume characteristics, as indicated in Figure 4.

Hypoxia and Hypercapnia:

F) Not Ventilated: In 5 dogs the left lower lobe bronchus was ligated and the pulmonary circulation to the lobe was kept intact. The pulmonary arterial blood gas tensions were: $PO_2 < 47$ mm Hg, $PCO_2 > 70$ mm Hg and $pH < 7.18$. Since lobes become atelectatic within 1/2 to 1 hour after bronchial ligation, the collapse was prevented in two dogs. During the entire experiment these lobes were kept constantly inflated by a slow and continuous flow of pure nitrogen into the lobe, via a T tube. One limb of the T tube was kept 4 to 5 cm. under water to prevent a rise in inflation pressure beyond 4 to 5

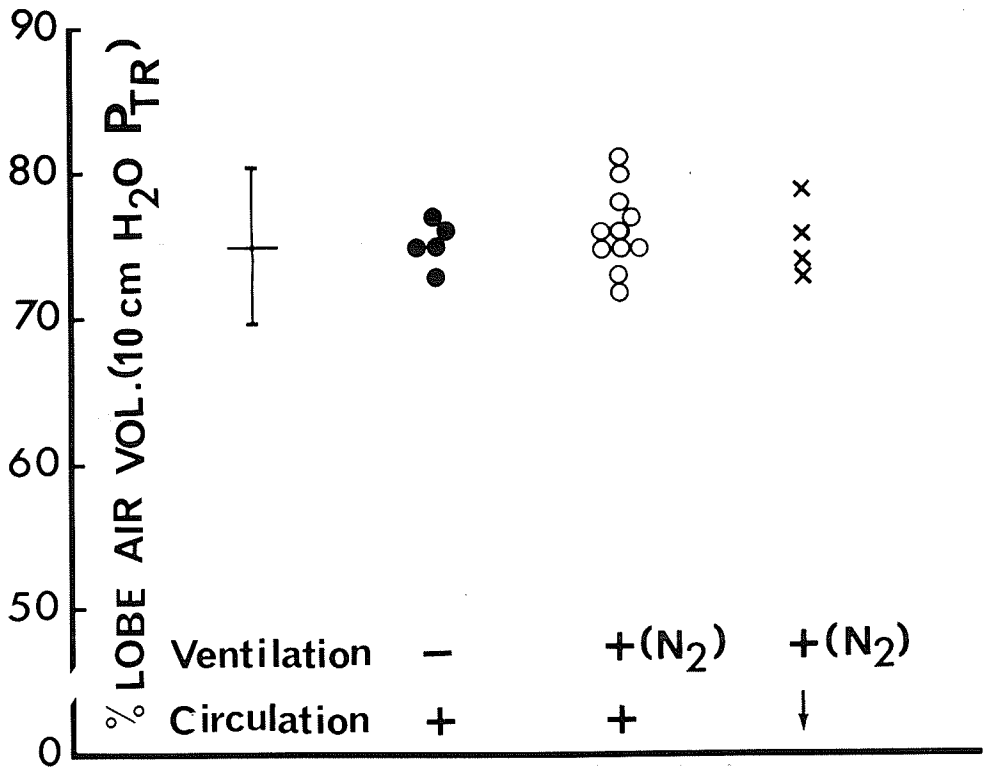

Figure 4. The effect of hypoxia and eucapnia on pressure-volume characteristics ($V\%_{10}$) of lobes. The lobes are perfused with blood containing a $PO_2 < 48$ mm Hg., $PCO_2 < 46$ mm Hg and pH > 7.25.

● = Lobes not ventilated (atelectatic).
O = Lobes ventilated with pure nitrogen.
× = Lobes ventilated with pure nitrogen, and blood flow reduced to 15% of the control values.

The $V\%_{10}$ of the controls is indicated with one standard deviation to either side of the mean for comparison.

cm. H_2O pressure. These lobes were also perfused with hypoxic, hypercapnic blood. At the end of 4 hours, their pressure-volume measurements also indicated a decrease in percent air volume remaining at different transpulmonary pressures. The $V\%_{10}$ of these lobes differed significantly ($p < .001$) from the controls (Fig. 5).

G) Ventilated: In 10 dogs, the left lower lobe was ventilated with pure nitrogen and perfused with hypoxic, hypercapnic blood. The pulmonary arterial blood gas tensions were: $PO_2 < 50$ mm Hg, $PCO_2 > 64$ mm Hg and pH < 7.21. Their percent lobe air volume at 10 cm. H_2O transpulmonary pressure were significantly ($p < .001$) lower than those of the controls (Fig. 5). The $V\%_{10}$ of the ventilated lobes were also significantly different ($p < .05$) from the non-ventilated lobes perfused with hypoxic, hypercapnic blood (F).

Hypercapnia in the Presence of High PO_2:

H) Not Ventilated: In 4 dogs the left lower lobe bronchus was ligated. The pulmonary arterial blood entering the lobe contained a $PO_2 > 60$ mm Hg, $PCO_2 > 77$ mm Hg and a pH of < 7.20. These lobes completely collapsed within 1/2 to 1 hour following the ligation of the bronchus. Their pressure-volume curves, at the end of 4 hours, were within the normal range (Figure 6), and they were significantly different ($p < .001$) from those of the non-ventilated lobes perfused with hypoxic, hypercapnic blood (F).

I) Ventilated: In 6 dogs the left lower lobe was perfused with hypoxic, hypercapnic blood ($PO_2 < 48$ mm Hg, $PCO_2 > 71$ mm Hg, and pH < 7.15) and ventilated with room air. The lobes showed pressure-

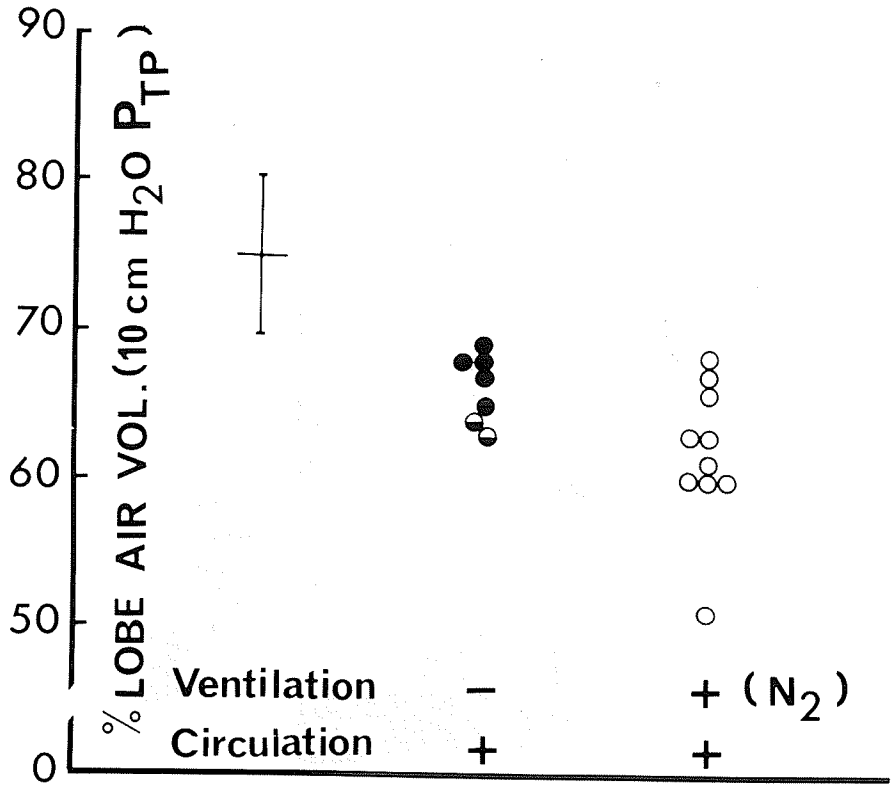

Figure 5. The effect of hypoxia and hypercapnia on P-V characteristics ($V\%_{10}$) of lobes. The lobes are perfused with blood containing a PO_2 < 50 mm Hg, PCO_2 > 64 mm Hg and pH < 7.21.

● = non-ventilated lobes (atelectatic).
◒ = lobes kept constantly inflated with pure nitrogen at a transpulmonary of 4-5 cm H_2O.
O = lobes ventilated with pure nitrogen.

