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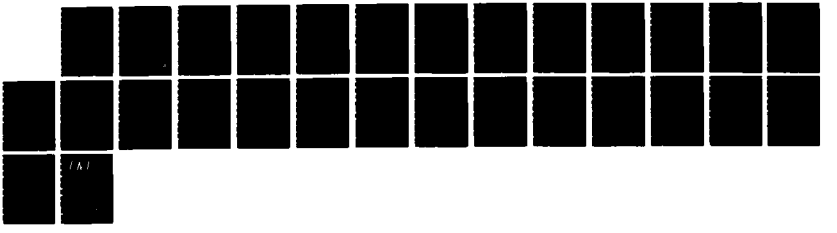

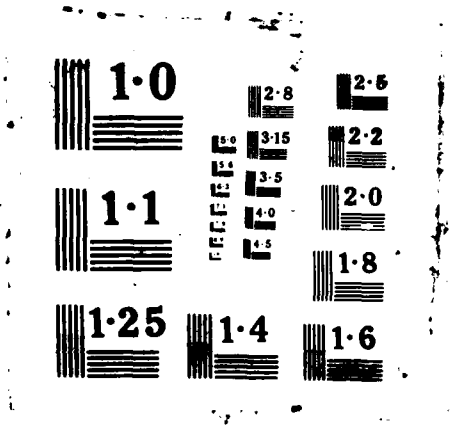
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FINAL REPORT

SCINTIGRAPHY FOR PULMONARY CAPILLARY PROTEIN LEAK

Harvey J. Sugerman, M.D., James L. Tatum, M.D.
Jerry I. Hirsch, Pharm.D., and Alfred M. Strash, Ph.D.

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20. ABSTRACT (Continue on reverse side if necessary and identify by block number)

Computerized scintigraphy, employing the gamma camera, has been used in this contract to study the dynamics of the pulmonary capillary membrane leak of ^{99m}Technetium-tagged human serum albumin (Tc-HSA). We first studied a number of agents in an attempt to prevent oleic acid-induced pulmonary microvascular injury. Following a series of five control dogs, five dogs each were studied with each of the following agents: methylprednisolone (30 mg/kg) ibuprofen (12 mg/kg), the superoxide radical scavenger, MK-447 (4 mg/kg), and,

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20. Abstract (Continued)

in three dogs, calcium gluconate (140 mg/kg). None of these agents was able to alter the rise in lung:heart radioactivity ratio following oleic acid injury. In another study, we have administered 0.1 N hydrochloric acid, 2 ml/kg, into the trachea of dogs in the right lateral decubitus position and have found an acute and reproducible "slope of injury" similar to that seen with oleic acid. Neither pharmacologic doses of methylprednisolone (30 mg/kg) or ibuprofen (12.5 mg/kg) were able to alter the SI in 5 dogs each when given 5 minutes prior to instillation of 2 ml/kg 0.1 N HCl into the right mainstem bronchus. Infusion of prostacyclin (PGI_2), 0.3 mcg/kg/min, produced a 50% increase in the slope index.

A septic pig model was developed for study of bacterially induced ARDS. Following the i.v. infusion of 5×10^9 Pseudomonas organisms at 0.3 ml/20 kg/min, a marked rise in SI was seen. Control animals, infused with saline alone, had no rise in SI. Thromboxane and 6-keto-$PGF_{1\alpha}$ were measured, as well as SI and extra-vascular lung water (EVLW) using the thermal-cardiogreen technique, pulmonary artery pressure (PAP), systemic arterial pressure (SAP) and cardiac output (CO). The effects of ibuprofen (12.5 mg/kg) given at 20 and 120 minutes after Pseudomonas (P) was studied. At 3 hours Pseudomonas produced marked ($\overline{p<0.05}$) increases in PAP, TxB_2, 6-keto $PGF_{1\alpha}$, EVLW and SI with decreases in PaO_2, CO and SAP. I caused a rapid clearing of TxB_2 and 6-keto $PGF_{1\alpha}$ associated with a transient decrease in PAP; PaO_2 was considerably improved compared to Pseudomonas; however, CO, SAP, EVLW and SI were unaffected. In conclusion, prostaglandin blockage temporarily ameliorated the pulmonary hypertension and markedly improved oxygenation in this porcine septic ARDS model, but failed to alter increased permeability, confirming other studies that the increased pulmonary shunt in ARDS is not totally dependent upon capillary leak. The next study was designed to examine a combination of other possible mediators of ARDS. Ibuprofen (I) 12.5 mg/kg, methylprednisolone (M) 30 mg/kg, cimetidine (C) 150 mg, diphenhydramine (D) 10 mg/kg and ketanserin (K) 0.2 mg/kg (Poly 5) were given i.v. at 20 and 120 minutes after Pseudomonas. Poly 5 prevented the fall in PaO_2 and CI and the early, but not the late, rise in PAP and maintained SAP at control levels until three hours post Pseudomonas. Multiagent pharmacological blockage of several possible mediators of septic ARDS (arachidonic acid metabolites, histamine H_1 and H_2 and serotonin receptors) were effective in ameliorating the hypoxemia, the marked increase in capillary permeability and cardiovascular deterioration associated with i.v. Pseudomonas.

