Observations on the Pathogenesis of Pleural Effusions

by

W. J. H. Leckie
M.B., Ch.B., M.R.C.P.E.

A thesis for the degree of
Doctor of Medicine
in the University of Edinburgh

December, 1963

TABLE OF CONTENTS

PREFACE

"Pleural effusion" in its current usage is a term which lacks precise definition. For instance, it is taught that there is a critical level of protein concentration (3.5g%) in the pleural fluid above which effusions are "exudates" and below which they are "transudates". For many reasons this definition is not satisfactory and requires qualification. It is obviously unlikely that such a critical point exists, for the relevance of protein concentration in this context is its osmotic pressure, and it is well known that the proportions of the individual proteins within pleural fluid can vary substantially, and that one of them, albumin, is of by far the greatest osmotic importance in relation to the capillary membrane. Also the concentration of proteins in the plasma may vary and, if the plasma colloid osmotic pressure is reduced, an effusion would be maintained by a pleural fluid colloid osmotic pressure lower than if the plasma proteins were normal. If then the intention of this determination is to ascertain whether or not local protein accumulation is responsible for the effusion, it is to the relative concentrations of proteins in the two compartments - pleural and vascular - that attention must be paid. Even the

terms "exudates" and "transudates" seem to be incorrectly applied. The fact that a high concentration of protein collects in a region of the body does not mean that there is copious capillary exudation. For example, the cholesterol pleural effusion has a high colloid osmotic pressure in relation to that of the plasma and yet it has an extremely low protein turnover rate. As is shown in many of the pleural effusions described in this thesis, obstruction to protein drainage rather than increased capillary exudation is quite often the cause of protein accumulation. The term "hydrothorax" is also a rather puzzling one, as the "effusion" with which it is being contrasted also usually contains at least 93% of its weight of water.

These anomalies disclosed some uncertainty in our knowledge of the pathogenesis of effusions, and it was felt that a truer understanding of this would perhaps clarify the nomenclature and assist rational teaching of the subject. It was from this point of view that the investigation was planned.

The thesis is presented in chapters. After the introductory chapter, in which the physiology of the pleural space and the aims and method of the investigation are fully outlined, the chapters follow a set pattern. Each deals with a group of patients with a similar diagnosis and particular attention is paid to the diagnostic evidence in each case, the results, the pathology of the condition and the correlation

between pathology and results. In addition to the discussion and summary in each chapter there is a final chapter concerned with the correlation of the findings in each group.

Acknowledgments

I would like to express most sincerely my indebtedness to Dr. I. W. B. Grant and Dr. N. W. Horne for their close interest and encouragement in this investigation and to Professor A. G. Macgregor for providing the initial stimulus to my interest in isotopic work. The great majority of the patients included in the investigation have been under the care of Dr. Grant and Dr. Horne but I would also like to thank Professor J. W. Crofton, Dr. J. McC. Murdoch, Dr. J. D. Ross, Mr. J. D. Wade and Mr. P. R. Walbaum for permission to investigate their patients.

Miss A. Gregor has been of great technical assistance in carrying out many of the estimations of radioactivity and Dr. J. Crombie and Dr. C. P. Stewart arranged for protein estimations in routine laboratories under their charge. Dr. J. A. Lorraine kindly undertook the urinary oestrogen estimation and Dr. G. E. Moar and Dr. G. W. Ashcroft the estimation in ascitic fluid of bradykinin and the hydroxy-indoles respectively in one patient.

I am also indebted to the Trustees of the Royal Victoria and Associated Hospitals Endowments Funds for allocating a

generous grant for the purchase of radioactivity measuring equipment and radioactive materials necessary for this investigation.

Finally, I would like to thank most warmly Miss E. Nelson for her invaluable secretarial assistance, and above all for her patience.

CHAPTER I

Introduction

A. Physiological mechanisms controlling fluid volume within the pleural space.

The pleural cavity is part of the extravascular extracellular space, but it is unique in that, in common with other structures within the thoracic cavity, it lies within a negative pressure compartment. The remainder of the extracellular space has a positive interstitial tissue pressure, the difference between the hydrostatic pressure in the two situations being about 10 mm.Hg.. If a negative pressure, of a degree similar to that within the thoracic cavity, were to be exerted on this interstitial tissue, assuming that osmotic forces did not change, fluid would collect at the site (Starling, 1896). Nevertheless, fluid does not accumulate in the subpleural interstitial tissue of the lungs and Yamada (1933) has shown in a group of healthy soldiers that the normal amount of fluid within the pleural cavity varies between a drop and 20g., the larger amounts being found only following heavy exercise.

In the pleural space, as with the remainder of the interstitial space, water and electrolytes, other than the larger protein molecules, pass freely to and from the adjacent capillaries (Pappenheimer, 1953)(Prentice, Siri & Joiner, 1952). Conflicting opinions on the blood supply of the visceral pleura have been expressed. Miller (1937) stated that this layer of

pleura was supplied largely by capillaries of the systemic circulation through the bronchial arteries. However, Hayek (1953) describes "giant" capillaries in the convex and diaphragmatic surfaces of the visceral pleura supplied by vessels from the pulmonary circulation, which arrive at the pleura 1-1.5 cm. apart. Some of the remaining portions, the mediastinal and diaphragmatic surfaces, are occasionally supplied by the bronchial arteries. The latter description now appears to be generally accepted and it would appear that the capillary supply to the interstitial tissue immediately underlying the visceral pleura is predominantly from the pulmonary circulation.

Figure I shows diagrammatically the hydrostatic and osmotic forces which exist both inside and outside the visceral and parietal pleural capillaries. In any capillary there must be a point along its length at which hydrostatic and osmotic forces are in equilibrium (Starling, 1896). If such a point is selected on a pulmonary capillary, the net outward hydrostatic pressure, produced by the intraluminal pressure of approximately 8 mm.Hg. and the mean negative interstitial pressure of 4 mm.Hg., is 12 mm.Hg.. This will be balanced by a net absorptive force, acting inwards across the capillary wall, of 12 mm.Hg.. The plasma protein osmotic force is about 30 mm.Hg. using the nomogram devised by Wells, Youmans & Miller (1933), and so the osmotic force exerted in the interstitial space will be 30 less 12 mm.Hg., that is, approximately 18 mm.Hg.. Applying the same

FIGURE I

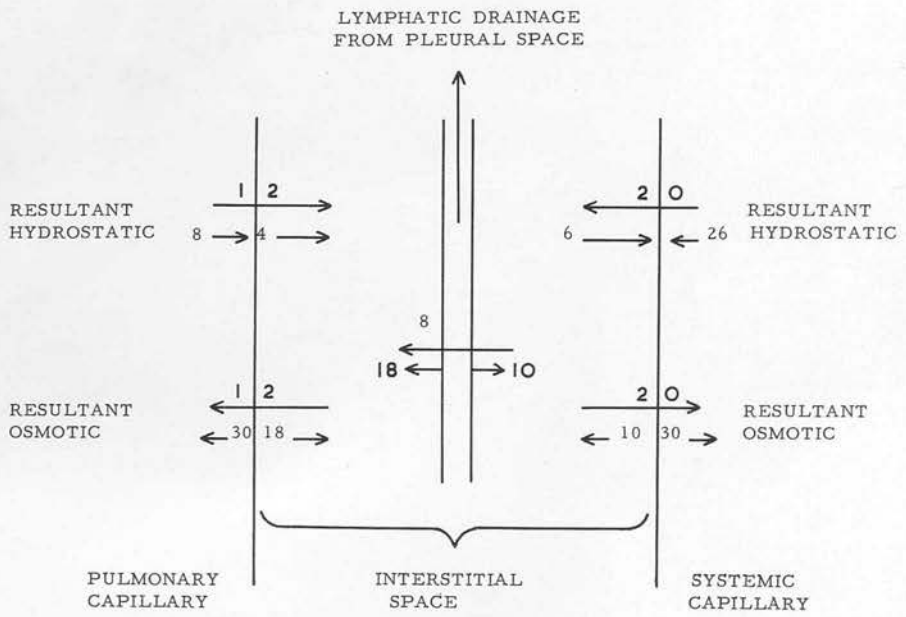


DIAGRAM ILLUSTRATING THE HYDROSTATIC AND OSMOTIC FORCES OPERATING IN THE PULMONARY AND SYSTEMIC CIRCULATIONS.

NET ABSORPTION FORCE = DIFFERENCE BETWEEN INTERSTITIAL COLLOID PRESSURES (8) + MEAN NEGATIVE INTRATHORACIC PRESSURE (4) = 12 mm. Hg.

argument to the systemic capillary, the net hydrostatic force outwards will be 26 less 6 mm.Hg., that is 20 mm.Hg., so that equilibrium will only be reached if there is an osmotic pressure in the interstitial space of 30 less 20 mm.Hg., that is 10 mm.Hg.. These figures are of course approximations but they make accurate representation of the principles governing fluid interchange at the pleural surfaces.

Thus, there exists in interstitial spaces separated by the two pleural layers an osmotic pressure difference which results in a net absorptive force of at least 8 mm.Hg.. (This figure also takes into account the different hydrostatic pressures within the two spaces.) Agostoni, Taglietti & Setnikar (1957) have shown experimentally in the dog that there is a net absorption force of about 15 cm. H_2O across the visceral pleural surface. They point out that this is the force which maintains the pleural space clear of free fluid and the visceral pleura apposed to the parietal against the elastic traction force of the lung. Adhesive and cohesive forces often described in the past as responsible for this phenomenon cannot possibly exist, as the membranes to which these terms are being applied are semipermeable. The visceral pleura therefore forms the surface at which this essential "sucking" force is created. It is obviously necessary, if the force is to be maintained, that the colloid osmotic pressure within the pleural space should be related to the colloid osmotic pressure in the interstitial

space surrounding the systemic capillary, so it is not surprising to find that lymph is drained from the pleural space almost exclusively by parietal lymphatics (Lemon & Higgins, 1932; Courtice & Morris, 1953).

It is known that the capillaries show modified permeability to plasma proteins, the ratio of the rate of leak of albumin to globulin being approximately 1.8:1 (Field & Drinker, 1931). The fraction of serum albumin exchanged with extravascular albumin in 24 hours has been estimated by different workers to lie between 0.62 and 0.74 (Walker, Ross & Hammond, 1960) (Schoenberger, Kroll, Eckert & Kark, 1956). The rate of albumin leak into the pleural space in man has not been estimated, but in anaesthetised ventilated dogs it was found that the right lymph duct, which drains at least 80% of the extravasated peritoneal and intrathoracic protein (Courtice & Simmonds, 1949; Courtice & Steinbeck, 1950), carried 3.6% of the total intravascular protein (average 55 g.) in 24 hours (Courtice, 1951). If these results apply at all to man, it is unlikely that more than ½-1% of the total intravascular albumin leaks into each pleural space in 24 hours. In health there is good evidence that passage of protein through the capillary wall is unidirectional and that protein does not return from the interstitial space to the blood-stream through the capillary wall in any significant quantity (Drinker & Field, 1931). Protein is returned to the blood-stream by the lymphatics and it is mainly the lymphatics

of the lower intercostal spaces (Cooray, 1949; Courtice & Simmonds, 1954) and of the lower mediastinal folds (Kampmeier, 1928) which clear the pleural space of protein. Particulate material is largely absorbed from sites in close relationship to the diaphragm, particularly Kampmeier's foci (Cooray, 1949; Courtice & Morris, 1953). There remains some controversy concerning the direction of lymph flow in the area of lung immediately underlying the pleura. Miller (1937) claims that the lymphatics of the visceral pleura are so valved that the lymph flow is towards the pleura but this is contested by other authors, notably Simer (1952), who concluded that the pulmonary lymph flow is entirely centripetal. It is not now thought that the parietal lymphatic channels drain from actual stomata in the parietal pleura. The exit of protein molecules and some particulate matter from the pleural space is apparently facilitated by dehiscence of adjoining mesothelial cells which occurs during respiration (Allen & Vogt, 1937). Cooray (1949) concluded that particles could pass either between the mesothelial cells or even through their cytoplasm. In the systemic interstitial space the protein molecules are cleared by a network of lymphatics and the rate of clearance is dependent, to quite a large degree, upon the rate and depth of respiratory movements (Courtice & Morris, 1953; Stewart & Burgen, 1958).

B. Causes of fluid accumulation within the pleural cavity.

Possible causes of fluid accumulation within the pleural cavity may be adduced from the foregoing physiological data. They may be divided into two major groups:

(1) General

Those in which there is a disturbance of total body water, sodium or protein balance, and

(2) Local

Those in which there is a disturbance of the pulmonary circulation or disease of the lungs and pleura.

In the general group are included: hypoproteinaemia, hyperaldosteronism, water intoxication, and in the local: left atrial failure, pulmonary infarct, carcinoma or inflammatory conditions affecting the pleural surfaces, primary pleural disease or intrathoracic lymphatic leakage. Theoretically, fluid would collect within the pleural cavity if the pressure within it became sufficiently negative, and certainly alterations of the intrathoracic pressure could well produce changes in effusion volume and in protein content. Separation of the pleural surfaces by air can also prevent satisfactory clearance of protein by the parietal lymphatics and so result in a small pleural effusion.

C. Aim of investigation.

It is with the local group that this investigation has been primarily concerned. In most instances the pleural surfaces themselves have been involved and in all other than acute left heart failure, the effusion is due to protein accumulation within the pleural space. In the absence of cardiac failure, the bigger the effusion, the higher is its colloid osmotic pressure, and albumin concentration varies between 1.8 and 3.5g%. Of course, the significance of this level depends also on the serum albumin concentration. Albumin may accumulate in the pleural space in higher than normal concentration either because it is entering at a rate in excess of the lymphatic capacity to clear it, or because there is obstruction to pleural lymphatic drainage. The relative importance of these factors in pleural effusions of different aetiology has not previously been determined. It was felt that the estimation of the rate of entry of albumin into an effusion and of its departure from it might indicate a characteristic pattern of albumin movement for each condition. It was also thought that the pattern might change in some conditions following local corticosteroid administration and that this additional information might prove of diagnostic help.

D. Patients investigated

Forty-two patients were investigated and Table I indicates the number in each diagnostic group. More detailed information about the individual patients, including age, sex, side of effusion, and the diagnostic evidence is listed in Appendix A. In the majority of patients this study was the first to be carried out and prior interference with the pleural surfaces was avoided if possible. Every effort was made to determine precisely the condition responsible for the effusion.

E. (1) Materials and Methods

Previous studies, comparing untreated with radioiodinated human serum albumin (R.I.H.S.A.) using zone paper electrophoresis, have shown only very slight differences with some higher mobility and "tailing" of the R.I.H.S.A. (Gabrieli, Goulian, Kinersly & Collet, 1954). It has been assumed, therefore, in common with many other workers, that R.I.H.S.A. behaves, at least from the aspect of its capillary and lymphatic transfer rates, similarly to normal human serum albumin. The R.I.H.S.A. has been prepared at the Radiochemical Centre, Amersham by the United Kingdom Atomic Energy Authority initially using dialysis and more recently an ion exchange method. The method of preparation of the R.I.H.S.A. is not described as such, but the use of the oxident (chloramine-T) is detailed by Greenwood,

TABLE I

Numbers investigated in each diagnostic group

Pleural Tuberculosis	7
Pulmonary Embolism	8
Lung Carcinoma	9
Secondary Carcinoma other than Lung	
(1) Breast	5
(2) Lymphosarcoma	1
Disseminated Lupus Erythematosus	4
Miscellaneous	
(1) Subphrenic Abscess	1
(2)"Pseudo-Meigs'" Syndrome	1
Congestive Cardiac Failure	6
Total	42

Hunter & Glover (1963).

At the time of supply the proportion of free radioactive iodide to that combined with albumin was always less than 2% and each sample was used within the time stipulated by the United Kingdom Atomic Energy Authority. In three patients whose thyroid gland uptake of iodine was blocked by the administration of potassium iodide there was no peak of urinary radioactivity at the time of intrapleural administration of R.I.H.S.A. to suggest the presence of an undue amount of free radioiodide in the sample. Also, the effusion volume as estimated by the isotope dilution technique tallied well with the quantity subsequently aspirated. If anything, the former estimation tended to be slightly lower, again suggesting that a significant quantity of free radioiodide was most unlikely to be present.

(2) Measurement of radioactivity

The radioactivity of 5 ml. liquid samples was measured with a sodium iodide well-type crystal scintillation counter which had an efficiency of 42%. The co-efficient of variation for a single sample was 2% for a total of over 5,000 counts. This was regarded as the minimum number of counts and a count rate of at least 50 per second was aimed at with all samples. Each sample was counted twice and the mean of the two counts accepted.

(3) Measurement of protein

Estimations of albumin and total protein concentrations were carried out on each sample by two laboratories, one

employing largely the micro-Kjehldahl technique, and the other using zone paper electrophoresis. The difference in the results obtained from the two laboratories, both of which were taking particular care, was no more than 5% in over 90% of the samples.

(4) Procedure of investigation.

In all but one of the patients the initial determination of the amount of albumin entering and leaving the effusion was made over 48 hours. The exception was a patient (Case 42) with severe congestive cardiac failure whose effusions were accumulating so rapidly that aspiration at 24 hours became necessary. In investigations carried out later in the series, where a satisfactory effusion volume remained at the end of 48 hours and where there was no obvious contra-indication, prednisolone was administered intrapleurally over the next four days and the pattern of albumin turnover again estimated. In Figure II is outlined the full scheme of investigation as it was carried out in each patient. The volume of the pleural effusion was determined at 0 and 48 hours, and, if the second part of the investigation was undertaken, at the end of six days. A sample R.I.H.S.A. solution was weighed to the nearest microgram and diluted to a known volume (500 ml.). The activity of this "standard" solution was then compared with the activity of the effusion following the intrapleural injection of 2-5g. of the prepared R.I.H.S.A. solution again weighed to the nearest microgram.

FIGURE II

Scheme of Standard Investigation

Day	Hours	Pleural Effusion	
		In	Out
1	0	R.I.H.S.A.	
	2		1
	12		2
2	24		3
	36		4
	48	R.I.H.S.A.	5
	50	20 mg. Pred.	6
3	72	20 mg. Pred.	7
4	96	20 mg. Pred.	8
5	120	20 mg. Pred.	9
6	144		10
		R.I.H.S.A.	
	146		11
7	156		12
	168		13

R.I.H.S.A. = Radioiodinated human serum albumin in aqueous solution weighed to nearest microgram

Pred. = Prednisolone disodium phosphate

It was found that adequate mixing of the injected sample of R.I.H.S.A. was achieved in one hour by a short burst of hyperventilation and a change of position every five minutes. At the same time as the volume estimations, specimens of pleural fluid were sent for determination of total protein and albumin concentrations and the intrapleural hydrostatic pressure was measured. The latter was estimated by the insertion of a needle connected to a water filled manometer into the pleural space at the level of the eighth intercostal space below the angle of the scapula. The patient was positioned with the axis of the dorsal spine as vertical as possible. It was found that even slight changes of position altered the manometer reading as did changes in the level at which the needle was inserted, and these factors were maintained as constant as possible during the three observations on each individual.

At 1, 12, 24, 36 and 48 hours after the injection of R.I.H.S.A., samples of pleural fluid for measurement of radioactivity were withdrawn through a different region of the chest wall from that used for the injection in order to avoid any possibility of contamination. An indwelling catheter was not used due to the increased risk of introducing infection intrapleurally. It was thought that the patients with thin chest walls would have been particularly liable to this. In no instance was infection introduced by the repeated needle aspirations and the discomfort using an intramuscular needle was minimal. When the investigation was continued to the second

phase, samples were also withdrawn at the beginning of the 4th, 5th, 6th and 7th days, and 20 mg. prednisolone introduced intrapleurally daily on the 3rd, 4th, 5th and 6th days.

(5) Calculation of results.

The method of estimation of effusion volume has been described above:

$$Vol(E) = \frac{Y \times 500 \times c(X)}{X \times c(Y)}$$

where X is g.R.I.H.S.A. in 500 ml. standard solution

Y is g.R.I.H.S.A. injected intrapleurally

c(X) is counts/5ml./sec. standard solution

c(Y) is counts/5ml./sec. pleural fluid at time 0

c(Y) was obtained by extrapolation to time 0 on semilogarithmic graph paper of the straight line joining points plotting effusion activity against time.