The $V\%_{10}$ of the controls (mean ± 1 S.D.) is indicated for comparison.

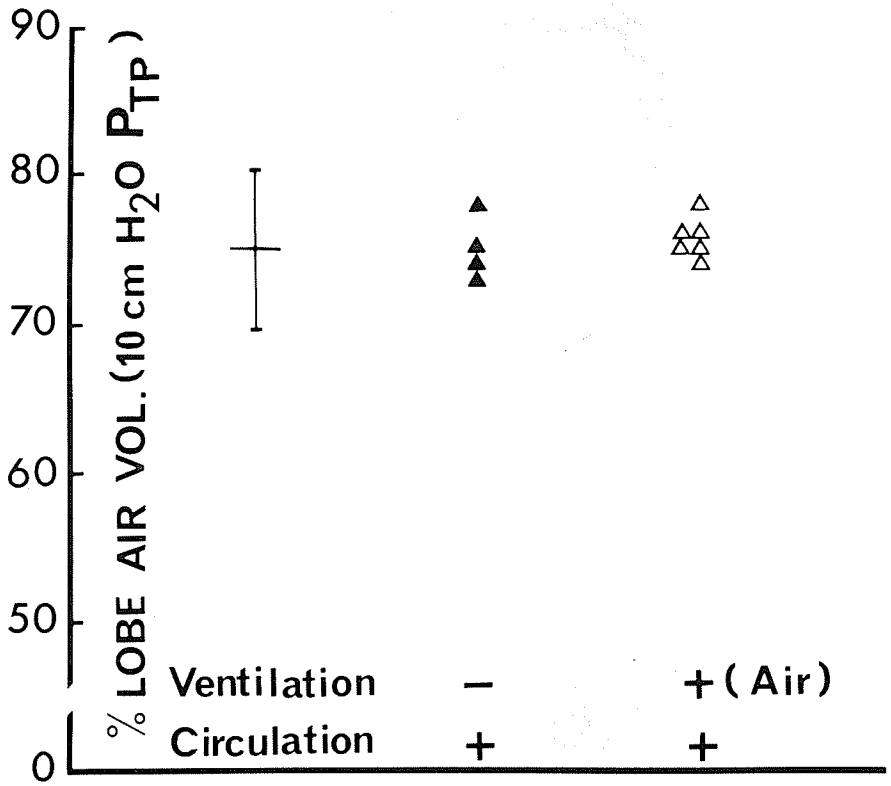

Figure 6. The effect of hypercapnia in the presence of high oxygen tension on P-V characteristics ($V\%_{10}$) of lobes.

▲ = Lobes not ventilated (atelectatic), and perfused with blood containing a $PO_2 > 60$ mm Hg, $PCO_2 > 77$ mm Hg and pH < 7.20.

△ = Lobes ventilated with air, and perfused with $PO_2 < 48$ mm Hg, $PCO_2 > 71$ mm Hg and pH < 7.15.

The mean $V\%_{10} \pm 1$ S.D. of the controls is shown for comparison.

volume curves within the normal range (Fig. 6). There was a significant difference ($p < .001$) between the $V\%_{10}$ of this group and the $V\%_{10}$ of the lobes ventilated with nitrogen and perfused with hypoxic, hypercapnic blood (G).

Blood Hydrogen Ion Concentration:

J) Hypoxia, Eucapnia and Acidosis: In 4 dogs the left lower lobe was ventilated with pure nitrogen. A continuous intravenous infusion of lactic acid (solution of 1/10 normal) or hydrochloric acid (1/10 normal) was administered in order to increase the hydrogen ion concentration of the blood. In these experiments the pulmonary arterial blood entering the left lower lobe contained a $PO_2 < 32$ mm Hg, $PCO_2 < 40$ mm Hg and a pH < 7.14. The pressure-volume curves of these lobes were within the normal range (Fig. 7).

K) Hypoxia, Hypercapnia and Normal pH: Five dogs received a continuous I.V. infusion of sodium bicarbonate or THAM (tris buffer) to prevent the rise in the blood hydrogen ion concentration due to the high CO_2 tension in the inspired gas of the right lung. The left lower lobe was ventilated with pure nitrogen and perfused with pulmonary artery blood containing a $PO_2 < 40$ mm Hg, $PCO_2 > 66$ mm Hg and a pH > 7.30. There was a decrease in percent lobe air volume at different transpulmonary pressures. The $V\%_{10}$ of these lobes were significantly different ($p < .001$) from those of the eucapnic, acidosis group (J) (Fig. 7). There was no significant difference between this group and the group of lobes ventilated with pure nitrogen and receiving hypoxic,

circulation were abnormal. The ventilated and non-ventilated lobes perfused with hypoxic, eucapnic blood had normal pressure-volume curves; and those perfused with hypoxic, hypercapnic blood had a significant increase in retractive forces which was reversible by perfusion with hypoxic, eucapnic blood. Non-respiratory acidosis had no effect on the mechanical properties of the lung. The percent of maximum air volume at 10 cm H_2O transpulmonary pressure correlated with the stability ratio of the bubbles expressed from the lung. Suggested mechanisms for the changes in the surface forces of the lung due to a combination of hypoxia and hypercapnia are alterations in lung tissue metabolism and in permeability of the vascular bed.

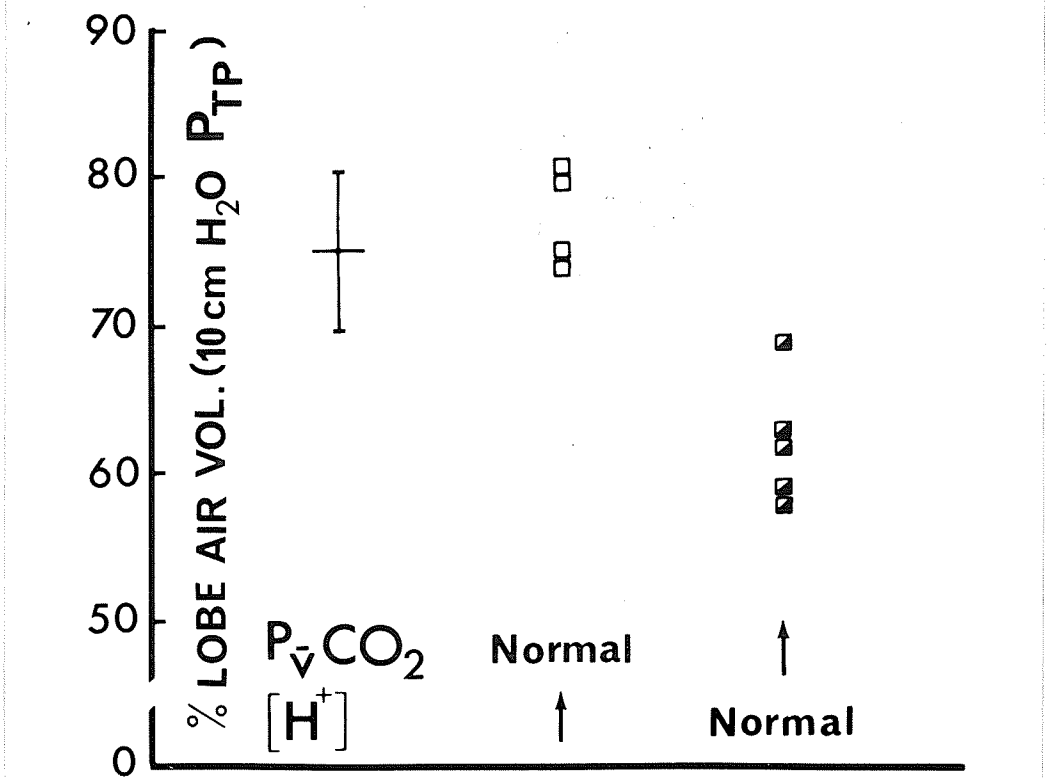

Figure 7. The effect of hypoxia, hypocapnia and acidosis, and the effect of hypoxia, hypercapnia and normal pH on the P-V characteristics ($V\%_{10}$) of lobes.

□ = ventilated with pure nitrogen, perfused with blood containing a $PO_2 < 32$ mm Hg, $PCO_2 < 40$ mm Hg and pH < 7.14 (continuous infusion of lactic acid or hydrochloric acid).