Septic-induced ARDS and multi-system organ failure are probably secondary to the systemic release of several mediators of inflammation, treatment will probably require a combination of anti-inflammatory agents. This should impact significantly on the mortality and morbidity of septic complications in traumatized combat soldiers.

Summary

Computerized scintigraphy, employing the gamma camera, has been used in this contract to study the dynamics of the pulmonary capillary membrane leak of ^{99m}Technetium-tagged human serum albumin (Tc-HSA). In preliminary canine studies, the severity of an oleic acid-induced albumin leak was proportional to the slope of lung:heart radioactivity ratio and was more sensitive than arterial blood gases or standard chest roentgenograms. We have called this rising ratio the "slope of injury" or "slope index" (SI). In the first 1-1/2 years of this contract, we studied a number of agents in an attempt to prevent oleic acid-induced pulmonary microvascular injury. Following a series of five control dogs, five dogs each were studied with each of the following agents: methylprednisolone (30 mg/kg), ibuprofen (12 mg/kg), the superoxide radical scavenger, MK-447 (4 mg/kg), and, in three dogs, calcium gluconate (140 mg/kg). Each of these agents was given five minutes prior to administration of oleic acid (0.05 ml/kg). None of these agents was able to alter the rise in lung:heart radioactivity ratio following oleic acid injury. In another study, we have administered 0.1 N hydrochloric acid, 2 ml/kg, into the trachea of dogs in the right lateral decubitus position and have found an acute and reproducible "slope of injury" similar to that seen with 0.05 ml/kg oleic acid. Neither pharmacologic doses of methylprednisolone (30 mg/kg) or ibuprofen (12.5 mg/kg) were able to alter the SI in 5 dogs each when given 5 minutes prior to instillation of 2 ml/kg 0.1 N HCl into the right mainstem bronchus. Infusion of prostocyclin (PGI_2), 0.3 mcg/kg/min, produced a 50% increase in the slope index. Methylprednisolone or ibuprofen failed to prevent the rise in pulmonary artery pressure or pulmonary vascular resistance or deterioration in PaO_2 following HCl instillation. PGI_2, as expected, decreased systemic arterial pressure and

preserved cardiac output following HCL. It was presumed that the increased rate of protein leak following the infusion of PGI_2 was due to preservation of flow to the injured pulmonary microvasculature.

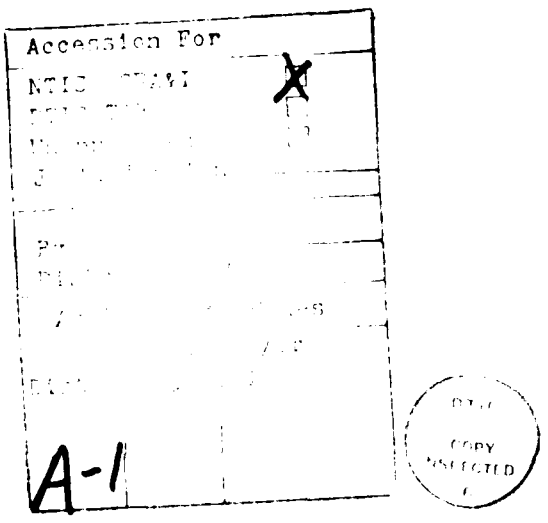
A septic pig model was developed for study of bacterially induced ARDS. Following the i.v. infusion of 5×10^9 Pseudomonas organisms at 0.3 ml/20 kg/min, a marked rise in SI was seen. Control animals, infused with saline alone, had no rise in SI. Thromboxane and 6-keto-$PGF_{2\alpha}$ (the stable metabolite of prostacyclin) were measured, as well as SI and extra-vascular lung water (EVLW) using the thermal-cardiogreen technique in pigs following i.v. pseudomonas, pulmonary artery pressure (PAP), systemic arterial pressure (SAP) and cardiac output (CO). The effects of ibuprofen (12.5 mg/kg) given at 20 and 120 minutes after Pseudomonas (P) was studied.