A simple model can be constructed (Figure III) to clarify the method of estimating the amount of albumin entering (Alb(E)) and the amount of albumin leaving (Alb(L)) the effusion in unit time. The unit of time in all the investigations was 24 hours. The factors which can affect Alb(E) and Alb(L) are also introduced in the diagram and have been referred to previously (p.7).

If Alb(L) were nil, the calculation of Alb(E) would be simple and would be represented by the product of the change of volume of the effusion and its albumin concentration, assuming that the latter remained constant throughout the investigation. This figure could also be obtained from a knowledge of the rate

FIGURE III

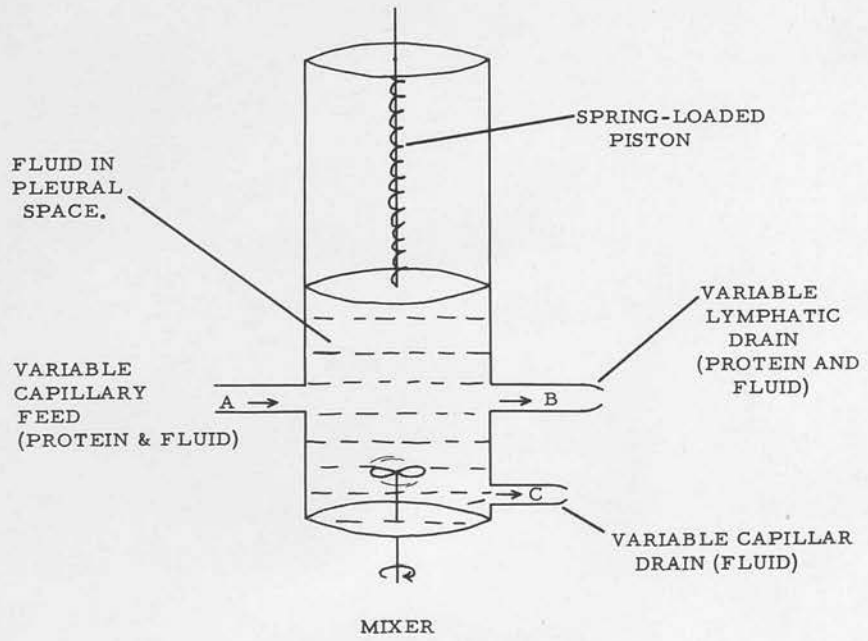


DIAGRAM SHOWING FACTORS RESPONSIBLE FOR FLUID AND PROTEIN MOVEMENT THROUGH THE PLEURAL CAVITY.

"Mixer" represents respiratory movement

The spring tension represents mean intrapleural pressure

of dilution of the initial fluid by the entering albumin solution. If Alb(E) were nil, the calculation of Alb(L) would be quite straight-forward and would be represented again by the product of the change in volume of the effusion and its albumin concentration. The albumin in the initial solution would not be diluted, and, if R.I.H.S.A. had been introduced, the level of activity would remain constant. The rate of dilution is, therefore, a function of the rate of albumin entering the effusion, although it is influenced by Alb(L) and changes in albumin concentration. If Alb(E) equals Alb(L), dilution occurs exponentially in relation to time, and, plotted on semilogarithmic graph paper will yield a straight line, the slope of the curve bearing a direct relationship to Alb(E). Provided that the rates of dilution and change of volume are constant and provided that the change of volume in unit time is no more than 25% of the initial volume, deviation from the straight line is negligible. Variations in Alb(L) will also affect the rate of fall of radioactivity. Calculation of the values for Alb(L) and Alb(E) is most easily made in the following manner.

The difference between the total counts in the fluid at t_1 and t_2 can be established from a knowledge of volume and count rate at these times and this value, less the counts removed in the samples, will represent counts leaving the compartment within this time.

$$A = \left[(Vt_1 \times ct_1) - (Vt_2 \times ct_2) \right] - cS$$

where A = counts leaving pleural space between t_1 and t_2

Vt_1 = vol. pleural fluid (mls.) at starting time

Vt_2 = vol. pleural fluid (mls.) at finishing time

ct_1 = count rate/ml. pleural fluid at starting time

ct_2 = count rate/ml. pleural fluid at finishing time

cS = counts withdrawn in samples

Related to the mean count concentration in the effusion over the time of the investigation and the albumin concentration in the effusion, this value will provide an estimation of Alb(L).

i.e. $Alb(L) = \frac{A \times a}{C_m \times t}$

where a = mean alb. concentration (g%) in pleural fluid

C_m = mean count rate in pleural fluid

t = hours of investigation $\times \frac{1}{24}$

Alb(E) is then a simple function of the change of albumin content of the effusion (also taking into account albumin removed in samples) and Alb(L).

$$Alb(E) = \left[(Vt_2 - Vt_1) + Vs \right] \times \frac{a}{t} + Alb(L)$$

where Vs = volume of samples (mls.)

CHAPTER II

Effusions Associated with Tuberculosis

TUBERCULOSIS

A. CLINICAL DATA

(1) General

There were seven patients, (Appendix A I), of whom two (Cases 5 and 6) could be classed clinically as acute, four (Cases 1, 3, 4 and 7) as subacute and one (Case 2) as chronic. The patients with acute pleurisy had a relatively short history and were systemically upset with fever, pain and high erythrocyte sedimentation rates. The patients classed as "subacute" had a longer history but the same systemic upset. The single patient with a chronic cholesterol effusion complained only of vague ill-health, comparatively mild night sweats and slight weight loss over six months. The effusion failed to clear with anti-tuberculous chemotherapy and the estimation of albumin turnover was repeated at a second admission, when the effect of intrapleural administration of prednisolone was also assessed.

The average colloid osmotic pressure of the effusions was 19.6 cm. H_2O, and of the sera 29 cm. H_2O, in the four patients in whom both were estimated.

(2) Diagnostic evidence

Proof of a tuberculous aetiology in Case 2 was finally obtained by histological examination following pleurectomy. Two "blind" punch biopsies of the parietal pleura had been previously obtained from this patient with the chronic cholesterol effusion but had proved negative. Of the remaining

six, proof of diagnosis was obtained in two (Cases 4 and 7) by isolation of tubercle bacilli, in two (Cases 1 and 6) by pleural biopsy and in another (Case 3) by a combination of pleural and scalene node biopsy. In the remaining patient (Case 5), a woman of 24 with a Mantoux test positive to 1:10,000 O.T., her acute systemic illness subsided shortly after the initiation of anti-tuberculous chemotherapy and aspiration of the lymphocytic effusion was only required twice. Pleural biopsy in this patient was "unsuccessful" but in three out of the four patients in this group in whom satisfactory tissue was obtained, a diagnosis of probable tuberculous infection of the parietal pleura could be made. Spriggs & Boddington (1960) state that a pleural effusion which contains frequent mesothelial cells can be assumed to be due to some cause other than tuberculosis. Only "occasional" mesothelial cells were seen in two of the seven patients, none being observed in the other five.

B. RESULTS

The results are grouped according to their clinical classification and are shown in Table II.

Alb(E) was variable, tending to be highest in the acute group.

Alb(L) was universally low during the initial assessment but in those patients whose response to intrapleural prednisolone administration was also measured (other than

TABLE II

Tuberculous Pleural Effusions - Results

Case	Side	A	B	C	Prot. (g%)	Alb. (g%)	Pre-Prednisolone		Post-Prednisolone	
							Alb(E)	Alb(L)	Alb(E)	Alb(L)
5	L	500	800			3.0	6.8	1.5		
6	R	2200	2650		5.7	3.0	10.6	2.9		
1	L	730	770	360	4.3	2.9	4.3	3.1	1.5	4.1
3	R	870	880	410	5.9	2.6	3.8	2.8	0.7	3.5
4	L	550	550	120	4.5	2.7	2.0	1.0	1.4	3.7
7	L	560	450	110	5.5	2.7	3.5	4.2	1.9	3.2
2(a)	R	1000	1000		5.5	2.2	1.9	1.3		
2(b)	R	1320	1260	1110	6.0	2.5	2.3	2.3	1.4	2.1

A = effusion volume (ml.) at 0 hours Alb(E) = albumin entering in g./24 hours
B = effusion volume (ml.) at 48 hours Alb(L) = albumin leaving in g./24 hours
C = effusion volume (ml.) at 144 hours

(a) = first admission
(b) = second admission

Case 2) quite marked increase in Alb(L) occurred along with decrease in Alb(E).

C. DISCUSSION

(1) Pathology and results (other than Case 2)

The pleural reaction in tuberculous pleural effusion most commonly involves the whole pleural surface, visceral and parietal, and the high diagnostic yield of thoracoscopy (Schuman, 1941) and blind parietal pleural biopsy (Mestitz, Purves & Pollard, 1958) are witness to this. Tuberculous pleural effusion occurs as a post-primary feature and there is a considerable weight of evidence to suggest that much of the "whole pleura" reaction is allergic in type. Paterson (1917) using guinea pigs was able to cause fatal infection by intra-pleural inoculation of tubercle bacilli but unable to bring about significant pleural effusion. However, in previously immunised animals similar treatment provoked an acute reaction of the pleura with effusion but limitation of systemic spread of the bacilli. Lemon & Feldman (1943) found that tuberculin placed into the pleural space of sensitised animals did not produce an effusion. They believed that this was due to rapid absorption and concluded that "a sensitised pleura and living bacilli in the pleural space are requisite factors in the production of a tuberculous pleural effusion. Myers (1955) goes so far as to say that "with rare exceptions pleurisy with

effusion is an allergic phenomenon". Pleurisy and polyserositis occur not only in early post-primary dissemination when allergy is most brisk: they may also be found in people over 45 when they may be associated with reactivation of old primary complexes. An effusion is most commonly the sequel, therefore, to eruption of a subpleural focus into the pleural cavity. Only a very few bacilli may be instrumental but, with special care, tubercle bacilli may be isolated from the fluid of as many as 90% of young adults (Close, 1946). Direct involvement of the visceral pleura by extension of bronchogenic tuberculosis or by haematogenous spread may give rise to pleural effusions. Even in these patients, although the generalised pleural reaction is not so acute, lymphocytic inflammation of the parietal pleura is almost invariable and tubercle bacilli may also be isolated from the fluid.

In either case, therefore, passage of protein from the pleural space would be expected to be greatly impaired. The rate of capillary leak of protein would probably be dependent on two factors - the degree of hypersensitivity of the pleura to tuberculoprotein and the extent of direct involvement of the pleura by tuberculous infection. As has been already mentioned, low Alb(L) figures were invariable in this group of patients. The only patients with high levels of Alb(E) were patients without significant parenchymal lung disease, with strongly positive Mantoux reactions and with presumed "allergic" effusions.

Prednisolone would be expected to effect a dramatic change on any allergic reaction of the pleura. Unfortunately, its action was not measured in either of the "acute" patients with this type of response.

However, there was quite marked relief of the obstruction to protein drainage in the "subacute" patients, and at least some of the inflammation in the parietal pleura appeared to respond rapidly to corticosteroid therapy. It has been shown in patients with pulmonary embolism (in whom the parietal pleura is not significantly involved) that Alb(L) tends to rise in relation to the volume of the effusion. The increase in Alb(L) following prednisolone administration in the tuberculous effusions is, therefore, even more significant as the effusion volume in each patient was falling. The average hydrostatic pressure fall was 3 cm. over the four day period and there was an average daily volume loss of at least 100 mls..

As might be expected there was a marked fall in Alb(E) following intrapleural prednisolone administration. Why corticosteroids affect capillary permeability is not known but that they do reduce it, particularly in inflammatory states, is a well documented fact (Dougherty & Schneebeli, 1950; Pickering, 1952). The speedy resolution of tuberculous pleural effusions treated by systemic corticosteroids and anti-tuberculous drugs is also well described (Fleishman, Coetzee, Mindel, Berjak, Lichter & Kerrich, 1960; Aspin & O'Hara, 1958).

(2) Cholesterol pleural effusion

This condition is well reviewed by Coe & Aikawa (1961) who also described two patients with cholesterol effusions in whom they had investigated albumin transfer into and from the pleural cavity and also cholesterol synthesis. No intrapleural synthesis of cholesterol from C^{14} - 2 - acetate could be detected in the cholesterol pleural effusion. C^{14} - 4 - cholesterol injected intrapleurally seemed to disappear very slowly in both a cholesterol and non-cholesterol pleural effusion. As the effusion volume was not estimated, it was not possible to obtain any accurate measurement of albumin turnover in the pleural space, but they noted that the fluid to plasma equilibration time was shorter for effusions of recent origin than for the more long-standing ones, both non-cholesterol and cholesterol in type.

Blomqvist reviewed this condition in 1951, finding 82 published cases and adding three new cases of his own. Many other authors on this subject, including Frew & Campbell-Fowler (1956) and Goldman & Burford (1950) agreed that a high cholesterol concentration in pleural effusion was more an index of chronicity than anything else. The latter authors considered that the following conditions are necessary for the production of a cholesterol effusion:
(1) a persistent exudate containing cholesterol and (2) a thick pleura with the normal interchange between effusion fluid and

blood decreased. In over half the patients, tuberculosis appears to be incriminated as the important aetiological agent and in the patient in this series with a cholesterol effusion, tuberculous caseation was demonstrated in the resected pleura. It seems most probable that cholesterol passes into the pleural cavity in soluble form as an $\alpha 1$ or $\alpha 2$ globulin complex (Wirta, 1936), or within leucocytes or erythrocytes, and there becomes trapped, some in insoluble form. Hypercholesterolaemia is rarely present in these patients and conversely a case of nephrosis with a serum cholesterol concentration of 700 mg.% had a pleural fluid cholesterol concentration of only 10 mg.%. The pleural fluid cholesterol concentration can reach very high values; it was 1,560 mg.% in the patient investigated in this series and a concentration of 2,165 mg.% has been reported (Goldman & Burford, 1950).

Alb(E) and Alb(L) were measured on two admissions separated by about nine months and found to be low. It was interesting that there was no evidence of accumulation of cholesterol in the nine months between the first admission when the effusion was aspirated and the second. The only drug therapy over this period was antimicrobial, P.A.S. and I.N.A.H. being administered in the usual dose.

During the second admission, i.e. after nine months of anti-tuberculous therapy, it was found that intrapleural prednisolone therapy produced only a slight fall in Alb(E) and

no significant change in Alb(L) (Table II).

The hydrostatic pressure (method of estimation p. 14) of the 1,250 ml. effusion was 2.5 cm. at the level selected and with a decrease in volume of approximately 200 mls. during prednisolone administration the pressure fell to 0.3 cm.. The pressure fall was disproportionate to the decrease in effusion volume and suggested that lung expansion was so restricted by visceral pleural thickening that, no matter how the pleural capabilities for handling protein were improved, the pleural space could not be closed.

The patient was, however, given a therapeutic trial of oral prednisolone for four months after this investigation in a dose of 15-20 mg. per day along with P.A.S. and I.N.A.H., but there was no real radiographic improvement and the patient continued to complain of vague ill-health and sweating attacks at night.

As Goldman & Burford (1950) described in their three cases, the effusion was eventually cleared by visceral pleurectomy with subsequent return to normal health.

D.

CONCLUSION

(1) Tuberculous pleural effusions are caused by accumulation of protein within the pleural space. There is obstruction to protein outflow by way of the parietal pleural lymphatics, which may be due to thickening of the parietal pleura with relative decrease in its permeability to the large colloid molecules, or

to primary lymphatic obstruction or to both. The rate of intrapleural protein, and therefore fluid accumulation, depends on the rate of entry of protein into the pleural cavity. The latter would appear to be related to the degree of exudative pleuritis present.

(2) In all but the chronic effusion, prednisolone quickly returned the abnormal protein handling by the pleura to normal.

(3) It was not possible to estimate the isolated effect of prednisolone on pleural handling of protein in the cholesterol effusion, as the restrictive visceral pleural disease prevented absorption of the effusion. It did appear, however, that prednisolone produced a decrease in capillary permeability even in this chronic condition as there was a small but definite fall in Alb(E).

CHAPTER III

Effusions associated with Pulmonary Embolism

PULMONARY EMBOLISM

A. CLINICAL DATA

(1) General

There were eight patients (Appendix A II), of whom three (Cases 23, 25 and 28) were in mild congestive failure. Case 23 also had marked emphysema, grade II pneumoconiosis and a history of chronic bronchitis. In Case 22 minimal pulmonary tuberculosis, on the same side as the effusion and affecting only the upper lobe, had been previously treated with standard chemotherapy. In the other four patients there was no clinical evidence of pre-existing pulmonary or cardiac disease.

The colloid osmotic pressure of the pleural fluid in the four patients without cardiac failure varied between 16.2 and 20.0cm. H_2O and in the three patients with cardiac failure it was 10.3cm., 13.7cm. and 14.5cm.. The average osmotic pressure of the protein in the effusions of these patients was 15cm. H_2O, and in the sera was 28.3cm. H_2O using the nomogram of Wells, Youmans & Miller (1933).

(2) Diagnostic evidence

There is no means of proving the diagnosis of pulmonary embolism, provided that the patient survives. However, if, during an episode of phlebothrombosis, the patient complains of pleuritic chest pain, has haemoptysis, becomes febrile and

has a blood-stained effusion, this diagnosis can be assumed. If, in addition, the condition clears satisfactorily with or without anti-coagulant therapy, leaving the tell-tale "linear" opacities on the chest radiograph, and the two-year follow-up does not reveal any other pathology, the diagnosis must be beyond reasonable doubt. Cases 25 and 28 satisfied all these criteria. None of the others had an obvious source of emboli, although in Case 22 embolism occurred shortly after a confinement. All the patients had chest pain, four had blood-stained sputum, five had fever, five had the typical "linear opacities" on chest radiograph, four effusions were eosinophilic. In fact, strongly presumptive evidence of the diagnosis was obtained in every case (MacLeod & Grant, 1954). Each patient has been followed for a minimum of two years and in none has fresh evidence arisen to suggest an alternative diagnosis. One has died subsequently (Case 28) with a leg vein thrombosis and a diagnosis of bronchopneumonia.

B. RESULTS

The results are presented in an order determined by effusion volume (Table IV). The volume varied between 100 mls. and 2,000 mls.. Most of the effusions were in a relatively constant state at the time of their investigation and Alb(E) approximates very closely to Alb(L) in each case. Both tend to

TABLE III

Pulmonary Embolism with Effusion - Results

Case	Side	A	B	Prot(g%)	Alb(g%)	Alb(E)	Alb(L)
26	L	2000	1770	5.3	2.7	7.3	9.5
22	L	1090	1050	4.9	2.7	5.5	4.9
25	R	800	590	4.7	1.8	3.4	4.8
27	L	725	680	5.2	2.6	6.5	6.3
23	R	680	610	4.3	2.2	4.9	5.0
28	R	390	360	3.3	1.6	2.6	2.7
24	R	130	90	4.8	2.5	3.4	3.2
21	L	100	50	4.4	2.6	1.9	1.9

A = effusion volume (ml.) at 0 hours

B = effusion volume (ml.) at 48 hours

Alb(E) = albumin entering in g./24 hours

Alb(L) = albumin leaving in g./24 hours

rise as the effusion volume increases and values for Alb(L) were higher in the larger effusions than they were in effusions of comparable size associated with any other pathology, excluding the single patient with pseudo-Meigs' syndrome.

C. DISCUSSION

(1) Pathology

There has been considerable experimental and autopsy investigation into the pathogenesis of pulmonary infarcts and the associated pleural reaction. Some of this work has considerable application to the results of this investigation and it is now reviewed.

Hampton & Castleman (1940), in a post mortem radiographic and autopsy study of 370 cases of pulmonary embolism, have shown that the pleura is always involved in pulmonary infarction. The area of most severe congestion lies immediately subpleurally and the thickness of lung involved is often no more than 1cm.. They suggest three categories of pulmonary embolism - simple embolism, incomplete infarction and complete infarction. In the first a pulmonary artery is obstructed but no pathological changes appear in the lung. In incomplete infarction there is severe congestion of the area of lung with rupture of capillaries and intra-alveolar haemorrhage, but it is only in complete infarction that lung tissue is actually destroyed.

