# Microradiography of Pulmonary Arterioles, Capillaries, and Venules of the Rabbit '

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ABSTRACT Pulmonary vessels of excised rabbit lungs were injected with a suspension of barium sulfate in gelatin. Slices 50  $\mu$  thick were radiographed at 5 kv and 2 ma using high resolution spectroscopic plates. When these plates were viewed through a microscope, pulmonary arterioles, venules and capillaries were identified. Arterioles show relatively regular branching at right angles. The capillary bed fills from short (10–20  $\mu$  long), thin (10–15  $\mu$  diameter) precapillaries arising at right angles from arterioles. The alveolar capillary network freely communicates with networks of adjacent alveoli. Several capillaries draining alveolar nets usually join forming a vessel which is broader at its origin than its insertion into a venule. These vessels, designated collecting venules join the venule at acute angles. Clear differentiation of small venous vessels from adjacent small arterial vessels is possible. The capillary network between an arteriole and venule appears to span at least two alveoli.

Miller ('37) conceived the capillary bed of the lung to be a rich network of freely anastomosing vessels around each alveolus. This has been confirmed by other investigators using a variety of techniques. (Wearn, Ernstene, Brower, Barr, German and Zschesche, '34; Irwin, Burrage, Aimar and Chesnut, '54; Krahl, '59, '62; Knisley, '60; Reid and Heard, '62; Staub, '63; Saunders and Carvalho '63; Weibel, '63.) It is now clear that a pulmonary arteriole may supply many alveoli, and conversely an alveolus may receive blood from more than one arteriole (Irwin et al., '54; Knisley, '60). The capillary networks of one alveolus communicate with those of adjacent alveoli (Staub, '63; Irwin et al., '54; Knisley, '60). Arterioles form the capillary bed by branching at right angles, (Krahl, '62). Pulmonary veins are located at maximum distance from arteries (Staub, '63), and receive tributaries at acute angles (Saunders, '63).

However, there does not appear to be a clear published description of the smallest radicles of the pulmonary venules. Yet, the anatomy of the terminal pulmonary capillary bed and the initial venous collection system is of physiologic importance since the final equilibration of alveolar air and pulmonary blood may occur here. We have shown (Quigley, Leathers and Reeves, '64) that capillary networks of air sacs with

their arterial supply may be identified in microradiographs of lung sections which are approximately one alveolus thick. Therefore, we have applied this technique to the study of small veins in the lung, hoping to differentiate these from the small arteries and therefore obtain a clearer definition of the limits of the pulmonary capillary bed.

#### METHODS

Healthy adult rabbits were given i.v. heparin 10 mgm/kg and then anesthetized with intraperitoneal pentobarbital 75 mgm/kg. The trachea was exposed and clamped, and the lungs with heart attached were carefully removed. The trachea was cannulated and the lungs inflated with air at a pressure of 2 cm H<sub>2</sub>O. A suspension of barium sulphate, gelatin, and water was injected into the pulmonary vasculature as previously described (Quigley et al., '64). The technique of injection was modified for three groups of rabbits in order to render radiopaque different parts of the pulmonary vasculature.

Group A, injection of pulmonary arterial tree and capillary bed. In six rabbits the barium sulphate suspension was injected at 50 mm Hg into the pulmonary arteries,

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whereas the pulmonary veins were cut and opened widely. Pulmonary arterial pressure of healthy rabbits averages 24 mm Hg (Reeves, Grover and Grover, '63).

Group B, injection of pulmonary venous system. In six rabbits the barium sulphate suspension was injected into the pulmonary veins at 10 mm Hg pressure after the pulmonary artery had been cut wide open.

Group C, injection of pulmonary arterioles, capillaries, and venules. In eight rabbits the barium sulphate suspension was injected at 50 mm Hg pressure into the pulmonary arteries. In the rabbit, pulmonary veins from both lungs join a common pulmonary vein before entering the left atrium. A glass tube with an outlet constricted to approximately 1 mm was tied securely in this common pulmonary vein. Partial obstruction of the pulmonary venous outflow was intended to distend the pulmonary veins.

Following injection of the barium sulphate suspension into the pulmonary vascular tree, the lungs were fixed in the inflated state by filling the airway with 10% formalin. The lungs were sliced and radiographed by conventional and by microradiographic techniques as previously described (Quigley et al., '64). For microradiography the lungs were sectioned 50  $\mu$  thick. Routine histologic sections from the same tissue blocks were taken for comparison with the microradiographs.

#### RESULTS

Irwin et al. ('54) list, in order, the components of the pulmonary circulation of living rabbits as pulmonary arteriole (diameters ranging 150 to 30  $\mu$ ), terminal arteriole (20 to 7  $\mu$ ), capillary (11 to 4  $\mu$ ), collecting venule (20 to 8  $\mu$ ) and pulmonary venule (160 to 30  $\mu$ ). We will follow this nomenclature. In addition, we will introduce and define the terms precapillary and confluence of venous capillaries.