At 3 hours Pseudomonas produced marked ($p<0.05$) increases in PAP (18 ± 7 to 46 ± 4 mmHg). TxB_2 (471 ± 513 to 9216 ± 3615 pg/ml), 6-keto $PGF_{1\alpha}$, EVLW (6.4 ± 1.4 to 14.6 ± 5.7 ml/kg), and SI (0.4 ± 0.2 to $1.7 \pm 0.5 \times 10^{-3}$ U/min) with decreases in PaO_2 (214 ± 47 to 101 ± 41 torr), CO and SAP. I caused a rapid clearing of TxB_2 and 6-keto $PGF_{1\alpha}$ associated with a transient decrease in PAP; PaO_2 was considerably improved compared to Pseudomonas; however, CO, SAP, EVLW and SI were unaffected. In conclusion, prostaglandin blockage temporarily ameliorated the pulmonary hypertension and markedly improved oxygenation in this porcine septic ARDS model, but failed to alter increased permeability, confirming other studies that the increased pulmonary shunt in ARDS is not totally dependent upon capillary leak. Because ibuprofen prevented hypoxemia, but failed to alter cardiovascular deterioration and increased capillary permeability in Pseudomonas-induced porcine ARDS, the next study was designed to examine a combination of other possible mediators of ARDS. Ibuprofen (I) 12.5 mg/kg, methylprednisolone (M) 30 mg/kg, cimetidine (C) 150 mg,

diphenhydramine (D) 10 mg/kg and ketanserin (K) 0.2 mg/kg (Poly 5) were given i.v. at 20 and 120 minutes after Pseudomonas.

Pseudomonas again produced a fulminant ARDS with significant ($p<0.05$) decreases in PaO_2, CI, SAP and significant ($p<0.05$) increases in PAP, EVLW and SI when compared to C. Pseudomonas produced a significant fall in serotonin which was unaffected by poly 5. Poly 5 prevented the fall in PaO_2 and CI and the early, but not the late, rise in PAP and maintained SAP at control levels until three hours post Pseudomonas. Multiagent pharmacological blockage of several possible mediators of septic ARDS (arachidonic acid metabolites, histamine H_1 and H_2 and serotonin receptors) were effective in ameliorating the hypoxemia, the marked increase in capillary permeability and cardiovascular deterioration associated with i.v. Pseudomonas.

Septic-induced ARDS and multi-system organ failure are probably secondary to the systemic release of several mediators of inflammation, treatment will probably require a combination of anti-inflammatory agents. This should impact significantly on the mortality and morbidity of septic complications in traumatized combat soldiers.



Foreword

In conducting the research described in this report, the investigators adhere to the "Guide for the Care and Use of Laboratory Animals," prepared by the Committee on Care and Use of Laboratory Animals of the Institute of Laboratory Animal Resources, National Research Council (DHEW Publication No. NIH 78-23, Rev. 1978).

Table of Contents

A. Problem

The acute respiratory distress syndrome (ARDS) is an ill-defined disorder with multiple etiologies which usually requires mechanical ventilation. Combat soldiers acquire this disorder from direct lung contusion, burn inhalation injury, inhalation of toxic substances as a result of chemical warfare, aspiration, multiple transfusions, as a complication of sepsis, etc. The National Heart, Lung and Blood Institute, Division of Lung Diseases, Task Force on Research in Respiratory Diseases estimated that 150,000 cases occur each year (1). Many of these are young, previously healthy persons. The overall mortality is impossible to assess, but must be quite high. Ninety-one percent of the 90 patients enrolled in the Extra-Corporeal Membrane Oxygenator study (ECMO), most of whom probably had ARDS, died (1). Data from the nine centers participating in the ECMO study showed that more than 75% of the 600 patients receiving inspired oxygen concentration (FiO_2) greater than 50% died (1). From 1973 to 1976, 119 patients were admitted to the Respiratory and Surgical Intensive Care Units of San Francisco General Hospital with a diagnosis of ARDS (7% of all their intensive care unit admissions) and of these, 53% died (1). Although the majority of these deaths were not solely due to respiratory failure, this probably affected their morbidity and mortality.

B. Background

The initial pathophysiologic event in ARDS is thought to be a leak in the pulmonary capillary membrane. This leads to an increase in pulmonary interstitial water and protein which is then removed by the pulmonary lymphatics. If the leak exceeds the lymphatic capacity, which can increase flow by a factor of 20, pulmonary interstitial edema occurs. When the interstitial compartment reaches a critical volume and pressure, the alveolar space abruptly fills. This causes an inhibition, or wash-out, of surfactant, which will

produce alveolar collapse and a reduced FRC. A ventilation-perfusion (V/Q) mismatch develops, with right-to-left pulmonary shunting and arterial hypoxemia.

