POSTNATAL CHANGES IN THE TERMINAL VASCULAR BED OF THE CANINE VENTRICULAR MYOCARDIUM

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Kenneth R. Holmes

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Kenneth R. Holmes

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To

Casper

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INTRODUCTION

Extensive research has been conducted in attempting to elucidate the nature of the intrinsic blood vascular system in both the normal and pathologic myocardium.

Investigators of the morphology of the coronary vasculature are generally aided by the use of injection materials which render the vasculature easily dissectable, or visable by corrosion or radiographic techniques. However, these macroscopic methods do not demonstrate the terminal vascular ramifications of the capillary bed.

The scarcity of publications regarding the morphological pattern of the myocardial microcirculation may be explained by the difficulty in injecting the capillaries of the heart muscle. Because many workers had failed to fill the myocardial capillaries, Nussbaum was led to say that complete injection of the heart capillaries was impossible (Wearn, 1928).

The first injection of the complete capillary bed of the heart was reported by Wearn in 1928. His research involved the calculation of capillary/heart muscle fiber ratios in the cat, rabbit and man. Research workers have subsequently exploited the capillary/muscle fiber ratio analysis to such an extent that the morphology of the heart microcirculation has been neglected.

The purpose of this paper is to present the results obtained with the injection of India ink into the vasculature of the isolated, beating heart of canine pups 12 hours to 7 weeks of age.

REVIEW OF LITERATURE

A search of the literature has revealed some information regarding the morphological pattern of the myocardial microcirculation. Prior to the method of injection reported by Wearn in 1928, Nussbaum stated that a complete injection of the myocardial capillaries was impossible (Wearn, 1928). Nearly two decades later Gregg (1946) stated that "comparatively few studies have been made on the capillaries of the heart muscle because of the great difficulty of their injection".

The first complete injection of the myocardial capillary bed is reported by Wearn (1928). Wearn's technique involved the injection of India ink or Berlin Blue into the isolated beating heart of the cat, rabbit, and man. His investigation of the capillary/heart muscle fiber ratio in the normal heart was extended by Shipley, *et al.* (1937) and Roberts and Wearn (1941) to include the hypertrophied heart. Angelakos, *et al.* (1964) used a modification of Wearn's injection technique to report the capillary-fiber ratio in the ventricles of the rat.

Bennett (1936) in his excellent study of the development of the myocardial vasculature in the pig embryo, described the formation of the myocardial capillary bed. He observed that the arterial primordia grow and branch abundantly in the subepicardium to form an expansive subepicardial capillary bed. This dynamic capillary network sends invading sprouts into the myocardium to form a system of anastomosing capillary loops. These loops are seen as dilated vessels with irregular lumens when immature, but have developed into "regular channels without embryonic dilatations" by the time the large coronary vessels have reached their adult patterns.

According to Roberts (1961), an additional method of heart capillary formation occurs with the constriction of most of the intertrabecular spaces or sinusoids of the embryonic spongy myocardium into capillaries and small sinusoids. The invading capillary loops from the subepicardial capillary plexus communicate with these capillaries and small sinusoids to form the myocardial capillary bed.

The presence of vascular sinusoids in the adult heart myocardium was noted by Truex and Angulo (1952). They noted that these sinusoids may function as capillaries in the myocardium. Young and Fell (1962) stated that the retention of embryonic sinusoids probably accounts for the presence of the Thebesian vein in the adult myocardium.

The arteriole, terminal arteriole, and metarteriole in the myocardium can be recognized histologically by the muscular nature of the intima (Reynolds, et al. 1958, Provenza and Scherlis, 1959a, 1959b). With segments of muscle lacking in the metarteriole, and the total lack of muscle cells in the true capillaries, Provenza and Scherlis (1959b) stated that a positive identification of the capillary or metarteriole is ascertained only if the vessels are longitudinally disposed, or if serial section reconstructions are made.

It is reported by Provenza and Scherlis (1959a) and Shmerling (1963), the veins of the heart surface are similar to those in other tissues. Reynolds, et al. (1958) noted that the venules are recognized by the multidirectional merging of capillaries which give rise to a rapidly increasing vessel lacking a muscular wall.

Provenza and Scherlis (1959a) and Shmerling (1963) also reported the lack of a muscle wall in the myocardial veins. Shmerling (1963) concluded that all the veins of the ventricular myocardium, with the exception of those on the heart surface, are sinusoids in shape and structure. He stated that the fibers of the myocardium "by their rhythmic constant contractions serve the same function as the missing muscular coat would ordinarily supply".

According to James (1961) and Shmerling (1963) the arteries of the myocardium are generally accompanied by a pair of veins in the dog. This was not observed in the human heart (James, 1961).

Using a modification of Wearn's injection technique, Brown (1965) reported the pattern of the microcirculatory bed in the ventricular myocardium of domestic mammals, including the adult dog. He described the arterioles as branching dichotomously with size reduction characteristics similar to those stated by Provenza and Scherlis (1959a). The capillary bed was described as being composed of long anastomosing meshes for which Brown presented a schematic drawing showing the pattern of the observed capillary anastomoses. Venules were formed by the confluence of capillaries resulting in an arborization which Brown reported as giving a "turnip root" appearance.

Brown pointed out the probability of a radical change in the morphological pattern of the myocardial capillaries as the animal develops from birth to adulthood.

MATERIALS AND METHODS

Injection Materials

A pilot program was undertaken to devise the optimum combination of events and ingredients which would be used in the collection of the specimens presented in this investigation. The technique which finally gave uniformly excellent results, as well as the technical failures will be described in detail to aid others who have experienced difficulty in obtaining complete myocardial capillary injections.

The selection of an injectable substance must be based on the ability of that substance to enter the capillary bed, show no extravasation, and withstand the processes of fixing, freezing, dehydration, and staining. The resulting product must be visible when used with conventional optics and photomicrography.

The India inks have been used with reported success by many investigators, and this type of visual material was used in this investigation. When properly executed, the injection of the isolated beating heart gave uniformly excellent capillary fillings. The procedure used was essentially that of Wearn (1928) with some modifications which rendered it more effective and useful.