# Pulmonary arterial tree (Group A)

Radiolucent (black) areas are airways and the radiopaque (white) areas represent contrast media within pulmonary vessels. The arterioles (fig. 1) tend to branch at right angles. When capillary networks lying in the walls of alveoli are seen "on edge" they appear as white curvi-

linear boundaries of the air sacs. Branching of smaller vessels is also at right angles (fig. 3). Here an arteriole (diameter,  $40\,\mu$ ) forms a right angle "Y," each limb of which ( $25\,\mu$ ) distributes several right angle branches to the same capillary bed. These limbs as well as the branches proximal to the "Y" probably are terminal arterioles (fig. 4). The short right angle branches (diameter,  $10{-}15\,\mu$ ) which feed the capillary bed are designated precapillaries. The terminal arterioles have a "knobby" appearance because of their dilation at the origins of precapillaries. The capillaries form a true network of freely anastomosing vessels.

# Pulmonary venous system (Group B)

The pulmonary venous system (fig. 2) may be compared to the pulmonary arteriolar system (fig. 1). Pulmonary veins are relatively short and they often receive tributaries at acute angles and more irregular intervals. Their branching pattern appears therefore less orderly. A pulmonary venule (diameter 50 µ) at higher power (fig. 5) shows tributaries joining at acute angles. The tributaries frequently are broad at the periphery and narrow at the point of junction with the venule. The broad end appears to represent the gathering of capillaries into a single channel, and we have called this portion of the vessel the confluence (fig. 6). That part of the venous system which appears to collect most of the capillary drainage is called the collecting venule. The collecting venule then, is distinguished by its location between capillaries and venules, its broad distal end, the confluence, its narrow point of junction, and frequently, its acute angle of junction with the venule.

# Pulmonary arterioles, capillaries and venules (Group C)

At least two pulmonary venules and four arterioles may be identified (fig. 7). Pulmonary venules in this and the following radiographs are less opaque than pulmonary arteries. Contrast medium reaching the pulmonary vein is diluted by the blood remaining in the lung and is pink tinged on emerging from the pulmonary veins. A lesser opacity of veins than ar-

teries has consistently aided differentiation of the supply and drainage systems. The arterioles tend to show regular right angle branching into vessels of progressively smaller size, whereas in the venous system, capillaries gather into confluences of the collecting venules which then narrow before joining venules. An enlargement of the central portion of this field (fig. 8) shows a venule, upper right, with the bulbous confluences of the capillaries at the origins of the collecting venules. Examination of figure 8 also suggests that contrast media crossed three alveolar nets in transit from arterial to venous systems. A long pulmonary venule at the left and an arteriole to the right are seen in figure 9. When the central portion is enlarged as in figure 10, the knobs of the terminal arterioles at the origins of the precapillary vessels are prominent. More than one arteriole contributes to this capillary bed. The contrast also appears to cross more than one arterial network before reaching the venous system.

Two venules and several arterioles are seen in figure 11. Note the hand like configuration of the venule in the upper center of the figure and the greater opacity of the arterial system. An arteriole feeding a "Y-like" collecting venule across several alveolar nets is seen in figure 12.

In these microradiographs and others examined, a pulmonary arteriole was not observed to communicate with a venule directly or across only one alveolar net. Rather, capillary networks receiving drainage from the arterial system through the precapillary vessels appeared to supply only other alveolar capillary nets. Conversely, capillary networks draining into the pulmonary venous system appeared to receive blood only from other alveolar capillaries. Thus, contrast media appeared to traverse two or more alevoli in transit from the arterial to venous system.

### DISCUSSION

This microradiographic technique resolves vessels as small as capillaries within the lung. The walls of air sacs may be identified by virtue of the vessels they contain. Fortunately, contrary to Barclay's early prediction ('51), the small vessels are not so delicate as to be incapable of

injection. Saunders and Carvalho ('63) were able to obtain good capillary filling in human lungs injected with fine barium sulphate suspensions. They also found that veins receive tributaries at acute angles. Published pictures of their air dried lungs suggested distortion of the vessels, however, making identification in the same section of arterioles and venules difficult. The major advantages of the technique in the present report are a clearer differentiation of small veins from arteries and the demonstration of these with their capillary beds.

