Effect of edema on pulmonary blood flow in the isolated perfused dog lung lobe

JAHAR BHATTACHARYA, KAZUYA NAKAHARA, AND NORMAN C. STAUB
Cardiovascular Research Institute and Department of Physiology, University of California, San Francisco, California 94143

BHATTACHARYA, JAHAR, KAZUYA NAKAHARA, AND NORMAN C. STAUB. *Effect of edema on pulmonary blood flow in the isolated perfused dog lung lobe.* J. Appl. Physiol.: Respirat. Environ. Exercise Physiol. 48(3): 444–449, 1980.—We determined the relationship between the amount of edema and changes in blood flow in the isolated, perfused, and ventilated lower lobe of dog lung. We held vascular pressure constant and measured lobe weight and flow continuously. Vascular pressures were set to produce minimal weight gain in four lobes (controls) and large weight gain in six lobes (edema). In all lobes, the outflow pressure exceeded alveolar pressure at end expiration (zone III conditions). The control lobes gained an average of 20% in weight over 4 h, but blood flow remained constant. They showed interstitial edema histologically and extravascular lung water was increased 38%. The edema lobes gained weight rapidly, ultimately tripling their weight. In these lobes, blood flow remained constant until lobe weight had doubled; then flow decreased progressively to low levels. These lobes showed extensive alveolar edema histologically and extravascular lung water was increased 238%. Pulmonary blood flow is not affected by interstitial edema, but is markedly reduced when alveolar flooding occurs.

vascular resistance; edema fluid proteins; lung water content; ultrasonic flow measurement

THERE IS CONSIDERABLE CONTROVERSY as to whether pulmonary edema affects lung blood flow and, even if it does, it is unclear how extensive edema has to be before flow is changed. West, Dollery, and Heard (13) produced high-pressure pulmonary edema in isolated perfused dog lungs and showed redistribution of flow from the lower to the upper lung regions. They noted that the redistribution occurred only at low driving pressures and attributed it to increased vascular resistance in the lower lung caused by interstitial edema (perivascular cuffs), which increased interstitial fluid pressure and decreased vascular transmural distending pressure (3, 13). However, they did not prove this hypothesis. In fact, their published histological pictures showed no evidence of vascular collapse.

At the opposite extreme, Hyman and associates (5) reported that in the perfused intact dog lung lobe even severe pulmonary edema had no effect on lobar vascular resistance.

Naimark and co-workers (8) and Muir and associates (7) reexamined the relationship between flow and edema. They concurred that edema affects vascular resistance, but only after alveolar flooding has developed. Interstitial edema alone did not cause blood flow redistribution. Very recently, Hogg (2) has presented evidence suggesting that both alveolar and interstitial edema are necessary for changes in pulmonary blood flow.

We have reexamined the relationship between flow and edema in the isolated dog lung lobe perfused with blood at constant pressure. We found that flow remained constant until lung weight had doubled and alveolar edema was evident; subsequently flow decreased progressively to very low levels.

METHODS

Surgical Procedures

We anesthetized 10 dogs (20–22 kg) with thiopental sodium (25 mg/kg, iv) and maintained anesthesia with pentobarbital sodium (60 mg iv, as required). The dogs were ventilated through an endotracheal tube with a positive-pressure pump at 12 breaths/min at a tidal volume of 450 ml. We inserted a plastic catheter into a femoral artery and withdrew approximately 300 ml blood into a plastic beaker containing 1,000 U heparin. We adjusted the hematocrit of this shed blood to 20–30% with 0.9% saline and filled the lobe perfusion circuit with it.

We opened the thorax widely by a sternum-splitting incision and injected 4,000 U heparin and 60 mg papaverine, intravenously. We made a second blood collection (approx 400 ml), which we stored in a water bath for use if required. A final blood sample (25 ml) was taken immediately prior to the removal of the lungs. This blood sample was used later in the determination of residual blood content of the right lower lobe.

We carefully removed the heart and lungs together and placed them in a stainless steel basin containing saline at room temperature. Then we cannulated the artery, vein, and bronchus to the left lower lobe and weighed the lobe.