There is a need for an accurate, sensitive, reproducible and noninvasive technique to measure the severity and duration of pulmonary capillary leakage in patients with ARDS. This will permit improvement in both the early diagnosis of this pathological condition as well as the objective evaluation of therapeutic interventions.

Attempts to document and quantitate the leakage of water and protein through the pulmonary capillary membrane have been fraught with frustration. Previous studies have been directed at attempts to measure pulmonary extravascular lung water (EVLW) using isotopic indicator dilution techniques. These methods, based on studies by Chinard and Enns (2) have been shown by Korsgren et al. (3) and Marshall et al. (4) to be inaccurate, since they are flow dependent. A thermodilution-indocyanine green dye technique has also been applied to measure EVLW (5,6). This method does not appear to be as sensitive to changes in cardiac output as the isotopic indicator dilution techniques because of the much greater diffusability of the thermal indicator (7,8). The method provides a static estimate of extravascular lung water and must be repeated frequently to determine the dynamics of the leak. Brigham et al. (9) have developed a technique requiring the measurement of concentration-time curves for four radioactive agents (^{51}Cr-erythrocytes, ^{125}I-albumin, ^{14}C-urea, and ^{3}H-water) from which the extravascular lung water and the ^{14}C-urea permeability-surface area product is calculated. This technique would also provide a static estimate of extravascular lung water, seems cumbersome to use, and would also be affected by alterations in pulmonary vascular recruitment. Using a gamma probe technique, Gorin et al. (10) demonstrated a leak of ^{113m}Indium transferrin from the chest of sheep following the intravenous injection of Pseudomonas

aeruginosa bacteria. The intensity of radioactive counts correlated with the directly measured accumulation of this isotope in lung lymph. The position and aim of the gamma probe would have to be unaltered for the technique to be reproducible. It requires gamma well-counter analysis of serum samples which may limit widespread clinical utilization.

Gamma Scintillation Camera Technique: In studies from our laboratory, the computerized gamma camera was able to record and quantitate the pulmonary capillary membrane leak of ^{99m}technetium-tagged human serum albumin (Tc-HSA) in dogs following intravenous oleic acid (11,12). This technique compares the change in radioactivity over the lung to that over the heart with the construction of a lung:heart radioactivity ratio.

Radioactive Tc-HSA distributes within the whole body blood pool after intravenous injection and remains essentially within the vascular compartment. Its distribution within the body can be imaged with the gamma camera. Using the computerized gamma camera, data are collected at one second intervals for 60 seconds and then at one-minute intervals for the duration of the study. During the initial pass of the radiopharmaceutical, it is possible to define the lungs and the heart anatomically for subsequent computer analysis and construction of lung:heart radioactivity ratios. This ratio remains constant unless a pulmonary microvascular membrane injury is present when a rising ratio is present. We have called this rising ratio the "slope index" (SI).

In previous canine oleic acid studies, we have found that the SI was proportional to the severity of injury and was more sensitive than either arterial blood gas analysis or standard chest roentgenograms (12). Using this method in the oleic acid model, it was found that the leak of Tc-HSA was much greater than the leak of ^{99m}Tc-tagged RBC's (13), that PEEP did not alter the rate of pulmonary capillary protein leak (14), that altered pulmonary vascular

recruitment did not produce a rising radioactivity ratio following hemodynamic equilibration (15) and that multiple doses of Tc-HSA were associated with reproducible SI's over six hours following oleic acid administration (16). The method is noninvasive and has been used clinically to determine the severity and duration of non-cardiogenic pulmonary edema (17).

C. Approach to the Problem

The animals were anesthetized, intubated and placed beneath a Pho-Gamma IV scintillation camera fitted with a low energy, parallel hole collimator. Data were collected on "floppy disc" using a DEC mobile gamma acquisition system and transferred to a DEC medical computer for the determinations of regions of interest. Data were collected at one frame per second for 60 seconds following the Tc-HSA and then at one frame per minute for the duration of the study. Lung:heart radioactivity ratios were performed on a Xerox Sigma-5 computer. SI's were calculated from 15 to 45 minutes following administration of Tc-HSA. Animals subsequently found to have a significant SI during the control period were presumed to have a primary pulmonary illness (i.e., viral pneumonia) and were deleted from the study group. Intravascular pressures were measured with a Brush-Gould, Model 2400, 4-channel recorder utilizing a Statham strain-gauge transducer. Cardiac outputs were measured with a Kim-Ray, Model 3500 E thermodilution cardiac output computer.