▨ = ventilated with pure nitrogen, perfused with $PO_2 < 40$ mm Hg, $PCO_2 > 66$ mm Hg and pH > 7.30 (continuous infusion of sodium bicarbonate or THAM).

The $V\%_{10}$ of the controls with one standard deviation to either side of the mean is indicated for comparison.

hypercapnic blood with a high concentration of hydrogen ions (G).

In Figure 8 the relationship between the pulmonary arterial blood PCO_2 and hydrogen ion concentration is plotted. Each point is the average of all the measurements done on one animal during the experiment.

When the $V\%_{10}$ of all experimental conditions were plotted against the corresponding pulmonary arterial blood PCO_2 (Fig. 9), three distinct groups were formed. 1) The hypoxic, eucapnic (or hypocapnic) group with normal pressure-volume curve, 2) the hypoxic, hypercapnic group with an abnormal pressure-volume curve and 3) the hypercapnic, high PO_2 group with normal pressure-volume curve.

Recovery of the Pressure-Volume Curve:

L) Recovery from the Effects of Pulmonary Artery Ligation: In 3 dogs the pulmonary artery to the left lower lobe was ligated and the lobe was ventilated with 5% O_2 in nitrogen for 4 hours (the same experimental condition as outlined in "B"). At the end of 4 hours the ligation to the pulmonary artery was released and the left lower lobe was ventilated with pure nitrogen with a tidal volume equal to 5 ml./Kg. of body weight and at a rate of 24/min. The pulmonary arterial blood gas tensions were: $PO_2 < 47$ mm Hg, $PCO_2 < 41$ mm Hg and pH was > 7.31. Three hours later the animal was exsanguinated. The deflation pressure-volume curves of these lobes were not different from the controls (Fig. 10A), but were significantly different ($p < .05$) from those of group "B" which were subjected to pulmonary arterial ligation and ventilated with 5% oxygen in nitrogen.

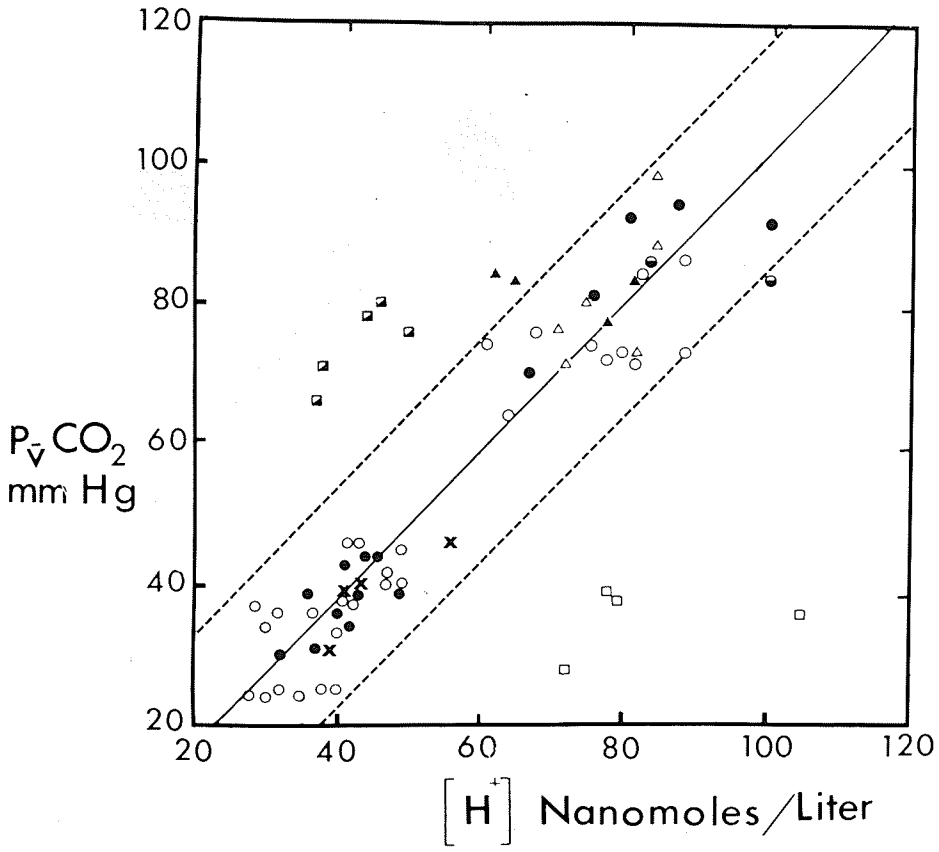

Figure 8. Relationship between the left pulmonary artery blood PCO_2 (mm Hg) and the hydrogen ion concentration (Nanomoles/liter) for all experimental conditions. Each point is the average of all measured PCO_2 and hydrogen ion concentrations during one experiment. The solid line is the regression line for all points except for □ (lactic acid or HCl perfusion) and ◪ (sodium bicarbonate or THAM perfusion) which are plotted on this graph after the calculation of the regression line is made. Regression line was calculated by the method of least squares. Dashed lines represent two standard deviations to either side of the mean. For description of the symbols please refer to the previous figures.

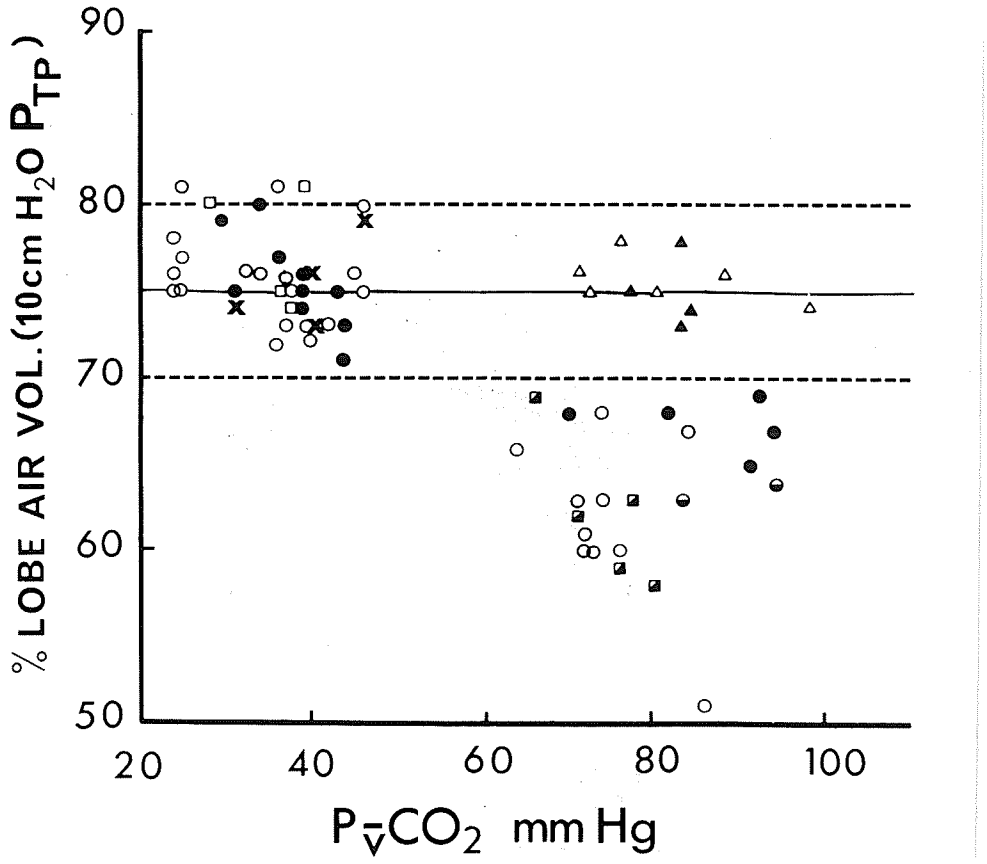

Figure 9. Relationship between the lobe air volume at 10 cm H_2O transpulmonary pressure ($V\%_{10}$) and the left pulmonary arterial blood PCO_2 for all experiments. The solid horizontal line is the mean $V\%_{10}$ of the control lobes. The broken lines are one standard deviation to either side of the mean. For description of symbols please refer to the previous figures.

