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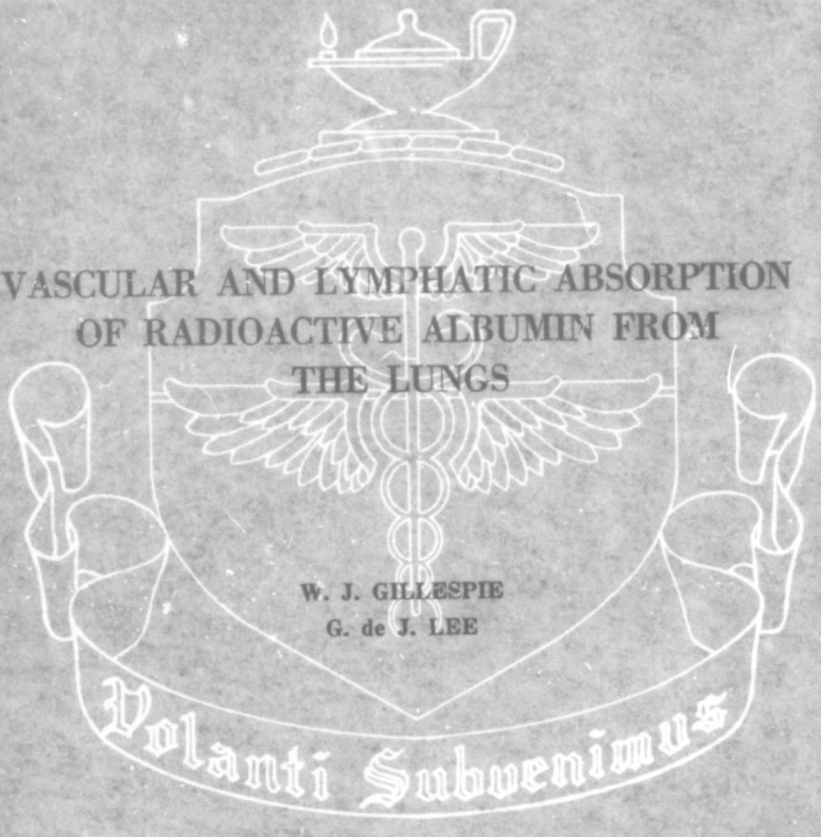

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VASCULAR AND LYMPHATIC ABSORPTION OF RADIOACTIVE ALBUMIN FROM THE LUNGS

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FOREWORD

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The experiments reported herein were conducted according to the "Principles of Laboratory Animal Care" established by the National Society for Medical Research.

This report has been reviewed and is approved.

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ABSTRACT

A study was made of the absorption of radioactive iodinated serum albumin (RISA) into the plasma after its instillation into the lungs of normal dogs and dogs with pulmonary edema in an effort to develop an indirect method for measuring lung lymph flow.

Absorption of protein from the lung alveoli was initially slower in dogs with pulmonary edema compared with normal dogs; however, by 24 hours, the percentage of the instilled dose of RISA present in the plasma was higher in the dogs with pulmonary edema than in the normal dogs.

Absorption of RISA occurred equally well from the lung into the plasma whether or not the pulmonary artery to the lung was occluded, thus indicating that protein absorption could take place by means of the bronchial circulation.

Bronchopneumonia was associated with a greatly accelerated entry of RISA from the lung into the plasma, showing a large increase in alveolar and capillary permeability in pneumonia.

Measurements of changes in plasma concentration of RISA after its instillation into the lung cannot be used to assess the rate of lymph formation in the lung since most of the protein is absorbed directly into the plasma both in normal animals and in those with pulmonary edema.

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VASCULAR AND LYMPHATIC ABSORPTION OF RADIOACTIVE ALBUMIN FROM THE LUNGS

I. INTRODUCTION

In man the development of pulmonary edema, manifest in its severest form by the production of copious frothy sputum, is invariably associated with an acute rise in the left atrial and pulmonary venous pressures. Its clinical manifestation, however, is not an invariable feature in patients whose left atrial pressures are chronically elevated to levels which often exceed the estimated plasma oncotic pressure. This is especially true in patients with chronic venous hypertension associated with mitral stenosis. It has been postulated that in these latter circumstances the lung lymphatics become dilated and are able to remove sufficient fluid transudate from the interstitial spaces of the lungs to prevent the development of clinically overt edema, and indirect evidence to support this view in patients with mitral stenosis has been obtained by Rossall and Gunning (10).

In addition to possible changes in lung lymphatic behavior, it is likely that the Starling hypothesis concerning the movement of fluid and solutes between the interior of blood vessels and the interstitium of the lung is too simple a concept. Evidence is accumulating which indicates that the transport of substances across the capillary wall is accomplished by two main systems. The first system permits free movement of small molecules with consequent rapid exchange of solutes but restricts movement of large molecules, such as the plasma proteins, responsible for maintaining fluid equilibrium across the capillary wall in the manner described by Starling. In addition, a second transport system permits the slow passage of large molecules across the capillary wall, either as a passive, but restricted, diffusion process or by some active mechanism (8). It is the latter process which may be concerned with the local distribution of antibodies, hormones, and other large molecular substances within the body. Drinker and Field (5) had reasoned that the compositions of lymph and interstitial fluid were identical; Warren and Drinker (18) had shown that lung lymph was removed almost exclusively by the right lymphatic duct. Courtice and Simmonds (4) concluded that protein, after intratracheal injection, was absorbed from the terminal respiratory tree by lymphatic pathways alone.

Because of the intimate relationship between the alveoli and capillaries within the lungs, it seemed useful to re-examine the permeability of membrane barriers to large molecules in the lung microcirculation and at the same time to re-examine the lung lymphatic circulation by measuring the rate of entry of radioiodinated albumin into the systemic circulation after its instillation into the lung alveoli.

We report here some preliminary studies in dogs to test this possibility in normal animals and in animals with chronic pulmonary vascular congestion. We hoped that similar methods could be applied later to man in an effort to study possible regional changes in lymph flow and capillary permeability within the lungs, which may be taking place during heart failure.