Subsequently, one of us removed the right lower lobe, inflated it to 5 cmH_2O, and weighed and rapidly froze it in liquid nitrogen for analysis of extravascular lung water.

Experimental Apparatus

Perfusion circuit. Our perfusion circuit is modified from that of Snashall and associates (11). It provides for continuous flow measurement by an ultrasonic flowmeter (model 806A, Parks Electronics, Medford, OR) and for constant perfusion pressures (Fig. 1). Briefly, blood is

supplied to the lobe from an arterial reservoir with overflow; blood returning from the lobe drains into a venous reservoir from which it is pumped back to the arterial reservoir. By adjusting the heights of the two reservoirs, we can set target pressures in the vein and artery. Vascular pressures were measured by catheters placed at the hilum of the lung and were recorded alternately on the same strain gauge and relative to the level of the lung hilum.

We placed thermistors at various points in the perfusion circuit to monitor blood temperature, which was maintained at 38°C as it entered the lung. All tubing was of silicone rubber (hospital-grade Silastic, 5/16 in. ID, Dow Corning, Midland, MI).

Lobe weighing apparatus. The perfused lobe was suspended from one arm of a balance beam in a warm humidified Lucite chamber. The other arm of the beam was connected to a force-displacement strain gauge (model FT 10, Grass Instrument, Quincy, MA). We estimated the initial increase in lobe blood volume at the onset of perfusion by the difference in lobe weight before and 10 min after the start of perfusion. We called the weight after 10 min perfusion, the initial lobe weight. All subsequent changes in lobe weight referred to this initial weight.

The time between removal of the heart and lungs and the restarting of lobar blood flow was between 20 and 30 min. The lobe was continuously ventilated at an inspiratory pressure of 12 cmH_2O and an end expiratory pressure of 5 cmH_2O at a frequency of 5 breaths/min using 100% oxygen. These pressures maintained the lobe well inflated during the entire experiment.

The outputs of the force displacement strain gauge, the pressure transducers, and the ultrasonic flow meter were recorded on a direct writing polygraph (model 5 Grass Instrument). Each channel was calibrated before and after the experiment.

Experimental Protocol

In all our experiments, we held pulmonary venous pressure above end-expiratory airway pressures; i.e., the lung was in zone III. In four control lobes, we set the arterial (Ppa) and venous (Ppv) pressures at approximately 10 and 7 cmH_2O, respectively, relative to the hilum of the lung. Thus, outflow pressure was 2 cmH_2O above airway pressure (P_{Alv}) at end expiration. Because the inflated lobe was approximately 6 cm high, more than 80% of the lobe was in zone III. We selected these pressures to keep lung weight from increasing more than 25% above initial weight over a 4-h observation period. In six edema experiments, we set Ppa and Ppv at about 18 and 15 cmH_2O, respectively, relative to the hilum. Under these conditions, the lobe was in zone III throughout the experiment and throughout all phases of ventilation. The lobe gained at least twice its initial weight over 6 h. The experiment was terminated with the appearance of edema foam.

We collected samples of airway fluid in five experiments for measurement of hematocrit and total protein. At the same time, we obtained 25 ml of the perfusion blood for measurement of hematocrit and total protein, and for determining the residual blood volume of the lobe. Finally, the lobes were frozen in liquid nitrogen and stored at −70°C until we could study the lung histologically. After microscopic examination of frozen specimens, the lobes were homogenized for measurement of lung water by the methods of Selinger and associates (10).

Total protein and albumin were determined by the biuret and BCG methods, respectively, in an automated system (AA II, Technicon, Tarrytown, NY).