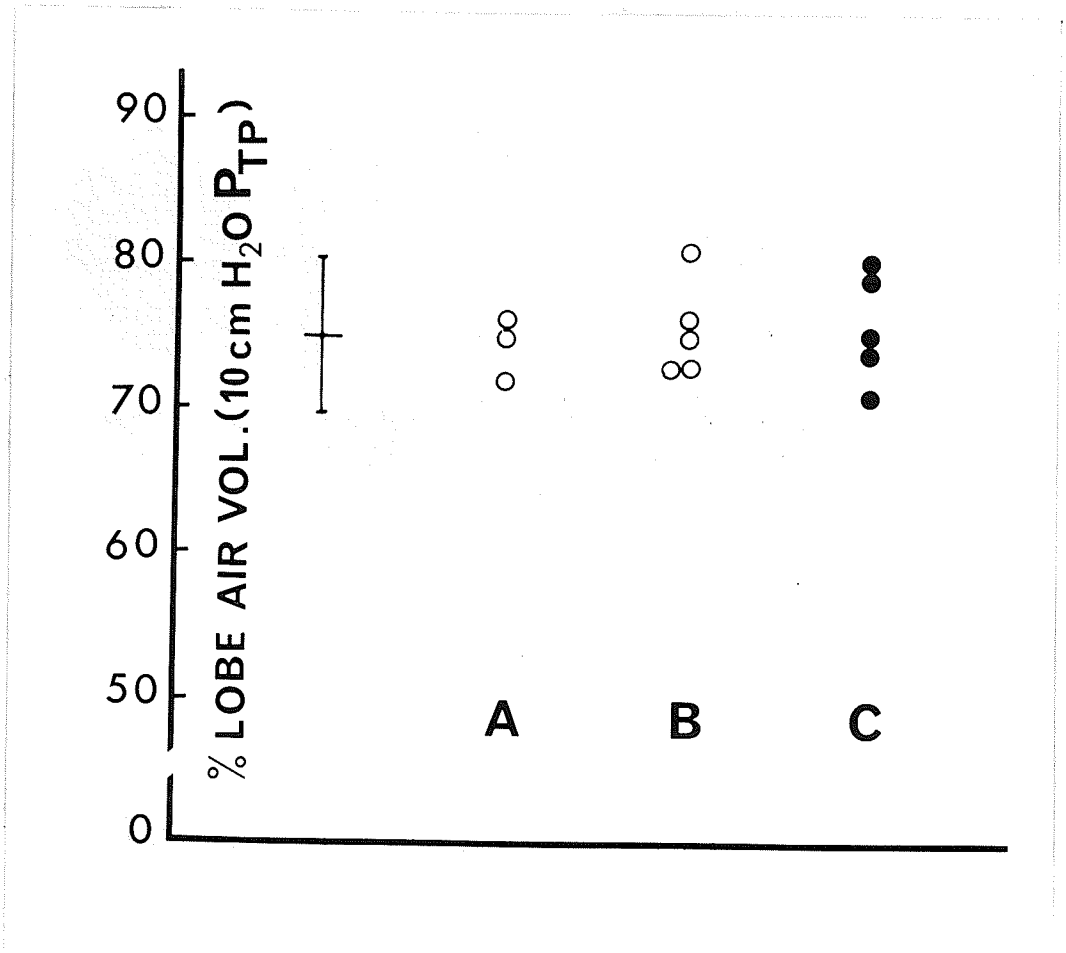

Figure 10. A - Recovery from the effects of pulmonary artery ligation.
B - Recovery from the effects of hypoxia and hypercapnia in ventilated lobes.
C - Recovery from the effects of hypoxia and hypercapnia in non-ventilated (atelectatic) lobes.
O = lobes ventilated with pure nitrogen.
● = lobes not ventilated (atelectatic).
The V%10 of the controls with one standard deviation to either side of the mean is indicated for comparison.

Recovery from the Effects of Hypoxia and Hypercapnia:

M) Ventilated: The left lower lobes of 5 dogs were ventilated with pure nitrogen and perfused with hypoxic, hypercapnic blood ($PO_2 < 46$ mm Hg, $PCO_2 > 63$ mm Hg and pH < 7.20 - similar to the experiments outlined in "G"). After 4 hours, the pulmonary arterial blood gas tensions were changed to a $PO_2 < 40$ mm Hg, $PCO_2 < 43$ mm Hg and the pH to > 7.33. Ventilation of the left lower lobe with pure nitrogen was continued using a tidal volume equal to 5 ml./Kg. of body weight and at a rate of 24/minute. After 3 hours, the pressure-volume curves were obtained. These were within the normal range (Fig. 10B). Their $V\%_{10}$ were significantly different ($p < .001$) from those of the hypoxic, hypercapnic group (G).

N) Not Ventilated: In 5 dogs the left lower lobe bronchus was ligated for 7 hours. During the first 4 hours the lobe was perfused with blood with a $PO_2 < 49$ mm Hg, $PCO_2 > 81$ mm Hg and pH < 7.08. Then in the last 3 hours the blood gas tensions were changed to $PO_2 < 45$ mm Hg, $PCO_2 < 44$ mm Hg and the pH to > 7.34. At the end of 7 hours the pressure-volume curves of these non-ventilated lobes were within the normal range (Fig. 10C). Their $V\%_{10}$ were significantly different ($p < .001$) from those of lobes perfused with hypoxic, hypercapnic blood for 4 hours (F).

Static Inflation of Excised Lobes:

O) Three of the 8 lobes subjected to pulmonary artery ligation and ventilated with 5% oxygen in nitrogen for 4 hours (Group "B") were

placed in a high humidity chamber at room temperature (25 - 26°C). The lobes were kept constantly inflated with room air at a transpulmonary pressure of 30 cm H_2O for 3 hours. At the end of 3 hours they were degassed and a deflation pressure-volume curve was obtained. The $V\%_{10}$ of these lobes (Fig. 11A) were not different from those taken prior to the static inflation.

P) Three lobes previously perfused with hypoxic, hypercapnic blood and ventilated with nitrogen (from Group "G") were statically inflated for 3 hours with room air. These lobes did not recover also. The $V\%_{10}$ was essentially the same before and after the static inflation (Fig. 11B).

Q) Three of the non-ventilated lobes, perfused with hypoxic, hypercapnic blood (From Group "F") were subjected to static inflation with room air for 3 hours. The pressure-volume characteristics of these lobes reverted to the normal range and their $V\%_{10}$ were significantly different ($p < .001$) from those obtained prior to the static inflation (Fig. 11C).

The results of all experiments are summarized in Table II.

Pressure-Volume Characteristics versus Stability Ratio of Bubbles:

In Figure 12 the $V\%_{10}$ of 22 lobes are plotted against the mean stability ratios of 40 bubbles obtained from each lobe. When a straight line was drawn through these points by the least-squares technique, the slope of the line was positive and significantly different from zero ($p < .001$). The numerical values and the experi-