The heart was perfused with Locke's solution prior to the injection of the ink to wash the heart vasculature free of blood and maintain a rhythmically beating heart. In the pilot study, the perfusate was first oxygenated, as indicated by Brown (1965) and Wearn (1928); however, it was found that better capillary filling resulted when the perfusate was not oxygenated. Therefore, this improvement led to the use of unoxygenated perfusate for the hearts obtained in this investigation. The pilot program showed that the addition of heparin* to the perfusate increased the number of successes of completely filled capillary beds. The addition of 2 ml of heparin per 1000 ml of Locke's solution was used in the course of this investigation. All chemicals were of reagent quality dissolved in distilled water and stored in Pyrex glass containers.

During the pilot program, ink injections were conducted using Higgin's India ink** or Pelikan ink***. Pelikan ink has a pH of 8.1, while Higgin's India ink is pH 9.5 (Peterson, et al. 1965). The pH of canine blood ranges from 7.4 to 7.6, with an average pH of 7.5. The best results were obtained with Pelikan ink and thereafter this was used exclusively in injecting the hearts in this investigation. The similarity of dog blood and Pelikan ink pH may account for the better results. The ink was used as it came from the bottle, being siphoned so as not to include any sediment.

Age, Numbers and Species Injected

The hearts of nineteen healthy mongrel puppies of both sexes were injected during the course of this investigation. The animals ranged in age from 12 hours to 7 weeks old. The hearts were taken at weekly intervals, starting with 12 hours post partum. The

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* Ammonium Salt, Scientific Products Division, American Hospital Supply Corp., Evanston, Ill.

** Higgin's Ink Co., Inc., Brooklyn 15, N.Y.

*** John Henshel and Co., Inc., 425 Park Avenue, South New York 16, N.Y.

following are the number of hearts studied for each age interval: 3 hearts at 12 hours, 5 hearts at 1 week, 2 hearts at 2 weeks, 2 hearts at 3 weeks, 2 hearts at 4 weeks, 2 hearts at 5 weeks, 2 hearts at 6 weeks, 1 heart at 7 weeks.

Injection Methods

During the first phases of the pilot injection study, the animals were killed by a blow on the head. Later, this method of sacrifice was abandoned in favor of the intraperitoneal injection of a suitable anesthetic. The anesthetic used in this investigation was Halatal* and the heart was removed as quickly as the animal was rendered unconscious.

The heart was approached through a sternal thoracotomy. During the pilot study, blood clots were found in the cut aortic lumen, and time was lost in removing what hopefully appeared to be the entire clot mass. Irregular or incomplete ink injections would result if a clot of even small proportions were to enter the coronary system with the perfusate. Therefore, immediately upon exposing the thoracic viscera, heparinized Locke's solution was used to fill the spaces around the intact viscera. This aided in preventing rapid blood clotting which was encountered during the heart removals of the pilot study. Using a hypodermic syringe, heparin was squirted into the chest cavity to bolster the action of the heparinized Locke's solution.

It was found that these young hearts were very susceptible to

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* Pentobarbital sodium, Jen Sal Laboratories, Kansas City, Missouri.

trauma, resulting in myocardial hematomas. Thus, during the entire removal, perfusion and injection, the heart was handled and manipulated by grasping the great vessels, avoiding unnecessary contact between foreign objects and the myocardium.

Following heart extirpation, a cannula was quickly tied into the aorta (Fig. 1) at a distance sufficient to insure that the normal coronary artery openings would not be blocked. This arrangement also allowed both coronary arteries to be injected simultaneously and at equal pressure. Injection pressure was developed and maintained by a hydrostatic pressure head of 55 cm, as indicated by Brown (1965).

Perfusion was always started before the heart had ceased beating. Air bubbles in the injection apparatus were avoided by allowing the perfusate to bypass the coronary system and pass freely out of the cut ends of the brachiocephalic and left subclavian arteries. During this time of purging, the pericardial sac was incised at the apex and folded back around the base of the heart. Hemostats were then placed on the cut ends of the arteries, thus forcing the perfusate into the coronary system. The myocardium quickly lost its deep red color and assumed a pale bloodless appearance, indicating that the vessels were filled with the perfusate and that none of the vessels were blocked with clots. If a heart area retained its reddish color, later investigation disclosed these areas to be devoid of the ink injection material. The perfusion was continued until such time when a strong heart beat was attained as evidenced by its rate, rhythm, and intensity. The flow of perfusate was then stopped and the flow of the ink was begun.

As reported by Wearn (1928) the heart literally injects itself by its beating action, becoming uniformly black as the vasculature fills with ink.

In the first pilot study injections, the heart was allowed to continue to beat until the contractions finally slowed, then ceased altogether. Hearts injected in this manner gave poor results with the capillaries and most of the greater vessels merely being outlined by residual ink particles clinging to the vessel wall. Wearn (1928) stated that permitting the heart to beat beyond the "time of the maximum injection of the capillaries" will result in a "milked out" condition, where the capillaries are nearly empty of the ink.

Prevention of this "milking out" process, caused by the slow cessation of the heart beat, was attempted by controlling the heart beat. During the pilot study, this "controlled" heart beat was attempted with a heart stimulator. The heart was forced to beat until such time as it was desirable for sudden and complete stoppage of the heart beat. The heart beat was pulsed until the time when removal of the stimulus would result in heart stoppage. However, the inability to determine the time when normal intrinsic pulsatile activity had ceased precluded its use in the final investigative procedure.

A long acting parasympathomimetic drug, Lentin* was found to be the most effective way to cause complete cessation of the heart during diastole. It was observed that this drug is equally effective if injected with the ink, directly into a ventricular heart chamber,

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* Merck and Co., Inc., Rakway, N.J.

or applied topographically on the epicardium. Regardless of how the drug was applied, the heart always stopped in sudden diastole and consistantly excellent capillary bed injections were obtained.

Tissue Preparation

Immediately following heart stoppage, a soft cord was tied around the great vessels at the base of the heart. The heart was placed in 5% neutral formalin and allowed to harden for 4 days prior to further work with the tissue. Tissue blocks were removed from the fixed heart as illustrated in Figure 2. Because hearts from younger animals were so small, it was not always possible to take all sections from the same heart.

The tissue blocks were placed on the freezing stage of a Sartorius-Werke* freezing microtome. Sections were cut at 10, 20 and 40 microns, the majority of which were placed on glass slides and coverslipped with 20% gelatin as the mounting medium. For examination under the conventional light microscope, tissue sections of this thickness did not require a clearing process (see Fig. 3).