II. SUMMARY

The absorption of radioactive iodinated serum albumin into the plasma after its instillation into the lungs has been studied in a normal group of dogs and in a group of dogs with mild chronic congestive pulmonary edema.

Absorption of protein from the lung alveoli normally occurred slowly and reached a plateau of concentration in the plasma. In normal animals, 6.6% was present in the plasma in 5 hours, 20.25% in 13 hours, and 20.95% in 24 hours. In dogs with pulmonary edema, the absorption of protein in the plasma was significantly slower, initially; 4.6% was present in the plasma in 5 hours and 12.47% in 13 hours. By 24 hours, however, 37.44% of the instilled dose of radioactive protein was present in the plasma.

These changes may be due to both an increase in the size of the pulmonary interstitial space as well as changes in lung capillary and alveolar membrane permeability in chronic congestive pulmonary edema.

Absorption of instilled radioactive albumin from the lung into the plasma occurred equally rapidly whether or not the pulmonary arterial inflow to that lung was prevented by a balloon catheter inflated in the appropriate pulmonary artery. This indicated that protein absorption from the lung could take place by means of the bronchial circulation.

Bronchopneumonia was associated with a greatly accelerated entry of labeled albumin from the lung alveoli into the plasma, indicating a large increase in alveolar and capillary membrane permeability in pneumonia.

Cannulation and total drainage of the lymph leaving the lungs through the right lymphatic duct and also of lymph leaving the thoracic duct did not prevent most of the protein absorbed from the lung alveoli from entering the plasma directly, both in normal animals and in animals with chronic congestive pulmonary edema.

Measurement of changes in plasma concentration of radioactive albumin after its instillation into the lung, therefore, cannot be used to yield information regarding the rate of lymph formation in the lung.

III. MATERIAL AND METHODS

Effect of pulmonary vascular congestion on the absorption of RISA from the lung

Mongrel dogs, weighing between 16 and 24 kg., were lightly anesthetized with intravenous pentobarbitone (27 mg./kg.). The trachea was intubated with a cuffed Magill tube, and the dogs were permitted to breathe naturally, lying on the right side.

A calibrated syringe was used to inject 50 μc. of 131-iodinated human serum albumin (RISA) in 5 ml. of normal saline solution directly into the pulmonary alveoli through a fine radio-opaque catheter (external diameter, 1.5 mm.) whose tip was wedged into a distal bronchus of the right lower lobe of the lung. The position of the catheter was confirmed fluoroscopically. The RISA was injected slowly, and after 2 minutes the catheter was removed. At no time did the dogs cough. The radioactive contents remaining in the injection syringe and catheter were counted to estimate the net dose of RISA administered to the lung. Thyroid uptake of radioiodine was blocked in each animal by the administration of 5 ml. of Lugol's iodine solution to the dogs' drinking water on the 2 days immediately before the study. The iodinated albumin solution contained approximately 4 mg. albumin per 50-μc. dose and less than 2% radioactivity as free iodine.

Serial venous blood samples were obtained at regular intervals for 15 hours with a final sample taken at 24 hours. Plasma radioactivity measurements were made in 1-ml. samples, using a Picker gamma spectrometer and automatic counter. Plasma volume estimations were also made in each animal using T1824 and a modification of the method of Chinard and Eder (2); the optical densities of the acetone-extracted plasma samples were measured in a Beckman D.B. spectrophotometer at 620 A.

From these measurements it was possible to express the radioactivity absorbed into the plasma at each time interval as a percentage of the total 131-iodinated albumin dose administered to the lung.

Six normal animals were studied in this way. The results which were obtained were compared with results obtained from six dogs in whom chronic pulmonary edema was produced experimentally, using the method of Uhley et al. (16). In this procedure, three weeks before the lung studies were undertaken an aortocaval fistula was produced surgically distal to the origin of the renal veins. The size of the fistula was critical. If it was too large, the animals died from congestive cardiac failure in a few hours; if it was too small, the animals remained unaffected. By experience it was found that a fistula diameter corresponding to 0.5 mm./kg. body weight was usually satisfactory, but to further overload the circulation it was usually necessary to administer 25 mg. DOCA trimethyl acetate thrice weekly and to supplement the animals' food with 5 gm. sodium chloride per day.

Six dogs with systemic signs of congestive cardiac failure were studied. In no case was this severe, however. The average weight gain of the animals was 2.3 kg. (range, 1.8 to 3.2 kg.), and they all had dependent edema. At postmortem two animals had a small amount of ascites; two had a little free fluid in the pericardium; and three animals had small pleural effusions. The lungs were only slightly increased in weight (average lung weight to body weight ratio, 1:5; upper limit of normal, 1:4 (16)). Histologic section of the lungs in three animals showed an increase in the peribronchial spaces and enlarged peribronchial lymphatics.

In three of the dogs on which operations had been performed, pulmonary vascular pressures were measured by cardiac catheterization immediately before postmortem examination at the termination of the study. The results shown in table I confirm that the operative procedure had produced some elevation of the pulmonary arterial and pulmonary wedge pressures.

Effect of pulmonary blood flow on the absorption of RISA from the lung

In one normal animal a balloon catheter was placed in the right main pulmonary artery,

TABLE I

Weight gain and pulmonary vascular pressures in three dogs with chronic aortocaval fistulas and systemic edema

Dog No.	Weight gain (kg.)	Pressure (mm. Hg)		
		P.A.	P.C.	R.A.
2692	1.9	55/24	12	10
2914	3.2	30/15	14	3
3095	2.8	32/12	7	5

P.A. = Pulmonary arterial pressure.

P.C. = Wedge pulmonary arterial pressure.

R.A. = Right atrial pressure.

and the balloon was inflated to completely occlude the right main pulmonary artery; correct position of the catheter was confirmed fluoroscopically. RISA (50 μc.) in 5 ml. of normal saline was then instilled into the pulmonary alveoli of the right lower lobe of the lung in the manner already described. Hourly systemic venous blood samples were then taken for 14 hours. The occluding balloon in the right main pulmonary artery was removed at the end of the 7th hour of study. When the right main pulmonary artery was occluded by the balloon, the main pulmonary arterial pressure was 62/25 mm. Hg. The main pulmonary arterial pressure fell to 30/12 mm. Hg when the occluding balloon was removed.