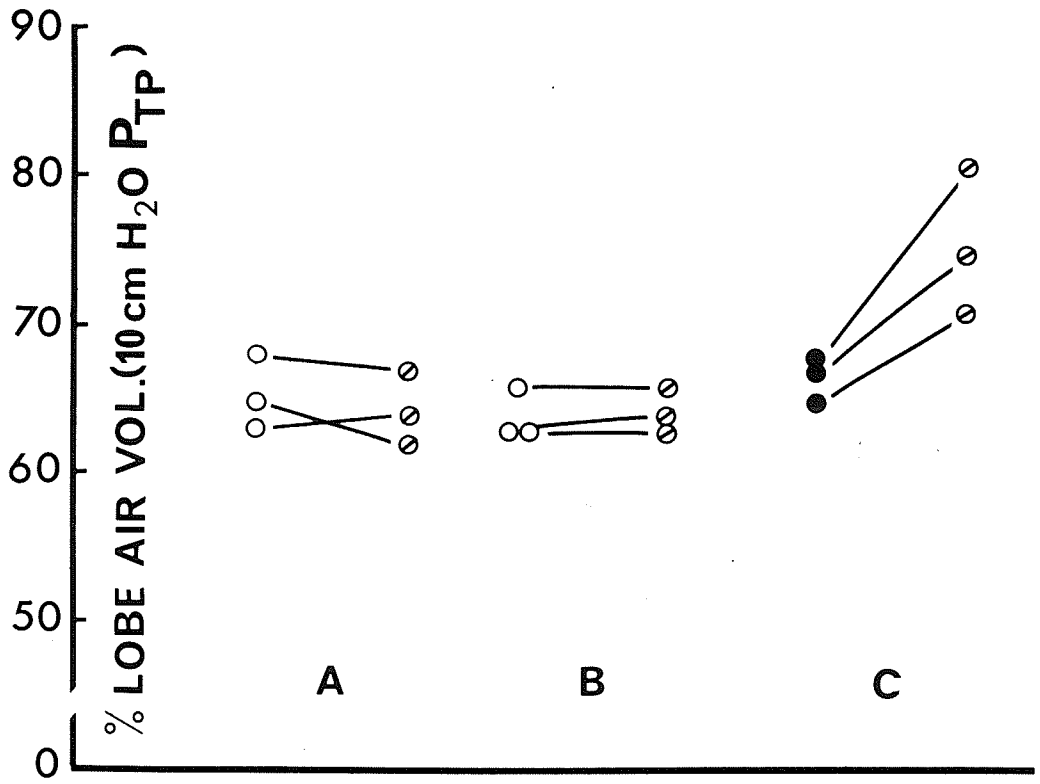

Figure 11. The effect of static inflation with room air in excised left lower lobes in different experimental conditions.
A - Lobes ventilated with 5% oxygen in nitrogen, and pulmonary artery ligated.
B - Lobes ventilated with pure nitrogen, perfused with hypoxic, hypercapnic blood.
C - Lobes not ventilated (atelectatic), perfused with hypoxic, hypercapnic blood.
The $V\%_{10}$ after three hours constant inflation is designated as ⊘ . The solid lines link the corresponding $V\%_{10}$ of each lobe before and after constant inflation with room air.

TABLE II

SUMMARY OF THE RESULTS

Experimental Cond. Vent.	Circ.	No. of animals	$P\bar{v}O_2$ mm Hg	$P\bar{v}CO_2$ mm Hg	pH	$V\%_{10}$
No	No	6	---	---	---	N
Yes (5% O_2, air)	No	8	---	---	---	↓*
No	Yes	5	<42	<44	>7.30	N
Yes (N_2)	Yes	11	<48	<46	>7.30	N
Yes (N_2)	Yes↓	4	<32	<46	>7.25	N
No	Yes	7	<47	>70	<7.18	↓*
Yes (N_2)	Yes	10	<50	>64	<7.21	↓*
No	Yes	4	>60	>77	<7.20	N
Yes (air)	Yes	6	<48	>71	<7.15	N
Yes (N_2)	Yes	4	<32	<40	<7.14	N
Yes (N_2)	Yes	5	<40	>66	>7.30	↓*
Recovery:						
Yes (5% O_2)	No	3	---	---	---	---
Yes (N_2)	Yes		<47	<41	>7.31	N
No	Yes	5	<49	>81	<7.08	---
No	Yes		<45	<44	>7.34	N
Yes (N_2)	Yes	5	<46	>63	<7.20	---
Yes (N_2)	Yes		<40	<43	>7.33	N

N = normal, i.e. within ±1S.D. from the mean of the control values.

↓ = below -1 S.D. from the mean of the control values.

* = significantly different ($p < .001$) from the controls.

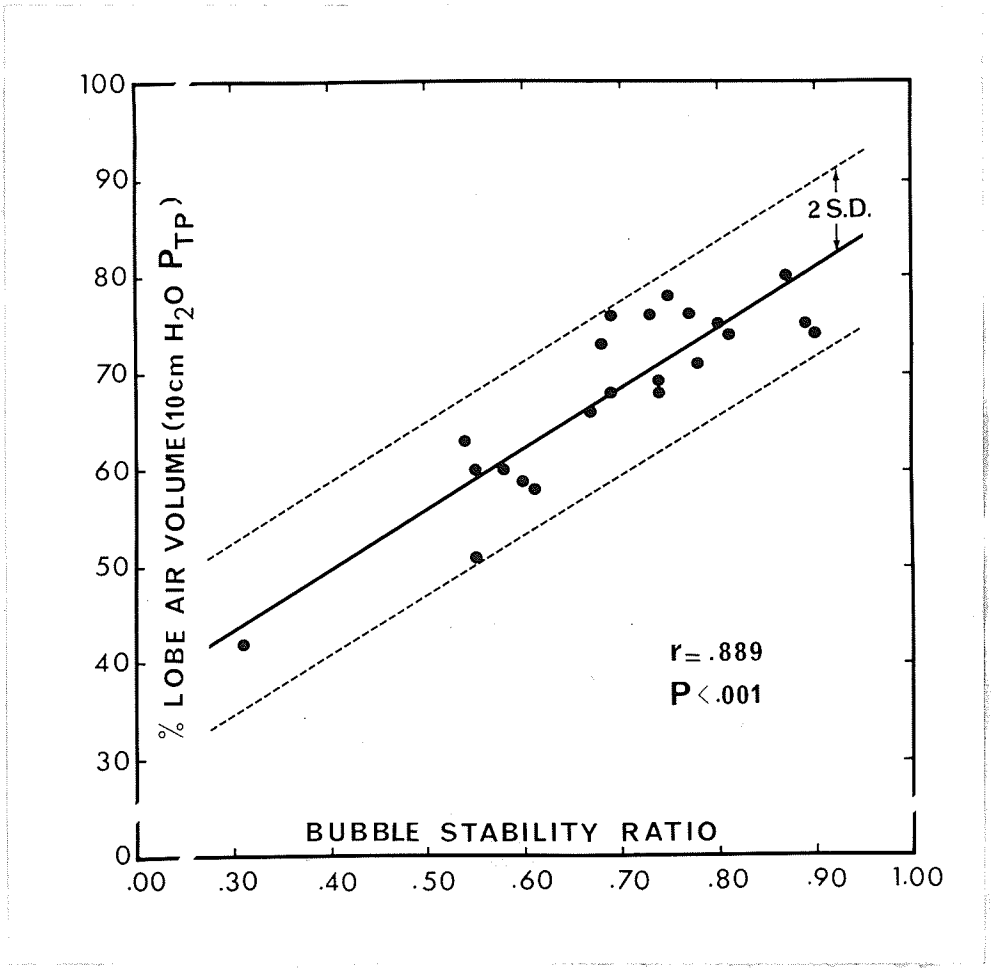

Figure 12. Relationship between the $V\%_{10}$ of lobes and the stability ratio of bubbles expressed from the lobes. Each point represents the mean value for 40 bubbles examined from four different areas of the lobe tissue. Regression line was calculated by the least squares technique. Broken lines represent two standard deviations to either side of the mean.

mental conditions for each point in Figure 12 are given in Table III.

In Figure 13 are shown the percentile distribution of the stability ratios of bubbles obtained from lungs with normal and abnormal pressure-volume characteristics. The percentile distribution of the two groups were tested by chi squares technique. A significant difference was found between the distribution of the two groups ($p < .001$).

Dry Weight/Wet Weight Ratio:

Thirteen lobes from the control animals, eight experimental lobes with normal pressure-volume curves and 18 lobes with abnormal pressure-volume curves were dried. The mean values ($\pm$ 1 S.D.) of dry weight/wet weight ratio x 100 were as follows: 23.22 ± 0.22 for the controls, 22.78 ± 0.62 for the lobes with normal pressure-volume curve and 23.16 ± 0.64 for the lobes with abnormal pressure-volume curve. There was no significant difference between the dry weight/wet weight ratios of these groups.

Left Lower Lobe Weight/Body Weight Ratio:

The mean value and standard deviation for 10 experimental lobes with normal pressure-volume curves and for 14 lobes with abnormal pressure-volume curves were 2.16 ± 0.25 and 2.33 ± 0.44 gm./Kg. body weight respectively. There was no significant difference between these two values.

Air Volume per Gram of Lung Tissue:

The air volume at 40 cm. H_2O transpulmonary pressure per gram

TABLE III

VALUES OF STABILITY RATIO OF BUBBLES WITH CORRESPONDING PERCENT LOBE AIR VOLUME AT 10 cm H_2O P_{TP} FOR DIFFERENT EXPERIMENTAL CONDITIONS.