The remaining sections were stained with hematoxylin and eosin, cleared in xylene, and coverslipped with Permount**. By the use of these stained sections, it was possible to histologically identify the arteries, arterioles, and veins. Having observed the characteristic pattern of branching for each vessel type, it was a simple matter to use this information in identifying these

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* Gottingen, Germany.

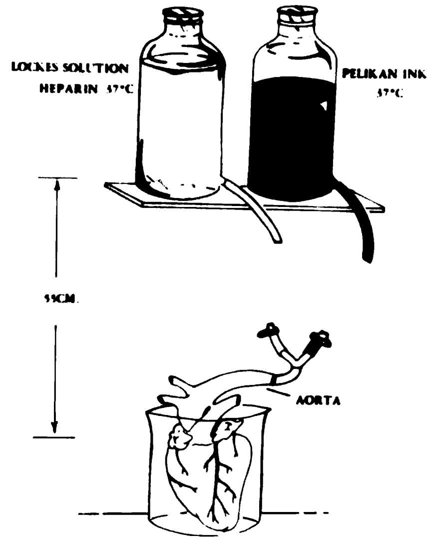
** Fisher Scientific Co., Chicago, Ill.

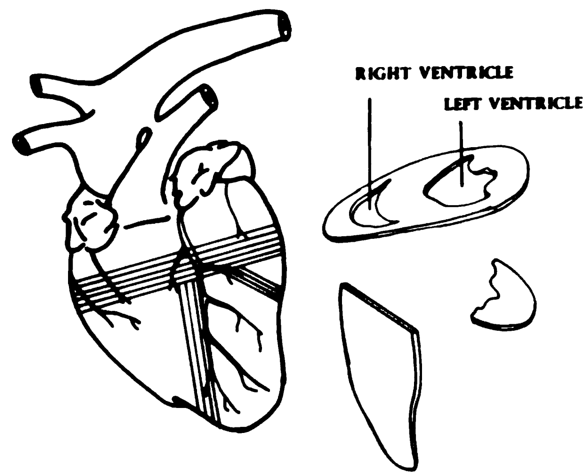
Figure 1. Perfusion and injection apparatus.

Figure 2. Scheme of sectioning.

Figure 3. Photograph of heart cross-sections prepared for investigation.

INJECTION APPARATUS





SCHEME OF SECTIONING

vessel types in sections which had not been stained. With the arteriole and/or vein identified the intervening terminal arteriole, capillary, post capillary venule, and venule are readily identifiable.

Optical and Photographic Equipment

The photomicrographs were made with a Carl Zeiss Photomicroscope with an automatic exposure setting device*. This instrument also served as the binocular, monobjective light microscope. The photographic film used was Adox KB14**. The 35 mm film has been enlarged in printing to a total magnification of 175, the noted exceptions being shown on the figures.

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* Carl Zeiss, Oberkochen, Wuerttemberg, Germany.

** Adox Fotowerke, Frankfurt, Germany.

OBSERVATIONS AND DISCUSSION

General Statement

Before presenting the data, some of the terms used throughout this paper will be defined. The phrase, terminal vascular bed, refers to the capillaries and that portion of the arteriole, venule, or vein at its junction with the capillary. The capillaries are those vessels which connect the smallest ramifications of the arterioles and venules. The arterioles and veins are identified histologically by the presence or absence of a muscle coat respectively (Fig. 31, 32). Once the definitive branching pattern of each vessel type has been identified in the stained preparation, this pattern is easily identified in the unstained preparation. The term, venous collection area, describes a region of dilated anastomosing venules. The term, capillary sinusoid, as used in this paper, refers to a vessel with the usual arteriole-capillary-venous relationship but having an irregular lumen of a generally larger diameter than that of a capillary.

Unless specifically stated otherwise, all descriptions of the terminal vascular bed that follow are typical of the right and left ventricular and septal myocardium.

Arterioles

The arteriole pattern of the terminal vascular bed is similar in all hearts studied, regardless of age, and will be discussed here to avoid duplication.

The arterioles in the ventricular walls branch in a dichotomous manner until the capillary bed is reached. Branching is accomplished with no reduction in the diameter of the daughter vessels (Fig. 13, 14, 16, 18, 24, 29, 31, 33, 34, 38, 41) with a reduction in both daughter vessels (Fig. 12, 15, 21, 29). The arterioles frequently terminate in a bifurcation which sends daughter arterioles in opposite directions (Fig. 12, 14, 16, 17, 27, 33). It is not uncommon to find small anastomotic loops associated with the termination of an arteriole (Fig. 16, 17). This is particularly the case in the younger hearts.

The arterioles in the interventricular septum form a somewhat different pattern. Although the branching characteristics in the septum are similar to the branching in the ventricular walls, the larger distributing arterioles are situated at nearly right angles to the direction of the capillary bed (Fig. 9, 25, 39), and form a type of arterial arcade.

Capillaries and Veins

12 Hour-Old Heart

The capillary bed consists of a tightly meshed network of anastomosing sinusoids (Fig. 10, 11). The sinusoidal nature of the capillary bed is typified by the large, irregular cross-sectional diameter of the capillaries (Fig. 5). The positive identification of a capillary or venule is confirmed only when the vessel in question can be followed to the arteriole or vein (Fig. 10).

1 Week-Old Heart

The capillary bed consists of a meshwork of anastomosing capillary sinusoids (Fig. 6, 13, 14, 15, 16). These anastomotic loops of sinusoids are longer than those of the 12 hour heart (Fig. 13, 16). A sinusoidal capillary is occasionally seen forming an arterio-venous anastomosis (Fig. 13).

A large area of tightly meshed anastomotic sinusoidal structures typifies the venous side of the terminal vascular bed (Fig. 13).

The length of the terminal vascular bed is relatively short when compared to that of the 7 week heart (Fig. 41, 42).

2 Week-Old Heart

The capillary component of the terminal vascular bed is sinusoidal in nature (Fig. 17, 18). The capillary lengths remain relatively short in comparison to the 7 week heart (Fig. 41, 42).

The venous collection area forms a more substantial configuration with the venule diameter being equal to or greater than the proximally located arteriole (Fig. 17, 18). In most instances the anastomotic sinusoidal looping is confined to the venous collection area (Fig. 19).