Ellis, Grindlay & Edwards (1951) have shown in an ingenious series of experiments using dogs that the bronchial arteries, which have only a supportive role in the peripheral bronchi and interstitial tissue of lung, play an important part in the pathologies produced by pulmonary embolism. Prior ligation of bronchial arteries to a region of lung prevented infarction following embolism in the same region of lung, whereas with intact bronchial arteries, an incomplete infarct was produced. Even in the absence of bronchial arterial supply, however, pulmonary infarction followed embolisation of an area of congested lung. They concluded that broncho-pulmonary arterial anastomoses, dilating in response to ischaemia, may be responsible for haemorrhage and congestion in an area of incomplete infarct and may be a contributory factor in the development of the complete infarct. Pump (1963), using corrosion models, has shown a remarkable number of anastomoses between bronchial and pulmonary vessels of larger size than previously described and also emphasised the importance of these anastomoses in pulmonary embolus.

MacLeod & Grant (1954) describe the pathological situation likely to be present in the three categories mentioned above. They suggest that the collateral pulmonary circulation is adequate in embolism without infarct, is not quite adequate in incomplete infarct and is quite inadequate in complete infarct. They also point out that linear opacities are uncommon as a

residue of pulmonary embolus in congestive cardiac failure. Most emboli produce complete infarcts in pulmonary congestion so that it would appear that the acute fibrinous exudate, thought to be most likely to give rise during their organisation to the linear opacities, is more likely to be produced by an incomplete infarct. Hampton & Castleman (1940) described an incidence of only 13% of pleural effusion in association with pulmonary embolism and were at a loss to explain why some "stimulated a serous reaction" while others did not. However, the degree of congestion and the extent of the convex surface of the visceral pleura involved are variable factors which must influence the effusion volume.

(2) High albumin entry rate

A high albumin entry rate was almost universally present in this group of patients and, in five out of the eight, the follow-up chest radiographs demonstrated the development of characteristic linear opacities. If, as seems very likely, bronchopulmonary arterial shunts develop in areas of lung to which the pulmonary arterial circulation is not quite adequate, already ischaemic pulmonary capillaries will be subjected to the increased intraluminal pressure of the systemic circulation. The raised capillary hydrostatic pressure will result in increased exudation of fluid and some increase in capillary permeability to plasma protein, particularly if capillary pore size is significantly increased by ischaemia. It may be that

a further factor in the high rate of albumin entry from a comparatively small area of diseased pleura is the liberation in the infarcted lung of capillary permeability agents such as hydroxytryptamine or bradykinin. Quantitative estimation of such substances was not made during this investigation but further study of this aspect is being undertaken. Whatever the role of these agents, Rabin & Meyer (1960) and Uhley, Leeds, Sampson & Friedman (1962) all found rapid increase in lymph flow from the right lymph duct when left atrial pressure was raised above 25 mm.Hg.. Prolonged increase in pressure resulted in considerable extravasation of red cells so that the lymph became blood-stained. This rupturing of lung capillaries at pressures well tolerated in muscle and skin can probably be attributed to the poorer support of the former and probably also to their greater intrinsic fragility.

It might be expected that the subpleural congestion, by causing a high rate of fluid leak, would produce an effusion of low albumin content. However, the excess fluid is presumably rapidly resorbed into capillaries supplying intact visceral pleura and the albumin concentration in the pleural fluid is in the upper range. It is noticeable, however, that even in the absence of any evidence of cardiac failure, the ratio of the colloid osmotic pressure of the pleural fluid to that of the serum is lower in pleural effusions due to pulmonary infarct than in those due to tuberculosis or to malignant disease.

(3) High rate of albumin leaving

There is very little evidence of obstruction to lymphatic drainage in effusions secondary to pulmonary embolism and Alb(L) tended to vary with the volume of the effusion. In one patient (Case 22) there was a history of pulmonary tuberculosis and the upper half of the pleural space was seen radiographically to be obliterated. This fact, and the possibility that the parietal pleura of the remainder of the pleural space had also been thickened by persisting tuberculous disease, may have been responsible for the low Alb(L) figure in this patient. The larger the effusion, the greater is the hydrostatic pressure within it, the greater in general is the ventilation and also the larger is the area of parietal pleura available for absorption. The last may not be so important as might be imagined, as lymphatic drainage of the pleural space is much more a function of the lower than the upper intercostal lymphatics and the film of fluid in the upper pleural space will be replenished as it is absorbed by ventilatory movement.

The pain of pulmonary embolism must also be a factor in the accumulation of pleural fluid as must be the common finding of a high, fixed and sometimes even paralysed diaphragm. Resulting decrease in diaphragmatic and intercostal movement is likely to reduce to some extent the rate of lymphatic absorption. Also it is known that red blood cells slow down the rate of lymphatic absorption of protein (Courtice & Simmonds, 1949).

Pleural effusions associated with pulmonary embolism are frequently blood-stained and in this series only three patients had effusions which were not macroscopically blood-stained (Cases 22, 23 and 24).

D. CONCLUSION

There is normally a high Alb(E) and Alb(L) in effusions secondary to pulmonary embolism and the albumin turnover increases with effusion volume.

This accords well with the known pathology of this condition.

Variation in Alb(L) from expected values in some patients may be due to pre-existing disease of the parietal pleura or to inhibition of respiratory movement, diaphragmatic or costal, by pain or to very heavy contamination of the pleural cavity by blood.

CHAPTER IV

Effusions associated with Lung Carcinoma

CARCINOMA LUNG

A. CLINICAL DATA

(1) General

There were nine patients (Appendix A III) and in all but one (Case 17) the effusions were persistent and required treatment with radiotherapy or mustine hydrochloride. The effusions were generally of large size (average volume 1,200 ml. approximately).

The albumin content of all but two of the effusions (Cases 17 and 19) was over 2 g.%. In Cases 17 and 19, although the concentration of serum albumin was reduced, the colloid osmotic pressure of the pleural fluid was over 40% that of the serum. In Case 19 the pleural fluid albumin concentration was, in fact, only 1.2 g.% but the effusion would still appear to have been maintained largely by osmotic forces. The very low serum protein concentration in Case 19 was attributed to four prior aspirations but, on the whole, the plasma colloid osmotic pressure in the patients with carcinoma tended to be lower than that of patients in the other groups (Table XI).

(2) Reasons for diagnosis

All the patients have now died. The diagnosis was proved histologically in all and was confirmed in the five in whom autopsy examination was carried out (Cases 12, 13, 14, 16 and 18).

Cytological examination of the pleural fluid was positive for cancer cells in six patients. Bronchoscopy was carried out in five patients and was positive in one. "Blind" pleural biopsy, using Abrams' needle, was positive in five patients (Cases 12, 15, 18, 19 and 20).

This comparatively high yield of diagnostic information from cytological examination of the pleural fluid is well reported. Spriggs (1957) and Leuallen & Carr (1955) found malignant cells in the pleural fluid in over 50% of effusions associated with carcinoma of the bronchus, although in Robertson's (1954) series, only 24% of malignant effusions could be "diagnosed with absolute certainty by cytological examination". The percentage of positive diagnostic material varies to some extent with the frequency of examination and in most of these patients three or more specimens were examined.

B. RESULTS

The results are detailed in Table IV.

Apart from one patient (Case 13) in whom Alb(L) was 6.3 g./24 hours, the rate of albumin loss from the pleural space tended to be low. Unlike the results obtained in the patients with pulmonary embolism (Table III), Alb(E) and Alb(L) in these patients have no significant correlation with effusion volume and appear to be quite haphazard.

In two patients (Cases 12 and 20) there was quite a

TABLE IV

Case	Side	A	B	C	Prot(g%)	Alb(g%)	Pre-Prednisolone		Post-Prednisolone	
							Alb(E)	Alb(L)	Alb(E)	Alb(L)
14	R	1900	2350		5.8	2.1	5.5	0.5		
15	L	560	570		4.7	2.3	2.6	1.8		
18	L	340	350		4.9	2.5	3.9	3.2		
19	R	370	1180		2.1	1.2	6.3	1.1		
12	L	1540	1920	2180	3.6	2.1	7.2	2.5	6.3	4.1
13	R	2160	2140	1970	3.7	2.1	6.4	6.3	0.9	1.4
16	R	630	610	410	4.5	2.6	2.2	1.5	2.2	3.0
17	R	420	410	100	4.2	1.9	3.7	3.2	1.8	3.4
20	L	2000	2170	1730	3.8	2.5	3.5	0.9	1.8	4.9

A = effusion volume (ml.) at 0 hours
B = effusion volume (ml.) at 48 hours
C = effusion volume (ml.) at 144 hours

Alb(E) = albumin entering in g./24 hours
Alb(L) = albumin leaving in g./24 hours

In Case 13 the intrapleural hydrostatic pressure fell 5.1 cm. between B and C. During the other estimations there was no significant change in intrapleural pressure.

dramatic increase in Alb(L) and in Cases 12, 13, 17 and 20 there was a definite fall in Alb(E) after prednisolone administration. One rather anomalous result occurred in Case 13. Alb(L) in this patient dropped following prednisolone from 6.3g./24 hours to 1.4g./24 hours. There was a concomitant drop in the hydrostatic pressure in the pleural fluid of 5 cm. H_2O and yet the chest radiograph did not show any significant change in the size of the effusion. It did, however, suggest mediastinal shift towards the side of the effusion and it seemed likely that pulmonary collapse occurred during the period of investigation and accounted for at least part of the change in the dynamics of the effusion. Complete collapse of the right lung was found at autopsy three weeks later. Changes in the hydrostatic pressure of the other effusions were consistent with changes of effusion volume.

C. DISCUSSION

(1) Pathology

Lung tumour is the commonest source of pleural metastases. However, pleural involvement by tumour is not invariable in effusions associated with lung tumour. Sometimes the effusion is associated with pneumonia distal to an obstructed bronchus involving a section of visceral pleura. Indeed, in these circumstances, when cancer cells are not identified in the pleural fluid and when there is no evidence of parietal pleural

involvement, the effusion will not alone form a contra-indication to thoracotomy.

Lung tumour, particularly the oat-cell variety, frequently invades neighbouring blood vessels. By direct pressure or by involving hilar lymph nodes it may cause pulmonary venous obstruction (Simpson, 1929). Also branches of the pulmonary artery may be involved and Ballantyne, Clagett & McDonald (1957) found pulmonary arteries invaded in 16.5% of resected specimens and veins in 88%. However, macroscopic arterial invasion has been reported to occur much more frequently by some other authors, notably Pryce & Walter (1960) (43% of 183 resected specimens). Areas of haemorrhagic congestion and inflammatory exudation are, therefore, common in the lung surrounding bronchogenic carcinoma. The visceral pleura may also be involved by direct extension of the primary tumour, by vascular metastases or by lymphatic permeation. Sometimes spread may occur by direct growth along lymphatic channels from a centrally-placed tumour. Cancer cells in general show loss of cohesiveness (Coman, 1944) and, having reached the pleural space, distribution of individual cells or clumps of cells is further facilitated by the constant respiratory movements. The malignant cells may then become implanted in miliary fashion over the pleural surfaces, the parietal pleura usually being more extensively involved than the visceral. The frequency of survival of malignant cells exfoliated in this manner is

unknown. Moore, Sako, Kondo, Badillo & Burke (1961) did, however, find a correlation between the finding of malignant cells in thoracic cavity washings and the resectability rate in 125 patients of whom 82 had malignant disease. No false positives were obtained. From this work and experimental work showing the ready implantation of malignant cells within serous cavities in animals it is likely that the finding of malignant cells in the pleural fluid is strong presumptive evidence of parietal pleural involvement. Although it is only the serous surface which is involved initially, permeation into the intercostal lymphatics and tumour embolisation to regional lymph nodes may then occur.

(2) Correlation of pathology with results

A consistent pattern of albumin turnover was not found in this group. There were two patients, e.g. Cases 17 and 18, with effusions of comparatively small volume (420 ml. and 340 ml.) in whom Alb(E) and Alb(L) were much as would be predicted from a study of the results in the pulmonary embolism group. In one (Case 17) the response to intrapleural prednisolone was assessed and produced the changes anticipated in an "inflammatory" effusion. In this patient the diagnosis was made by biopsy of a lymph gland in the contra-lateral scalene triangle. After a course of antibiotics the effusion cleared and did not recur and the patient died six months later following myocardial infarction. It seems most likely that this patient's effusion

was due to pulmonary infection.

Autopsy was never carried out sufficiently near the time of the investigation for autopsy findings necessarily to be related to conditions obtaining during the investigation. It was hoped to ascribe the high rate of accumulation of the effusion in Case 19 to pulmonary venous obstruction but permission for autopsy was not granted and the exact pathology could not be ascertained. In the other three patients with relatively high Alb(E)(Cases 12, 13 and 14) Case 13 was the only one in whom pleural biopsy was negative and at autopsy Case 13 was also exceptional in that the pleural surfaces were not widely involved by tumour, whereas Cases 12 and 14 were extensively studded with tumour metastases. The reason for the high Alb(E) in Case 13 was not immediately apparent at autopsy as only a comparatively small area of visceral pleura was involved by the peripherally situated primary tumour. It is, however, tempting to rationalise this result by postulating the presence of small areas of congestion secondary to local arterial or venous obstruction, or possibly the elaboration of a capillary permeability factor in high local concentration by the tumour. The importance of contemporaneous estimation of intrapleural hydrostatic pressure is also demonstrated by this case and the fall of 5 cm. hydrostatic pressure with only slight change in volume appeared to have a marked effect on Alb(L), the drop in Alb(E) being not inconsistent with the effect of

corticosteroids observed in the other four patients.

The fundamental reason for the increased Alb(E) in the patients with lung carcinoma may, therefore, be attributed to three possible causes, two indirect and one direct:

Indirect (a) It is suggested that both local or major pulmonary vascular involvement may produce the pattern seen in pulmonary infarct in which the increase in Alb(E) has already been fully discussed.

(b) Pneumonitis distal to the tumour may involve an area of visceral pleura and produce a completely reversible pleural effusion.

Direct Extensive involvement of the pleural surfaces by carcinoma can probably also be responsible independent of the indirect causes described above. Little is known about the effect of tumour cells on capillary permeability. However, there is evidence which suggests that they can affect capillary endothelium in much the same way as histamine. Wood (1958) and Wood, Holyoke & Yardley (1961), using the rabbit ear chamber, have recorded the complete series of events which takes place when tumour cells become adherent to capillary walls and emphasise the importance of the underlying endothelial injury. This injury results in the accumulation of leucocytes about it and their

penetration of the endothelium at the intercellular junction. After one or more leucocytes has passed through the capillary wall, the endothelial defect is maintained and provides a means of egress for the amoeboid tumour cells. Under these circumstances, capillary permeability must also be increased and it may be that the factors bringing about the endothelial injury witnessed by Wood and others are responsible.

Cases 12 and 20 showed increases of 4.4 and 4 g. in Alb(L) following intrapleural prednisolone. Although this was a good response, the effusions were both of large volume (1920 ml. and 2,170 ml.) and Alb(L) even after this rise was still not at its maximum, although further improvement could possibly have occurred. In both patients carcinoma cells were identified in the fluid and biopsy of the parietal pleura was also positive. In no instance were pathogenic bacteria isolated from the pleural fluid and it would appear that the products of inflammatory exudates combined on some occasions with direct metastatic involvement of the visceral pleura can produce an inflammatory reaction in it which is at least partly reversed by prednisolone. It appears to be uncommon to find such extensive carcinomatous involvement either of the pleura itself or of the lymphatics underlying it as to be entirely responsible for the low Alb(L) results commonly found in this condition.

D. CONCLUSION

(1) A mixed pattern of albumin turnover is seen in connection with effusions associated with bronchogenic carcinoma. This is consistent with the varied lung and pleural pathology found in association with this condition.

(2) In most cases the intrapleural administration of corticosteroids will reduce the rate of accumulation of intrapleural albumin.

CHAPTER V

Effusions associated with

1. Mammary Carcinoma
2. Lymphosarcoma

MAMMARY CARCINOMA

A.

(1) Clinical data

There were five patients (Appendix A IV) and in all, the effusions were persistent and required hormonal or mustine therapy for their control. In all but one patient (Case 53) there had been prior mastectomy for mammary carcinoma on the same side as the effusion.

Two of the patients are still alive. Case 56 has no clinical evidence of recurrence of tumour activity after intrapleural mustine hydrochloride and oestrogen therapy. Case 54 was improved initially by prednisolone but after six months' treatment she developed multiple bone secondaries and underwent adrenalectomy. The metastases one year later have completely regressed and there is no evidence of tumour activity.

The plasma protein osmotic pressure was perhaps slightly reduced in this group, averaging 26.8 cm. H_2O, and the effusion osmotic pressure was over 60% that of the plasma.

(2) Reasons for diagnosis

There was no doubt that these patients' effusions were all due to malignant disease.

In only one patient, Case 53, was there any reasonable doubt as to whether primary tumour arose in the breast. This patient, who died at home, did not have an autopsy. No mass was ever palpable in the breast and bronchoscopy was negative. Malignant

cells thought to have been exfoliated from an adenocarcinoma were found in the pleural fluid. Biopsy at thoracoscopy of parietal pleura revealed infiltration by cells derived from adenocarcinoma and prescalene gland biopsy also revealed adenocarcinoma. It was considered by the pathologist that the site of origin was most probably breast.

B.
LYMPHOSARCOMA

(1) Clinical data

There was only one patient (Case 51) in this sub-group. He was a man aged 79 admitted with bilateral pleural effusions and mild cardiac failure. The investigation was carried out after treatment of the heart failure and indeed the effusion colloid osmotic pressure was over 60% that of the plasma at the time of investigation. The effusions were persistent and shortly prior to his death he also developed ascites.

(2) Reasons for diagnosis

Although doubtful malignant cells were seen in the pleural fluid and the fourth "blind" biopsy, using Abrams' needle, revealed infiltration with anaplastic cells, the diagnosis of lymphoblastic lymphosarcoma was only made post mortem. At autopsy a soft homogeneous mass extended retroperitoneally enveloping the pancreas, adrenals, aorta and inferior vena cava. There was also moderate, fairly-well delineated lymphocytic infiltration of the visceral and parietal subpleural zone.

E. Results

The most striking feature of the group as a whole is the low Alb(L) - average 2.2 g. - for the size of the effusions - average 1,290 ml. (Tables V and VI). Even in the large effusions Alb(E) was never higher than 5 g..

In only three patients (Cases 51, 53 and 55) was the response to intrapleural prednisolone assessed. In all three there was quite a marked reduction in Alb(E). In contrast to the results in all the other groups, in which prednisolone response was measured, Alb(L) was also reduced without any significant change in the hydrostatic pressure within the effusion.

In Case 55 the rate of fall of radioactivity in the effusion during prednisolone administration did not occur exponentially and the plot of activity did not fall on a straight line when plotted against time on semilogarithmic graph paper. This curve was resolved into two portions, the first giving values for Alb(E) and Alb(L) in the first 48 hours and the second giving values for the second 48 hours. (Figure IV) Possible reasons for this isolated finding are discussed below.

In Case 51 the effusions were bilateral and samples of pleural fluid from the contralateral effusion and at the same time samples of blood were withdrawn for comparison. These results suggested that there was no direct lymphatic pathway between the two pleural cavities.