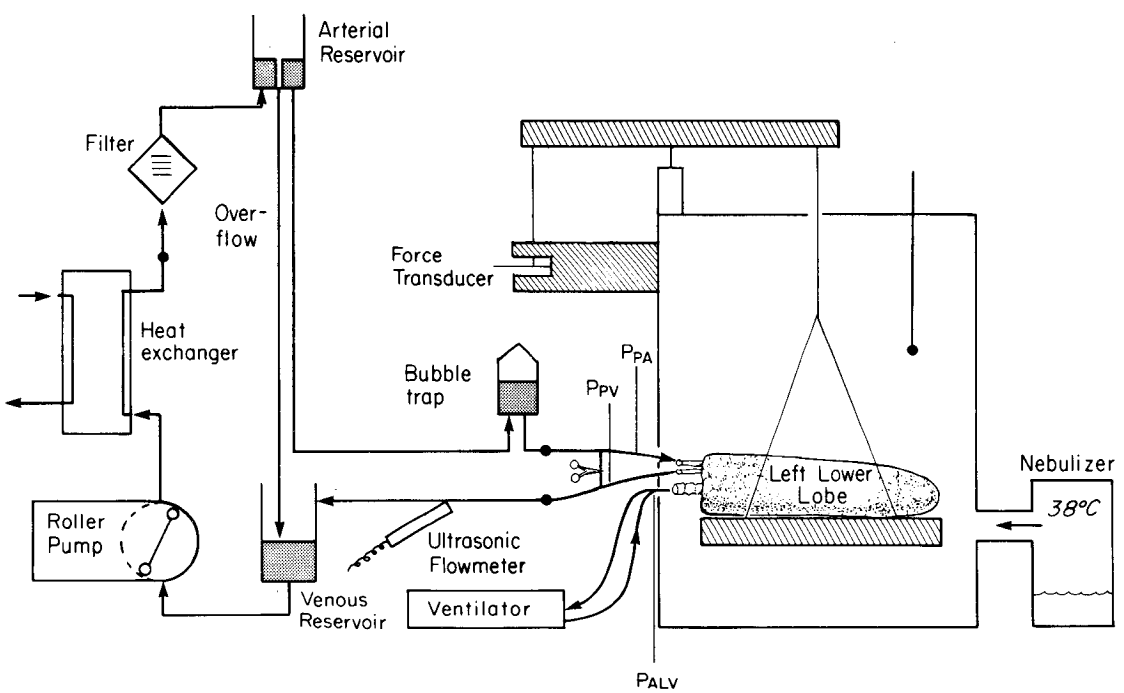

FIG. 1. Schema of apparatus for perfusion and continuous weighing of isolated dog lung lobe. Of special interest is our use of ultrasonic blood flow probe over venous outflow line, which permits continuous recording of lobe blood flow.

Data Analysis and Statistics

We recorded pressures and flows at end expiration as these vary cyclically with ventilation (see Fig. 2). In Figs. 3–5, comparing blood flow and lobe weight change over time, we have normalized the data as percent of the initial flow and weight. We averaged the data over 10–15 min. In Table 1, the control and edema experiment