As a result we suggest as has Knisley ('60) that in the rabbit, the pulmonary arterial blood perfuses more than one alveolus in transit from arteriole to venule. Published pictures of lungs in living rabbits (Irwin et al., '54) and of lungs quickly frozen (Staub, '63) suggest there may be more than one alveolus between artery and vein. Injections of red and blue latex into pulmonary arteries and veins respectively resulted in many all red, and many all blue alveolar nets but few with both red and blue color, (Oderr, Dauzat and Montamat, '63). These authors suggested that "arterial" blood passes several adjacent alveoli networks before entering the venous side of the capillary system. The concept that blood perfuses alveoli in series differs from the traditional view that ventilation of the blood is limited to its passage through one alveolus. One wonders if alveoli in series have differing oxygen tensions. Thus the physiologic significance of serial perfusion of alveoli requires examination.

Certain reservations must be borne in mind with our microradiographic studies. (1) Since these are excised lungs, injected with foreign materials, and fixed in formalin, distortion must occur. (2) The technique ignores the spontaneous opening and closing of living vessels (Wearn et al., '34; Irwin et al., '54). (3) When an alveolar wall with its capillary net is seen "on edge," the nature of the vessels in that wall remains obscure. (4) Single alveoli could be supplied and drained by vessels out of the plane of section. However, the results have been reproducible under the described conditions. In examining many radiographs of sections of varying thickness  $(35-150\,\mu)$  we expected to observe arterial supply to and venous drainage from a single alveolus if this occurred frequently. It is concluded that microradiography offers a promising and valid approach to a continuing problem, namely visualization of the small blood vessels of the lung and their relation to the air sacs.

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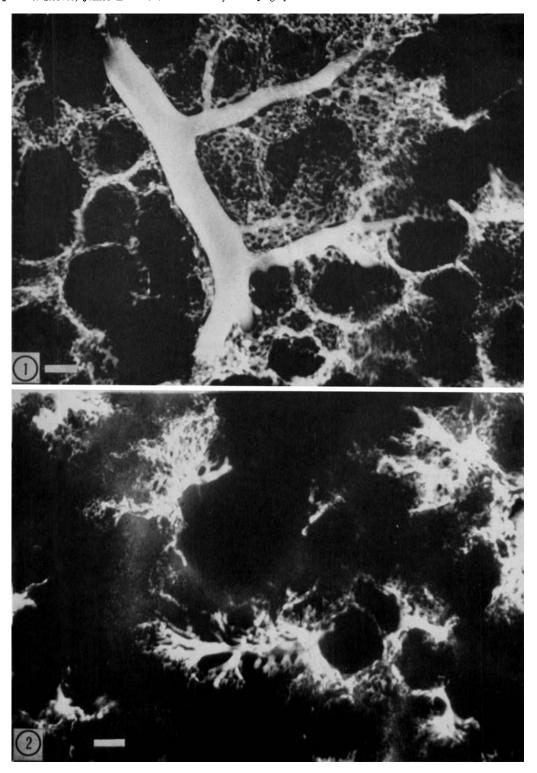
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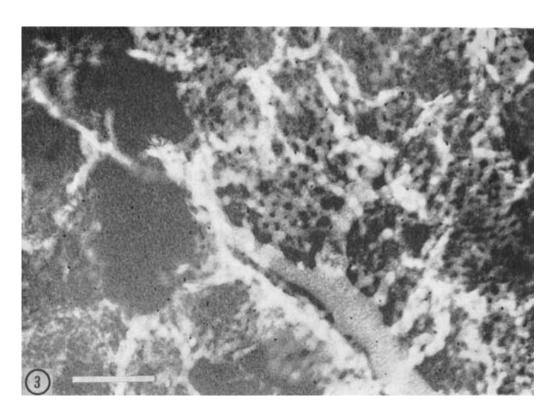
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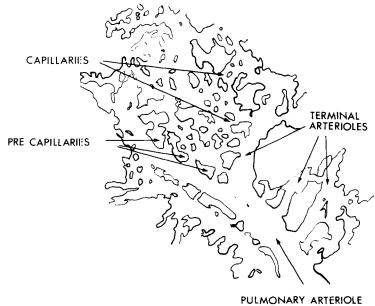
#### PLATE 1

- 1 Microradiograph of pulmonary arteriole in a rabbit (75 $\times$ ). There is some capillary filling. On this and subsequent plates the white bar in the left lower corner is 100  $\mu$  long.
- 2 Microradiograph of pulmonary venules in a rabbit  $(75 \times)$ .