$V\%_{10}$	Stability Ratio*	Experimental Condition **
80	.87	D
78	.75	D
76	.77	I
76	.73	D
76	.69	A
75	.89	D
75	.80	J (lactic acid)
74	.90	J (HCl)
74	.81	H
73	.68	C
71	.78	Control
69	.74	K ($NaHCO_3$)
68	.74	B (5% O_2 in N_2)
68	.69	G
66	.67	G
63	.54	K ($NaHCO_3$)
60	.58	G
60	.55	B (5% O_2 in N_2)
59	.60	K (THAM)
58	.61	K (THAM)
51	.55	G
42	.31	B (air)

* The mean stability ratio of 40 bubbles.

** For full explanation of the experimental condition, please refer to the corresponding group in "Results".

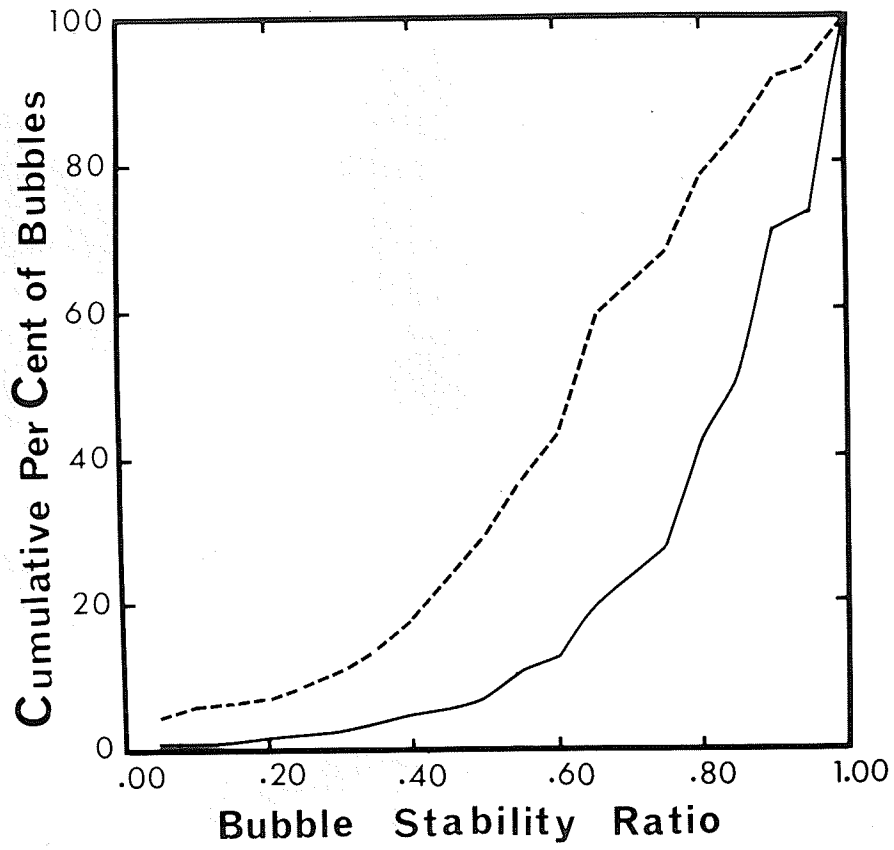

Figure 13. Percentile distribution of stability ratio of bubbles for the experimental lobes. The solid curve is the percentile distribution for lobes with "normal" P-V characteristics ($V\%_{10}$ between 70 and 80 percent). The broken curve is for lobes with $V\%_{10}$ lower than 70 percent.

of the lung tissue were calculated for 15 lobes with normal and 15 lobes with abnormal pressure-volume curves. The mean values ($\pm$ 1 standard deviation) were: 14.22 $\pm$ 2.82 and 13.58 $\pm$ 1.86 ml./gm. of lung tissue respectively. No significant difference was found between these two values.

In almost all experiments the left lower lobe was grossly normal except in 4 out of the 11 lobes subjected to pulmonary artery ligation and ventilation (Groups "B" and "L"). In one lobe, small areas of collapse and in the other 3 patchy areas of collapse and hemorrhage were noted.

During the pressure-volume studies the lobes were uniformly inflated at 40 cm H_2O transpulmonary pressure, and on deflation the air volume appeared to be uniformly distributed throughout the lobe. Areas of premature closure were observed in a few lobes at transpulmonary pressures of 10 cm H_2O or lower.

D. Discussion

A meaningful method of analyzing deflation pressure-volume data for alterations in retractive forces is a comparison of the percentage of maximum air volume present at various transpulmonary pressure, as suggested by Johnson et al (6). This method is concerned only with inflatable alveoli, which are the ones appropriately to be taken into account. In addition, it is a method that is independent of lung size or lung weight. In previous studies (7) it was found that the change in the deflation pressure-volume curve was most evident at 10 cm H_2O transpulmonary pressure and there was an excellent correlation with surface tension measurements. In the present study the air volume at 10 cm H_2O transpulmonary pressure expressed as percentage of the total lobe air volume at 40 cm H_2O pressure ($V\%_{10}$) is used to compare the effects of different experimental conditions on the mechanical properties of the lung. A decrease in $V\%_{10}$ could be due to: 1) premature closure of units above 10 cm H_2O pressure; 2) increased retractive force of all units containing air at 10 cm H_2O pressure; or 3) a combination of these factors.

The positive correlation between the percentage air volume at 10 cm H_2O transpulmonary pressure and stability ratio indicates that alterations in pressure-volume curves are associated with changes in the surface activity of lung lining material. If .71, as suggested by Pattle (3),is taken as the mean value of stability ratio for normal

lungs, then only two out of 11 lobes with abnormal pressure-volume curves (Table III) have stability ratios greater than the mean, in contrast to the lobes with normal pressure-volume curves, in which 9 out of 11 have stability ratios above the mean. This difference is statistically significant ($\chi^2 = 6.54$, $p < .025$). Even though there is a significant increase in the number of less stable bubbles from lungs with an abnormal pressure-volume curve, it is noted that a large number of bubbles have a high stability index. This suggests that, whatever mechanisms are involved for alterations in the activity of the lung lining material, do not act uniformly throughout the lung. This results in a wide range of stability index for a given $V\%_{10}$, since random samples taken for the measurement of stability ratio may include areas with or without altered surface forces. Therefore, the pressure-volume studies might be a better method of quantifying elevations in lung surface tension than studies of bubble stability ratio performed on focal areas, since the pressure-volume data afford a means of estimating the anatomical extent of such focal changes. This method, then, furnishes a rough index of the functional significance of focal changes.

It is important to emphasize, however, that these pressure-volume studies are quantitative and not qualitative. Pressure-volume changes could possibly result from other factors in addition to altered alveolar surface tension. For example, alterations in tissue elasticity might account for similar increases in retractive forces.

If changes in the lung tissue elasticity have been brought about by hypoxia and hypercapnia we can only assume that these are temporary and direct, since studies of the recovery process indicate that the lung is not permanently damaged. In the measurements of deflation pressure-volume curves whatever direct effect hypoxia and hypercapnia have on the lung elastic tissue is probably eliminated by degassing the lobes and inflating them with room air prior to the procedure. In addition, the air volume at full inflation (40 cm H_2O PTP) per gram of lung tissue was not statistically different in the lobes with normal and abnormal pressure-volume curves, indicating that tissue retractive forces are unaltered. The similarity of the air volume per gram of lung tissue was not due to differences in the lobe weight, since the lobe weight per Kg. of body weight and also the dry weight/wet weight ratio were not different in both groups.

The presence of intra-alveolar fluid resulting in a decrease in radii of alveolar gas-liquid surfaces (8) seems unlikely to be a factor for the increase in retractive forces in these experiments. The lobes were not edematous, were not heavier than normal lungs and their dry weight/wet weight ratios were not significantly different from the control lungs.

Thus it seems most likely that the pressure-volume changes seen in lobes in this study are primarily due to an elevation in alveolar surface tension.

The major findings can be summarized as follows:

a) Acute cessation of the pulmonary artery circulation was detrimental for pressure-volume characteristics in ventilated lobes.
b) As long as the lobe was perfused with normal pulmonary arterial blood, there was no significant alteration in the pressure-volume measurements. This was independent of ventilation or reduction of pulmonary blood flow to 15% of the control values.
c) When lobes received hypoxic, hypercapnic blood, there was a decrease in the percent lobe air volume at a given transpulmonary pressure. This was also independent of ventilation.
d) Alterations in the blood hydrogen ion concentration were not a cause for alterations in pressure-volume curves.
e) The effect of hypoxia and hypercapnia, and pulmonary artery ligation upon the pressure-volume curves were reversed if the lobes received blood with normal gas tensions for three hours.