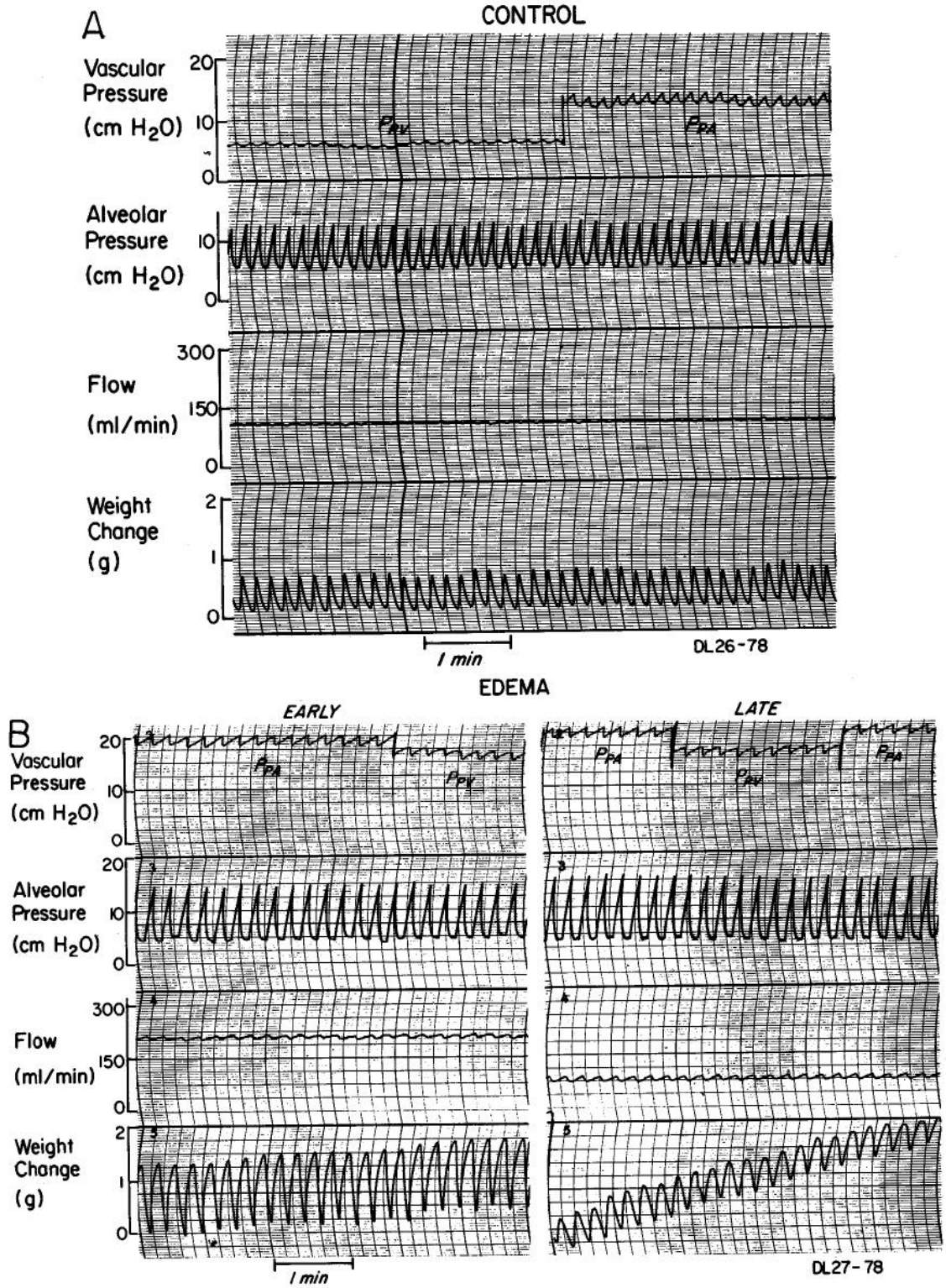

FIG. 2. *A*: tracings from control lobe; *B*: early (1st hour) and late (final hour) polygraph tracings from edematous lobe. Both pulmonary arterial (Ppa) and venous (Ppv) pressures were recorded alternately on same pressure transducer. Lung weight changed rhythmically with ventilation due to blood volume shifts. Slope of weight gain curve is much steeper in early phase of *B* than in *A*. It is much steeper in late phase of *B* than in early phase.

groups were compared by an unpaired t test. In our statistical analysis, we accepted $P < 0.05$ as indicating significance. The data are presented as the mean ± SD.

RESULTS

Control Experiments

Figure 2*A* shows segments of the polygraph record in one control experiment. The relation between lobe weight and blood flow for one experiment is shown in Fig. 3. Summary data for the four control lobes are shown in the first line of the table.

TABLE 1. *Effect of edema on blood flow in isolated perfused dog lung lobes*

Condition	No.	Lobe Wt, g		Rate of Weight Gain, g/min	Pulmonary Hemodynamics			
					Pressure, cmH_2O		Blood Flow, ml/min	
		Initial	Final		Pulmonary artery	Pulmonary vein	Initial	Final
Control	4	77.3 ±16.3	97.1 ±25.7	0.08 ±0.04	10.4 ±2.3	6.6 ±0.3	134 ±50	137 ±67
Edema	6	67.3 ±11.5	248.6 ±69.2*	0.37 ±0.13*	18.4 ±0.8*	15.2 ±2.0*	210 ±26*	30 ±27

Values are means ± SD. *Significant at $P < 0.05$ compared to control group.