Effect of pneumonia on the absorption of RISA from the lung

In one dog an aortocaval fistula was made of insufficient size to affect the animal. It recovered from its abdominal operation, but neither developed edema nor gained weight in spite of a salt-supplemented diet and DOCA trimethyl acetate injections. Six weeks later it developed severe distemper which did not respond to large doses of penicillin. On the 14th day of this illness 50 μc. RISA were instilled into the right lower lobe of the lung by means of a bronchial catheter in the manner already described. Half hourly systemic venous samples were taken for 2 hours. The dog

was then killed and a postmortem examination was made. The lungs showed confluent bronchopneumonia.

Amount of RISA entering the plasma from the lungs by way of the right lymph duct and thoracic duct

Lymph collections were made from the right lymph ducts and thoracic duct in one animal from the normal group and in two of the animals with chronic aortocaval fistulas. As soon as the 24-hour blood samples had been collected from the animals, they were re-anesthetized using intravenous pentobarbitone (27 mg./kg.); 5 ml. of 1% T1824 in water were instilled into the lung by means of a fine bronchial catheter in order to stain the lung lymphatics and to aid subsequent dissection. The thoracic duct was then exposed in the neck at its entry into the venous system at the junction of the internal and external jugular veins and was cannulated with a 1-mm. bore polyethylene tube.

Lymph from the right lymph ducts enters the venous system in the dog at several points in the vicinity of the junction of the right external jugular, internal jugular, and axillary veins. The area was carefully dissected without damaging the lymphatics, and a venous collection pocket was created and cannulated in the manner described by Uhley et al. (17). The lymph draining from each duct was measured by collection into the heparinized graduated centrifuge tubes.

In three other normal animals, the right lymph duct and thoracic ducts were cannulated and all the lymph collected in a similar manner during a control period and subsequent to the instillation of 50 μc. RISA in 5 ml. of saline into the right lower lobe of the lung by means of a fine bronchial catheter. Plasma and lymph specific activities were measured at hourly intervals and compared. The proportions of RISA administered to the alveoli which had entered the plasma, right lymph duct, and thoracic duct lymph were calculated.

In one animal (No. 41), the lymph collection was continued for 8 hours after the lung instillation of RISA. In a second animal, dissection was technically difficult and incomplete, and the animal died 4 hours after the commencement of the study. In a third animal, the right lymph duct pocket could not be produced in a manner which would yield bloodless lymph. Collection from the right lymph pocket was therefore abandoned, and the lymphatics entering the area were cauterized so that only lymph from the thoracic duct was collected in this case. It was deeply stained with Evans blue (T1824), thus indicating the lung lymph, prevented from its normal escape by way of the right lymph ducts, was able to enter the thoracic duct by way of anastomotic channels. One animal (No. 2914) with an aortocaval fistula and mild pulmonary congestion was also studied in a similar manner.

IV. RESULTS

Effect of pulmonary vascular congestion on the absorption of RISA from the lung

Table II and figure 1 show the quantities of radioactive iodinated human serum albumin (RISA) which appeared in the plasma at increasing intervals of time after its instillation in 5 ml. of saline into the alveoli and terminal bronchi of the right lower lobe of the dog's lung. The quantities appearing in the plasma, expressed as a percentage of the total dose of RISA instilled into the lungs of six normal dogs (group A, fig. 1), are compared with those obtained from six dogs with clinical, histologic, and hemodynamic evidence of mild chronic pulmonary vascular congestion (group B, fig. 1).

In the normal animals (group A), RISA slowly appeared in the plasma after its instillation in the lung; 1.56% $\pm$ 0.16% of the lung dose had entered the plasma at the end of 1 hour. This had increased to 6.60% $\pm$ 0.57% at the end of 5 hours, and a plateau of concentration had been reached in the plasma by 13 hours, accounting for 20.25% $\pm$ 3.96% of the original protein administered to the lung. No significant further increase in plasma radioactivity took place subsequently, and autopsy studies in one dog showed that most of the

TABLE II

Percentage of radioactive iodinated human serum albumin (RISA) present in the plasma after its instillation in 5 ml. of saline into the lung alveoli

Subjects	Time after bronchial instillation (hr.)								
	1	3	5	7	9	11	13	15	24*
	Mean and S.E.								
Group A: Six normal dogs	1.56 ± .16	3.90 ± .13	6.60 ± .57	9.99 ± .93	14.20 ± 1.90	18.30 ± 2.81	20.25 ± 3.96	20.5 ± 4.05	20.95 ± 3.81
Group B: Six dogs with fistulas	1.15 ± .15	2.66 ± .28	4.60 ± .12	6.49 ± .59	8.95 ± 1.02	10.40 ± .53	12.47 ± 1.56	16.95 ± 1.83	37.44 ± 10.37
P	.1	.02	.01	.01	.01	.01	.01	.5	.15

*At 24 hours only three animals were studied in each group.