The complex arrangement of muscle fibers at the junction of the ventricular walls and the interventricular septum results in a complex pattern of capillary sinusoids (Fig. 20).

3 Week-Old Heart

The capillary sinusoids are now fairly uniform in cross-sectional area (Fig. 7) but they continue to remain a prominent feature of the capillary bed (Fig. 21, 22, 23, 24). The capillary length is generally longer than that of the 2 week heart (Fig. 21, 22) but areas of short capillaries frequently are found (Fig. 24).

Anastomosing capillary loops seldom are seen outside the venous collection area. The large veins are now a prominent feature in the venous side of the terminal vascular bed with the capillaries frequently entering directly into their large lumen (Fig. 21).

4 Week-Old Heart

The large capillary sinusoid of the 12 hour heart generally is replaced by a vessel of smaller diameter, but of larger diameter than the capillaries of the 7 week heart (Fig. 25, 26, 27, 28, 29, 30, 41, 42). However, the capillary sinusoid characteristic of the younger hearts is frequently found in all areas of the 4 week heart. The distance between the arterial and venous side of the terminal vascular bed continues to increase over that of the 3 week heart (Fig. 27, 29), with areas of short capillaries frequently found (Fig. 27, 28, 30).

The venous collection area retains the anastomotic nature of the 12 hour heart (Fig. 30). The sinusoid loops are now confined entirely to the venous portion of the terminal vascular bed. (Fig. 30).

The junction of the ventricular wall and interventricular septum results in a complex pattern of capillaries (Fig. 26). The capillaries in this area reflect the change in size of the vasculature in the ventricular walls and septum (compare with Fig. 20).

5 Week-Old Heart

The capillaries continue to decrease in diameter and closely approximate the shape of the capillaries in the 7 week heart (Fig. 31, 32, 33, 34). The capillaries are longer but remain shorter than the 6 week or 7 week heart capillaries.

The venous collection area generally is replaced by venules draining directly into large veins, with a nearly complete absence of anastomotic vessels (Fig. 33, 34).

6 Week-Old Heart

The sinusoidal nature of the younger heart capillaries is no longer evident in the 6 week-old heart. In its place are the long thin capillaries as seen in the adult myocardium (Brown, 1965) (Fig. 35, 36, 37, 38). The capillaries bifurcate many times prior to venule formation (Fig. 35, 37) and the network thus formed appears similar to that reported by Brown (1965).

The number of venous collection areas is less than in a younger heart with venous drainage of the terminal vascular bed accomplished by the junction of the capillary or venule with the vein (Fig. 36, 37).

7 Week-Old Heart

The terminal vascular bed is similar to the 6 week heart with the exception of a further increase in the capillary length (Fig. 39, 40, 41, 42). The capillary bifurcations are increasingly evident over that of the younger hearts (Fig. 41, 42).

The uniform diameter of the capillary component of the terminal bed is seen best in cross-sections (Fig. 8). The capillary sinusoid of the younger heart is no longer a component of the terminal vascular bed of the 7 week heart.

General Discussion

The postnatal changes in the ventricular myocardial terminal vascular bed are schematically illustrated in Figure 4. The schematic