At an average microvascular pressure of 8 cmH_2O, these lobes gained weight at a rate of about 5 g/h. The weight gained over the 4-h observation period averaged 20% of the initial weight (range 9–29%). In no experiment was there any significant change in blood flow over the

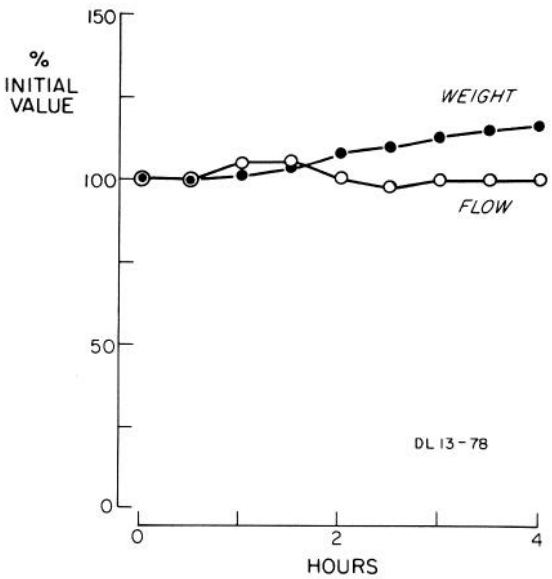

FIG. 3. Time course of change in lobe weight (*closed circles*) and blood flow (*open circles*) in one control experiment. On *ordinate*, 100% represents base-line (initial) values. This lobe (initial wt 84 g) had initial flow of 186 ml/min at Ppa = 10.5 cmH_2O and Ppv = 6.5 cmH_2O.