1. Sheep Studies

Computerized gamma scintigraphy has compared favorably to wet to dry lung weight ratios, alveolar epithelial membrane permeability, canine lymph flow, standard radiography and light microscopy for the measurement of pulmonary microvascular permeability to albumin. The effects of altered pulmonary vascular recruitment and positive end-expiratory pressure on the scintigraphic

lung:heart radioactivity have also been studied. Currently, measurement of volume and protein concentration of lymph from the right caudal efferent lymph duct of sheep is the accepted model for the study of pulmonary permeability edema (18). It is, therefore, necessary to compare our scintigraphic technique to this model. We believed that an increased pulmonary microvascular permeability, associated with an increased lung lymph flow and protein concentration would be accurately detected by alterations in the scintigraphic "slope index." In addition, it should be possible to quantitate the amount of technetium-tagged albumin appearing in the lung lymph using a "well" counter. Several attempts were made to scan awake sheep suspended beneath the gamma camera in the upright position. However, the sheep could not be kept immobilized nor were posterior images adequate for accurate lung:heart radioactivity ratios. Therefore, the sheep were anesthetized, intubated, and placed in the supine position for anterior imaging and ventilated at a tidal volume of 20 ml/kg with 50% O_2 and 5 cm PEEP.

Initially, the sheep were subjected to thoractomy and right caudal thoracic lymph duct cannulation shortly before scanning and administration of endotoxin, the first pertubation we planned to investigate. However, sheep had very high SI's during the "control" period (see Results). These data led us to hypothesize that the "acute" lung lymph sheep model was associated with significant pulmonary injury, which would make it difficult to interpret a pertubation such as endotoxin. We, therefore, elected to scan five anesthetized sheep for one hour following 10 mCi Tc-HSA, perform a thoracotomy and right caudal thoracic lymphatic duct cannulation (which is associated with retraction of the right lung for 30 to 60 minutes) close the chest and re-expand the lung, inject another 10 mCi Tc-HSA and scan again for one hour. These results were compared to two groups of five sheep each who were either scanned before and after

thoracotomy only, without lung retraction or lymph duct cannulation or sheep who underwent anesthesia alone.

2. Canine Studies

Dogs weighing approximately 20 kg, were anesthetized with 30 mg/kg sodium pentobarbital, intubated, and ventilated at a tidal volume of 20 ml/kg with 50% O_2 and 5 cm H_2O positive end-expiratory pressure (PEEP).

a. Oleic Acid Injury: This model was chosen because of our extensive previous experience with it as well as the possible relationship of ARDS to free fatty acids in pancreatitis and the traumatic pulmonary fat embolism syndrome. We attempted to block the rising SI seen with oleic acid pulmonary microvascular injury with the following agents administered to five dogs each five minutes prior to 0.05 ml/kg oleic acid: methylprednisolone (30 ml/kg), the non-steroidal anti-inflammatory prostaglandin blocker, ibuprofen (12 mg/kg), the superoxide radical scavenger, MK-447 (4 mg/kg), and calcium gluconate (140 mg/kg). If any agent proved effective, we planned to administer it at various time intervals following oleic acid. However, no agent was found to reduce the rising SI after oleic acid (19).

b. HCl: Aspiration of gastric contents is a known complication of traumatized individuals as well as a well-recognized postoperative complication. In this study, eight dogs were anesthetized with sodium pentobarbital, intubated, ventilated at a tidal volume of 20 ml/kg, and placed beneath a computerized gamma camera. Two ml/kg 0.1 N HCl were instilled into the right mainstem bronchus of dogs placed in the right lateral decubitus position for 30 minutes. Second and third doses of 10 mCi Tc-HSA were given 1/2 hour and 1-1/2 hours after HCl, respectively. Five animals received HCl instillation alone and five animals each were pretreated with methylprednisolone (30 mg/kg), ibuprofen (12.5 mg/kg) 5 minutes prior to acid instillation or prostacyclin

(PGI_2) as a continuous infusion (0.3 mcg/kg/min) begun 30 minutes prior to HCl instillation and continued for 3 hours after administration of acid.