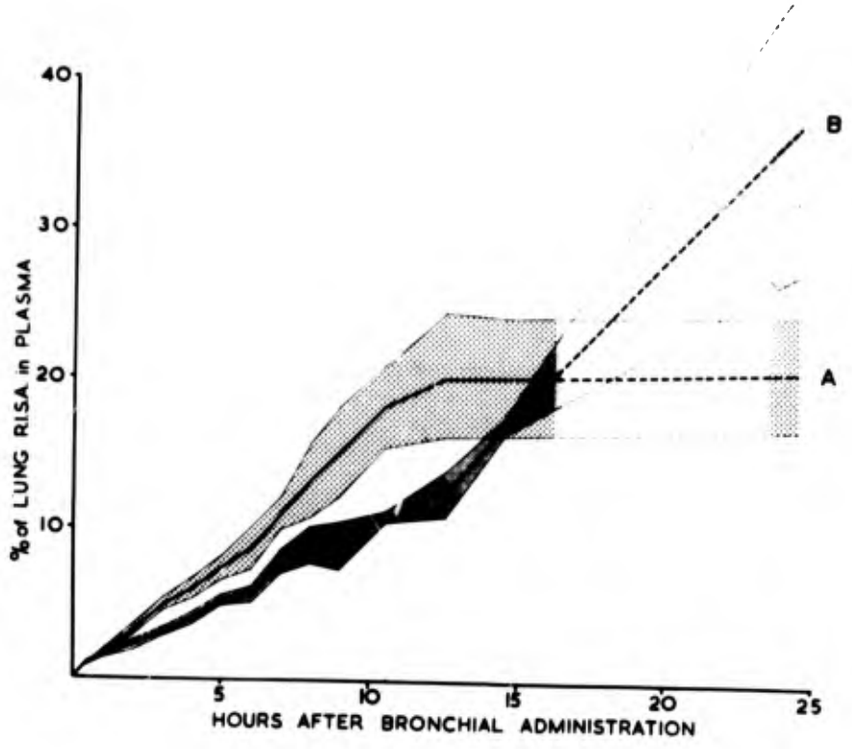

FIGURE 1

The percentage of RISA entering the plasma from the lung alveoli. Group A: Six normal dogs. Group B: Six dogs with aortocaval fistulas and pulmonary congestion. Shaded areas indicate the standard errors for each group.

remainder of the administered isotope was still present in the right lung, with appreciable amounts in the mediastinal lymph glands, liver, kidneys, and urine, but little in the gut contents.

The absorption of RISA into the plasma in the six animals with chronic aortocaval fistulas (group B) was significantly different from that of the normal group. The rate of entry of radioactive material into the plasma from the lungs was initially slower. At 5 hours, only 5.60% ± 0.12% of the lung dose was present in the plasma, and this had risen to only 12.47% ± 1.56% by the 13th hour. Unlike the normal group of animals, however, the plasma concentration of radioactivity continued to rise thereafter and never reached a plateau of concentration so that at the 24th hour, 37.44% ± 10.37% of the radioactive material initially placed in the alveoli was detectable in the plasma, compared with 20.95% ± 3.81% in the normal dogs at the same time interval.

Effect of pulmonary blood flow on the absorption of RISA from the lung

Figure 2 shows the results of a single study in which an occluding balloon was inflated in the main right pulmonary artery of a normal dog before instilling 50 μc. of RISA into the alveoli of the right lower lobe of the lung. The percentage of RISA entering the animal's plasma at serial intervals of time after its instillation into the lung was no different from that taking place in the normal group of animals in which pulmonary blood flow was unobstructed. Removal of the occluding balloon from the right pulmonary artery at the 7th hour of study made no appreciable difference to the rate of entry of RISA into the plasma, compared with the normal group.

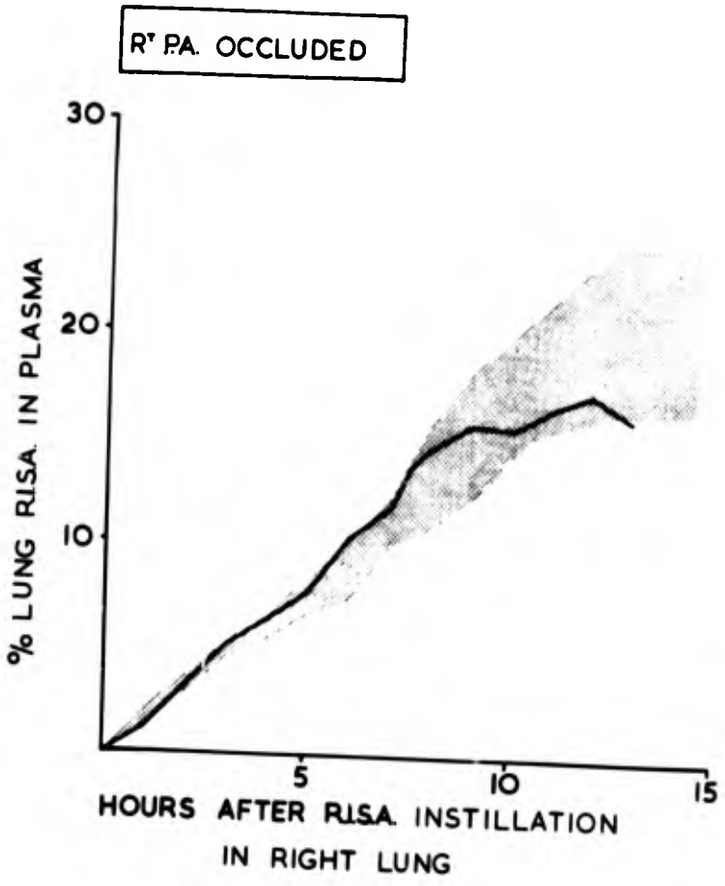

FIGURE 2

The percentage of RISA entering the plasma of a dog after instillation into the alveoli of the right lower lobe with the main pulmonary artery to the right lung occluded by a balloon catheter. The balloon was deflated at the 7th hour. The shaded area represents the results obtained from six normal animals previously studied (group A, fig. 1)

Effect of pneumonia on the absorption of RISA from the lung

Figure 3 shows the results of a study in one animal suffering from distemper with post-mortem evidence of extensive broncho-pneumonia. RISA (50 μc.) in 5 ml. of saline was instilled into the animal's right lower lobe by means of a fine bronchial catheter wedged into a peripheral bronchus. The saline contained 1% Evans blue, and the lung section showed extensive staining of alveoli and terminal bronchioles with spread of the dye through the pneumonic areas.

In contrast to the slow absorption of RISA into the plasma which took place in the normal animals, 26.1% of the dose of RISA instilled into the pneumonic dog's lung had entered the plasma in 2 hours.