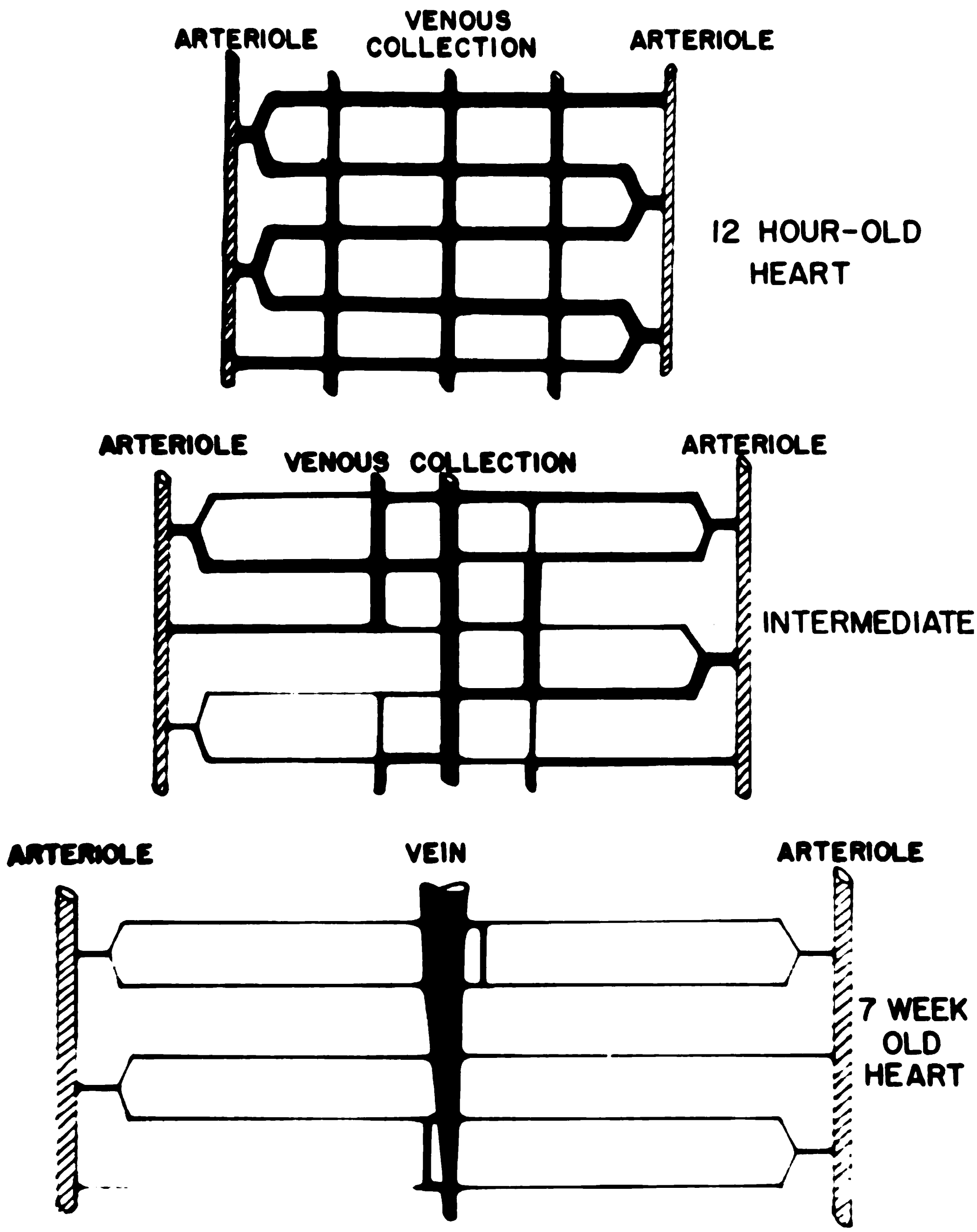


Figure 4. Schematic drawing showing the progressive transition from the anastomosing sinusoidal capillary pattern and diverse venous collection areas of the 12 hour heart, to the non-anastomosing, long capillary, narrow venous collection area of the 7 week heart.

nature of the figure must be emphasized, and no attempt is made to show a pattern of capillary branching.

Three features of the terminal vascular bed are developmentally active as the heart increases from 12 hours to 7 weeks of age. The dynamic aspects of the terminal vasculature are; 1. The luminal size of the capillary, 2. The area of venous collection, and 3. The distance between the arteriole and venous sides of the terminal bed.

The capillary lumen in the 12 hour heart is sinusoidal in nature. In the 12 hour heart, the capillary cannot be distinguished from a venule unless a direct connection to the arteriole or vein is observed. Anastomoses are numerous forming a tight meshed network in the terminal vascular bed of the 12 hour heart. As the age increases, the capillaries gradually lose their sinusoidal shape and at 7 weeks give an adult appearance as reported by Brown (1965). The anastomotic loops disappear from the capillary bed at a slightly faster rate, being seen only rarely after the age of 4 weeks.

The area of venous collection is difficult to differentiate from the capillary sinusoid in the 12 hour heart, as noted previously. However, the venous side of the vascular bed is clearly evident in the 1 week heart as an area of anastomosing sinusoidal vessels. The loops formed by the anastomoses is relatively smaller than the vascular bed loops observed in the 12 hour heart. As the age of the heart increases, the loops coalesce to form the venule patterns, which at 7 weeks agree with those patterns reported by Reynolds *et al.* (1938) and Brown (1965) for the adult dog.

The fact that the myocardial capillary structures lengthen (the distance between the arteriole and venous sides increases) with age over the 7 weeks of growth studied in this paper, may be explained by the following sequential events. During the development of the coronary vasculature there may be a time when a further increase in the number of arterioles developing in the myocardium no longer occurs. Thus the relative positions of the arterioles become fixed in the myocardium. If the heart muscle continued to grow and enlarge, as the heart does from 12 hours to 7 weeks, it may result in the effective separation of the arterioles, one from the other. It seems logical that the capillaries "fill in" this separation by a lengthening process as seen when comparing Figures 10 and 41.

The heart muscle extracts about 70 percent or more of the oxygen presented to it (Rushmer, 1961). Brown (1965) stated that the length of the myocardial capillaries may account for this high rate of extraction. It is left to some future investigator to examine the relationship between the short, sinusoidal nature of the myocardial capillary observed in the 12 hour heart, and the percentage of oxygen extraction in this age of heart.

SUMMARY AND CONCLUSIONS

The postnatal changes in the terminal vascular bed of the canine ventricular myocardium were studied microscopically following India ink injection through the coronary arteries of the isolated beating heart. The observations were based on a study of 19 normal dogs ranging in age from 12 hours to 7 weeks old.

The vascular bed was visualized by using a procedure similar to that reported by Wearn in 1928. Tissue blocks from the ventricular myocardium were sectioned on a freezing microtome and mounted with a gelatin solution for microscopic examination and photography. The phrase, terminal vascular bed, as defined for this paper included the capillaries and that portion of the arteriole, venule, or vein at its junction with the capillary.

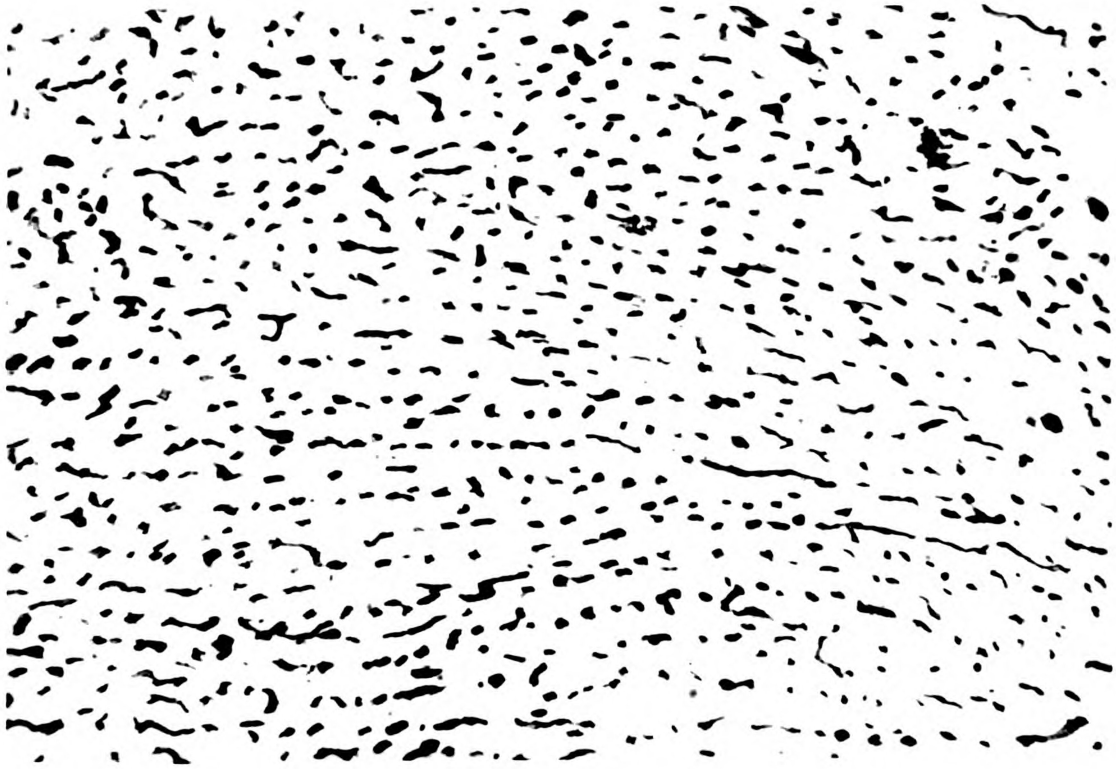
Three features of the terminal vascular bed were developmentally active as the heart grew from 12 hours to 7 weeks of age. The capillaries of the 12 hour heart consisted of anastomosing sinusoidal structures. As the heart aged, the sinusoidal and anastomotic nature of the system was lost and at 7 weeks the capillary bed was that of the adult dog.