3. Porcine Studies

It is not surprising that injuries produced by intravenous oleic acid or transbronchial instillation of HCl were not prevented with corticosteroids or prostaglandin blockers, since these appear to be direct, non-mediated injuries to the pulmonary alveoli and microvasculature. It is quite probable that ARDS secondary to sepsis is, in part, mediated by the release of prostaglandins (22) and complement (23) and is associated with both leukocyte (24) and platelet (25) aggregation in the pulmonary microvasculature. Septicemia is also one of the leading causes for fulminant ARDS and might be, theoretically, ameliorated with pharmacologic agents. We have, therefore, studied a porcine Pseudomonas septicemia model which has been shown to be associated with a fulminant ARDS (26). We have compared the SI, EVLW (measured by the thermal-cardiogreen technique), thromboxane and 6-keto-$PGF_{2\alpha}$ production, pulmonary and systemic hemodynamics in pigs given a saline infusion to pigs given an infusion of 5 x 10^9 CFU Pseudomonas (PAO strain) organisms at a rate of 0.3 ml/20 kg/min.

D. Results

1. Sheep

a. Five sheep were found to have high SI's during the "control" period following thoracotomy and lymph duct cannulation. The effect of endotoxin administration in these animals could not be interpreted.

b. Eight sheep were studied prior to and following thoracotomy without lung retraction or lymphatic duct cannulation. Three animals had a high SI during the control period and were deleted from the study. The SI's in the remaining five sheep were normal during the control period. In two sheep

the SI was normal and in three the SI was > 1 x 10-3 U/min following thoracotomy. This suggests that thoracotomy with lung collapse and re-expansion alone may produce an increased albumin flux associated with an elevated SI. Re-expansion following pneumothorax has been shown to be associated with an increased lung water (27).

c. Seven sheep were studied prior to and following thoracotomy, lung retraction, and lymphatic duct cannulation. Two animals had a high SI during the control period and were deleted from the study. The mean control SI in the remaining five sheep was $0.6 \pm 0.6 \times 10^{-3}$ U/min. Following thoracotomy, lung retraction and lymphatic duct cannulation, the SI rose significantly ($p<0.01$) to $1.3 \pm 0.6 \times 10^{-3}$ U/min. The acute lymphatic duct cannulation model, therefore, appears to have a significant injury to the lung according to the scintigraphic pulmonary capillary protein leak technique. Similar results have been noted by other investigators (28).

2. Dogs

a. Oleic Acid Injury: Of 28 dogs studied, 5 dogs had an SI greater than 0.6×10^{-3} U/min during the control period and were deleted from the study. The following data were obtained (29):

Study Group	SI ($\times 10^{-3}$ U/min)
Control oleic acid (OA) 0.05 ml/kg	2.9 ± 1.0
Methylprednisolone (30 mg/kg) + OA	3.8 ± 1.9
Ibuprofen (12 mg/kg) + OA	3.8 ± 0.8
MK 447 (4 mg/kg) + OA	2.9 ± 0.3
Ca^{++} Gluconate (140 mg/kg) + OA	3.0 ± 0.4

Therefore, none of the agents tested were able to prevent scintigraphic evidence of an oleic acid pulmonary microvascular injury.

b. HCl Studies: Instillation of 2 ml/kg 0.1N HCl into the right mainstem bronchus of dogs produced an acute increase in pulmonary vascular resistance, from 164 $\pm$ 58 to 281 $\pm$ 67 dynes-sec-cm^{-5}, and SI, from 0.4 $\pm$ 0.4 to 2.0 $\pm$ 1.0, within 30 minutes which persisted for the next 90 minutes. These changes were not ameliorated with pretreatment, 5 minutes prior to acid instillation, with either 30 mg/kg i.v. methylprednisolone or 12.5 mg/kg i.v. ibuprofen (30). Infusion of PGI_2 at 0.3 mcg/kg/min produced a significant increase in both cardiac output and slope index following acid instillation over control values, but did not alter pulmonary artery pressure. The increased albumin flux as determined by the increased SI following PGI_2 was presumably due to increased flow to the injured pulmonary microvasculature.

3. Porcine Studies

Pigs, weighing 20 to 30 kg, underwent anesthesia with ketamine and pentobarbital (30 mg/kg), intubation, insertion of arterial and Swan-Ganz catheters. Control scintigraphic studies were obtained over 3 hours and the SI remained 0.4 x 10^{-3} U/min following 2 injections of Tc-HSA. Pigs were given an i.v. infusion of 5 x 10^9 CFU Pseudomonas organisms at a rate of 0.3 ml/20 kg/min. This was associated with a marked and sustained increase in pulmonary artery pressure (PAP), fall in cardiac index (CI) and systemic arterial pressure (SAP) and a significant fall in arterial oxygen tension. A marked increase in SI and thermal-cardiogreen EVLW were also noted. The increased SI preceded the rise in EVLW (31).