Amount of RISA entering the plasma from the lungs by way of the right lymph duct and thoracic duct

Although many right lymph duct collections were attempted, technical competence was achieved in only a few. Table III shows the rate of lymph flow per hour from the right

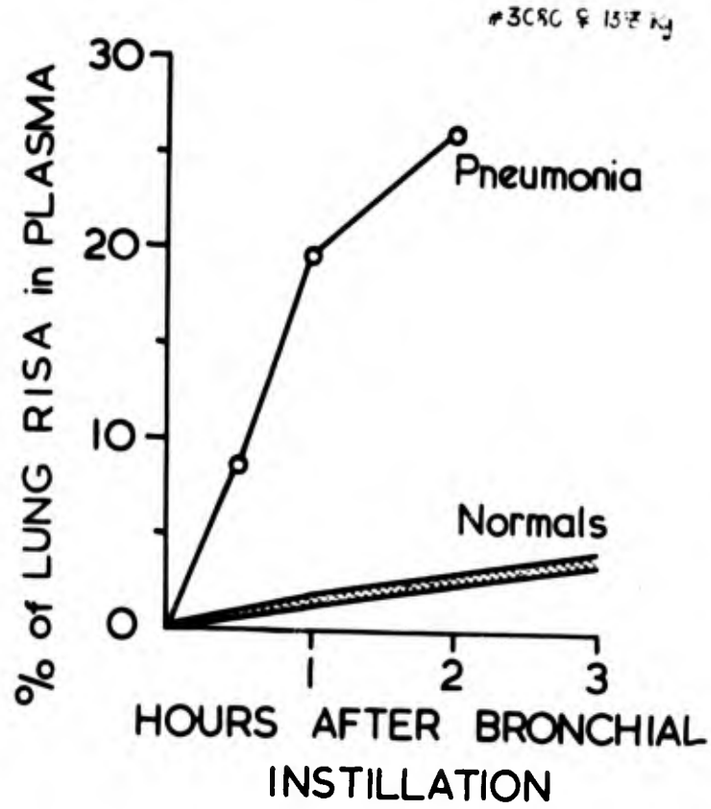

FIGURE 3

The percentage of RISA entering the plasma after its instillation into the lung of a dog with pneumonia, compared with the results obtained from six normal animals.

lymph duct (RLD) and thoracic duct (TD) in one normal dog and two dogs with chronic aortocaval fistulas in which lymph cannulation studies were made at the 24th hour after RISA instillation into the lung. The percentage of radioactive material which had entered the plasma of the normal dog at 24 hours was 16%, compared with 46.5% in one dog with a chronic aortocaval fistula and 12.0% in the other (average, 29.25%). RLD lymph flow in the normal dog was 4.0 ml./hr., compared with an average RLD lymph flow of 47.25 ml./hr. in the two animals with chronic aortocaval fistulas.

TD lymph flow in the normal dog was 16 ml./hr., compared with 73.0 ml./hr. and 20 ml./hr. (average, 46.5 ml./hr.) in the two dogs with aortocaval fistulas.

The specific activities of plasma, RLD lymph, and TD lymph were 2,949, 10,092, and 1,009 c.p.m./ml., respectively, in the normal dog. The specific activities of plasma, RLD lymph, and TD lymph varied considerably in the two dogs with aortocaval fistulas. There was a high and comparatively uniform activity from all three sites in the dog with large RLD and TD lymph flows. There was less activity in plasma and lymph in the other dog with an aortocaval fistula and a normal RD lymph flow. When averaged, the specific activities of plasma, RLD lymph, and TD lymph in the two animals with aortocaval fistulas were 22,942, 21,693, and 18,091 c.p.m./ml., respectively.

Studies of a similar nature were also performed as acute experiments in dogs in which no previous 24-hour plasma absorption studies

TABLE III

Lymph flow from the acutely cannulated right lymph duct and thoracic duct during the 24th to 25th hour after instillation of 50 μc. RISA into the right lung of three dogs

Dog	Right lymph duct			Thoracic duct			Plasma	
	Flow (ml./hr.)	Specific activity (c.p.m./ml.)	Lung RISA (%) in 24- to 25- hr. collection	Flow (ml./hr.)	Specific activity (c.p.m./ml.)	Lung RISA (%) in 24- to 25-hr. collection	Specific activity (c.p.m./ml.) 24-hr. sample	Lung RISA (%) in plasma by 24th hr.
No. 2692: Female; 18.1 kg.; aortocaval fistula	32.0	30,866	1.10	73.0	26,440	2.10	35,175	46.5
No. 2540: Female; 19.2 kg.; aortocaval fistula	62.5	12,521	0.10	20.0	9,743	0.20	10,809	12.0
Av.	47.25	21,693	0.60	46.5	18,091	1.15	22,942	29.25
No. 2236: Female; 14.3 kg.; normal	4.0	10,092	0.01	16.0	1,009	0.005	2,949	16.0

had been performed. Many right lymph duct (RLD) collections were attempted in this group also, but again technical competence was achieved in only a few.

Table IV and figure 4 show an example of a study performed on a normal dog (No. 41). During dissection of the venous pocket for the collection of RLD lymph the pleural space was inadvertently opened. The subsequent study was therefore undertaken with the animal artificially ventilated with a Palmer positive-pressure pump.

Before the lymphatic dissection was made in the neck, 5 ml. of 1% Evans blue in water were instilled by means of a bronchial catheter into the alveoli of the lower lobe of the left lung. Control collections were made from the right lymph duct and thoracic duct for 1 hour; 50 μc. of RISA in 5 ml. of saline were then instilled into the alveoli of the lower lobe of the right lung by means of a bronchial catheter, and plasma and lymph samples were collected for 8 hours thereafter.

The average lymph flow from the right lymph duct was 3.9 ml./hr. with an average of 10,028 c.p.m./ml. due to the presence of RISA. The average lymph flow from the thoracic duct was 29 ml./hr., with an average of 1,075 c.p.m./ml. In the plasma, the average count was 620 c.p.m./ml. Thus, the concentration of RISA in RLD lymph was approximately eleven times that found in TD lymph, and seventeen times that of the plasma.