TABLE V
&
TABLE VI

Secondary Carcinoma and Effusions - Results

V
Mammary Carcinoma

Case	Side	A	B	C	Prot(g%)	Alb(g%)	Pre-Prednisolone		Post-Prednisolone	
							Alb(E)	Alb(L)	Alb(E)	Alb(L)
52	R	1660	1850		5.4	3.0	4.0	0.5		
54	R	580	620		4.3	2.4	1.9	0.6		
56	L	1600	1500		4.8	2.7	2.4	3.1		
53	R	1200	1440	1800	3.4	2.1	4.3	1.3	2.8	0.6
55	R	880	890	990	5.0	2.5	4.6	3.7	3.6_a 2.2_b	2.0_a 1.8_b

a = estimations made between 48 and 96 hours b = estimations made between 96 and 144 hours

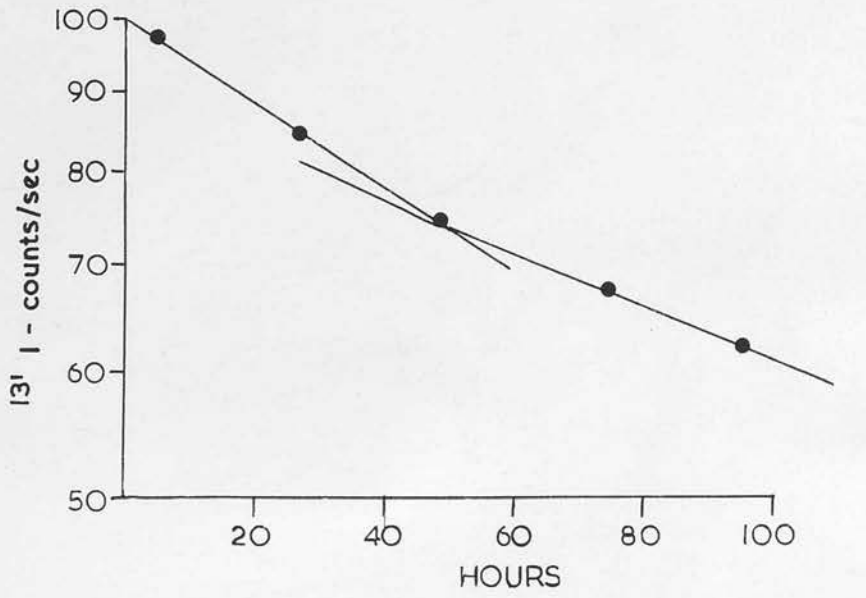
VI
Lymphosarcoma

Case	Side	A	B	C	Prot(g%)	Alb(g%)	Pre-Prednisolone		Post-Prednisolone	
							Alb(E)	Alb(L)	Alb(E)	Alb(L)
11	R	1220	1220	1160	3.6	2.0	4.7	3.9	2.6	2.7

A = effusion volume (ml.) at 0 hours
B = effusion volume (ml.) at 48 hours
C = effusion volume (ml.) at 144 hours

Alb(E) = albumin entering in g./24 hours
Alb(L) = albumin leaving in g./24 hours

FIGURE IV



GRAPH SHOWING CHANGE IN RATE OF ALB(E) DURING INTRAPLEURAL PREDNISOLONE ADMINISTRATION IN CASE 55 (See text)

F. Discussion

(1) Pathology

(a) Mammary Carcinoma

Mammary carcinoma commonly metastasises to lung and Abrams, Spiro & Goldstein (1950) report an incidence of 77% in patients dying of the condition.

It may be responsible for pleural effusion, either by metastasis to the subpleural region of the lung or by direct invasion of the parietal pleura by way of the intercostal lymphatics. Sometimes the tumour tissue extends in a sheet in the connective tissue underlying the parietal pleura and, not infrequently, spread to the lung is transpleural. Willis (1952) has shown that the majority of metastases spreading transpleurally are deposited on the dependent parts of the lung. Usually, a high percentage of these effusions contain malignant cells and "blind" pleural biopsy is also more commonly positive than in effusions associated with lung carcinoma. In this small group cytology was positive in three (Cases 52, 53 and 56) out of the five patients and pleural biopsy, which was carried out in four, showed malignant invasion in two (Cases 53 and 56) and thickened fibrous pleura in the other two (Cases 52 and 54).

It is probable that the effusions in this small group were due to involvement more of the parietal than the visceral pleura by direct extension. In none was there radiographic evidence of intrapulmonary metastases. In three (Cases 53, 54 and 55)

there was invasion of the chest wall with rib erosion on the same side as the effusion and, as already mentioned, mastectomy had been carried out previously in four patients on the same side as the effusion.

The general remarks made concerning the spread of carcinoma in Chapter IV apply also to this chapter but there are some additional features in breast carcinoma due to the continuing influence on its cells in many instances of the endocrine system. Hall, Dederick, Nevinny & Kelley (1960) show that the growth of a proportion of mammary carcinomata will be controlled at least for a time by either androgens, oestrogens or corticosteroids. They also endeavoured to predict patient response to therapy from observations of age, dominant lesion and cortical stromal hyperplasia. Lemon (1960) states that prednisolone is rarely effective in patients under the age of 65 who have intact ovaries. He suggests that prednisolone increases gonadotrophin production by the pituitary and it is only when the ovaries are absent or can no longer be stimulated that prednisolone is effective by depressing oestrogen or in some cases androgen excretion by the adrenals. Accordingly, in assessing the prednisolone effect on albumin turnover in effusions secondary to these tumours its action in depressing both capillary permeability and tumour growth would need to be taken into account.

(b) Lymphosarcoma

The lung is frequently involved in secondary lymphosarcoma although it is rather an uncommon primary site for this tumour. The literature has been reviewed by Rose (1957) and pleural deposits occur more commonly in the generalised disease, pulmonary involvement being found in about 50% of these patients at autopsy (van Hazel & Jensik, 1956). Although lymphosarcoma can occur at any age, it is uncommon over the age of 70. The secondary deposits are often confined initially to the sub-pleural region of lung but, by the time that pleural effusions occur, it has usually spread to involve the parietal pleura. In this patient the effusions were bilateral and were probably in part due to direct extension of the tumour by way of the diaphragmatic lymphatics to involve the parietal subpleural space and in part to blood-borne spread to the visceral sub-pleural space.

(2) Correlation of Pathology with Results

(a) Mammary Carcinoma

From the above discussion a universally low value for Alb(L) would be expected and, taking into account the size of the effusions, this was found in all but Case 55, where it fell in the intermediate range - 3.7 g. for an effusion of average volume 890 ml.. In this patient at the time of the investigation a portion of the right postero-lateral chest wall,

approximately 8 cm. in diameter, was palpably involved by tumour. Two ribs appeared completely destroyed radiographically over about 4 cm.. However, three specimens of pleural fluid examined at separate times did not contain malignant cells. Pleural biopsy was not done for fear of inducing local spread of the cancer to involve skin. It is possible, therefore, that only a comparatively small area of parietal pleura in this patient was involved and that much of the intercostal lymphatic system remained intact.

Alb(E) was in the intermediate range in most patients and there was no very marked evidence of increased capillary permeability.

The fall in Alb(E) in both patients in whom the response to intrapleural prednisolone was assessed is not easy to explain. There is no doubt that prednisolone produced considerable symptomatic improvement and it may be that ventilation was considerably reduced thereby. In Case 53 it is particularly surprising as the effusion volume in this patient increased from 1,440 ml. to 1,800 ml. over the four days of the investigation. It would be reasonable to assume that the effect of prednisolone on capillary permeability was almost immediate. It is rather surprising therefore to find in Case 55 that Alb(E) drops markedly in the second as compared to the first 48 hours of the investigation in Case 55. This patient in fact happened to be the only one over the age of 65 in this group

and some control of tumour growth by prednisolone might have been expected. It may be that this was responsible for the fall of Alb(E) from 3.6 to 2.2 g. in the second 48 hours, while Alb(L) remained almost constant.

(b) Lymphosarcoma

It appears in this patient that the involvement of the parietal pleura was patchy and malignant invasion was certainly not found in the first three "blind" biopsies of the parietal pleura. This Alb(L) was a little higher than expected. It may be that Alb(E) was contributed to by some ascitic protein, but ascites was certainly not demonstrable clinically at the time of the investigation. Intrapleural prednisolone appeared to have an effect on Alb(E) which could be explained solely in terms of its action on capillary permeability. Reduction in dyspnoea following the prednisolone was obvious and could well have brought about the slight drop in Alb(L).

G.
CONCLUSION

The patients in this group with mammary carcinoma probably had largely parietal pleural involvement at the time of the investigation and the average value for Alb(L) in this group is much lower than that of any other group.

In one patient a further change in Alb(E) occurred during the intrapleural prednisolone phase. It is suggested that this might have been the result of hormonal action on tumour growth

superimposed upon the initial depressant effect of prednisolone on capillary permeability.

CHAPTER VI

Effusions associated with Connective Tissue Disorders

SYSTEMIC LUPUS ERYTHEMATOSUS

A. Clinical data

(1) General

There were four patients (Appendix A V) whose bilateral effusions were considered to be due to systemic lupus erythematosus. Two, Cases 31 and 33, had comparatively small effusions, the other two patients having moderate sized effusions, but the size of the effusion did not seem to be a measure of the severity of the systemic illness. Cases 32 and 33 were both suffering from acute exacerbations of their condition with severe pleurisy and polyarthralgia, the L.E. cell phenomenon being readily exhibited in every specimen of serum at the time of this investigation. Cases 31 and 34, though both pyrexial and with E.S.R.s of over 40 mm. in the first hour, were not acutely upset by their disease. Chest pain, though present, was not particularly disturbing; they were not breathless and did not have joint symptoms.

The colloid osmotic pressure of the effusions was 57% that of the serum (Table XI) and the gamma globulin fraction was very prominent, making up more that 30% of the protein in the effusions (42% in Case 32).

(2) Reasons for diagnosis

All the patients had systematised disease characterised

by fever, bilateral pleural effusions, high erythrocyte sedimentation rate (over 40 mm. in the first hour), raised serum gamma globulin and antinuclear factor in the serum. In addition, the sera of the two patients with polyarthralgia exhibited the L.E. cell phenomenon. Full investigations, including chest radiograph, cytological and bacteriological examination of the fluid, pleural biopsy and general clinical examination have failed to reveal any other aetiological agent in the effusions. In the three patients (Cases 31, 32 and 33) treated with prednisolone, there was an immediate response to therapy and complete control of the condition has been achieved. In Case 34 the condition was not thought to be of sufficient severity to warrant commitment to long-term corticosteroid therapy and, when this patient left the neighbourhood six months after admission to hospital, the effusions, though slightly reduced in size on radiographic examination, were still present. A mild pyrexia disappeared, however, during the period of the investigation when prednisolone was administered and the patient also described considerable improvement in well-being at this time.

It is unfortunate that the serum in both Case 31 and 34 was examined for L.E. factor on only two occasions but, despite the negative results, it was felt that the diagnosis was secure on clinical grounds.

The absence of L.E. cells on repeated testing, although rather uncommon, has been reported in as many as 20% of

clinically diagnosed patients (Dubois, 1956; Rupe & Nickel, 1959).

B. Results

Although Cases 32 and 33 both had clinically acute S.L.E., Alb(E) was only 4.7 and 4.4 g. respectively. In Case 32, although the effusion was a large one (1,900 ml.), Alb(L) was only 3.3 g.. In Case 33, a much smaller effusion but increasing in size (average volume 300 ml.), Alb(L) was 1.3 g.. In the clinically less acute, Cases 31 and 34, Alb(E) was 2.7 g. and 3.4 g. respectively and Alb(L) 3.3 g. and 1.7 g.. The latter was again a low value for Alb(L) for an effusion of 1,100 ml..

The response to intrapleural prednisolone was most striking in Case 32 where Alb(E) was reduced by 66% and, despite a rapid reduction in effusion volume, Alb(L) increased by 27%. In the less acute patient, Alb(E) was reduced by 30% and Alb(L) increased from a very low value by over 100%.

C. Discussion

(1) Pathology

The lungs and pleura are commonly involved in systemic lupus erythematosus and different authors give rates of involvement varying from 50 to 80% (Dubois, 1953; Alarcon-Segovia & Alarcon, 1961). The majority of the pleural effusions are small, and a large volume, such as was found in Case 32, is comparatively rare

TABLE VII

Pleural Effusions Associated with Connective Tissue Disorders

Results

Case	Side	A	B	C	Prot(g%)	Alb(g%)	Pre-Prednisolone		Post-Prednisolone	
							Alb(E)	Alb(L)	Alb(E)	Alb(L)
31	R	420	320		5.1	2.6	2.7	3.3		
33	R	200	400		5.3	2.6	4.4	1.3		
32	L	1820	1900	1300	4.8	1.9	4.7	3.3	1.6	4.2
34	R	1080	1190	910	3.9	2.0	3.4	1.7	2.4	3.6

A = effusion volume (ml.) at 0 hours

B = effusion volume (ml.) at 48 hours

C = effusion volume (ml.) at 144 hours

Alb(E) = albumin entering in g./24 hours

Alb(L) = albumin leaving in g./24 hours

(Myhre, 1959). Pleural effusions are quite commonly associated with lung lesions (Harvey, Schulman, Tumulty, Conley & Schoenrich, 1955) and Teilum (1946) referred to these as a form of "focal allergic pneumonia". Baggenstoss (1952) describes the microscopic appearances of the lung as showing an interstitial mucinous oedema in the alveolar walls. Both found the changes particularly in the subpleural and interlobular connective tissue and the subpleural nature of the lesions is also commented upon by Thorell (1952). Teilum (1946) remarked on the evidence of widespread vasculitis occurring both in pulmonary arterioles and in the subpleural veins. The "pneumonia" which usually involves the lower lobes is not commonly bacterial, although in the acute phase it may often resemble organising bacterial pneumonia and the chest radiograph frequently shows linear opacities. Many authors have referred to these as areas of plate atelectasis (Myhre, 1959; Israel, 1953), but they have much more the appearance of the "linear opacities" of pulmonary infarction (Alarcon-Segovia & Alarcon, 1961; MacLeod & Grant, 1954). It has been suggested (Ellman & Cudkowitz, 1954; Ellman, 1956) that it is the bronchial arteries which suffer particularly in S.L.E. but this would not explain the acute pulmonary hypertension which develops in some patients.

Auto-immune processes are thought to play a major part in the vasculitis which is an essential feature of the lesions

found in the lungs and pleura in S.L.E.. Vaccari, Baldini & Fontana (1962) have injected a saline solution of pulped tibial artery intradermally into six newly diagnosed patients with S.L.E. and into 210 general medical patients. Peptone solution was used as a control. A strong local reaction to the artery solution was produced in all the patients with S.L.E.. Although some of the "general" patients reacted weakly to the artery solution, they also reacted to the control solution. This would appear to support the concept of "auto-immunity" although as Burrell (1963) has suggested, the initial trigger to the disease may originate from genetically determined "sick" cells, which, by releasing organic antigens by hydrolysis of cell protein, may excite an antibody response.

In considering the effect of "vasculitis" on the formation of a pleural effusion, it is important to know whether this affects only pulmonary vessels and, accordingly, solely the visceral pleura, or whether it also has an effect on the parietal pleura or the lymphatics. It is known that capillaries and arterioles, apart from those in viscera, may be affected. Smith & Kurban (1962) describe characteristic changes to be found in the capillaries of the skin overlying the hypothenar eminence and zygomatic arch and Dubois & Arterberry (1962) describe gangrene of the fingers and/or toes in five patients with S.L.E.. There has never, however, been reported a study of the histology of the parietal pleura

in this condition. In two of the patients in this investigation (Cases 32 and 33) the pleural biopsies showed a non-specific inflammatory response with some fibrin deposition on the surface of the pleura and round cell infiltration in the sub-pleural zone. In the other two patients, apart from slight pleural thickening, no abnormality was noted.

(2) Correlation of Pathology with Results

The pulmonary lesions as described above, often lying subpleurally and histologically resembling pneumonitis or pulmonary infarct, might be expected to leak very high quantities of protein into the pleural space but in none of the four patients in this investigation was this so. Alb(E) was never more than moderate and was considerably less than the figures achieved in "acute" pleural tuberculosis. On the other hand, Alb(L) was considerably lower when related to effusion volume than the rates found in pulmonary embolism. The low Alb(L) was quite significantly raised by prednisolone and it is difficult to escape from the conclusion that, certainly in the large effusions, either the parietal pleura or the underlying lymphatics were affected by the disease and so slowed down the rate of absorption of protein. The overall dramatic effect of corticosteroids in this condition, where vasculitis and increased capillary permeability are combined with non-bacterial tissue inflammation, was to be expected and is well substantiated by experimental and clinical data (Mackay & Burnet, 1963).

As in the other groups, it is not yet known which of the presently known vasoactive compounds are responsible for the increased capillary permeability in this condition but it seems likely that histamine, the "globulin-peptide" capillary permeability systems and possibly also xanthosine (Spector & Willoughby, 1957a) play the most prominent part.

D. Conclusion

The pleural effusions in systemic lupus erythematosus appear to be due in part to increased pleural capillary permeability and in part, certainly in some patients, to decrease in protein absorption.

Both abnormalities are corrected by the administration of prednisolone. The results are consistent with the pathological concept of pleural vasculitis.

CHAPTER VII

Effusions associated with

1. Subphrenic Abscess
2. Granulosa Cell Tumour of Ovary

SUBPHRENIC ABSCESS

A. Clinical data

Case 61 was a man of 50 who was admitted to hospital with a six-hour history of severe upper abdominal pain of sudden onset, without vomiting or diarrhoea. There was no previous history of dyspepsia and he did not complain of shoulder pain. On examination there was marked guarding of the upper abdomen and bowel sounds were absent. He was thought to have a perforated ulcer but abdominal X-ray did not demonstrate gas under the diaphragm and there was no leak of gastrografin. The abdominal symptoms improved but he developed, after ten days, an intermittent pyrexia and a right pleural effusion followed a week later by an effusion on the left and a small haemoptysis. There was no chest pain.

The pleural fluid was clear straw-coloured. It contained numerous lymphocytes and a few polymorphs, red cells and serosal cells but no malignant cells. The colloid osmotic pressure of the fluid was 17.0 cm. H_2O (alb. conc. 2.3 g.%) and of the plasma was 30.1 cm. H_2O (alb. conc. 3.35 g.%). The colloid osmotic pressure of the fluid was therefore 57% that of the plasma.

The liver edge gradually descended further below the costal margin and felt firm and slightly irregular. Liver biopsy was normal. It was at this stage, when pulmonary embolism and carcinoma were the favoured possible diagnoses, that the

albumin turnover studies were undertaken, the response to intrapleural prednisolone also being assessed.

Immediately after the investigation was completed, repeat X-ray of the diaphragmatic area revealed a fluid level between liver and diaphragm. The abscess was drained following exploration of the subphrenic space, about a pint of foul-smelling pus being released, and the patient made an uneventful recovery. A profuse growth of B. coli was obtained from pus aspirated prior to incision of the abscess. Subsequent barium meal examination showed deformity of the duodenal cap - cholecystogram was normal.

B. Results

Alb(E) was 4.9 g. during the initial assessment and was reduced to 3.0 g.% during the time that prednisolone was administered intrapleurally. Alb(L) remained virtually constant at about 4.5 g.. The effusion volume was constant at 800 ml. initially, falling to 500 ml. at the end of the second phase. The hydrostatic pressure fell from 5.2 cm. to 4.0 cm. during this latter phase.

C.
DISCUSSION

(a) Pathology

Subphrenic abscess may lie between the liver or the stomach or spleen and the diaphragm. In most cases the

original focus is in the upper part of the abdomen, stomach, duodenum or liver, and in most cases, other than the liver, appendix or kidney, the pus lies within the peritoneal cavity but isolated by adhesions. Sometimes it may rupture into the peritoneal cavity causing peritonitis and sometimes through the diaphragm. Frequently it is associated with pleural effusion but the pleural space rarely becomes infected unless the abscess ruptures into it.

(b) Correlation of Pathology with Results

The inflamed oedematous diaphragm supplied by systemic capillaries at systemic capillary pressure would be expected to leak protein at fairly high rates into the pleural cavity and 4.9 g./24 hours is quite a substantial leak, taking into consideration the comparatively limited area affected by the abscess.

Alb(L) is perhaps surprisingly high for an effusion of this size (not being far outwith the predicted value obtained in effusions associated with pulmonary infarction), as it would seem likely that the lower mediastinal area is involved to some extent in the inflammatory process. Also the diaphragm is paralysed though, of course, it remains mobile. This effusion had been established for three weeks prior to the investigation and this may have given time for some lymphatic adaptation in the higher intercostal spaces to the increased drainage flow required of them. All cultures of pleural fluid were sterile

and there is no reason to suppose generalised parietal pleural inflammation. If that had been so, Alb(L) would have been very much lower and would have been considerably increased by prednisolone. As it was, only Alb(E) was altered significantly by prednisolone. However, even Alb(E) remained at the raised level of 3 g. in contrast to the greater reduction found in patients with tuberculosis and disseminated lupus erythematosus and even in many patients with carcinoma.