Pulmonary Artery Ligation:

From experiments with excised lobes of dogs and rats (7, 9) it is known that pressure-volume characteristics of the lobes are not altered if they are not ventilated. However, ventilation depletes the surfactant, resulting in a decrease in the percent lobe air volume at a given transpulmonary pressure. Depletion is directly related to the tidal volume and inversely related to the end expiratory pressure. In the present studies, the non-perfused lobes were ventilated with a large tidal volume (equal to about 30% of the MLV) and with no end expiratory pressure. Four out of 11 ventilated non-perfused lobes had patchy atelectasis or hemorrhagic areas. It is

possible that ventilation depleted surfactant and that when alveolar surface forces increased, exudation of plasma fluid and blood into the alveoli followed (10). There is evidence to support this assumption: 1) the non-ventilated, non-perfused lobes were not hemorrhagic and their pressure-volume characteristics were within normal range. 2) Giammona et al found no alterations in the pressure-volume curves and no changes on gross and microscopic examination of the lungs subjected to pulmonary artery ligation and ventilation for 4 hours (11). However, their non-perfused lobes were not ventilated with a tidal volume as large as used in the present studies. In excised lobes ventilation with small tidal volumes and with end expiratory pressure does not deplete surfactant (7).

When the lungs are over-inflated using peak pressures of 26 - 32 cm. H_2O tearing of the alveolar and capillary walls occurs. This causes pulmonary edema and hemorrhage and alterations in surface forces only after 24 hours (12). The inflation pressures in the present experiment were below 8 cm H_2O and changes in surface forces were seen in 4 hours, suggesting that there was no anatomical disruption of lung tissue.

Atelectasis:

The results of pressure-volume curves of atelectatic lobes with pulmonary artery ligation or of lobes perfused with hypoxic, eucapnic blood indicate that atelectasis per se is not deleterious to the surfactant and the lung mechanics. This is in agreement with

findings of Levine and Johnson (13) who showed normal deflation pressure-volume curves and a highly surface active material in the tracheal foam of lungs collapsed for 90 minutes to 8 days.

Hypoxia and Hypercapnia:

It has been shown that alveolar hypoxia causes pulmonary vasoconstriction in the lungs of living anesthetized dogs (14, 15, 16, 17) and in isolated lobes of dogs (18) and cats (19). Pulmonary vasoconstriction results in an increase in pulmonary vascular resistance, a rise in pulmonary arterial pressure, and a decrease in pulmonary blood flow (20). Pulmonary vasoconstriction and rise in pulmonary arterial pressure are also observed with hypercapnia (21) and with elevations in blood hydrogen ion concentration (22).

In 3 dogs the effect of ventilation with air or nitrogen, perfusion with hypoxic-eucapnic, and hypoxic-hypercapnic blood on pulmonary arterial blood flow to the left lower lobe, was measured directly by means of a magnetic flow meter. The maximum reduction observed under such experimental conditions was 40% of initial blood flow. At the same time further experiments show that a pulmonary arterial blood flow of 15% of the control value is sufficient to maintain the normal pressure-volume characteristics of lobes even under extreme hypoxia. This evidence suggests that reduction of pulmonary blood flow per se does not alter surfactant.

Measurements of the left pulmonary arterial pressure showed that when the left lower lobe was ventilated with nitrogen, perfusion of the lobe with hypoxic, eucapnic blood was associated with a greater

rise in pulmonary blood pressure than perfusion with hypoxic, hypercapnic blood. If elevation of blood pressure was a factor for alterations in surface forces of the lung, one would expect abnormal pressure-volume curves in lobes ventilated with nitrogen and perfused with hypoxic, eucapnic blood. However, this was not the case. The alterations were noted in lobes perfused with hypercapnic, hypoxic blood.

It should be noted that a rise in pulmonary arterial blood pressure may be due to a pre or post-capillary constriction. The particular vascular segment, or segments, involved in the pulmonary vasoconstriction in response to hypoxia, hypercapnia and acidosis is not yet agreed upon. However, it seems that both the precapillary and postcapillary small vessels can constrict if exposed to a sufficient degree of hypoxia (23), or hypercapnia (24) and that the postcapillary segments would be more affected by hypoxia than the precapillary segments since mixed venous blood is ordinarily low in oxygen tension (23).

From 4 conditions tested in this study (1 - hypoxia, eucapnia; 2 - hypoxia, hypercapnia; 3 - hypoxia, acidosis; and 4 - hypercapnia in the presence of high P_{O_2}) only the combination of hypoxia and hypercapnia increased the retractive forces of the lung.

If pulmonary vasoconstriction is responsible for the observed changes, it should have occurred only with or more severely with hypoxia and hypercapnia. Let us assume that the site of vasoconstric-

tion is in the precapillary segments. If vascular constriction is not uniform throughout the lobe, some areas may be deprived completely of blood flow, while others may continue to receive sufficient blood flow. Ventilation of non-perfused areas in such an instance would result in a depletion of surfactant and an increase in retractive forces of the lobe. However, this does not explain the findings in the non-ventilated lobes perfused with hypoxic, hypercapnic blood where the surface forces are also altered.

If the site of vasoconstriction is primarily in the post-capillary vessels, capillary pressure would rise causing an exudation of plasma from the capillaries onto the alveolar surface, which would inhibit the activity of surfactant (25). This could occur independent of ventilation.

It is possible that hypoxia and hypercapnia exert their effects on mechanical properties of the lung through a combination of a cessation in blood flow to some areas (precapillary vasoconstriction) and an increase in capillary pressure in others (postcapillary vasoconstriction).

It is stated that the effects of CO_2 on pulmonary circulation depend on the degree of acidosis that it produces (26). However, in the experiments with hypoxia-acidosis the mechanical properties of the lung remained unchanged. This raises the possibility that mechanisms other than or in addition to pulmonary vasoconstriction are

responsible for the changes observed.

A mechanism to be considered is a reduction in the metabolic activity of the cells responsible for the production of surfactant. Either a decrease in the rate of production of surface-active material, or a decrease in the rate of its delivery onto the surface could explain the present findings. If surfactant were continuously depleted, the amount present on the alveolar surfaces would diminish and eventually reach levels insufficient for maintaining normal surface tension. The rate of depletion itself may be normal or accelerated.

The effects of hypoxia and hypercapnia, if any, on the metabolic activity of the lung tissue are reversible by perfusion with hypoxic, eucapnic blood.

Another possible mechanism is the presence of an inhibitor of surfactant on the alveolar surface. Plasma fluid does inhibit the activity of surfactant (25). An increase in the permeability of capillary vessels could cause exudation of plasma fluid from the capillaries onto the alveolar surface. Boonyaprakob et al (27) have shown an increase in the clearance rate of the plasma proteins in the pulmonary capillaries of puppies ventilated with low concentrations of oxygen. This increase in the permeability of vessels has not yet been shown for adult dogs. If alterations in the permeability of capillary vessels account for the changes observed in the lung mechanics then it must be assumed that in adult dogs it occurs only upon exposure to a combination of hypoxia and hypercapnia. Recovery

from the effect of hypoxia, hypercapnia following a period of hypoxic, eucapnic perfusion could be explained on the basis of a return of capillary permeability to a normal state and the removal of the inhibitor from the surface. The concept of an inhibitor is supported by the following observations.

In previous studies (7) it has been shown that in excised lobes of dogs the changes in pressure-volume curves revert to normal if lobes are kept at static inflation with room air for 3 hours. This recovery phenomenon is not seen in lobes instilled with plasma (25).

In the present studies the ventilated lobes subjected to pulmonary artery ligation or perfused with hypoxic, hypercapnic blood did not recover when excised and statically inflated with room air (Fig. 11). This suggests the presence of an inhibitor on the alveolar surface. If these lobes were not excised but were perfused in vivo with hypoxic, eucapnic blood the pressure-volume curves reverted to normal, possibly because re-establishment of the circulation with normal blood removed the inhibitors.

In non-ventilated lobes the pressure-volume curves revert to normal after static inflation. Since ventilation per se depletes surfactant, perhaps the difference between the ventilated and non-ventilated lobes in the process of recovery is the relative amount of inhibitor and surfactant present.