Figure 4 also shows that in spite of an attempt to complete drainage of lymph from the thorax and abdomen by way of the thoracic and right lymph ducts, most of the RISA instilled into the lung still entered the plasma by way of the bloodstream. Thus, at the end of 4 hours, approximately 7% of the RISA instilled into the alveoli had entered the plasma at a time when only 1.6% had drained by way of the right lymph duct and 1.2% by way of the thoracic duct, respectively. At the end of 8 hours the plasma contained 11.1% of the total dose of RISA which had been instilled into the lung alveoli; an additional 4.1% had drained by way of the right lymph duct and 3.1% by way of the thoracic duct. This resulted in a total movement of 18.3% of RISA from the lung alveoli. This amount is more than was found to enter the plasma from the alveoli in our six intact normal dogs, breathing spontaneously (6.6% at 5 hours, and 14.2% at 9 hours).

TABLE IV

*Specific activity of lymph from the right lymph duct, thoracic duct, and plasma; percentage of RISA entering plasma and lymph after instillation of 50 μc. RISA into the alveoli of the right lung of a normal dog**

Time (hr.)	Right lymph duct		Thoracic duct		Plasma	Percentage of lung dose of RISA entering—			
	Volume (ml.)	Specific activity (c.p.m./ml.)	Volume (ml.)	Specific activity (c.p.m./ml.)	Specific activity (c.p.m./ml.)	Plasma	RLD lymph	TD lymph	Plasma + lymph
0 – 1	4.6	1,506	38.0	168	118	1.0	0.1	0.1	1.2
1 – 2	3.7	13,128	39.2	663	517	4.2	0.5	0.3	5.0
2 – 3	4.3	13,404	33.4	983	912	7.1	1.2	0.6	8.90
3 – 4	4.6	13,440	42.0	1,554	824	6.7	1.6	1.2	9.5
4 – 6	7.3	18,751	19.0	2,708	1,236	10.1	2.8	1.7	14.6
6 – 8	6.8	19,994	61.5	2,625	1,357	11.1	4.1	3.1	18.6
Av.	3.9	10,028	29.0	1,075	620				

*No. 41; female weighing 17.6 kg. Plasma volume, 778 ml.; open-chested, positive-pressure ventilation.

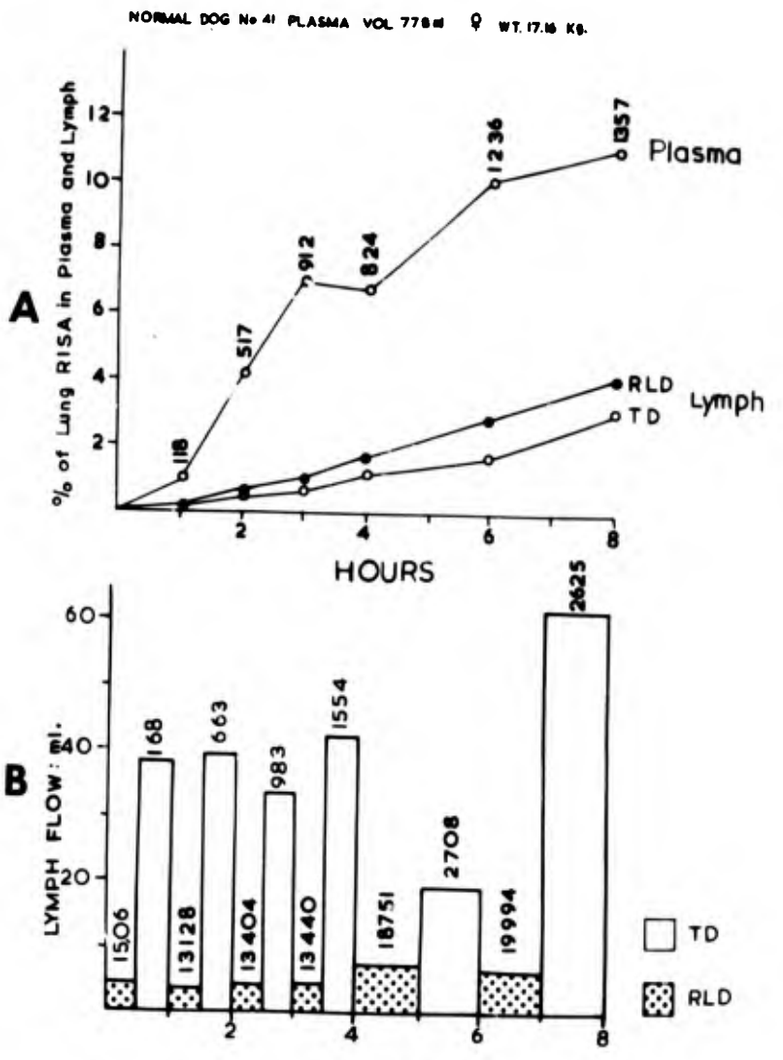

FIGURE 4

A: *Percentage of RISA entering the plasma and draining in lymph from the cannulated right lymph duct (RLD) and thoracic duct (TD) in a normal dog (No. 41) after the instillation of 50 μc. RISA into the right lung by means of a bronchial catheter. The figures over each plasma sample indicate the radioactivity in counts per minute.*

B: *The lymph flow and specific activity of each lymph sample (counts per minute) collected from the right lymph duct (RLD) and thoracic duct (TD) in a normal dog (No. 41).*

Table V and figure 5 show the results of a similar study performed on an animal with mild pulmonary venous congestion resulting from an aortocaval fistula produced two weeks previously. The animal had gained 3.2 kg. and was edematous, with some ascites. The pulmonary arterial pressure was slightly elevated (30/15 mm. Hg), and the wedge pulmonary arterial pressure was 14 mm. Hg. Lymph flow from the right lymph duct averaged 8.7 ml./hr., more than double the amount obtained in the normal animal described above (3.9 ml./hr.). The average thoracic duct lymph flow was similar, being 23.3 ml./hr. compared with 29 ml./hr. in the normal animal.