Ibuprofen (12.5 mg/kg), given 20 and 120 minutes after the Pseudomonas infusion was begun produced a marked and sudden fall in PAP. However, by 180 minutes the PAP had again risen toward the untreated value. The PaO_2 remained near control levels throughout the three-hour study period; however, ibuprofen had no effect on EVLW or SI. Nor did it alter the fall in CI and SAP.

Thus, ibuprofen prevented hypoxemia without altering increased permeability following Pseudomonas. This suggests that ARDS is not just secondary to a pulmonary capillary leak but more likely a result of bronchospasm (31).

In an attempt to improve other aspects of Pseudomonas injury unaffected by ibuprofen, a combination of several possible therapeutic agents was studied. These drugs were also given 20 and 120 minutes after the Pseudomonas infusion was begun and included: ibuprofen 12.5 mg/kg, methyl-prednisolone 30 mg/kg, the H_2 blocker, cimetidine 150 mg, the H_1 blocker, diphenhydramine 10 mg/kg, and ketanserin 0.2 mg/kg, a serotonin blocker.

Results: Pseudomonas produced a fulminant ARDS with significant ($p<0.05$) decreases in PaO_2, CI, SAP and significant ($p<0.05$) increases in PAP, EVLW and SI when compared to C. Pseudomonas produced a significant fall in serotonin which was unaffected by poly 5. Poly 5 prevented the fall in PaO_2 and CI and the early, but not the late, rise in PAP and maintained SAP and EVLW at control levels until three hours post Pseudomonas. The SI remained low in the poly 5 animals at 75 minutes but rose at 145 minutes to a level significantly greater than C but less than Ps.

	PaO_2 (torr)		EVLW (ml/kg)		SAP (mmHG)	
Time (min)	0	180	0	180	0	180
C	174 ± 9	203 ± 15	4.1 ± 0.9	4.3 ± 1.2	107 ± 8	128 ± 5
Ps	223 ± 15	93 ± 17*	6.8 ± 0.6	11.0 ± 2.2*	101 ± 6	71 ± 1*
I	230 ± 2	179 ± 5	5.7 ± 0.9	13.4 ± 2.0*	115 ± 5	85 ± 7*
Poly 5	206 ± 18	210 ± 17	7.5 ± 1.5	7.4 ± 0.4	110 ± 6	104 ± 11

*P<0.05 vs. 0 min or C; mean ± SEM

Conclusions: Multiagent pharmacological blockage of several possible mediators of septic ARDS (arachidonic acid metabolites, histamine H_1 and H_2 and

serotonin receptors) were effective in ameliorating the hypoxemia, the marked increase in capillary permeability and cardiovascular deterioration associated with IV Pseudomonas.

E. Conclusions

The sheep studies showed a significant rise in SI following thoracotomy, lung retraction and lymphatic duct cannulation. Furthermore, thoracotomy alone produced a rise in SI in several animals. Therefore, the acute lung lymph cannulation model appears to be associated with a significant pulmonary microvascular injury.

The oleic acid dog studies showed that none of the agents tested (methylprednisolone, ibuprofen, MK-447 or calcium gluconate) were able to alter the scintigraphic evidence of oleic acid induced pulmonary microvascular injury when given prior to the administration of oleic acid. Several studies have suggested that methylprednisolone will prevent oleic acid injury (33,34); whereas, other studies have found no effect on lung water with this medication (35). The data from our study are quite clear that pharmacologic doses of methylprednisolone had no effect on the pulmonary capillary leak of albumin following oleic acid injury.

Similar negative results were seen with pretreatment of dogs following acid instillation with either pharmacologic doses of methylprednisolone or ibuprofen. Infusion of prostacyclin (PGI_2) at 0.3 mcg/kg/min produced an increase in SI compared to control dogs as well as an increase in cardiac output. Presumably the increased albumin flux noted with PGI_2 was secondary to an increased flow across the injured pulmonary microvasculature.

Studies in a porcine model of Pseudomonas-induced septic ARDS reproduced most of the pathologic changes noted in patients wih septic ARDS including progressive hypoxemia, pulmonary hypertension, and increased capillary

permeability. Ibuprofen markedly ameliorated the rise in PAP, but this was a transient phenomenon. Ibuprofen also prevented hypoxemia but had no effect on increased pulmonary capillary permeability. A combination of anti-inflammatory agents was, however, able to block the increased permeability associated with Pseudomonas infusion. Septic-induced ARDS is probably secondary to the release of multiple inflammatory mediators, treatment will probably require a combination of therapeutic agents.