It would appear that the increased capillary permeability found in association with an accumulation of pus such as this is only partially reversible with prednisolone, certainly when prednisolone is administered in this dose.

D.
CONCLUSION

A subphrenic abscess causes pleural effusions by increasing the rate of protein entry into the pleural cavity and does not have much effect on the rate of protein leaving.

Prednisolone only partially supresses the increased capillary permeability in these circumstances.

MEIGS' SYNDROME

A. Clinical and pathological data

Case 62 was a female patient, aged 54, who presented with a fixed pelvic mass, ascites and a right pleural effusion. The mass was proved at autopsy to be a malignant granulosa cell tumour of the ovary and malignant cells were seen in the ascitic fluid. There was no evidence at autopsy of tumour seeding in the lymphatic drainage of the peritoneal cavity or in mediastinal glands. Tumour cells were not found in the pleural fluid during life, nor was any tumour tissue identified at autopsy in the lungs or in the walls of the right pleural cavity. Cells thought to be tumour cells were seen in one out of four specimens of ascitic fluid sent for cytological examination.

Meigs' syndrome is strictly defined as occurring in association with (a) a benign solid tumour with the gross appearance of a fibroma, (b) ascites and (c) pleural effusion. A further condition for acceptance under the strict definition is that the ascites and pleural effusion must disappear with removal of the fibroma (Meigs, 1954).

This patient cannot, therefore, be included as a case of Meigs' syndrome. Nevertheless, in view of the autopsy findings, which did not show any evidence of lymphatic, pleural, or lung involvement, and of the fact that the ascitic and pleural fluid was never blood-stained, it is likely that the results of this

investigation throw further light on the pathogenesis of the ascites and pleural effusion in Meigs' syndrome.

B. Review of theories and experimental work

The condition has so far been most inadequately investigated and explanations for the fluid collections have been largely confined to speculation based on gross and microscopic examination of the tumour and an investigation carried out by Meigs, Armstrong & Hamilton in 1943.

Microscopic examination has shown that, although fibroma is the most common tumour responsible, thecomata, granulosa cell tumours, cystadenomata, Brenner's tumour and papillomata of the uterine tube have also been described in association with the syndrome. Uterine tumours have also been responsible for the fluid collections as has pancreatitis, and pancreatic tumours, and malignant ovarian tumour without evidence of metastases. Although occurring in association with tumours of widely differing cell type, it remains surprising, therefore, that by far the largest number occur in association with the comparatively uncommon ovarian fibroma. Pathologists have commented on the vascularity of the tumour, its marked interstitial oedema and the frequent finding of cyst formation within it (Calmenson, Docherty & Bianco, 1947; Lawson, 1950; Marshall, 1949; Rubin, Novak & Squire, 1944). Lawson (1950) and Meigs (1954) and others have demonstrated a weight loss of between 30% and

75% in 24 hours in the tumour when it is placed in a dry pot following resection.

The protein and albumin concentration of the ascitic and pleural fluids are usually very similar. They seem to be at concentrations between those of exudates and transudates where they are described in the literature, but the serum protein levels are not often given and it is, therefore, difficult to make an accurate physiological evaluation. It does appear, however, that the serum albumin tends to be reduced in this condition and that the effusions can be regarded as exudates and not transudates as Meigs (1954) suggested. Certainly this patient's effusion fell into the category of exudate (serum albumin 4 g.%, ascites albumin 2.8 g.%, pleural effusion albumin 3.1 g.% and colloid osmotic pressure 28.8, 16.6 and 17.1 cm. H_2O respectively).

In 1943 Meigs, Armstrong & Hamilton injected carbon particles into the peritoneal cavities of two patients and found an almost equal concentration within the pleural cavity after 24 hours. They felt that this demonstrated a current of flow from peritoneal to pleural cavity. Meigs concluded that "fluid" created by the presence of the fibroma was delivered either through sub- and supra-diaphragmatic lymphatics or even through interstices between the cells of the diaphragm. Lemming (1960) described two patients with ascites and pleural effusion, one with a myoma and the other with an ovarian cystadenoma, in a

paper entitled "Meigs' syndrome and the pathogenesis of pleurisy and polyserositis". The title is perhaps a little unfortunate as there is no evidence that "serositis" has any part to play in this condition. Nevertheless, he stresses the importance of the diaphragmatic lymphatics in clearing fluid and protein from the peritoneal cavity and suggests that a factor in producing the pleural effusions may be the integrity of the mediastinal lymphatic drainage. If the latter is affected by past disease, such as primary tubercle, "spill" into the pleural cavities would be increased. There is experimental evidence of a capacity for very rapid transfer of ascitic fluid through the diaphragmatic lymphatics of the cat (Courtice & Steinbech, 1950). These authors also showed that the ratio of lymph clearance from the cat's peritoneal cavity, right lymph duct to thoracic duct, was 4:1, and also (Courtice & Steinbech, 1951) that obstruction of parasternal lymphatics resulted in oedema of the mediastinum and fluid collection in the pleural cavities. This latter work supported the theory of one-way permeability of capillaries and two-way permeability of lymphatics. The results are confirmed in man in a report by Stephanopoulos & Doucas (1962) who noted a rapid increase in ascites following operation on the pleural effusion of a patient with an ovarian cystadenoma. They considered that diaphragmatic lymphatics had been injured at operation and ascribed the accumulation of ascites and disappearance of the pleural effusion to this.

Nairn (1957) considered that there was a direct trans-diaphragmatic fluid pathway between the peritoneal and the pleural cavities, probably provided by local lymphatics, and that fluid leaked into the pleural cavities due partly to overfilling of the lymphatics and partly to the mean negative intrathoracic pressure. He showed in rabbits and rats that pleural effusions occurred if ascites was induced by injecting Ringer's solution intraperitoneally, but not if it was injected subcutaneously. The flow was slowed down by intraperitoneal injection of graphite which blocked the lymphatics. Urinary clearance of the excess fluid was quicker if it was injected subcutaneously rather than intraperitoneally. This is presumably due partly to the greater capillary surface area available subcutaneously compared to that in the lining of the pleural and peritoneal cavities, and partly to the one cell layer increase in the partition between the ascitic and pleural effusion fluid and blood.

However, despite this fact, studies using tritium labelled water have shown remarkably high transfer rates, 40-80% per hour in the case of ascites (Prentice, Siri & Joiner, 1952) and at least 40% per hour in the case of pleural effusions (Clauss, Yacoubian & Barker, 1957) and it would appear that it is again the large colloid molecules which are of chief osmotic importance in the effusions of Meigs' syndrome rather than the rapidly diffusing small molecules such

as water, sodium, chloride and potassium. Such an accumulation of plasma proteins must take place either because excess protein leaks into the peritoneal space or because there is diaphragmatic lymphatic obstruction or both. Lymphatic obstruction at diaphragmatic level would exclude the above interpretation as the cause of the pleural effusions, and high protein entry into the peritoneal cavity would appear more likely. Investigation of Case 62 was, therefore, designed to elucidate the rates of Alb(E) and Alb(L) for both the ascites and the pleural effusion and to attempt to discover whether an excess of capillary permeability factors were present either in the ascites or the urine.

C. Procedure of investigation

The times of intraperitoneal and intrapleural injection of R.I.H.S.A. and withdrawal of samples of fluid and of blood are shown in Figure V. Three hours were allowed for mixing following intraperitoneal injection and the patient changed from side to side every ten minutes. A little over two hours was allowed for intrapleural mixing. Otherwise the basic technique was similar to that already described on pages 12-15.

D. Results

The results are tabulated in full in Appendix B and Table VIII.

FIGURE V

Granulosa Cell Tumour Ovary

Method of Investigation

Day	Hours	Peritoneal Cavity		Pleural Cavity		Blood	
		In	Out	In	Out	In	Out
1	0.0 3.0 17.0	2.013g.R.I.H.S.A.	1 2		2		2
2	3.0 17.5		3 4		3		3
3	2.5 2.55 5.5	1.914g.R.I.H.S.A.	5 6		5		5
4	5.0 20.75		7 8		7 8		7 8
6	4.5 4.55 7.0 17.5		9	2.385g.R.I.H.S.A.	9 10 11		9
7	4.3 17.5				12 13		
8	1.0 1.05 3.2		14	2.480g.R.I.H.S.A.	14 15		14
9	0.0 0.05 0.33 1.43 4.5					4.562g.R.I.H.S.A.	16 17 18 19

TABLE VIII

Case 62 - Results

Granulosa Cell Tumour Ovary

	Day	Volume(ml.)	Alb. Conc. g.%	Total Alb.	Alb(E)	Alb(L)
Ascites	1	4500	2.8	120	55	60
	3	4000				
Pleural Effusion	6	2350	3.1	70	27	28.4
	8	2200				
Plasma	9	3100	4.0	120		
Interstitial Space				120		
Total				430		

Alb(E) = albumin entering in g./24 hours
Alb(L) = albumin leaving in g./24 hours

Urine Tryptamine 347 µg./24 hours range (30-120)

The albumin turnover rate in the ascites was so high that re-entry of labelled protein had to be taken into account giving a final result of 55 g. for Alb(E) and 60 g. for Alb(L). Entry of labelled albumin from the ascites during the period of investigation of the pleural effusion also had to be taken into account and resulted in a value for Alb(E) of 27 g. and Alb(L) of 28.4 g..

On the assumption that the interstitial space (excluding the peritoneal and pleural cavities) contained approximately the same amount of albumin as the plasma, i.e. 120 g., the total extracellular albumin estimated to be present in this patient was 430 g., which is a remarkably high figure.

Table IX indicates the levels of urinary oestrogen and 5-hydroxy indole compounds. No 5-hydroxy indole compounds were detected in the ascitic fluid and the blood 5-hydroxytryptamine levels were below normal. The urinary oestrogens were at the upper limit of the normal range and the only real abnormality was the raised urinary tryptamine excretion. Bradykinin concentration in the ascitic fluid was estimated by the bioassay method described by Lahiri (1962) and was less than 0.4 nonogram/ml.

E. DISCUSSION

If Courtice's (1951) work on the dog's lymphatic clearance of protein applies at all to humans, it is likely that albumin

TABLE IX

Case 62

Special investigations carried out on Blood, Ascitic Fluid and Urine

Ascitic fluid	No 5-hydroxy-indole compounds detected No kinin activity detected			
				Normal range
Blood	5-hydroxytryptamine	18	m.ug/ml.	50-300
Urine	5-hydroxytryptamine	38	ug/24 hrs.	30-120
	Tryptamine	347	ug/24 hrs.	30-120
	Indol-3-ylacetic acid	1.5	mg/24 hrs.	5-12
	Indol-3-yl lactic acid	9.5	mg/24 hrs.	(?0-20)
	5-hydroxy-indole-3-ylacetic acid	2.5	mg/24 hrs.	2-9

			Mean
Oestrone	1.6 ug/24 hrs.	0.8-7.1	2.5
Oestradiol	0.5 ug/24 hrs.	0.0-3.9	0.6
Oestriol	4.7 ug/24 hrs.	0.6-8.6	3.3

turnover in the peritoneal cavity does not normally exceed 6 g./24 hours.

A raised capillary-ascites albumin transfer rate with a maximum of 24 g./day has been shown to occur in portal cirrhosis (Berson & Yalow, 1954; Schoenberger, Kroll, Eckert & Kark, 1956). The latter authors also showed a significant reduction in one patient after treatment. Although the ascites-plasma Alb I^{131} equilibration took over a month, using tritium water, as has been mentioned already, molecules have been shown to enter and leave the peritoneal cavity rapidly - 40-80%/hour (Prentice, Siri & Joiner, 1952). Normally, venous obstruction produces only a small increase in total protein leak from the congested tissue (White, Field & Drinker, 1933), although the lymphatic flow may be increased many times. In ascites due to portal hypertension stabilisation of the aqueous component may be almost instantaneous but even a slight increase in portal capillary permeability to protein could result in a disturbance of protein balance within the peritoneal cavity, causing its accumulation and ascites formation independent of general anasarca. It is probable that passive increase in portal capillary permeability due to capillary hypertension is only part of the reason for ascites formation in this condition and it is difficult to see how capillary congestion can have any significant part to play in the ascites of Meigs' syndrome. Torsion of the tumour itself might be expected to cause

compression of blood vessels in the pedicle but this is rare and could at best explain only a very small percentage of the effusions. It is inconceivable that pressure on pelvic systemic veins could produce ascites before producing oedema of the lower limbs.

Again therefore (see Pulmonary Embolism and Bronchial Carcinoma) the conclusion seems inevitable that some of these pelvic tumours secrete capillary permeability factors which, acting in vascular tumour, produce oedema in it and raised protein exudation into the peritoneal cavity, both from tumour capillaries and possibly also from capillaries on the peritoneal surface. There is no histological evidence of serositis as such. The results of estimation of urinary hydroxytryptamine and its vasoactive derivatives show a "raised" value (Table IX) for tryptamine. The significance of the raised urinary excretion of tryptamine is unknown. In the absence of this or any of the other 5-hydroxyindole compounds in the ascitic fluid it seems rather unlikely that it has significance in relation to the high rate of Alb(E). The blood 5-hydroxytryptamine level was, if anything, low.

Oestrogens are known to increase capillary sensitivity to bradykinin and for this reason urinary oestrogen levels were determined. These were at the upper limit of normal but were not significantly raised and there was only mild cystic hyperplasia of the uterus. It seems unlikely, therefore, that the

endocrine origin of the tumour cells in this patient necessarily prevent the results from having general applicability. Little is known about the role of bradykinin in controlling capillary permeability in physiological and pathological situations (Bhoola, Calle & Schacter, 1960; Ghosh, Banerjie & Mukherji, 1963). The level of bradykinin activity in the ascitic fluid of this patient was negligible. The negative result, however, does not necessarily exonerate bradykinin as a possible capillary permeability factor of importance in this patient, for, even if produced in excess by the cells of the tumour, bradykinin might well have been destroyed as soon as it entered the ascitic fluid.

F.
CONCLUSION

(1) Although the patient investigated had a malignant granulosa cell tumour, the mechanism of production of the ascites and pleural effusion was probably similar to that of the true Meigs' syndrome.

(2) The ascites was due to a very high rate of albumin entry into the peritoneal cavity, the cause of which has not been ascertained. The pleural effusion was due to leakage of lymph into the pleural space from lymphatics draining the peritoneal space.

CHAPTER VIII

Effusions associated with Congestive Cardiac Failure

CONGESTIVE CARDIAC FAILURE

A. Clinical data

(1) General

There were six patients (Appendix A VI) whose effusions were associated with congestive cardiac failure. In five the cardiac failure was chronic and the effusions were either persistent or very slow to clear and in one patient (Case 44) a small effusion was associated with left ventricular failure following myocardial infarction on a background of hypertension and this was clearing rapidly at the time of the investigation. This was the only patient in the whole series in whom there was evidence of a major change in the protein concentration of the effusion during the period of the investigation. In Case 42 the bilateral effusions were accumulating so rapidly that the investigation was limited to 24 hours.

The investigation was repeated in one patient (Case 46) in whom bilateral effusions recurred following discharge. They were resistant to digoxin and diuretic therapy on re-admission to the ward, despite only slightly raised jugular venous pressure and the complete absence of oedema.

In three patients (Cases 41, 42 and 43) the cardiac failure was associated with rheumatic heart disease and in the other three it was due to myocardial degeneration secondary to coronary arterial disease. In four patients the effusions were

bilateral but in Cases 43 and 45 they were confined to the right side. Case 43 had right-sided pleuritic chest pain on the day prior to admission and staphylococcus aureus was isolated from the sputum on admission. The chest radiograph suggested an area of consolidation at the right base and this may have predisposed to the unilateral fluid accumulation. Case 45 presented with markedly raised jugular venous pressure following a myocardial infarct, no peripheral oedema but a quite rapidly accumulating right-sided pleural effusion. After three aspirations at another hospital the fluid continued to accumulate and control was only achieved finally (following the investigation) by intercostal intubation and water seal drainage of the pleural space. It was considered clinically most likely that he had developed a right-sided pulmonary venous thrombosis which in the presence of cardiac failure predisposed to unilateral accumulation of fluid of low protein content, but this was, of course, no more than conjectural.

The colloid osmotic pressure of the pleural fluid in each case was never greater than 30% that of the serum and this on its own is regarded as sufficient evidence to justify inclusion in this group. However, it may still be that pulmonary arterial or venous occlusive disease played an additional part in the pathogenesis of the effusions. Atrial fibrillation was present in Cases 42 and 43 and the right atrium could have been a possible source of embolus. Case 41 developed leg vein

thrombosis subsequent to the investigation and then a pulmonary embolus with severe pleuritic pain.

Autopsy examination in Case 42 carried out two weeks after the investigation revealed left pulmonary arterial thrombosis but it could not be satisfactorily determined to what extent this was present at the time of the investigation.

However, it remains true that in every patient in this group cardiac failure was clinically manifest at the time of the investigation and was the most important cause of the pleural effusions in each case.

B. Results

The results are presented in an order determined by effusion volume (Table X). The volume varied between 2,750 ml. and 740 ml.. In contrast to the pulmonary embolism group there tends to be, if anything, an increase in Alb(L) with decreasing effusion volume. In one patient (Case 42), with the very rapidly accumulating left pleural effusion, there was a very high rate of Alb(E) - 8.2 g. - and as has been mentioned in the preceding paragraph, pulmonary artery thrombosis may have been present at the time of investigation.

In Case 49 Alb(E) was increased at the second admission when the effusions were accumulating more rapidly then previously. In this patient the effect of intrapleural prednisolone was also observed. Although the effusion continued to accumulate at

TABLE X

Pleural Effusions Associated with Congestive Cardiac Failure

Results

Case	Side	A	B	C	Prot(g%)	Alb(g%)	Pre-Prednisolone		Post-Prednisolone	
							Alb(E)	Alb(L)	Alb(E)	Alb(L)
42	L	2750	3600		1.0	0.6	8.2	2.8		
45	R	1800	2026		2.3	1.6	5.7	3.4		
46(1)	R	1080	1090		1.5	0.8	2.7	2.4		
46(2)	R	1020	1140	1470	1.7	1.0	4.5	3.6	2.5	2.0
43	R	1060	1000		1.9	1.0	2.4	2.4		
41	R	1020	780		1.0	0.7	3.7	4.2		
44	L	740	360		1.8_a 3.3_b	1.6_a 2.5_b	3.9	4.6		

A = effusion volume (ml.) at 0 hours
B = effusion volume (ml.) at 48 hours
C = effusion volume (ml.) at 144 hours

Alb(E) = albumin entering in g./24 hours
Alb(L) = albumin leaving in g./24 hours

1 = First admission
2 = Second admission

a = prot. and alb. concentration at 0 hours
b = prot. and alb. concentration at 48 hours

about the same speed, both Alb(E) and Alb(L) were reduced by about half.

C. Discussion

(1) Pathology

Cardiac failure is associated with total body water increase. This increase takes place largely in the extracellular space and is associated with sodium retention by the kidney. As increase in weight and increase in blood volume can be shown to occur before there is any rise in central venous pressure (Warren & Stead, 1944), it would seem that sodium retention by the kidney is one of the earliest features of cardiac failure. A central venous pressure rise which occurs usually as the cardiac failure becomes clinically detectable is also bound to contribute to extracellular fluid accumulation (Starling, 1896; Landis, 1927). The accumulation of extracellular fluid in cardiac failure does not appear to be associated with much increase in capillary permeability to protein, certainly as it occurs outside the lungs. White, Field & Drinker (1933) showed that raising the venous pressure in a dog's leg from 14mm.Hg. to 26mm.Hg. resulted in about a twofold rise in lymph flow but only a very slight increase in protein clearance. If, however, the pressure was further increased to about 40mm.Hg., there was approximately a fourfold increase in lymph flow and almost a 40% rise in protein clearance. It would appear then that at lower intra-

luminal pressures there is little increase in capillary permeability to protein, but that this is definitely increased when the capillaries are more severely distended at higher pressures.