Again, most of the RISA which left the lung alveoli entered the plasma by way of the

TABLE V

Specific activity of lymph from the right lymph duct, thoracic duct, and plasma; percentage of RISA entering plasma and lymph after instillation of 50 μc. RISA into the alveoli of the right lung of a dog with pulmonary congestion*

Time (hr.)	Right lymph duct		Thoracic duct		Plasma	Percentage of lung dose of RISA entering—			
	Volume (ml.)	Specific activity (c.p.m./ml.)	Volume (ml.)	Specific activity (c.p.m./ml.)	Specific activity (c.p.m./ml.)	Plasma	RLD lymph	TD lymph	Plasma + lymph
0 – 1	7.10	318	16.8	259	352	2.25	0.02	0.02	2.29
1 – 2	6.3	1,442	25.9	664	411	2.65	0.07	0.09	2.81
2 – 3	11.6	4,896	35.6	1,118	947	6.10	0.31	0.25	6.66
3 – 4	9.7	7,495	15.0	1,622	1,028	6.60	0.61	0.35	7.56
Av.	8.7	3,538	23.3	916	684				

No. 2914: female weighing 28 kg. Mild heart failure; peripheral edema. Pulmonary arterial pressure, 30/15 mm. Hg; wedge pulmonary arterial pressure, 14 mm. Hg.

bloodstream (6.6% in 4 hours) at a time when only 0.6% of the instilled RISA had drained by way of the right lymph duct and 0.35% by way of the thoracic duct. The concentration of RISA in RLD lymph, expressed in counts per minute per milliliter of lymph, was approximately one-third that of the normal animal (3,538 c.p.m./ml. compared with 10,028 c.p.m./ml. in the normal animal), although the concentration of RISA in the thoracic duct lymph was very similar to that found in the normal animal studied (916 c.p.m./ml. compared with 1,075 c.p.m./ml. in the normal animal). Plasma counts were also similar in the edematous animal (684 c.p.m./ml. compared with 620 c.p.m./ml. in the normal animal).

V. DISCUSSION

The curves which we have obtained of increasing plasma radioactivity resulting from the absorption of radioiodinated albumin solution instilled into the lungs in a group of normal dogs are similar in every way to those obtained by Schultz and his colleagues (11) under similar circumstances. They confirm that the removal of albumin from the lung is slow, but that a considerable quantity is absorbed during the first 5 hours after instillation into the lung and that a plateau of concentration is also obtained in the plasma by 13 hours in our studies. Protein-bound radioactivity was detectable in blood within an hour after RISA instillation into the lungs.

In the past, Robertson (9), Drinker and Hardenbergh (6), and Courtice and Simmonds (4) had indicated that little or no protein absorption took place from the lung alveoli into the plasma within the first 5 hours. Drinker and Hardenbergh (6) had concluded that it was necessary for protein to be first broken down before it could be absorbed directly into the bloodstream; however, they also showed that albumin was present in lymph, presumably having entered it by way of the interstitial space from the capillaries.

Schultz et al. (12) have also performed radioiodinated protein absorption studies from isolated dog lungs. These showed a very similar rate of increase in plasma radioactivity to that obtained from the lungs of intact dogs. Plasma fractionation experiments in their isolated lung studies also showed that over half the absorbed radioactivity occurred as intact albumin and that a lesser proportion had entered the plasma from the alveoli in smaller form. These studies indicated that protein could pass directly from the alveoli across the alveolar and capillary membranes to enter the pulmonary circulation directly, for lung

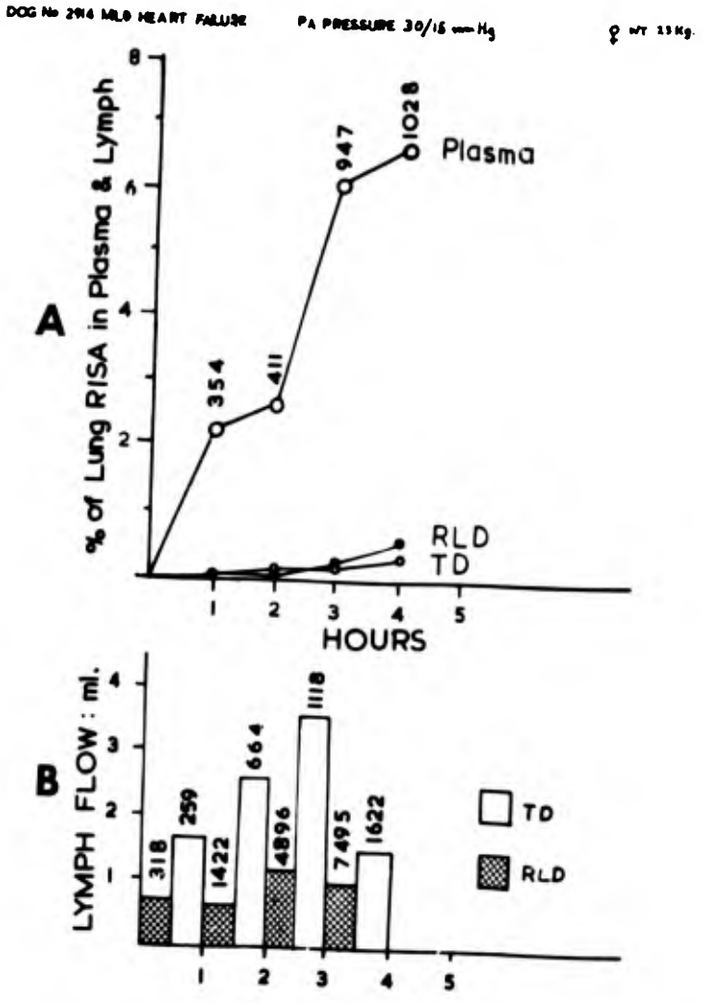

FIGURE 5

A: *Percentage of RISA entering the plasma and draining into the lymph from the cannulated right lymph duct (RLD) and thoracic duct (TD) in a dog (No. 2914) with heart failure after the instillation of 50 μc. RISA into the right lung by means of a bronchial catheter. The figures over each plasma sample indicate the radioactivity in counts per minute.*

B: *The lymph flow and specific activity of each lymph sample (counts per minute) collected from the right lymph duct (RLD) and thoracic duct (TD) in a dog (No. 2914) with heart failure.*

lymphatic drainage was interrupted in their isolated lung studies and there was no communication between the pulmonary lymphatics and the blood perfusion system.