F. Recommendations

Pulmonary contusion and the acute respiratory distress syndrome were major complications of the Korean conflict, where it was termed the "Traumatic Wet Lung Syndrome," and Viet Nam, where it was called "Da Nang Lung" (36-39). A major cause of death following trauma is sepsis which leads to multi-system organ failure. In a 1977 study by Eiseman et al. (40), 42 patients were found with multiple system organ failure of whom 29 were septic and 19 died. Average hospital costs, excluding physician's fees, were conservatively estimated at $21,000. Fry et al. (41) found that sepsis was the most common cause of multiple system organ failure and this was the most common fatal expression of severe sepsis. Of 553 consecutive emergency surgical patients, 55 died post-operatively. Of these, infection was the cause of death in 32. Thirty-four of 123 septic patients had multiple system organ failure. The lung is the most common organ injured in the septic patient.

There has not previously been an accepted, objective, clinical measurement of a pulmonary capillary protein leak. The thermal-cardiogreen method will measure the amount of lung water that has already leaked (5-8), but does not determine if an active protein leak is occurring. The method of computerized pulmonary gamma scintigraphy is conceptually simple, noninvasive,

reproducible and should permit the objective evaluation of the presence and duration of ARDS, and its response to therapeutic interventions. Should a treatment be found to be efficacious, the severity of ARDS should be reduced. This would include a reduced need for prolonged mechanical ventilation as well as a decrease in the attendant morbidity associated with the leakage of proteinaceous fluid with its associated bacterial pneumonia and pulmonary fibrosis. This should improve the resuscitation of the severely injured combat soldier so that he can be returned sooner to active duty or be more rapidly rehabilitated.

The scintigraphic results suggest that the acute sheep lung lymph model may be an unreliable model since it is associated with severe lung injury prior to any perturbation. No agents yet studied were able to reduce the increased albumin flux in animals given i.v. oleic acid or transbronchial instillation of HCl. These results are not surprising since both oleic acid and HCl probably produce a direct microvascular injury and do not act through the release of noxious intermediates. It is more likely that the injury following bacteremia is produced by the release of intermediates such as prostaglandins, complement activation, and super-oxide radicals. The Pseudomonas porcine model holds promise for studying various blocking agents in modifying the rate of albumin flux in sepsis. Preliminary studies suggest that a combination of anti-inflammatory agents will be required and will be effective in reducing the severity of injury.

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Abstracts and Presentations

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1. Sugerman, H.J., Blocher, C.R., Hirsch, J.I., Tatum, J.L., and Strash, A.M.: Failure of methylprednisolone, ibuprofen and MK-447 to alter scintigraphic measurement of oleic acid permeability edema. Circ. Shock 9:197(A), 1982. Presented at Fifth Annual Conference on Shock, Smuggler's Notch, Vermont, June, 1982.

2. Sugerman, H.J., Blocher, C.R., Hirsch, J.I., Tatum, J.L., and Strash, A.M.: Failure of methylprednisolone, ibuprofen and MK-447 to alter scintigraphic measurement of oleic acid permeability edema. Circ. Shock 9:197(A), 1982. Presented at The Association of Academic Surgery, San Diego, California, November, 1982.

3. Sugerman, H.J., Buchman, S.R., Tatum, J.L., et al.: Failure of methylprednisolone, ibuprofen or prostacyclin (PGI_2) to reduce HCl induced pulmonary albumin leak in dogs. Presented to the Society of University Surgeons Meeting, Chicago, Illinois, February, 1984.

4. Lee, C.C., Sugerman, H.J., Tatum, J.L., et al.: Effects of ibuprofen on a pig pseudomonas ARDS model. Presented at The Association for Academic Surgery, Cincinnatti, Ohio, November, 1985.

5. Harvey, C.F., Sugerman, H.J., Tatum, J.L., et al.: Ibuprofen and methylprednisolone in a pig pseudomonas ARDS model. Presented at the Ninth Annual Conference on Shock, The Shock Society, Scottsdale, Arizona, June, 1986.

6. Sugerman, H.J., Sielaff, T.D., Tatum, J.L., et al.: Multi-agent pharmacological treatment of porcine pseudomonas ARDS. To be presented at the American Association for Surgery of Trauma Meeting, Honolulu, Hawaii, September, 1986.

7. Sielaff, T.D., Sugerman, H.J., Tatum, J.L., et al.: Multi-agent pharmacological treatment of porcine pseudomonas ARDS. To be presented at the Surgical Forum, American College of Surgeons Meeting, New Orleans, Louisiana, October, 1986.

Articles

1. Sugerman, H.J., Blocher, C.R., Hirsch, J.I., et al.: Failure of various agents to decrease oleic acid pulmonary albumin leak. J. Surg. Res. 34:156, 1983.

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