It might be thought that oedema would compress lymphatic channels and result in a vicious circle of retention of tissue fluid. However, lymphatics are, in fact, dilated by oedema. (Pullinger & Florey, 1935; McMaster, 1947). McMaster (1937) has investigated lymph flow in cardiac oedema by intracutaneous injection of dye and shown ready filling of the local lymphatic plexus, but a comparative absence of "streamers" suggested diminished overall lymphatic drainage. The protein content of oedema fluid found in patients with cardiac failure is normally less than 0.5 g.% (Stead & Warren, 1944), so that it is unlikely that reduced lymph flow is ever a significant factor in the production of the oedema of cardiac failure on its own. However, if the capillary hydrostatic pressure is raised, reduced lymphatic drainage will tend to increase oedema formation and the protein concentration in these circumstances will not necessarily be high.

From the work of Ross, Walker & Hammond (1960) it would seem that in cardiac failure the transcapillary exchange of albumin is generally reduced. In three patients the transcapillary exchange rate of radioiodinated human serum albumin was also estimated after recovery from cardiac failure. It was

found that with return of plasma volume and intravascular albumin mass to normal there was an increase in the R.I.H.S.A. transcapillary exchange rate so that it appears that the process is reversible.

The discussion so far has been concerned with factors responsible for extracellular fluid accumulation in cardiac failure as it may occur at any situation in the body. Sometimes, however, pulmonary oedema may occur, and pleural effusions also, in the absence of clinical evidence of extracellular fluid accumulation at any other site. Also pleural effusions may persist after all clinical evidence of cardiac failure has disappeared. The effusions have a low colloid osmotic pressure and are referred to as transudates. It is surprising that such transudates can persist without evidence of pulmonary oedema but such a clinical situation does exist. There would appear to be two possible reasons for this. Firstly, the influence of mean negative intrathoracic pressure on capillary fluid exchange (Figure I) will be most marked where it is most negative, i.e. at the visceral pleural surface. Secondly, pulmonary capillaries receive most of their oxygen supply directly from the alveolar air (Drinker, 1945) and subpleural alveolar filling with transudate will tend through anoxia to increase permeability of the local capillaries (Warren, Peterson & Drinker, 1942) so that a vicious cycle of increasing anoxia and oedema will be established. It is likely also that under these circumstances a degree of

shunt will develop between the bronchial and pulmonary vessels (Pump, 1963) so that the subpleural pulmonary capillaries will become perfused with blood from the systemic circulation at systemic capillary pressure. However, if pulmonary capillary permeability is increased in left-sided heart failure, it never appears to be increased to the extent at which there is a significant increase in permeability to protein, as the diagnostic criterion is a low colloid osmotic pressure of the effusion fluid.

In many patients, therefore, retention of body sodium results in accumulation of extracellular fluid at a site which depends on the intracapillary hydrostatic pressure and the surrounding tissue pressure. Only very rarely does the capillary pressure rise to such an extent in true cardiac failure as to make a significant difference to capillary protein leak but this situation can occur in lung. It is also well known that pulmonary arterial and venous thrombosis may occur when blood flow through the pulmonary vessels is reduced (Best & Heath, 1958). Magidson & Jacobson (1955) have shown that it is always difficult to be sure whether the arterial thrombosis occurs as a primary condition or is secondary to arterial embolisation. It could be, therefore, that many of the pleural effusions occurring in association with cardiac failure have coincidental pulmonary emboli or vascular thromboses. Again, however, it must be emphasised that,

although these vascular lesions may predispose to effusions, they will not cause a significant accumulation of fluid of low colloid osmotic pressure relative to plasma unless cardiac failure is also present.

(2) Correlation of Pathology with Results

In all the patients investigated Alb(E) was increased above the normal which probably lies between 0.5 and 2 g.. However, in four patients the increase was only very slight, and yet three had effusions of over 1,000 ml. volume. The effusions were relatively static and venous pressure was not markedly raised. In none of the four was there marked oedema.

The apparent contradiction between these results for Alb(E) and those of Ross, Walker & Hammond (1960) is not a real one, being presumably due to the fact that these authors were dealing with generalised capillary reaction to cardiac failure, whereas this investigation is concerned with a very restricted capillary bed.

Two patients, Cases 42 and 45, had very large effusions which were accumulating at the time of the investigation. In Case 42 the colloid osmotic pressure of the effusion fluid was 23% that of the plasma and Alb(E) was 9.2 g. suggesting that capillary pressure was generally high (venous pressure - 8 cm.; generalised oedema) and that capillary permeability was also quite markedly increased. It was not clear at autopsy in this patient whether the pulmonary arterial thrombosis noted on the

investigated side had been present at the time of the investigation but it is presumed that the increased capillary permeability was mainly in the visceral capillaries. In Case 45 the colloid osmotic pressure of the effusion fluid was 26% that of the plasma, indicating that the pulmonary capillary pressure was probably also fairly high (venous pressure 5 cm.; no generalised oedema) and Alb(E) was 5.7 g., showing moderately increased capillary permeability to protein. This was a chronic effusion which was unilateral and it seemed likely that it was associated with pulmonary vascular occlusion, whether venous or arterial.

In the patients with lower Alb(E) it appeared probable that the effusions were more due to general extracellular fluid accumulation than to local capillary hypertension and clinically this seemed to be the case. Alb(E) was interestingly high (3.9 g.) in Case 44, in whom a relatively small effusion was diminishing rapidly in size and in whom the general improvement in heart failure with a negative sodium balance was reflected in a rising concentration of intrapleural protein from 1.8 to 3.3 g.% during the 48 hours of the investigation. It is assumed that in this patient energetic diuretic therapy had resulted in sodium excretion but that some left ventricular failure with quite markedly raised pulmonary capillary pressure remained. The other possibility is that there was an underlying pulmonary infarct, but there was no evidence of this radiographically.

Alb(L) was generally low except in Cases 41 and 44 where energetic treatment of the cardiac failure had resulted in fall of systemic venous pressure. It would appear that the lymphatic clearance of the pleural space under the circumstances of a markedly raised systemic venous pressure is reduced. It seems rather unlikely that there is actual obstruction to the right lymph duct by raised venous pressure in the neck as the lymph duct is capable of building up a pressure of 50 cm. H_2O. It appears more probable that the apparently low lymphatic output of protein is secondary to the local retention of fluid, but in view of the intercostal drainage of lymph through systemic and not pulmonary tissue, this is still not an entirely satisfactory explanation.

The effect of intrapleural prednisolone was only assessed in one patient who had persistent pleural effusions. In this patient prednisolone brought about quite a marked reduction in Alb(E). However, the effusion continued to increase in volume at approximately the same rate as previously due to an almost equal reduction in Alb(L). The fall in Alb(E) would be expected as a direct effect of prednisolone on capillary permeability but the fall in Alb(L) is more difficult to explain. During the four days of intrapleural prednisolone administration there was quite a marked fall in urine output and rise in body weight. This increase in sodium retention, presumably due to prednisolone, brought about slight reduction in effusion protein concentration.

It would appear unlikely, however, that this degree of fluid retention could be the sole cause of such a fall in Alb(L). It is possible that the effect of prednisolone in increasing Alb(L) in inflammatory effusions is due solely to its effect on the parietal pleura and that, in fact, its prime effect on lymphatic capillaries is to decrease their permeability to protein. In cardiac failure such an effect would produce a decrease in Alb(L).

D.

Summary

The effusions of heart failure have a colloid osmotic pressure which is less than 30% that of the plasma.

When occurring unilaterally or as the sole evidence of sodium retention in cardiac failure, it is likely that there is some pulmonary vascular pathology. However, it has been suggested that the subpleural area of lung is likely to be most ischaemic under these conditions and the subpleural capillaries, therefore, the most permeable.

Alb(E) in congestive failure is variable. It is usually slightly raised but is only high in rapidly accumulating effusions or in effusions associated with concomitant pulmonary vascular thrombosis.

Alb(L) tends to be low except when the effusions are receding with recovery from cardiac failure.

It is suggested that corticosteroids may have a direct effect on lymphatic as well as on blood capillaries in

reducing their permeability to protein.

TABLE XI

Colloid Osmotic Pressure($cm.H_2O$)

No. of Cases	Diagnosis	Colloid Osmotic Pressure($cm.H_2O$)		Effusion osmotic pressure expressed as percentage of plasma osmotic pressure
		Effusion	Plasma	
5	Tuberculosis	20.0	29.1	68
6	Embolism	15.9	28.9	55
7	Lung Carcinoma	14.6	25.1	58
4	Mammary Carcinoma	16.3	26.8	61
1	Lymphosarcoma	11.5	18.1	64
4	Systemic Lupus Erythematosus	16.7	29.4	57
6	Congestive Cardiac Failure	5.4	23.9	23

CHAPTER IX

Final Discussion

FINAL DISCUSSION

A. The Method of Investigation

As with all investigations the results and, therefore, the inferences drawn depend entirely on the validity of the method used to obtain them. There has been some work of a similar nature but unfortunately it has been very limited and has been mainly carried out on animals. Burgen & Stewart (1958) described a method for measuring "the turnover of fluid in the pleural and other serous cavities" using T-1824 labelled protein and R.I.H.S.A. and again Stewart & Burgen (1958) in a paper entitled "The turnover of fluid in the dog's pleural cavity" employed a method embodying many of the principles involved in this investigation. Also in later work, incidentally published after this investigation was begun, Stewart (1963) used an almost identical method, but with T-1824 labelled effusion protein, to calculate fluid turnover in a small number of effusions occurring in humans. It is unfortunate that his results are expressed in terms of "fluid formation and lymphatic removal" and not as albumin turnover, as this is based on a misconception that albumin and fluid turnover are directly related. Water is, of course, handled quite differently from protein by both the blood and the lymphatic capillaries. Results are also expressed in ml./kilo body weight. The patients in the present investigation appeared to

have greater variance in weight than in the volume of their thoracic cage and it was felt that it would probably be more misleading to correct the results for weight than otherwise. However, the mean of the results for each group has been expressed in this way (Table XII) A disturbing feature of Stewart's investigation was introduced when he stated that T-1824 was never detected in the circulation and, therefore, re-entry of dye from the circulation into the effusion (which would lead to a falsely low albumin entry value) did not have to be taken into account. This suggested two possibilities; firstly, that the biochemical estimation of dye in the plasma was not sufficiently sensitive, and secondly, that in the physiological situation the dye was becoming detached from albumin and either excreted from the body or bound in it so that negligible amounts remained in the blood stream. In either event, the validity of the results must be in some doubt.

Table XIII expresses the mean results for each group of the present investigation in mg. alb./kg./hr., and, on the same basis as in Stewart's work, the conversion of "albumin leaving" to "lymphatic flow". It will be seen that the "lymphatic flow" in cardiac failure, determined in this investigation with radioiodinated human serum albumin is considerably lower than the values obtained by Stewart. In view of the slight doubts about the stability of the dye-protein complex, it is likely that the lower results are more correct.

TABLE XII

No. of Cases		Av. Vol.	Column I		Column II	Av. Weight	Column III	
			Alb(E)	Alb(L)	Alb(L)		Alb(E)	Alb(L)
7	Tuberculosis	920	4.7	2.4	2.6	54	87	48
8	Pulmonary Embolism	740	4.4	4.8	6.5	60	73	108
9	Lung Carcinoma	1100	4.6	2.3	2.1	67	69	31
5	Mammary Carcinoma	1200	3.4	1.8	1.5	56	61	27
4	S.L.E.	880	3.8	2.4	2.5	64	59	39
7	Cardiac Failure	1350	4.4	3.3	2.4	56	79	43

Column I Alb(E) = albumin entering in g./24 hours (average of group)
Alb(L) = albumin leaving in g./24 hours (average of group)

Column II Alb(L) = albumin leaving in g./24 hours corrected for average volume of group

Column III Alb(E) = albumin entering in mg./kilo/24 hours
Alb(L) = albumin leaving in mg./kilo/24 hours corrected for average volume of group

TABLE XIII

Comparison of Results of this Investigation with those of Stewart (1963)

	No.of* Cases	Average* Effusion Volume	Alb(L)* mg./alb./ kg./hr.	Lymphatic* flow ml./ kg./hr.	Lymphatic° flow ml./ kg./hr.	No.of° Cases	Average° Effusion Volume
Tuberculosis	7	920	2.0	0.07	0.20(0.21)	1	1037
Pulmonary Embolism	8	740	4.5	0.20	-	-	
Lung Carcinoma	9	1100	1.3	0.06	0.20(0.30)	1	1687
Secondary Carcinoma	5	1200	1.1	0.04	0.07(0.16)	1 (Ca.Ovary)	2260
S.L.E.	4	880	1.6	0.07	-	-	
Cardiac Failure	7	1350	1.8	0.16	0.37(0.40)	7	1050

* Results refer to this investigation

° Stewart's (1963) investigation

All results corrected for effusion volume except those bracketed

Whether the results of this investigation, using an artificial compound (R.I.H.S.A.) despite the great volume of work which has expressed its physiological stability, can yet be regarded as an accurate estimate of true albumin turnover must remain in slight doubt, but they must be a reasonably close approximation to it.

As described in the introductory chapter (page 7) the possible variables affecting protein turnover, such as pressure (Stewart & Burgen, 1958), volume, and protein concentration, which could alter during the course of the investigation, have been taken into account. One factor which it was not considered possible to record is ventilation. That this could change during the course of the investigation and so affect albumin absorption by the parietal lymphatics has already been recognised. It may well be that some of the reduction in Alb(L) which occurred in a few patients after prednisolone administration was due to a soothing effect of the drug on respiratory distress.

The only other authors who have tried to form an estimate of albumin turnover in pleural effusions are Coe & Aikawa (1961), who in two patients with cholesterol effusions and in six with other types of effusions, worked out a plasma to pleural fluid equilibration time after injecting radioiodinated human serum albumin intravenously. No attempt was made in their investigation to measure any change in volume of the effusion, so

that relative rates of albumin entering and leaving the effusion could not be calculated. They found the equilibration time varied from about five to fifteen days, the longer equilibration time tending to be associated with the more chronic effusions.

The rate of capillary to ascites transfer of albumin has been estimated by intraperitoneal introduction of radioiodinated human serum albumin by Berson & Yalow (1954) and Schoenberger, Kroll, Eckert & Kark (1956) but again account was not taken of change in volume of the ascites, so that no calculation could be made of the rate of albumin leaving the ascites.

Re-entry of radioiodinated human serum albumin into the effusions was only considered to be of importance in the patient with the "pseudo-Meigs'" syndrome in association with a malignant ovarian granulosa cell tumour. In this patient albumin turnover in the pleural effusion was over three times more than the highest turnover found in any other patient and about a seventh of the total albumin pool was passing in and out of the ascitic fluid each day. In the other effusions albumin leaving the effusion never made up more than 5% of the total exchangeable albumin and usually very much less. As metabolism of albumin with loss of its radioactive label takes place at a rate of about 6.7%/day of the total exchangeable albumin pool (Sterling, 1951), it would appear impossible for re-entry to affect the results by more than 4%. In the majority of patients with an albumin turnover of about 3.5g./day the results would be altered

by no more than 2%, even in the smaller effusions. In two patients with bilateral effusions of approximately similar volume radiographically, Case 32 and Case 51 whose effusions were due to S.L.E. and lymphosarcoma respectively, these calculations were verified. The rise in radioactivity of the effusion on the contralateral side to the investigation amounted to approximately 1%/day of the mean radioactivity on the investigated side in Case 32 and less than 1%/day in Case 51.

B. Results

Table XII shows the mean results in each group tabulated in three columns. In the first column Alb(E) and Alb(L) are expressed in grams/24 hours. In the second Alb(L) is corrected for effusion volume, and in the third Alb(E) and Alb(L) are expressed in mg./24 hours/kilo body weight and Alb(L) remains corrected for volume. It would appear desirable, when comparing group results, to correct Alb(L) in this way, for it will be remembered that, in the pulmonary embolism group in which the parietal pleura was not thought to be involved and therefore protein absorption not affected by the disease process, the value for Alb(L) tended to vary with the effusion volume.

The striking feature which emerges from the results grouped in this way is the consistently low figure for Alb(L) in all other than the pulmonary embolism group. There would appear to be a tendency in all the other groups, and particularly in the

mammary carcinoma group, for parietal pleural reaction to the disease to slow down absorption of protein from the pleural space. It must be remembered, however, that, although the pleural effusions associated with tuberculosis have a rather higher mean value for Alb(L) than those associated with lung carcinoma, individually they consistently showed low values for Alb(L) in contrast to the lung carcinoma group in which the individual results were very variable. The reasons for the variability in the lung carcinoma group have already been fully discussed in Chapter IV and were considered to be consistent with the very variable pleural pathology which could be found in this condition.

One of the rather surprising features is the moderately high rate of Alb(E) in the cardiac failure group but here again coincident pulmonary vascular disease was thought to contribute to the abnormally high values obtained in three patients of this group. It is in fact likely that, when pleural effusions persist in patients with cardiac failure, there is some pulmonary vascular embolism or thrombosis present.

C. Correlation of Pathology with Results

In each group the results have corresponded well with the pleural pathology commonly found in the condition under investigation. However, the ultimate control of albumin entry and loss from the pleural cavity, in biochemical terms, has not

been evaulated.

Albumin Entry

There has been a great deal of attention focused on this aspect of the problem of inflammation in recent years. Spector (1958) reviews the greater part of the literature concerned with substances which affect capillary permeability. The substances are all derived from amino-acids and nearly all cause contraction of smooth muscle preparations. The better known are histamine, 5-hydroxytryptamine, peptides such as the kinins, hyaluronidase and oestrogens. Florey (1962), Majno & Palade (1961) and Palade (1961) describe the structural changes in the capillary injured by histamine and serotinin as seen under the electron microscope. They have not been able to confirm Pappenheimer's (1953) pore theory in terms of anatomical fact as no true pores have been identified. There appear to be two ways by which macromolecules, such as ferritin, can be transferred across the endothelial boundary: (a) by micro-phagocytosis (Zweifach, 1962) the endothelium can form micro-vesicles or "pinocytes" along its inner surface and these may then move across the cytoplasm to be discharged under the basement membrane, or (b) under the influence of capillary permeability factors, such as histamine and serotinin, which may cause partial disconnection of the endothelial cells along their intercellular junctions. The basement membrane of the capillary remains intact even although complete disconnection of

endothelial cells is present and must have a role in restricting protein exit from the capillary after the endothelial barrier is breached. The precise part which it plays and whether its permeability to macromolecules is changed either by vasoactive substances or by purely mechanical dilatation is not known. Majno, Palade & Schoefl (1961) have pointed out that the most marked effect of histamine and serotinin on the dog's capillaries is on the venous side of the circulation and the heaviest deposits of subendothelial dye occur in the vessels of 20-30 μ size. The capillaries below 7 μ in size are largely spared.

The effect of corticosteroids in reducing increased capillary permeability varies to some extent according to the factor(s) responsible for the increase. It is generally effective in suppressing the increased capillary permeability caused by certain peptides, histamine and 5-hydroxytryptamine (Menkin, 1940; Bangham, 1951; Somani & Arora, 1962). It may also diminish the local production of these capillary permeability factors by preventing the effect of the antigen-antibody combination on the capillary wall.

Considerable experimental work has also been carried out in animals to try to relate concentrations of vasoactive substances in inflammatory exudates to the rate of formation of exudate and to its response to substances such as salicylate. The extreme rapidity with which the peptides can be inactivated

by enzymes makes the evaluation of their action in inflammation extremely difficult. The work of Spector & Willoughby (1962) suggests, in that salicylates depress the effect of kallikrein but not synthetic bradykinin, that in inflammation of the type sensitive to salicylates, there is activation of a specific enzyme-substrate system, and that salicylates interfere with the activator. In pleural effusions artificially induced in rats by turpentine (Spector & Willoughby, 1957b; Spector & Storey, 1958) increased concentrations of histamine and 5-hydroxytryptamine have been found. Histamine can be found only very early in the injury and 5-hydroxytryptamine, using selective inhibition techniques, was thought not to have any significant effect on capillary permeability in this situation.