The exact mechanisms whereby hypoxia and hypercapnia affect lung mechanics are far from elucidated. Nevertheless, it appears from

these studies that high concentrations of oxygen prevent the effects of hypercapnia. Reversal of the ill effects of hypoxia-hypercapnia is not achieved by correction of blood pH with $NaHCO_3$ or THAM but by a reduction in PCO_2.

E. Summary

The effect of acute alterations of pulmonary blood flow, pulmonary arterial blood gas tensions and alveolar gas tensions on the mechanical properties of lungs were studied in left lower lobes of open-chest dogs. Static deflation pressure-volume measurements and the stability ratio of bubbles expressed from the lung were used to assess changes in the retractive forces of the lung.

The major findings are:

a) In non-ventilated lobes, pulmonary artery ligation had no effect on pressure-volume characteristics of the lobes.

b) In ventilated lobes, acute cessation of pulmonary blood flow caused an increase in retractive forces of the lobe.

c) When lobes were perfused with hypoxic, eucapnic blood, there was no alteration in pressure-volume measurements regardless of whether these lobes were ventilated or their pulmonary blood flow was reduced to 15% of the control values.

d) When lobes received hypoxic, hypercapnic blood ($P\bar{V}_{O_2} < 45$ mm Hg, $P\bar{V}_{CO_2} > 60$ mm Hg) there was a decrease in the percent lobe air volume at different transpulmonary pressures. This was independent of ventilation.

e) The effects of hypoxia-hypercapnia, and pulmonary artery ligation upon the pressure-volume curves were reversed if the lobes received hypoxic, eucapnic blood for 3 hours.

f) Non-respiratory acidosis (infusion of lactic acid or hydrochloric acid) had no effects on pressure-volume curves. This suggested that changes in the mechanical properties of the lung are probably due to some other effect of CO_2 rather than changes in the blood hydrogen ion concentration.

g) Hypercapnia in the presence of high PO_2 did not alter the pressure-volume characteristics of the lung.

h) There was a significant correlation between the $V\%_{10}$ and the bubble stability ratio.

It is suggested that changes in the surface activity of the lung lining material and mechanical properties of the lung in the presence of hypoxia with hypercapnia may be due to any one or a combination of the following factors: 1) multiple focal pulmonary vasoconstriction, 2) decrease in the rate of production of surface-active material, 3) increase in the permeability of the vascular bed with the concomitant exudation of plasma inhibitors of surfactant.

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G. Bibliography

1. Gribetz, I., Frank, N.R. and Avery, M.E.: Static volume-pressure relations of excised lungs of infants with hyaline membrane disease, newborn and stillborn infants. J. Clin. Invest. 38: 2168-2175, 1959.

2. Radford, E.P.: Mechanical stability of the lung. Arch. Environ. Health 6: 128-133, 1963.

3. Pattle, R.E.: Surface lining of lung alveoli. Physiol. Review 45: 48-79, 1965.

4. Standard Mathematical Tables (10th. edition). Cleveland: Chemical Rubber Publishing Co. 1956. p. 239.

5. Faridy, E.E., Kidd, R., Milie-Emili, J.: Topographical distribution of inspired gas in excised lobes of dogs. J. Appl. Physiol. 22: 760-766, 1967.

6. Johnson, J.W.C., Permutt, S., Sipple, J.H. and Salem, E.S.: Effect of intra-alveolar fluid on pulmonary surface tension properties. J. Appl. Physiol. 19: 769-777, 1964.

7. Faridy, E.E., Permutt, S. and Riley, R.L.: Effect of ventilation on surface forces in excised dogs' lungs. J. Appl. Physiol. 21: 1453-1462, 1966.

8. Cook, C.D., Mead, J., Schreiner, G.L., Frank, N.R. and Craig, J.M.: Pulmonary mechanics during induced pulmonary edema in anesthetized dogs. J. Appl. Physiol. 14: 177-186, 1959.

9. McClenahan, J.B. and Urtnowski, A.: Effect of ventilation on surfactant, and its turnover rate. J. Appl. Physiol. 23: 215-220, 1967.

10. Pattle, R.E.: Properties, function and origin of the alveolar lining layer. Proc. Roy. Soc. London Ser. B. 148: 217-240, 1958.

11. Giammona, S.T., Mandelbaum, I., Foy, J. and Bondurant, S.: Effects of pulmonary artery ligation on pulmonary surfactant and pressure-volume characteristics of dog lung. Circulation Research 18: 683-691, 1966.

12. Greenfield, L.J., Ebert, P.A. and Benson, D.W.: Effect of positive pressure ventilation on surface tension properties of lung extracts. Anesthesiology 25: 312-316, 1964.

13. Levine, B.E. and Johnson, R.P.: Effects of atelectasis on pulmonary surfactant and quasi-static lung mechanics. J. Appl. Physiol. 20: 859-864, 1965.

14. Aviado, D.M., Ling, J.S. and Schmidt, C.F.: Effects of anoxia on the pulmonary circulation reflex pulmonary vasoconstriction. Am. J. Physiol. 189: 253-262, 1957.

15. Bergofsky, E.H., Bass, B.H., Ferretti, R. and Fishman, A.P.: Pulmonary vasoconstriction in response to pre-capillary hypoxemia. J. Clin. Invest. 52: 1201-1215, 1963.

16. Duke, H.N.: Observations on the effects of hypoxia on the pulmonary vascular bed. J. Physiol., London 135: 45-51, 1957.

17. Dugard, A. and Naimark, A.: Effect of hypoxia on distribution of pulmonary blood flow. J. Appl. Physiol. 23: 663-671, 1967.

18. Lloyd, T.C. Jr.: Pulmonary vasoconstriction during histotoxic hypoxia. J. Appl. Physiol. 20: 488-490, 1965.

19. Duke, H.N.: The site of action of anoxia on the pulmonary blood vessels of the cat. J. Physiol., London 125: 373-382, 1954.

20. Kato, M. and Staub, N.C.: Response of small pulmonary arteries to unilobar hypoxia and hypercapnia. Circulation Research 19: 426-440, 1966.

21. Duke, H.N.: Pulmonary vasomotor responses of isolated perfused cat lungs to anoxia and hypercapnia. Quart. J. Exptl. Physiol. 36: 75-88, 1951.

22. Bergofsky, E.H., Lehr, D.E. and Fishman, A.P.: The effect of changes in hydrogen ion concentration on the pulmonary circulation. J. Clin. Invest. 41: 1492-1502, 1962.

23. Fishman, A.P.: Respiratory gases in the regulation of the pulmonary circulation. Physiol. Revs. 41: 214-280, 1961.

24. Hyde, R.W., Lawson, W.H. and Forster, R.E.: Influence of carbon dioxide on pulmonary vasculature. J. Appl. Physiol. 19: 734-744, 1964.

25. Johnson, J.W.C., Faridy, E.E. and Riley, R.L.: Factors affecting pressure-volume and surface tension properties of excised lungs (abstract). Fed. Proc. 23: 155, 1964.

26. Fishman, A.P.: Dynamics of the pulmonary circulation. In Handbook of Physiology, Circulation. Vol. 2. pp 1667-1743, 1963.

27. Boonyaprakob, U., Taylor, P., Watson, D., Waterman, V. and Lopata, E.: Hypoxemia and protein clearance from the pulmonary vascular bed of pups. Ped. Research 1: 200, 1967.

28. Collier, C.R., Hackney, J.D. and Rounds, D.E.: Alterations of surfactant in oxygen poisoning. Dis. Chest 48: 233-238, 1965.

29. Kennedy, J.H.: Hyperbaric oxygenation and pulmonary damage. The effect of exposure at two atmospheres upon surface activity of lung extracts in the rat. Med. Thorac. 23: 27-35, 1966.

30. Schaefer, K.E., Avery, M.E. and Bensch, K.: Time course of changes in surface tension and morphology of alveolar epithelial cells in CO_2 induced hyaline membrane disease. J. Clin. Invest. 43: 2080-2093, 1964.

31. Finley, T.N., Tooley, W.H., Sevenson, E.W., Gardner, R.E. and Clements, J.A.: Pulmonary surface tension in experimental atelectasis. Am. Rev. Respirat. Diseases 89: 372-378, 1964.

32. Edmunds, L.H.Jr. and Huber, G.L.: Pulmonary artery occlusion. J. Appl. Physiol. 22: 990-1016, 1967.