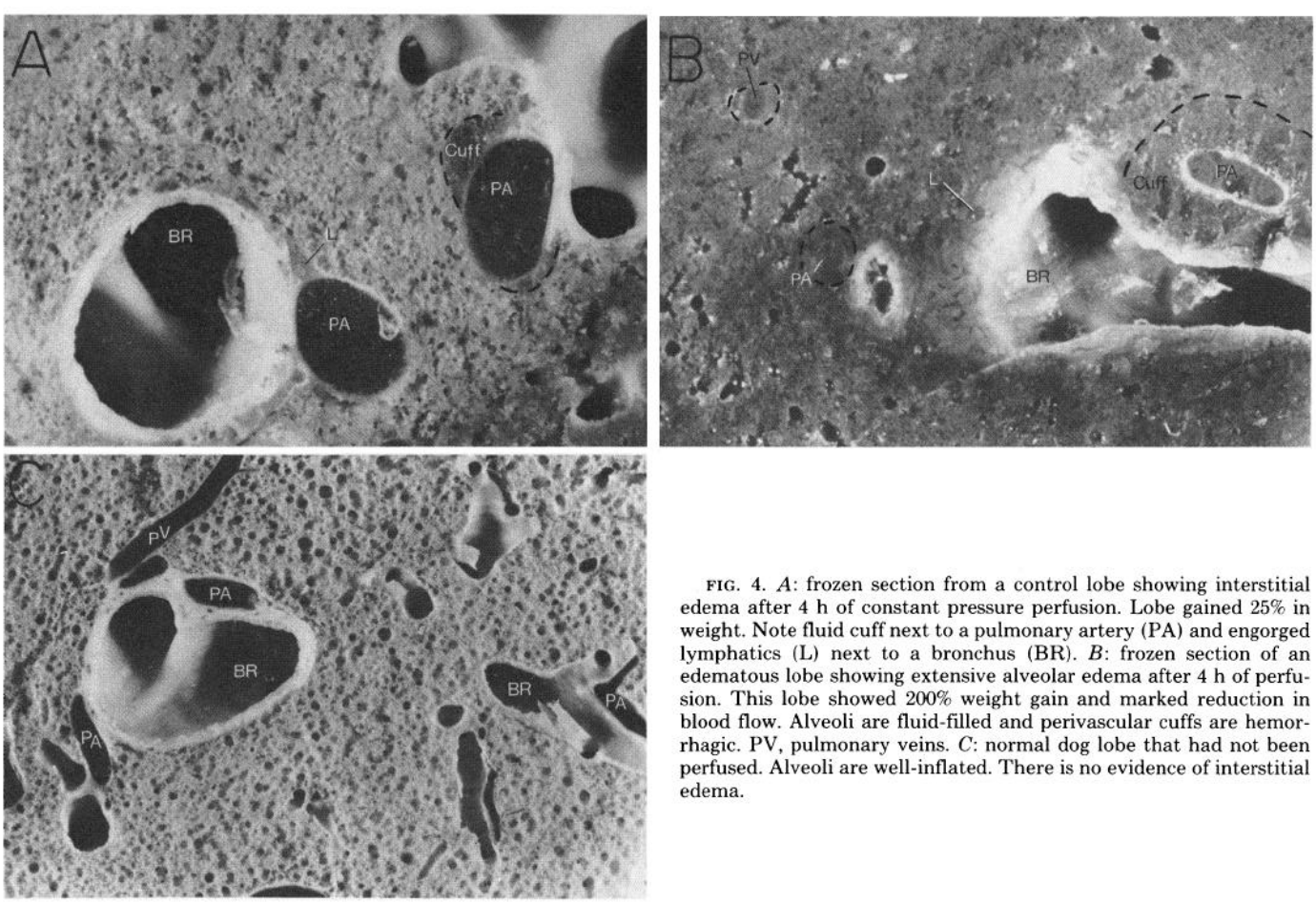

FIG. 4. *A*: frozen section from a control lobe showing interstitial edema after 4 h of constant pressure perfusion. Lobe gained 25% in weight. Note fluid cuff next to a pulmonary artery (PA) and engorged lymphatics (L) next to a bronchus (BR). *B*: frozen section of an edematous lobe showing extensive alveolar edema after 4 h of perfusion. This lobe showed 200% weight gain and marked reduction in blood flow. Alveoli are fluid-filled and perivascular cuffs are hemorrhagic. PV, pulmonary veins. *C*: normal dog lobe that had not been perfused. Alveoli are well-inflated. There is no evidence of interstitial edema.

4-h period. Compared to the right lower lobe in which extravascular water content averaged 3.1 ± 0.2 g/g dry lung, the perfused lobe's extravascular water was increased approximately 38% to 4.3 ± 0.8 g/g dry lung. This compares favorably with the measured weight gain over the 4-h experiment because 20% of initial weight is 30% of blood-free weight if lobe blood content initially was 0.3. The average hematocrit of the perfusate blood was 26 ± 9% at a total protein of 3.3 ± 1.2 g/100 ml.

Histologically, the frozen lobes were well inflated and of uniform color except at the extreme margins. There were small cuffs of fluid around the large pulmonary vessels and airways. An example is shown in Fig. 4*A*. The control lobe that had gained 29% of its initial weight showed a small amount of alveolar edema.

Edema Experiments

Tracings from the early and late phases of an edema experiment are shown in Fig. 2*B*. In the early phase, the rate of weight gain was faster than for the control lobes. In the late phase, in spite of constant vascular pressures, blood flow decreased markedly and the rate of weight gain increased. The relation between lobe weight and blood flow for one experiment is shown in Fig. 5. Summary data for the six edema lobes are shown in the second line of the table.

These lobes gained weight right from the beginning with the rate of weight gain increasing with time. Still, blood flow remained absolutely constant at first, even though the rate of weight gain averaged 22 g/h. In all six lobes, when the lobe had doubled its initial weight (avg increase of 101 ± 27%) blood flow began to decrease rapidly reaching about one-fifth of base-line flow within 1 h. Terminally, the lobes were almost four times as heavy as initially (avg increase of 269%).