Our measurement of increasing plasma radioactivity as a result of the absorption of iodinated albumin from the alveoli of the right lung when the pulmonary arterial blood flow to the lung had been prevented by balloon occlusion of the right pulmonary artery supplements the isolated lung experiments performed by Schultz et al. (12). The balloon study indicated that protein absorption from the lung

may also be accomplished effectively by way of the bronchial circulation or by way of anastomoses between it and the pulmonary venous system.

The pulmonary lymphatic capillary wall appears to be much more permeable to large molecules than that of the blood capillary (3). Wasserman et al. (19) have used this knowledge to study the permeability of the lung capillaries to plasma proteins and other macromolecules. They used small infusions of dextran fractions and radioiodinated albumin and obtained plasma-to-lymph concentration ratios which were directly proportional to their molecular weight. They also showed that the restriction to passage of albumin in the lungs was somewhat greater than in certain other tissues such as the liver. Shirley et al. (13) extended this work and showed that the concentration gradient between plasma and lymph for a specific molecular weight decreased if the plasma volume was expanded by transfusions. They explained this increase in capillary permeability in terms of stretching the capillary pores postulated to exist by Pappenheimer (7) to account for the permeability of capillaries to large molecules.

The permeability of the lung alveolar membrane has also been extensively studied by measuring the rate of movement of water and electrolytes between the alveoli and plasma. This is reviewed by Chinard (1). Taylor et al. (14) compared the permeability of the lung alveolar membrane of the dog to potassium, urea, sodium, glucose, deuterium, and dinitrophenol. They showed that the alveolar membrane presented a transport barrier to solutes many times more formidable than that of the capillary membrane. It is, therefore, perhaps surprising that protein should be able to traverse the alveolar-capillary barrier as readily as we and others before us have demonstrated. Increases both in lung vascular permeability and in lung lymph flow might be expected to take place in heart failure for the reasons already described in considering the work of Shirley et al. (13). Uhley and his colleagues (15, 16) have demonstrated a large increase in lung lymph flow when pulmonary venous hypertension is produced acutely or chronically, and we have confirmed this in chronic pulmonary edema produced in the manner described by Uhley.

In man, acute left ventricular failure may be associated clinically with the production of copious frothy sputum with a high protein content, indicating transudate of plasma constituents into the alveoli. We have attempted to examine the changes in overall permeability of the alveolar-capillary membrane to albumin by studying the absorption of RISA into the plasma after its instillation into the lung in animals with chronic lung edema produced hemodynamically in the manner described by Uhley et al. (16). We found that the normal slow absorption of protein into the plasma from the lungs was initially even more retarded when pulmonary edema was present. In studies where lymph cannulation experiments were being undertaken simultaneously there was an associated increase in lung lymph flow in animals with heart failure. Radioactive protein concentration in lymph draining from the lungs was lower than normal in animals with heart failure, although the total amount of radioactive protein entering the lymph was similar in the studies of normal animals and those with heart failure.

Although the initial rise in radioactivity in the plasma of the animals with heart failure was slower than in the normal group, the concentration of radioactivity in the plasma of the group with heart failure was higher at 24 hours than in the normal group and appeared to be continuing to rise. This slow initial rise in plasma radioactivity in the face of a subsequent continuous rise in plasma activity could indicate the presence of an increased interstitial lung volume in heart failure in which the radioactive albumin first became distributed before entering the plasma. The ultimate increase in plasma radioactivity could indicate some overall increase in membrane permeability affecting both the capillary and alveolar membranes. In this connection, it would be of interest to study the effect of chronic hypoxia on the plasma absorption of protein from the lung.

The most dramatic change in RISA absorption into the plasma from the lung took place

in one dog studied while suffering from bronchopneumonia. In this animal, radioactive albumin instilled into the lower lobe of the right lung was absorbed some twenty times faster than in the normal group of animals so that approximately 26% of the instilled protein was present in the plasma within 2 hours. This very rapid absorption indicated a great increase in permeability of both the alveolar and capillary membranes to large molecules under such circumstances. This finding needs to be substantiated by further study, but it suggests possible important implications regarding the permeability of tissues to bacterial products, on the one hand, and the delivery of circulating antibodies and antibiotics by the bloodstream to the tissues, on the other.

Finally, our studies on the effects of cannulating both the right lymphatic duct and the thoracic duct before the instillation of RISA into the lung indicate that only a minor proportion of the protein traversing the alveolar membrane enters the lymphatic system and that the larger proportion diffuses directly into the bloodstream. The volume of lymph flow from the right lymphatic duct was greater in animals with pulmonary edema than in the normal group, although the percentage of radioactive protein leaving the lung by way of the lymph in animals with pulmonary edema appeared to be less. These cannulation experiments confirm the conclusions reached by Schultz and his colleagues (12) in isolated lung experiments that most of the protein absorbed from the alveoli enters the plasma directly. Our studies also suggest that in conditions leading to pulmonary edema, the concentration of labeled protein entering the lung lymphatics from the alveoli may either rise or fall so that no indirect semiquantitative assessment of lung lymph flow is therefore possible from measurement of plasma radioactive albumin absorption from the lungs.

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13. ABSTRACT

A study was made of the absorption of radioactive iodinated serum albumin (RISA) into the plasma after its instillation into the lungs of normal dogs and dogs with pulmonary edema in an effort to develop an indirect method for measuring lung lymph flow.

Absorption of protein from the lung alveoli was initially slower in dogs with pulmonary edema compared with normal dogs; however, by 24 hours, the percentage of the instilled dose of RISA present in the plasma was higher in the dogs with pulmonary edema than in the normal dogs.

Absorption of RISA occurred equally well from the lung into the plasma whether or not the pulmonary artery to the lung was occluded, thus indicating that protein absorption could take place by means of the bronchial circulation.

Bronchopneumonia was associated with a greatly accelerated entry of RISA from the lung into the plasma, showing a large increase in alveolar and capillary permeability in pneumonia.

Measurements of changes in plasma concentration of RISA after its instillation into the lung cannot be used to assess the rate of lymph formation in the lung since most of the protein is absorbed directly into the plasma both in normal animals and in those with pulmonary edema.

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