The venous system coalesced from the expansive venous collection area of the younger hearts to a venous system which, at 7 weeks, was that of the adult dog.

The length of the capillary continued to increase from the short vessel of the 12 hour heart, to a longer vessel in the 7 week heart. At 7 weeks the capillary had not reached its adult length.

Figure 5. 12 hour-old heart - Cross-section of capillary bed. X175.

Figure 6. 1 week-old heart - Cross section of capillary bed. X175.



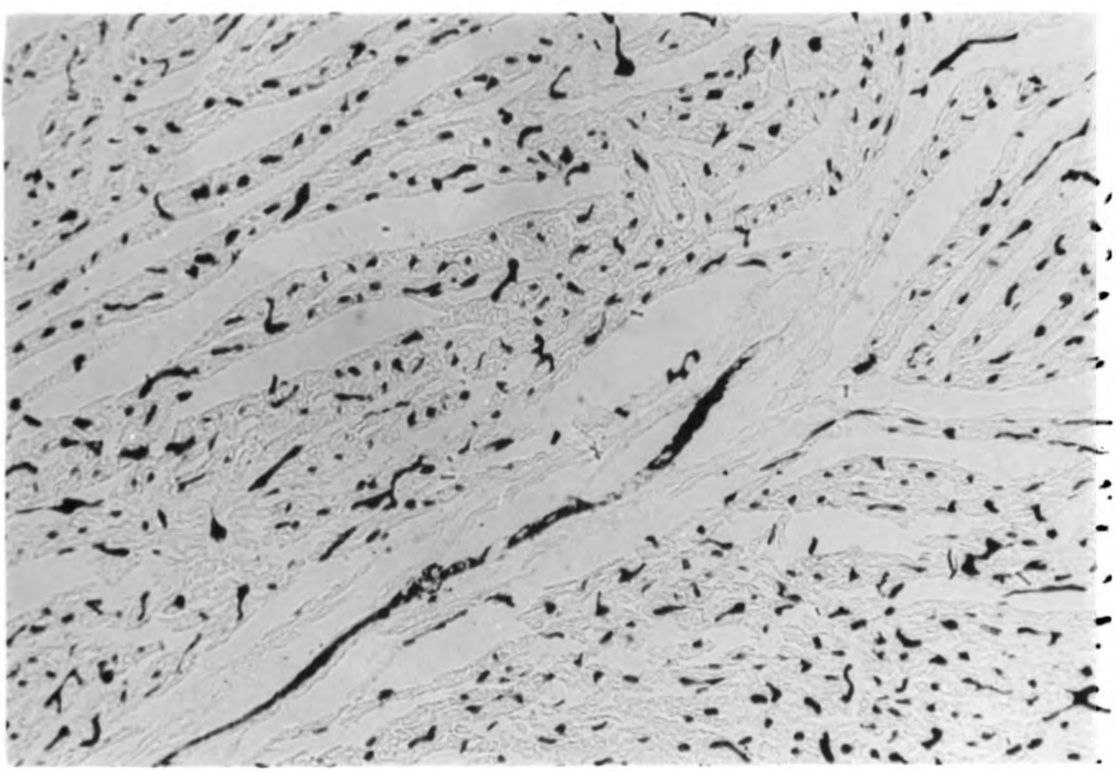


Figure 7. 3 week-old heart - Cross-section of capillary bed. X175

Figure 8. 7 week-old heart - Cross-section of capillary bed. X175.