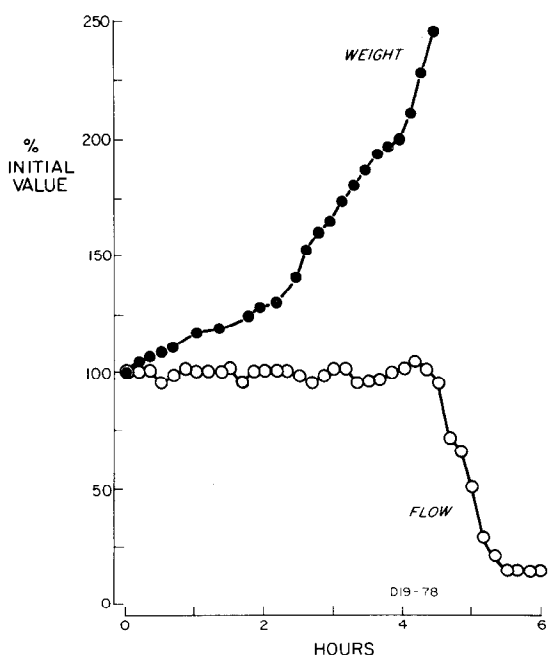

FIG. 5. Time course of change in lobe weight and blood flow in one edema experiment. This lobe (initial wt 61 g) had an initial blood flow of 179 ml/min at Ppa = 18 cmH_2O and Ppv = 12.5 cmH_2O.

The time of the onset of decreasing blood flow ranged between 1.5 and 4.5 h (avg 3.1 h). The reduction in flow depended only on the relative gain in lung weight, not on the time over which the weight gain took place.

Compared to the right lower lobe, in which extravascular water content averaged 3.6 ± 0.7 g/g dry lung, the perfused edematous lobes' extravascular water was increased 238% to 12.2 ± 0.4 g/g dry lung. This also compares favorably with the measured weight gain over the 6-h experiment.

The hematocrit of the airway fluid averaged 3.4% compared to 27% in the perfusing blood. The protein concentration of the airway edema fluid averaged 75% of that of the circulating perfusate plasma. The albumin fraction in the airway fluid was increased compared to blood in four of five experiments.

Histologically, the edematous lobes were congested with large perivascular and peribronchial fluid cuffs and extensive alveolar edema. Figure 4*B* is a photograph of one such lobe. Figure 4*C* shows what a normal dog lobe should look like for comparison.

DISCUSSION

Our control experiments indicate that the isolated perfused lung is a reasonably stable preparation. We were able to maintain steady blood flow in these lobes for 4 h; therefore the changes in blood flow that occurred in the edema lobes could not have been due to deterioration of the preparation with time. Nevertheless, these lobes, subjected to handling and interruption of blood flow as well as being deprived of lymphatics and innervation, are not normal. But for the purpose of this study, which was to quantify the effects of edema fluid accumulation on blood flow, the isolated lobe is ideal because extraneous factors have been eliminated.

Our control lobes increased in weight by 10–30% over 4 h in spite of the low vascular pressures. All these lobes showed histological evidence of interstitial edema and extravascular lung water was increased. This has been observed by others (6). It is of the greatest importance, however, to stress blood flow did not change! A remote possibility is that, in spite of constant pressure and continuous flow measurement, interstitial edema may have caused flow redistribution within the lobe. Such an event is very unlikely, because if localized interstitial edema caused local increases in vascular resistance then surely generalized edema up to 100% weight gain should have caused total resistance to increase.

In contrast, the edema lobes were arranged so that they would gain weight; they did. But blood flow remained constant until lobe weight had doubled. Although we did not stop our experiments at the point where flow reduction became evident, it is clear from our prior experience, as well as that of others (12), that a 100% increase in lung weight is always accompanied by alveolar flooding. We conclude that alveolar edema is a prerequisite for the observed reduction in blood flow.

Terminally, the lungs were grossly edematous and blood-tinged froth welled up from the main lobar bronchus. We determined the protein concentration of the edema fluid and found it to be about 75% of the plasma

concentration. Thus, we had induced an increased permeability edema by a modest increase in pulmonary microvascular pressure, something that does not occur in the intact animal's lung (1). This confirms the view of Snashall, Nakahara, and Staub (11) that the isolated perfused lung is excessively leaky to proteins.

In spite of its vigorous defense (4), there is little experimental evidence to support the hypothesis that interstitial edema compresses lung blood vessels and increases local vascular resistance (13). This is not to deny the phenomenon of redistribution of blood flow in edema.