Another rather interesting fact in relation to Meigs' syndrome is that oestrogens in the experimental animal can have a remarkably powerful effect in increasing capillary permeability in the mouse uterus (Spector & Storey, 1958). Oestrogens are also known to increase capillary sensitivity to the kinins. It will be remembered that there was no evidence of excessive urinary excretion of oestrogen in Case 62, although it is still possible that local tissue concentrations were excessive.

Much is known, therefore, about a range of substances which in the experimental situation can increase capillary permeability in certain regions of the body, but little is known of their relative importance in the various disease processes

(some of which are included in this thesis) associated with increased capillary leak of protein.

Albumin Loss

It has been shown that the ability of the pleural surface to absorb the albumin is reduced to a greater or lesser extent in all the diseases investigated other than pulmonary embolism and high protein entry ascites. Doubt has, however, already been expressed as to the precise site of obstruction. In most of these conditions the pleural mesothelium is itself thickened by fibrin deposition and it is not necessary to postulate lymphatic dysfunction. It is not now believed that the lymphatics open into the pleural space by stomata but that the macromolecules pass both through and between the mesothelial cells to be collected by the subpleural lymphatic plexus. Passage through the mesothelium is bound to be inhibited by the deposition of a fibrin barrier, or even by mesothelial oedema and inflammation. Normally, lymphatic flow is increased in inflammation. The lymphatics at the site of the inflammation become dilated and like blood capillaries more permeable (McMaster & Hudack, 1932). This permeability may exist in both directions unlike blood capillaries but lymph capillary structure demonstrated by electron microscopy (Casley-Smith & Florey, 1961) is very similar to the appearances seen in blood capillaries.

It might be expected, therefore, that lymphatic capillary

permeability would be affected by much the same agents as blood capillary permeability and corticosteroids might lower lymphatic permeability and slow down rather than increase the protein removal from an inflamed site. In fact, decreased albumin clearance following intrapleural prednisolone administration was seen in a patient with cardiac failure in whom there was no evidence of any inflammatory pleural condition. This would accord with the suspicion that the corticosteroid effect on albumin removal from effusions is composed of a balance between diminished parietal pleural inflammation, if that is present, and decreased lymphatic absorption.

In conclusion it may be pointed out that a pleural effusion provides an excellent experimental device for an attempt to elucidate which of the three groups of capillary permeability factors - histamine, 5-hydroxytryptamine and the kinins - are particularly associated with increased capillary permeability in different conditions. By the method described in this thesis Alb(E) can be determined and should provide an accurate estimate of capillary permeability. Provided that the above permeability factors are released into the fluid and not immediately inactivated, their pattern of concentration in the light of the estimated capillary permeability and the diagnosis might provide further insight into the tissue responses in inflammation. It is proposed to undertake such a study in the coming year.

D.

Clinical Application

If the method described in the Introductory Chapter is strictly adhered to, accurate and repeatable estimations of albumin entry into and loss from the pleural cavity can be made. In view of the relative ease with which the investigation can be carried out, it is important to consider its diagnostic scope in clinical practice.

In approximately four out of every five patients with pleural effusions the diagnosis can be made securely on clinical, bacteriological, biochemical or histological grounds. However, in about 20% of effusions associated with lung or pleural disease the diagnosis is not certain and often lies between pulmonary embolism, tuberculosis and primary or secondary carcinoma. In this small group of patients the investigation, although its limitations in providing positive diagnostic information are apparent, may be of definite help.

For example, if there is no evidence of previous pleural disease, a low rate of albumin loss will tend to exclude a diagnosis of pulmonary embolism. On the other hand, the diagnosis of a tuberculous pleural effusion would be excluded if a low rate of albumin loss in the initial part of the investigation was not increased, or at least maintained, following prednisolone administration. The latter statement would only apply to effusions of over 750 mls.

As with all investigations which provide this type of information, care is needed to avoid over-interpretation of the results.

E.
Nomenclature

On the question of nomenclature I would make a plea for simplification. All fluid collections within the pleural cavity should be termed "pleural effusions". If the effusion colloid osmotic pressure is less than 30% of that of the plasma, the effusion is secondary either to cardiac failure, to renal disease, to portal hypertension or to overhydration and should be so qualified. If it is above 40%, the effusion is secondary to pleural or lung disease and should be qualified as particularly as diagnostic evidence allows. When the value is between 30 and 40%, the effusion is due to pleural or lung disease occurring in association with cardiac failure or one of the other conditions mentioned above.

SUMMARY

1. Factors which affect fluid accumulation within the pleural cavities have been discussed, particular emphasis being laid on the derangement in the handling of protein which may occur at serous surfaces.

2. A method of estimating albumin turnover in serous effusions has been described.

3. Forty-two patients, whose diagnoses had been established, were included in the investigation. The evidence for the diagnosis in each case has been presented in detail and a clinical report on each patient has been given in Appendix A.

4. The diagnostic groups examined were: tuberculosis, pulmonary embolus, lung carcinoma, secondary carcinoma and congestive cardiac failure. In each group the rate of albumin entry into, and loss from, the pleural cavity has been estimated and the results expressed individually and collectively. These have shown that the rate of albumin loss tended to be low in all conditions other than pulmonary embolism. In pulmonary embolism, where no significant reduction in this value is thought to exist, the rate of albumin loss bore a direct relationship to effusion volume.

In some instances, the change in the albumin turnover pattern following intrapleural prednisolone administration was also measured. These results have been expressed in grams

albumin entering or leaving the pleural cavity in 24 hours. The mean results of each group have also been expressed in mg./kg./hour albumin and the "effusion fluid" loss in ml./kg./hour. The rate of albumin entry has been found to fall fairly consistently following prednisolone administration. The response in the rate of albumin loss has been more variable and this has been attributed to three factors:

(1) the volume of the effusion,

(2) the degree of reversibility of any inflammatory condition of the parietal pleura, if present,

(3) a corticosteroid effect in reducing lymphatic capillary permeability.

5. A patient with malignant granulosa cell tumour of the ovary associated with ascites and a right pleural effusion has also been investigated. The results, which are thought to apply to Meigs' syndrome, demonstrated that a very large amount of albumin was leaking into the ascites and some of this en route to the blood vascular system spilled into the right pleural cavity, presumably from either the diaphragmatic or mediastinal lymphatics.

6. The known pathology of the various conditions, as it is likely to affect the pleural space, has been reviewed and the results of the investigations correlated with it.

7. It has been suggested that there is a diagnostic application of the method when routine clinical, bacteriological,

biochemical and histological investigations have failed to secure the diagnosis.

8. The method has been shown to be particularly applicable to a study of the importance of certain capillary permeability factors in diseases associated with ascites and pleural effusion.

9. Finally, in the light of the findings, the present nomenclature of pleural effusions has been discussed and certain recommendations made regarding the definition of the terms "exudate" and "transudate".

Appendix A I Tuberculosis

Case	History	Weight (Kilos)	Temp. °F.	E.S.R.	Mantoux 1:1000	Pleural Fluid				Plasma		Pleural Biopsy	Response to Treatment
						Prot.	Alb.	Cytology	Bact.	Prot.	Alb.		
1 M.P. F. 33	L. Pleurisy 4/52. Fever 1/52	44	100	37	10 mm.	4.2	2.9	Lymphos. No serosal cells	-ve	6.0	3.0	T.B. granulation tissue	Only pleural thickening after 2/52. X-ray now normal
2a J.K. M. 56	'Flu 1/12 Tiredness and cough 3/12. Weight loss 6/12	53	99.4	41	7 mm.	5.5	2.2	R.B.C's Polys. Histiocytes	-ve	6.5	3.5	Inflammatory tissue	Control of fever Cholesterol (1,530 mg.%) effusion unchanged.
2b	Re-admitted for prednisolone assessment	53	98	25	-	6.0	2.5	Lymphos. ++ Few serosal cells	-ve	-	-	Acute inflammatory. Pleurectomy - caseating tuberculosis	
3 S.P. M. 63	R. pleuritic pain & cough 6/52. Occas. haemoptyses 6/52. Weight loss 3/12.	64	99.6	54	4 mm.	5.9	2.7	100% Lymphos. V. few serosal cells.	-ve	-	-	Follicles & giant cells tuberculosis Scalene biopsy: Sarcoid or tuberculosis	Control of fever and steady weight gain. Normal x-ray at 18/12.
4 M.A. F. 36	L.Pleurisy, fever & cough 2/52. Tiredness 2/12	45	100.2	58	12 mm.	4.5	2.7	Lymphos. Polys. No serosal cells	+ve sputum	-	-	-	Effusion gone in 5/52. Bilat. upper zone tuberculosis cleared satisfactorily.

Appendix A I Tuberculosis (Cont'd)

Case	History	Weight (Kilos)	Temp.	E.S.R.	Mantoux 1:1000	Pleural Fluid				Plasma		Pleural Biopsy	Response to Treatment
						Prot.	Alb.	Cytology	Bact.	Prot.	Alb.		
5 J.B. F. 24	L.pleurisy & breathlessness 4/52 Cough and slight haemoptysis 2/12 Febrile and ill 1/52	54	101	51	12 mm.	5.7	3.0	80% lymphos. No serosal cells	-ve	7.3	3.35	Unsuccessful	Quick response of fever and malaise to anti-tuberculous drugs. Discharged in 10 days. Normal x-ray at 1 month
6 A.N. F. 47	Tiredness & increasing breathlessness 3/52 Febrile 2/52	57	100.6	27	10 mm.	5.7	3.0	80% lymphos.	-ve	7.3	3.72	Two folliculoid structures very probably tuberculous	The systemic illness responded rapidly to anti-tuberculous chemotherapy. The effusion and a small right upper zone tuberculous opacity both showed complete resolution in 4/12.
7 J.McP. F. 36	Weight loss 1/12. L.pleuritic pain 10 days Fever found during confinement	62	98.4	130	-	5.4	2.7	-	+ve	7.6	3.8	-	Gradual improvement in well-being. Complete clearing of pleural opacity and substantial clearing lung opacities 4/12

Appendix A II Pulmonary Embolism

Case	History	Weight (Kilos)	E.S.R.	W.B.C. /cu.mm.	Pleural Fluid			Plasma		Pleural Biopsy	Response to Treatment
					Prot.	Alb.	Cytology	Prot.	Alb.		
21 W.H. M. 59	L.chest pain and mild breathlessness $2\frac{1}{2}$/52.	67	32	7,600	4.4	2.6	Serosal Cells +	-	-	-	Satisfactory resolution without anticoagulants. Follow-up 18/12. Typical linear opacity
22 M.L. F. 29	Minimal L. T.B. in 1960 treated with 18/12 good therapy. L.pleuritic pain 1/52 and cough and breathlessness. Fever	64	58	10,500	4.9	2.7	Serosal cells. Lymphos. +	6.9	3.6	Granulation tissue with some lymphos.	Settled with anticoagulants. Small linear opacities on x-ray. Recurrance after pregnancy 9/12 later.
23 J.C. M. 67	Malaise. R.pleuritic pain - 2 days. Haemoptysis Fever	54	48	8,600	4.3	2.2	Eosins. Few lymphos.	6.8	3.8	-	Settled with anticoagulants. Cardiac failure now requires treatment with digoxin and diuretics. Linear opacities on x-ray.
24 W.F. M. 40	R.pleurisy - 8 days. Haemoptyses - 7 days.	59	30	7,700	4.8	2.5	Few R.B.C's Lymphos.	6.5	3.8	-	Satisfactory resolution with anticoagulant therapy. Follow-up 15/12.
25 J.M. M.	L.chest ache - 1/52 Breathlessness 2/52. Slight ankle swelling 2/12	64	40	22,000	4.7	1.8	R.B.C's ++ Blood stained fluid	6.3	3.6	-	Atrial fibrillation treated with digoxin. Effusion gradually disappeared. Residual linear opacities. Follow-up 18/12.

Appendix A II Pulmonary Embolism (Cont'd)

Case	History	Weight (Kilos)	E.S.R.	W.B.C. /cu.mm.	Pleural Fluid			Plasma		Pleural biopsy	Response to Treatment
					Prot.	Alb.	Cytology	Prot.	Alb.		
26 A.W. M. 55	Increasing breathlessness 1/12. L.pleuritic pain 2/52	75	86	3,900	5.3	2.7	R.B.C's ++ Eosins. +	7.6	3.8	Eosins + Mild inflammatory reaction	Rapid resolution without specific treatment other than aspiration. 2 yr. follow-up. Normal chest x-ray.
27 J.M. M. 37	Malaise 10 days Initial improvement in cough with antibiotics. Sudden L. pleuritic pain on day before admission. Fever.	49	65	15,000	5.2	2.6	R.B.C's ++ 90% Polys.	-	-	Normal pleura	Only antibiotic treatment. Pleural effusion slowly resolved. Fever slowly settled Chest radiograph developed "linear opacities" typical of pulm. infarct. 20/12 Follow-up
28 G.V. M. 81	'Flu-like illness with R.pleurisy 2/52. Breathlessness and cough 1/52	52	70	6,950	3.3	1.6	R.B.C's++ No malignant cells (3 specs.)	6.4	3.4	Fibrocellular tissue. Thin fibrin coat.	Cardiac failure and calf vein thromboses settled with treatment - digoxin, diuretics anticoagulants - small linear opacity. Died 20/12 later - no evidence lung tumour; leg vein thrombosis and bronchopneumonia.

N.B. Full bacteriological examination of pleural fluid was negative.

Appendix A III Lung Carcinoma

Case	History	Weight (Kilos)	Pleural Fluid			Plasma		Pleural Biopsy	Bronchoscopy	Outcome
			Prot.	Alb.	Cyt.	Prot.	Alb.			
12 A.B. M. 78	Hoarseness 2/12 Cough and breathlessness 5/52	64.5	3.6	2.1	+ve	5.4	3.2	Infiltration Anaplastic cells	-	DIED after 7/52. Autopsy - anaplastic bronchogenic carcinoma. Both pleural surfaces widely involved by tumour. Liver metastases.
13 F.L. M. 87	Increasing breathlessness 4/52 Cough +	60	3.7	2.1	+ve	-	-	Mild chronic inflammatory reaction	-ve	DIED after 5/52. Autopsy - squamous bronchogenic carcinoma. No macroscopic tumour in pleural space. Adrenal metastases.
14 R.S. M. 64	Breathlessness & R. pleuritic pain 3/52	61	5.8	2.1	+ve	7.3	3.6	Chronic inflammatory tissue	-ve	DIED after 3/52. Autopsy - anaplastic large cell carcinoma with tumour studded pleural surfaces. Wide metastases. Death due to massive pulmonary embolus.
15 A.K. M. 57	Cough 1 year L. chest pain 6/12. Haemoptysis 1/52	76	4.7	2.3	-ve	7.9	3.6	Undiff. carcinoma	-ve	DIED after 10/52. Palliative radiotherapy to chest wall relieved pain. No autopsy.
16 J.W. M. 61	None related to respiratory system. Anorexia. Weight loss.	63.5	4.5	2.6	+ve	5.2	3.2	Blind -ve Thoraco-scopy +ve	-ve	DIED after 6/52. Effusion partly arrested by Mustine but chest wall involvement and steady deterioration. Autopsy - anaplastic "oat-cell" carcinoma - widespread metastases.
17 A.H. M. 65	Breathlessness & slight cough 4/12 Haemoptyses 4/12	72	4.2	1.9	-ve	6.2	3.4	-ve Scalene gland biopsy +ve	-	DIED after 6/12. Death due to myocardial infarction. Effusion showed rapid response to antibiotics and prednisolone. No autopsy.

Appendix A III Lung Carcinoma (Cont'd)

Case	History	Weight (Kilos)	Pleural Fluid			Plasma		Pleural Biopsy	Bronchoscopy	Outcome
			Prot.	Alb.	Cyt.	Prot.	Alb.			
18 J.McD. M. 54	Weight loss - tiredness 6/12 L.post. chest pain 5 days.	71	4.9	2.5	-ve	6.7	3.9	? Malignant invasion pleura	-ve	DIED after 5/12. Local Mustine led to L. fibrothorax. Steady deterioration. L. sac partially obliterated with small nodules and plaques on parietal surface.
19 J.M. M. 63	Cough and R. lower chest wall pain 2/12. One haemoptysis.	68	2.1	1.2	+ve	5.5	1.8	Anaplastic carcinoma	Anaplastic carcinoma intermediate bronchus	DIED after 4/52. Very rapid deterioration with pleural fistula. No autopsy.
20 A.O. M. 67	Increased breathlessness and cough 1/12. L. chest discomfort.	65	3.8	2.5	+ve	6.0	3.7	Anaplastic cell infiltration.	L. lower bronchus narrowed. Normal mucosa.	DIED after 6/52 following acute urinary retention. Pleural effusion satisfactorily controlled with Mustine.

N.B. Full bacteriological examination of pleural fluid was negative.

Appendix A IV Secondary Carcinoma

Case	History	Weight (Kilos)	Pleural Fluid			Plasma		Pleural Biopsy	Outcome
			Prot.	Alb.	Cytology	Prot.	Alb.		
51 J.B. M. 79	General deterioration - months. Cough and breathlessness 2/52.	53	3.6	2.0	Doubtful malignant cells. Lymphos.++	5.1	2.4	Fourth blind biopsy. Anaplastic cell infiltration	DIED after 2/12. No improvement in bilateral effusions with antituberculous chemotherapy. Autopsy - extensive lymphosarcoma involving retroperitoneal tissues in upper abdomen and spreading through diaphragm to involve parietal subpleural tissue. Also metastases to subpleural zone both lungs.
52 M.S. F. 53	R.mastectomy and radiotherapy 1958. (Admitted Nov. 1960). Breathlessness and cough - 5/52. R.trigeminal pain and weakness R. leg 5/52	70	5.4	3.0	Malignant cells 3 ex 5 specs.	7.0	3.8	Fibrous tissue only	DIED after 10/12. Cerebral, bone and hepatic metastases showed no improvement with oestrogens but 6/12 remission obtained with prednisolone. Intrapleural mustine later required and effective in suppressing effusion. No autopsy.
53 C.G. F. 63	Breathlessness 3/12 Weight loss 3/12 No palpable breast tumour	58	3.4	2.1	Cells from adenocarcinoma	5.6	3.2	At thoracoscopy - Adenocarcinoma prob.breast. Scalene biopsy - Adenocarcinoma prob.breast	DIED after 7/12. Initial control with prednisolone and diuretics. Later required intrapleural mustine - good result and palliative radiotherapy to rib. No autopsy.
54 M.P. F.	R.mastectomy and radiotherapy 1954 (Admitted July 1961). Breathlessness and cough 7/12	60	4.3	2.4	No malignant cells (1 spec.)	7.2	3.8	Fibrous tissue - no tumour	No improvement with stilboestrol. Improvement with prednisolone for 6/12, then rib involvement and multiple bone secondaries. Dramatic response to adrenalectomy (Mar.1962). Alive and well Oct. 1963.

Appendix A IV Secondary Carcinoma (Contd)

Case	History	Weight (Kilos)	Pleural Fluid			Plasma		Pleural Biopsy	Outcome
			Prot.	Alb.	Cytology	Prot.	Alb.		
55 J.R. F. 66	R.mastectomy and radiotherapy 1958 (Admitted Nov. 1962) Tiredness 4 years. R.chest pain and dyspnoea 6/52	40	5.0	2.5	Negative (3 specs.)	5.9	3.1	-	DIED after 3/12. Poorly controlled with oestrogens and then prednisolone. Rapid deterioration after 6/52. X-ray showed extensive rib involvement. No autopsy.
56 J.G. F. 60	L.mastectomy and radiotherapy 1959 (Admitted Jan. 1962) Breathlessness and slight cough 3/52	52	4.8	2.7	Neoplastic clumps of cells	-	-	Metastatic carcinoma probably arising from breast	Slight improvement with oestrogens. Then complete remission following intra-pleural mustine. Followed up to Oct. 1963.

N.B. Full bacteriological examination of pleural fluid was negative.