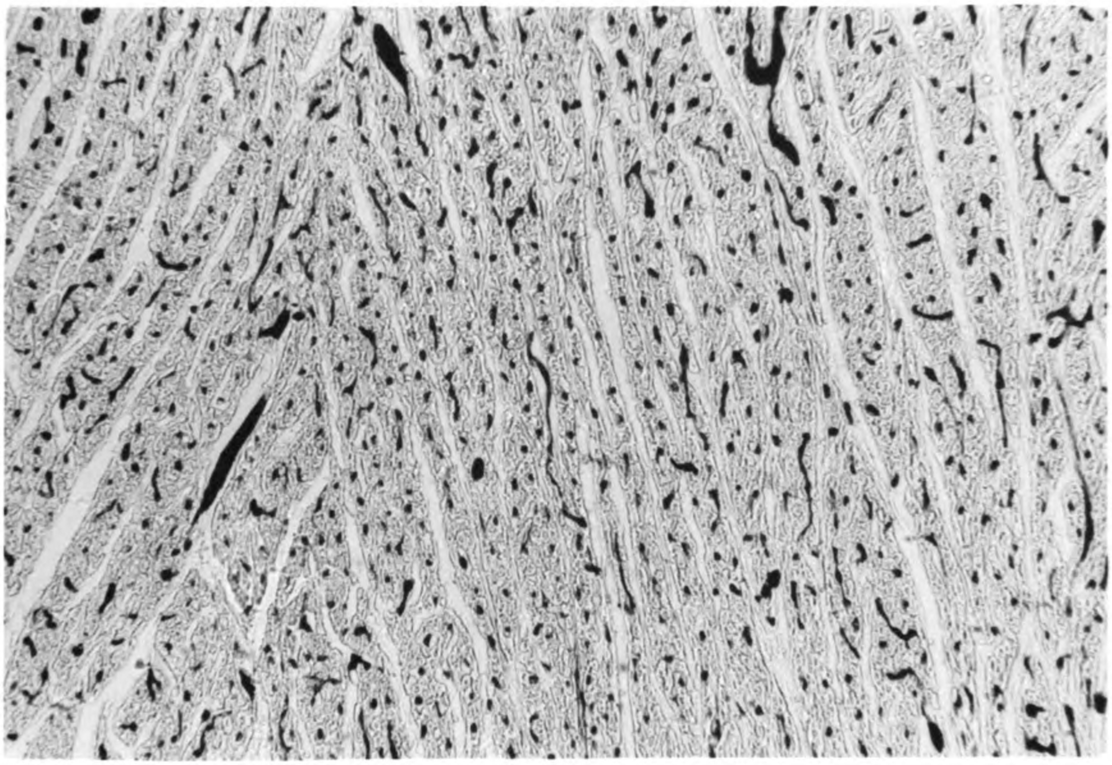
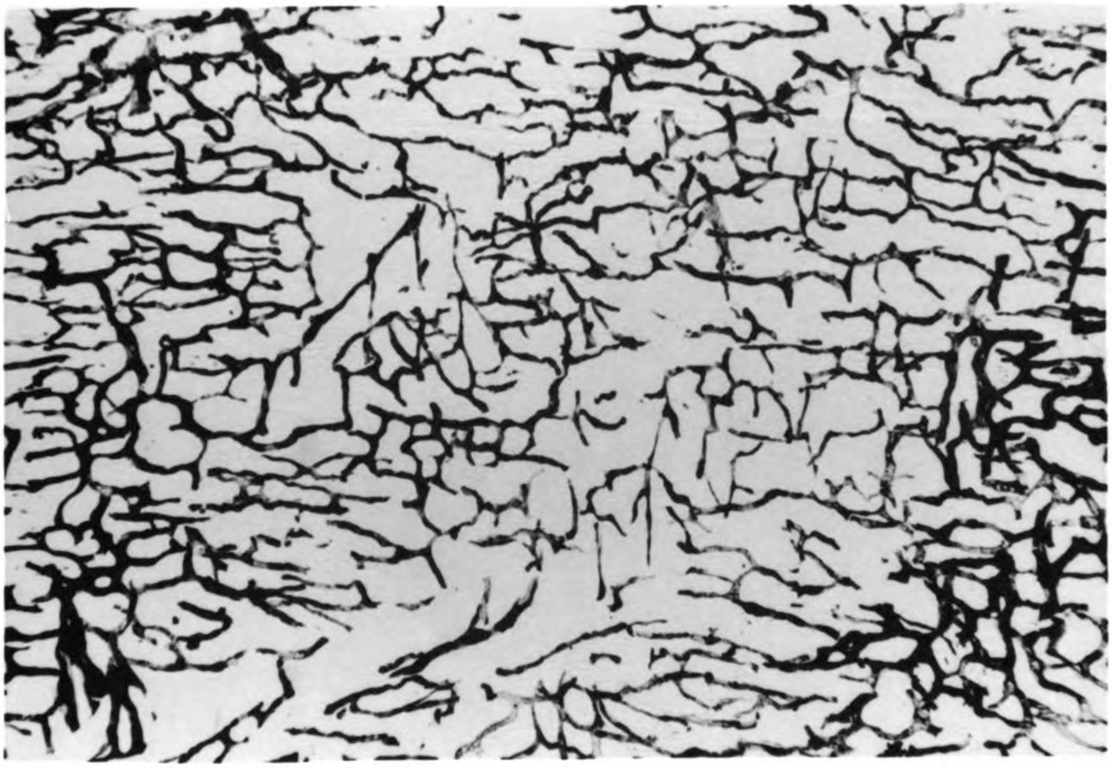


Figure 9. 12 hour-old heart - Vascular arcade peculiar to the interventricular septum. X175.

Figure 10. 12 hour-old heart - Meshwork of capillary sinusoids. Compare with Figures 40, 41. X175.