Naimark and co-workers (8) found flow redistribution away from those parts of the lungs with alveolar edema. Muir and associates (7) found the same thing and showed a good correlation between the loss of alveolar gas volume and the reduction in blood flow, indicating that the amount of alveolar flooding determined the magnitude of the flow change. Our findings confirm this view because blood flow reduction took place only after alveolar flooding had occurred. This is attributable to the weight of edema fluid compressing alveolar wall vessels.

The constant-pressure perfusion system we used should be very sensitive to changes in vascular resistance. Thus, our finding of no flow reduction in four lobes with weight gain averaging 25% and in six lobes with weight gains up to 100% confirms the fact that interstitial edema alone does not affect pulmonary blood flow.

We thank Oscar Osorio and Walter Battaglia for their technical assistance.

This study was supported in part by Public Health Service Program Project Grant HL-06285.

Current address of K. Nakahara: First Dept. of Surgery, Osaka University Medical School, Osaka, Japan.

Received 16 April 1979; accepted in final form 29 October 1979.

REFERENCES

1. ERDMANN, A. J., III, T. R. VAUGHAN, JR., K. L. BRIGHAM, W. C. WOOLVERTON, AND N. C. STAUB. Effect of increased vascular pressure on lung fluid balance in unanesthetized sheep. *Circ. Res.* 37: 271–284, 1975.
2. HOGG, J. C. Effect of pulmonary edema on distribution of blood flow in the lung. In: *Lung Water and Solute Exchange*, edited by N. C. Staub. New York: Dekker, 1978, chapt. 7, p. 167–182.
3. HUGHES, J. M. B. Pulmonary interstitial pressure. *Bull. Physio-Pathol. Respir.* 7: 1095–1123, 1971.
4. HUGHES, J. M. B. Effect of edema on distribution of blood flow. In: *Regional Differences in the Lung*, edited by J. B. West. New York: Academic, 1977, p. 403–409.
5. HYMAN, A. L., D. G. PENNINGTON, AND P. J. KADOWITZ. Effect of acute pulmonary edema on pulmonary artery pressure in intact dogs. *Am. J. Physiol.* 224: 35–39, 1973.
6. ILIFF, L. D., R. E. GREENE, AND J. M. B. HUGHES. Effect of interstitial edema on distribution of ventilation and perfusion in isolated lung. *J. Appl. Physiol.* 33: 462–467, 1972.
7. MUIR, A. L., D. L. HALL, A. DESPAS, AND J. C. HOGG. Distribution of blood flow in the lungs in acute pulmonary edema in dogs. *J. Appl. Physiol.* 33: 763–769, 1972.
8. NAIMARK, A., B. W. KIRK, AND W. CHERNECKI. Regional water volume, blood volume and perfusion in the lung. In: *Central Hemodynamics and Gas Exchange*, edited by C. Giuntini. Torino, Italy: Minerva Medica, 1971, p. 144–160.
9. RITCHIE, B. C., G. SCHNAUBERGER, AND N. C. STAUB. Inadequacy of perivascular edema hypothesis to account for distribution of pulmonary blood flow in lung edema. *Circ. Res.* 24: 807–814, 1969.
10. SELINGER, S. L., R. D. BLAND, R. H. DEMLING, AND N. C. STAUB. Distribution volumes of [^{131}I]albumin, [^{14}C]sucrose, and ^{36}Cl in sheep lung. *J. Appl. Physiol.* 39: 773–779, 1975.
11. SNASHALL, P. D., K. NAKAHARA, AND N. C. STAUB. Estimation of perimicrovascular fluid pressure in isolated perfused dog lung lobes. *J. Appl. Physiol.: Respirat. Environ. Exercise Physiol.* 46: 1003–1010, 1979.
12. STAUB, N. C. Pulmonary edema. *Physiol. Rev.* 54: 678–811, 1974.
13. WEST, J. B., C. T. DOLLERY, AND B. E. HEARD. Increased pulmonary vascular resistance in the dependent zone of the isolated dog lung caused by perivascular edema. *Circ. Res.* 17: 191–206, 1965.