Appendix A V

Systemic Lupus Erythematosus

Case	History	Weight (Kilos)	Temp. °F.	E.S.R.	L.E. Cells	A.N.F.	Pleural Fluid			Plasma		Pleural Biopsy	Response to Treatment
							Prot.	Alb.	Cytology	Prot.	Alb.		
31 A.C. M. 43	R.Pleurisy 4/12 L.Pleurisy 2/52 L.knee pain 6/12 R > L Bilat.effusions	73	99 to 100	65 *(440/cu. mm.)	- (2 specs)	+ve	5.1	2.6	Eosins. lymphos. No neoplastic cells	7.7	3.8	Fibrocellular tissue only	Initially anticoags, but after 2/52 no relief in pain or fall in fever. Dramatic response to prednisolone now for 2 years.
32 J.D. F. 35	Polyarthritis 2 years. Bilat. pleurisy 6/52. Bilat.effusions	50	98 to 101	120 *(800/cu. mm.)	+ve	+ve	4.8	1.9	V.few cells	7.9	2.7	Fibrocellular tissue	Very rapid response to prednisolone but relapse after dose reduced to 5 mgs.
33 W.A. M. 43	Polyarthritis 5 years. R.pleurisy - 10 days Breathlessness- 10 days Bilat.effusions R > L	73	99 to 100	93 *(650/cu. mm.)	+ve	+ve	5.3	2.6	Few eosins. and lymphos.	7.3	2.8	Non-specific pleural inflammation	Settled with salicylates after 3/52. 2/52 later re-admitted with fever, pericarditis, pulmonary arteritis. Controlled with prednisolone
34 A.H. F. 53	Bilat.pleurisy and increasing breathlessness - 6/12 R > L	59	98 to 100	50 *(960/cu. mm.)	-ve (1 spec)	+ve	3.9	2.0	V. few cells	6.3	3.6	Mild and non-specific inflammatory reaction	After 4/12 anti-tub. therapy - very little response in effusions. Hypersens. to P.A.S.

N.B. Full bacteriological examination of pleural fluid was negative

* Eosinophils per cu.mm. blood

Appendix A VI Congestive Cardiac Failure

Case	History	Weight (Kilos)	J.V.P. (cms.)	Pleural Fluid			Plasma		Pleural Biopsy	E.C.G.	Progress
				Prot.	Alb.	Cytology	Prot.	Alb.			
41 E.L. F. 63	4 attacks rheumatic fever aged 4-28 yrs. Increasing breathlessness 10 yrs. Treated cardiac failure due to aortic stenosis and mitral stenosis and incompetence. Moderate oedema.	63.5	5	1.0	0.7	Few serosal cells	5.5	2.5	Fibrocellular tissue	Moderate left ventricular hypertrophy	Sodium retention controlled but eventually died with output failure. Autopsy - myocardial fibrosis. Hypertrophied L. ventricle. Rheumatic M.S.,M.I.,A.S. & A.I. (minimal). Hepatic cirrhosis. Bronchopneumonia.
42 U.H. M. 63	Chronic asthmatic bronchitis - 40 yrs. Bronchiectasis L. lower. Increased breathlessness, ankle swelling and L. pleural effusion 4/12. O.E. Atrial fibrillation due to mitral stenosis. Moderate oedema.	50	8+	1.0	0.6	Eosins. Polys. Lymphos.	4.1	2.4	Normal serosa.	Atrial fibrillation. Severe R. ventric. strain P.mitrale	Rapid re-accumulation bilateral pleural effusions L > R despite energetic drug treatment including anticoagulants. Plasma infusions failed to reverse process and died after 6/52. Autopsy - M.S. with R. atrial failure and generalised congestion. L. pulmonary arterial thrombosis.
43 E.S. F.	Rheumatic fever aged 4 and 14 yrs. Breathless on exertion 20 yrs. Ankle swelling 5 yrs. R. pleuritic pain 4/12 and 2/52. Heart failure due to severe rheumatic aortic and mitral disease. Hepatomegaly. Slight oedema.	41	3	1.9	1.0	Very small cellular content	6.1	3.0	-	Atrial fibrillation. R. axis deviation	Staph.aureus isolated from sputum on admission and probable R. basal pulmonary opacity in addition to effusion. Some improvement with hospital treatment but died 3/12 after discharge. No autopsy.

Appendix A VI Congestive Cardiac Failure (Cont'd)

Case	History	Weight (Kilos)	J.V.P. (cms.)	Pleural Fluid			Plasma		Pleural Biopsy	E.C.G.	Progress
				Prot.	Alb.	Cytology	Prot.	Alb.			
44 G.S. M. 58	Increasing breathlessness 7/52. Nocturnal dyspnoea 2/52 Continuous dyspnoea 3 days. B.P. 140/110 Slight oedema	76	6 ↓ 3	1.8 ↓ 3.3	1.6 ↓ 2.5	Few polys. and eosins.	7.5	4.1	Normal pleura	Post.myocard. infarct. ? duration	Rapid improvement with digoxin and diuretics. At 18/12 follow-up, well and working. Chest radiograph - slight cardiomegaly.
45 H.M. M. 71	Myocardial infarction 5/12 previously followed by cardiac failure and persistent R. effusion. No oedema	58	5	2.3	1.6	Few lymphos.	6.4	3.6	Intact serosal lining-normal	Postero-lateral myocard. infarct.	After investigation of albumin turnover, intercostal tube inserted. Although J.V.P. remained raised, pleural space became obliterated. Died suddenly at home 14/12 later. No autopsy.
46 A.L. M. 76	Ant.chest pain 3/52. Breathlessness and tiredness 3/52 Bilateral pleural effusions. Oedema +	51.5	5	1.5	0.8	Lymphos.+ Polys. + Serosal cells	-	-		Antero-lateral myocard. infarct.	Rapid improvement with drug treatment and discharged after 4/52 clear of effusions and oedema.
	On re-admission 15/12 later ventricular tachycardia and recurrence pleural effusions	50	5	1.7	1.0	-	6.1	2.9		Ventric. tachycard.	No significant response to treatment. Slowly increasing forward output failure. Died after 5/12. No autopsy.

N.B. Full bacteriological examination of pleural fluid was negative.

J.V.P. = Jugular venous pressure

APPENDIX B

Detailed Results in Case 62

Granulosa Cell Tumour Ovary

Day	Hours	Peritoneal Cavity		Pleural Cavity		Plasma	
		Injected	Sample Radioactivity (counts/sec.)	Injected	Sample Radioactivity (counts/sec.)	Injected	Sample Radioactivity (counts/sec.)
1	0.0	2.013 g. R.I.H.S.A.					
	3.0		213				
	17.0		151		37		31
2	3.0		131		54		43
	17.5		106		72		59
3	2.5	1.914 g. R.I.H.S.A.					
	2.55		93				
	5.5		305				
4	5.0		212		130		120
	20.75		174		132		126
6	4.5		139	2.385 g. R.I.H.S.A.	129		122
	4.55				565		
	7.0				496		
	17.5						
7	4.3				452		
	17.5				386		
8	1.0		132	2.480 g. R.I.H.S.A.	323		194
	1.05						
	3.2				791		
9	0.0						250
	0.05					4.56 g. R.I.H.S.A.	
	0.33						860
	1.43						790
	4.5						757

REFERENCES

ABRAMS, H.L., SPIRO, R. & GOLDSTEIN, N. (1950) Cancer 3, 74.

AGOSTONI, E., TAGLIETTI, A. & SETNIKAR, I. (1957) Amer. J. Phys. 191(2), 277.

ALARCON-SEGOVIA, P. & ALARCON, D.G. (1961) Dis. Chest 39, 7.

ALLEN, L. & VOGT, E. (1937) Amer. J. Physiol. 119, 776.

ASPIN, J. & O'HARA, H. (1958) Brit. J. Tuberc. 52, 81.

BAGGENSTOSS, A.H. (1952) Proc. Mayo Clin. 27, 412.

BANGHAM, A.D. (1951) Brit. J. exp. Path. 32, 77.

BALLANTYNE, A.J., CLAGETT, O.T. & McDONALD, J.R. (1957) Thorax 12, 294.

BERSON, S.A. & YALOW, Rosalyn S. (1954) J. Clin. Invest. 33, 377.

BEST, P.V. & HEATH, D. (1958) J. Path. Bact. 75, 281.

BHOOLA, K.D., CALLE, J.D. & SCHACTER, M. (1960) J. Physiol. 152, 75.

BLOMQVIST, A.S.K. (1951) Nord. Med. 46, 1336.

BURGEN, A.S.V. & STEWART, P.B. (1958) J. Lab. Clin. Med. 52(2), 118.

BURRELL, R.G. (1963) Amer. Rev. Resp. Dis. 87, 389.

CALMENSON, M., DOCHERTY, M.B. & BIANCO, J.J. (1947) Surg. Gynec. Obstet.84, 181.

CASLEY-SMITH, J.R. & FLOREY, H.W. (1961) Quart. J. exp. Physiol. 46, 101.

CLAUSS, H., YACOUBIAN, J. & BARKER, H.G. (1957) Surg. Forum 7, 201.

CLOSE, H.G. (1946) Lancet 1, 193.

COE, J.E. & AIKAWA, J.K. (1961) Arch. Intern. Med. (Chic.) 108, 763.

COMAN, D.R. (1944) Cancer Res. 4, 625.

COORAY, G.H. (1949) J. Path. Bact. 61, 551.

COURTICE, F.C. (1951) Report Australian & New Zealand Ass. Adv. Sci. 28, 115. Government Printer, Brisbane.

COURTICE, F.C. & MORRIS, B. (1953) Austr. J. exp. Biol. Med. Sci. 31, 227.

COURTICE, F.C. & SIMMONDS, W.J. (1949) J. Phys. 109, 117.

COURTICE, F.C. & SIMMONDS, W.J. (1954) Physiol. Rev. 34, 419.

COURTICE, F.C. & STEINBECK, A.W. (1950) Austr. J. exp. Biol. Med. Sci. 28, 161.

COURTICE, F.C. & STEINBECK, A.W. (1951) Austr. J. exp. Biol. Med. Sci. 29, 451.

DOUGHERTY, T.F. & SCHNEEBELI, G.L. (1950) Proc. Soc. exp. Biol. N.Y. 75, 854.

DRINKER, C.K. (1945) Pulmonary Edema and Inflammation. Cambridge, Mass. Harvard Univ. Press.

DRINKER, C.K. & FIELD, M.E. (1931) Amer. J. Phys. 97, 32.

DUBOIS, E.L. (1953) Am. Intern. Med. 38, 1265.

DUBOIS, E.L. (1956) Am. Intern. Med. 45, 163.

DUBOIS, E.L. & ARTERBERRY, J.D. (1962) Proceedings of the Annual Meeting of the American Rheumatism Association.

ELLIS, F.H., GRINDLAY, J.H. & EDWARDS, J.E. (1951) Surgery 30, 810.

ELLMAN, P. (1956) Post-grad. Med. J. 32, 370.

ELLMAN, P. & CUDKOWICZ, L. (1954) Thorax 9, 46.

FIELD, M.E. & DRINKER, C.K. (1931) Amer. J. Physiol. 97, 40.

FLEISHMAN, S.J., COETZEE, A.M., MINDEL, S., BERJAK, J., LICHTER, A.I. & KERRICH, J.E. (1960) Lancet 1, 199.

FLOREY, H. (1962) General Pathology 3rd Edition. Lloyd-Luke (Medical Books) Ltd., London.

FREW, H.W.O. & CAMPBELL-FOWLER, C. (1956) Practitioner 176, 416.

GABRIELI, E.R., GOULIAN, D.Jr., KINERSLY, T. & COLLET, R. (1954) J. Clin. Invest. 33(1), 136.

GHOSH, M.N., BANERJIE, R.M. & MUKHERJI, S.K. (1963) Ind. J. Phys. Pharm. 7, 17.

GOLDMAN, A. & BURFORD, T.H. (1950) Dis. Chest 18, 586.

GREENWOOD, F.C., HUNTER, W.M. & GLOVER, J.S. (1963) Biochem. J. 89, 114.

HALL, T.C., DEDERICK, Margarida M., NEVINNY, H.B. & KELLEY, Rita M. (1960) Biological Activities of Steroids in relation to Cancer. Ed. Pincus, G. & Vollmer, E.P., Academic Press, New York & London.

HAMPTON, A.O. & CASTLEMAN, B. (1940) Amer. J. Roentgenol. 43, 305.

HARVEY, A.M., SCHULMAN, L.E., TUMULTY, P.A., CONLEY, C.L. & SCHOENRICH, E.H. (1955) Systemic Lupus Erythematosus: Review of the Literature and Clinical Analysis of 138 Cases. Williams & Wilkins, Baltimore.

HAYEK, H.V. (1953) Die menschliche Lunge. Springer, Berlin, Goettingen, Heidelberg. Transl. Krahl, V.E., Hafner Pub. Coy. Inc., New York (1960).

ISRAEL, H.L. (1953) Am. J. Med. Sci. 226, 387.

KAMPMEIER, O.F. (1928) Amer. J. Anat. 40, 413.

LAHIRI, S.C. (1962) Ph.D. Thesis, Edinburgh University. The Activation of Kinins by antigen-antibody reactions.

LANDIS, E.M. (1927) Amer. J. Physiol. 82, 217.

LAWSON, J.G. (1950) J. Obstet. Gynaec. Brit. Emp. 57, 595.

LEMMING, R. (1960) Acta Med. Scand. 168, 197.

LEMON, H.M. (1960) Biological Activities of Steroids in relation to Cancer. Ed. Pincus, G. & Vollmer, E.P., Academic Press, New York & London.

LEMON, W.S. & FELDMAN, W.H. (1943) Am. Rev. Tuberc. 48, 177.

LEMON, W.S. & HIGGINS, G.M. (1932) Amer. J. Med. Sci. 184, 846.

LEUALLEN, E.C. & CARR, D.T. (1955) New Eng. J. Med. 252, 79.

MACKAY, I.R. & BURNET, F. MacF. (1963) Autoimmune Disease, pp. 253-256. C. C. Thomas, Bannerstone House, Illinois.

MacLEOD, J.G. & GRANT, I.W.B. (1954) Thorax 9, 71.

McMASTER, P.D. (1937) J. exp. Med. 65, 373.

McMASTER, P.D. (1947) J. exp. Med. 86, 293.

McMASTER, P.D. & HUDACK, S. (1932) J. exp. Med. 56, 239.

MAGIDSON, O. & JACOBSON, G. (1955) Brit. Heart J. 17, 207.

MAJNO, G. & PALADE, G.E. (1961) J. Biophys. Biochem. Cytol. 11, 571.

MAJNO, G., PALADE, G.E. & SCHOEFL, G. (1961) J. Biophys. Biochem. Cytol. 11, 607.

MARSHALL, A.G. (1949) J. Obstet. Gynaec. Brit. Emp. 56, 86.

MEIGS, J.V. (1954) Amer. J. Obstet. Gynec. 67, 962.

MEIGS, J.V., ARMSTRONG, S.H. & HAMILTON, H.H. (1943) Am. J. Obstet. Gynec. 46, 19.

MENKIN, V. (1940) Amer. J. Physiol. 129, 691.

MESTITZ, P., PURVES, M.J. & POLLARD, A.C. (1958) Lancet 2, 1349.

MILLER, W.S. (1937) The Lung. Balliere, Tindall & Cox, London.

MOORE, G.E., SAKO, K., KONDO, T., BADILLO, J. & BURKE, E. (1961) Surg. Obstet. Gynec. 112, 469.

MYERS, J.A. (1955) Arch. Intern. Med. 96, 191.

MYHRE, J.R. (1959) Acta Med. Scand. 165, 55.

NAIRN, R.C. (1957) Brit. J. exp. Path. 38(1), 62.

PALADE, G.E. (1961) Circulation 24, 368.

PAPPENHEIMER, J.R. (1953) Physiol. Rev. 33, 387.

PATERSON, R.C. (1917) Am. Rev. Tuberc. 1, 353.

PICKERING, G.W. (1952) Brit. Med. J. 1, 1207.

PRENTICE, T.C., SIRI, W. & JOINER, E.E. (1952) Amer. J. Med. 13, 668.

PRYCE, D.M. & WALTER, J.B. (1960) J. Path. Bact. 79, 141.

PULLINGER, B.D. & FLOREY, H.W. (1935) Brit. J. exp. Path. 16, 49.

PUMP, K.K. (1963) Dis. Chest 43, 245.

RABIN, E.R. & MEYER, E.C. (1960) Circ. Res. 8, 324.

ROBERTSON, R.F. (1954) Edin. Med. J. 61, 37.

ROSE, A.H. (1957) J. Thorac. Surg. 33, 254.

ROSS, R.S., WALKER, W.G. & HAMMOND, J.D.S. (1960) Circulation 8, 1041.

RUBIN, I.C., NOVAK, J. & SQUIRE, J.J. (1944) Amer. J. Obstet. Gynec. 48, 601.

RUPE, C.E. & NICKEL, S.N. (1959) J. Amer. Med. Ass. 171, 1055.

SCHOENBERGER, J.A., KROLL, G., ECKERT, E.L. & KARK, R.M. (1956) J. Lab. Clin. Med. 47, 227.

SCHUMAN, C. (1941) Quart. Bull. Sea View Hosp. 7, 69.

SIMER, P.H. (1952) Anat. Rec. 113, 269.

SIMPSON, S.L. (1929) Quart. J. Med. 22, 413.

SMITH, E.W. & KURBAN, A. (1962) Bull. Johns Hopkins Hosp. 110, 202.

SOMANI, P. & ARORA, R.B. (1962) J. Pharm. Pharmacol. 14, 535.

SPECTOR, W.G. (1958) Pharm. Rev. 10, 475.

SPECTOR, W.G. & STOREY, E. (1958) J. Path. Bact. 75, 383.

SPECTOR, W.G. & WILLOUGHBY, D.A. (1957) J. Path. Bact. 73, 133.a

SPECTOR, W.G. & WILLOUGHBY, D.A. (1957) J. Path. Bact. 74, 57.b

SPECTOR, W.G. & WILLOUGHBY, D.A. (1962) Nature (London) 196, 1104.

SPRIGGS, A.L. (1957) The Cytology of Effusions. Heineman, London.

SPRIGGS, A.L. & BODDINGTON, M.M. (1960) Thorax 15, 169.

STARLING, E.H. (1896) J. Phys. 19, 312.

STEAD, E.A. & WARREN, J.V. (1944) J. Clin. Invest. 23, 283.

STEPHANOPOULOS, C. & DOUCAS, N. (1962) Brit. J. Dis. Chest 56, 96.

STERLING, K. (1951) J. Clin. Invest. 30, 1228.

STEWART, P.B. (1963) J. Clin. Invest. 42, 258.

STEWART, P.B. & BURGEN, A.S.V. (1958) J. Lab. Clin. Med. 52(2), 212.

TEILUM, G. (1946) Acta Med. Scand. 123, 126.

THORELL, I. (1952) Acta Radiol. 37, 8.

UHLEY, H.N., LEEDS, S.E., SAMPSON, J.J. & FRIEDMAN, M. (1962) Circ. Res. 11, 966.

VACCARI, F., BALDINI, E. & FONTANA, G. (1962) Acta Haemat. (Basel) 28, 352.

Van HAZEL, W. & JENSIK, R.J. (1956) J. Thorac. Surg. 31, 19.

WALKER, W.G., ROSS, R.S. & HAMMOND, J.D.S. (1960) Circ. Res. 8, 1028.

WARREN, M.F., PETERSEN, D.K. & DRINKER, C.K. (1942) Amer. J. Physiol. 137, 641.

WARREN, J.V. & STEAD, E.A. (1944) Arch. Intern. Med. 73, 138.

WELLS, H.S., YOUMANS, J.B. & MILLER, D.G. (1933) J. Clin. Invest. 12, 1103.

WHITE, J.C., FIELD, M.E. & DRINKER, C.K. (1933) Amer. J. Phys. 103, 33.

WILLIS, R.A. (1952) The Spread of Tumours in the Human Body. 1st Edition 1933, 2nd Edition 1952, Butterworth, London.

WIRTA, A.V. (1936) Duodecim. 52, 289.

WOOD, S. (1958) Amer. Med. Ass. Arch. Path. 66, 550.

WOOD, S.Jr., HOLYOKE, E.D. & YARDLEY, J.H. (1961) Canadian Cancer Conference 4, 167.

YAMADA, S. (1933) Ztschr. ges. exp. Med. 90, 342.

ZWEIFACH, B.W. (1962) Angiology 13, 345.