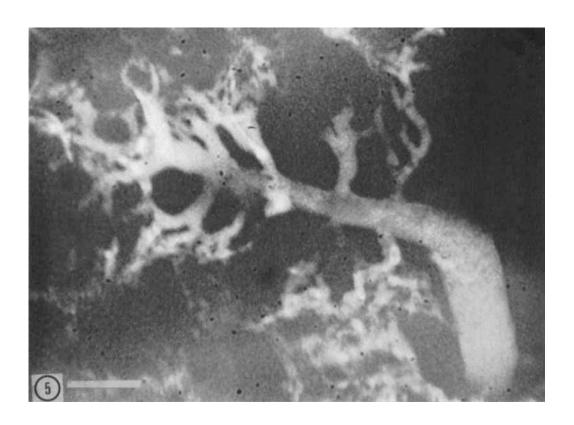


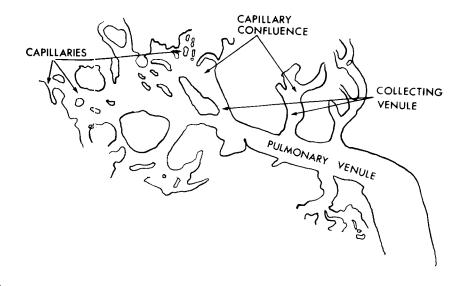
- 3 Microradiograph of pulmonary arterioles in a rabbit (200  $\times$ ).
- 4 Schema of figure 3 showing designation of vessels in the pulmonary arterial system.





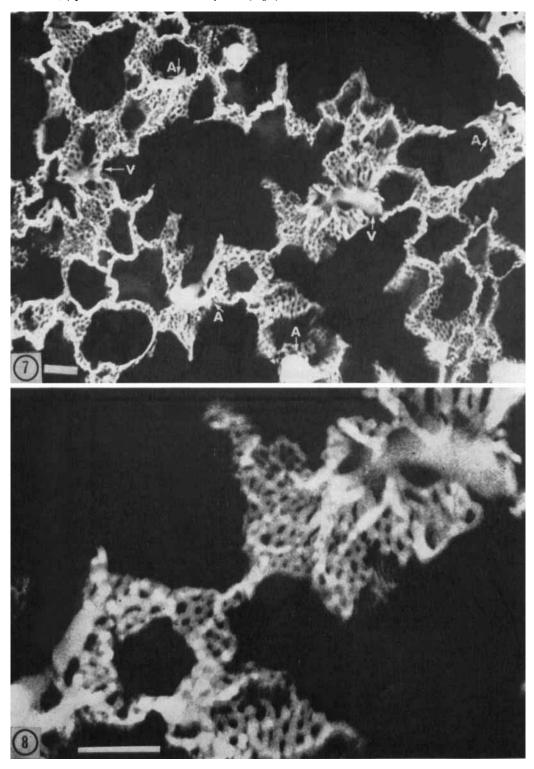
- 5 Microradiograph of a pulmonary venule in a rabbit (200  $\times$ ).
- 6 Schema of figure 5 showing designation of vessels in the pulmonary venous system.



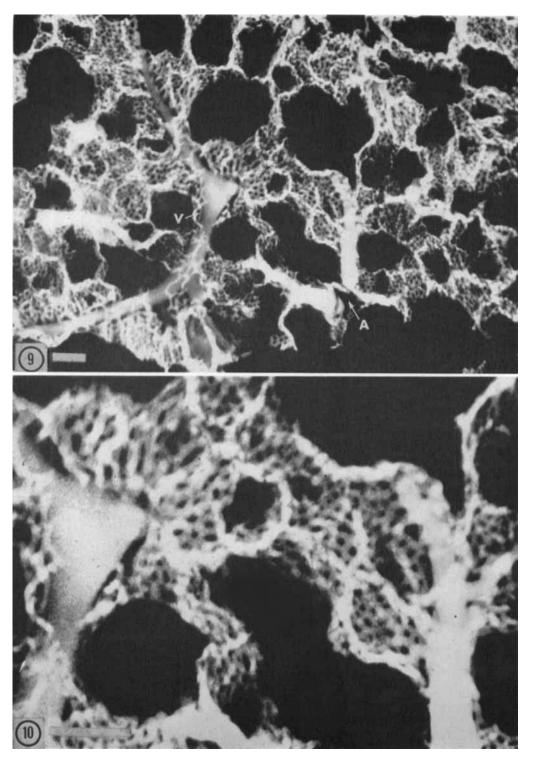


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- 7 Microradiograph of pulmonary arterioles (A), capillaries, and venules (V) in a rabbit  $(75\,\times).$
- 8 Enlarged center section of figure 7 (200  $\times$ ).



- 9 Microradiograph of pulmonary arterioles (A), capillaries, and venules (V) in a rabbit (75  $\times$ ).
- 10 Enlarged center section of figure 9 (200  $\times$ ).



- 11 Microradiograph of pulmonary arterioles (A), capillaries, and venules (V) in a rabbit (75  $\times$  ).
- 12 Microradiograph of pulmonary arterioles (A), capillaries, and venules (V) in a rabbit (75  $\times$  ).