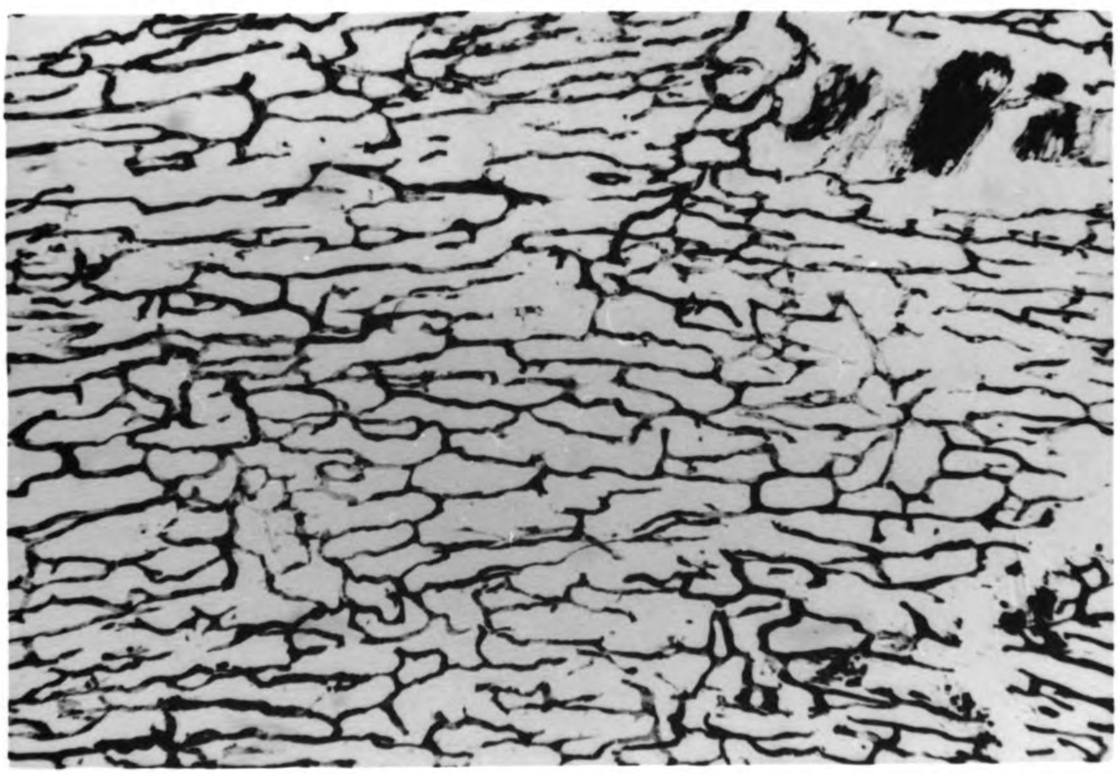
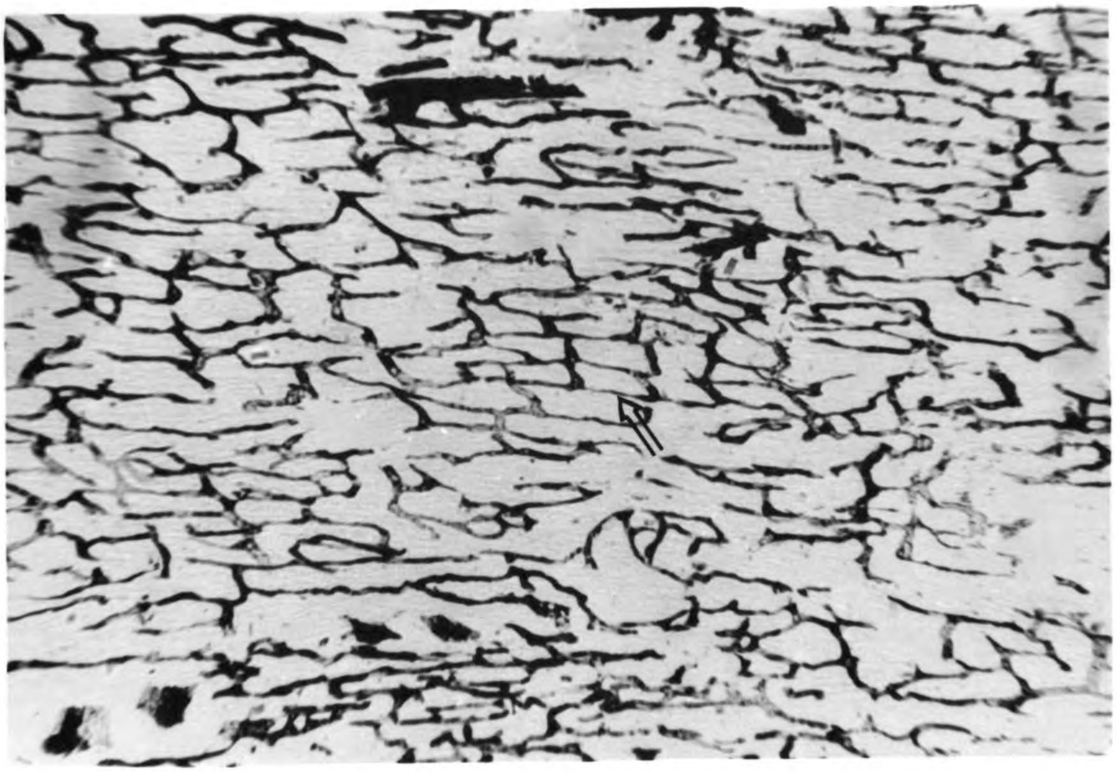


Figure 11. 12 hour-old heart - Meshwork of capillary sinusoids. Note regularity of anastomotic loops at arrow. X175.

Figure 12. 12 hour-old heart - Meshwork of capillary sinusoids. Note distinguishable arteriole at arrow. X175.



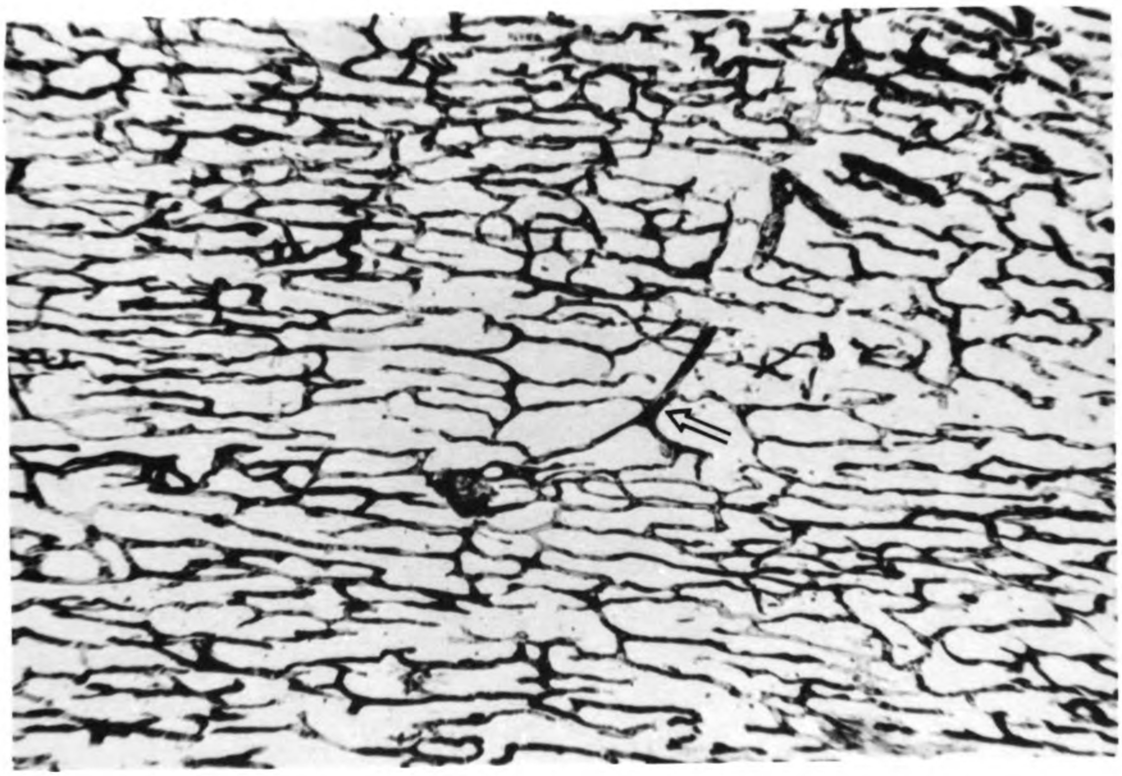
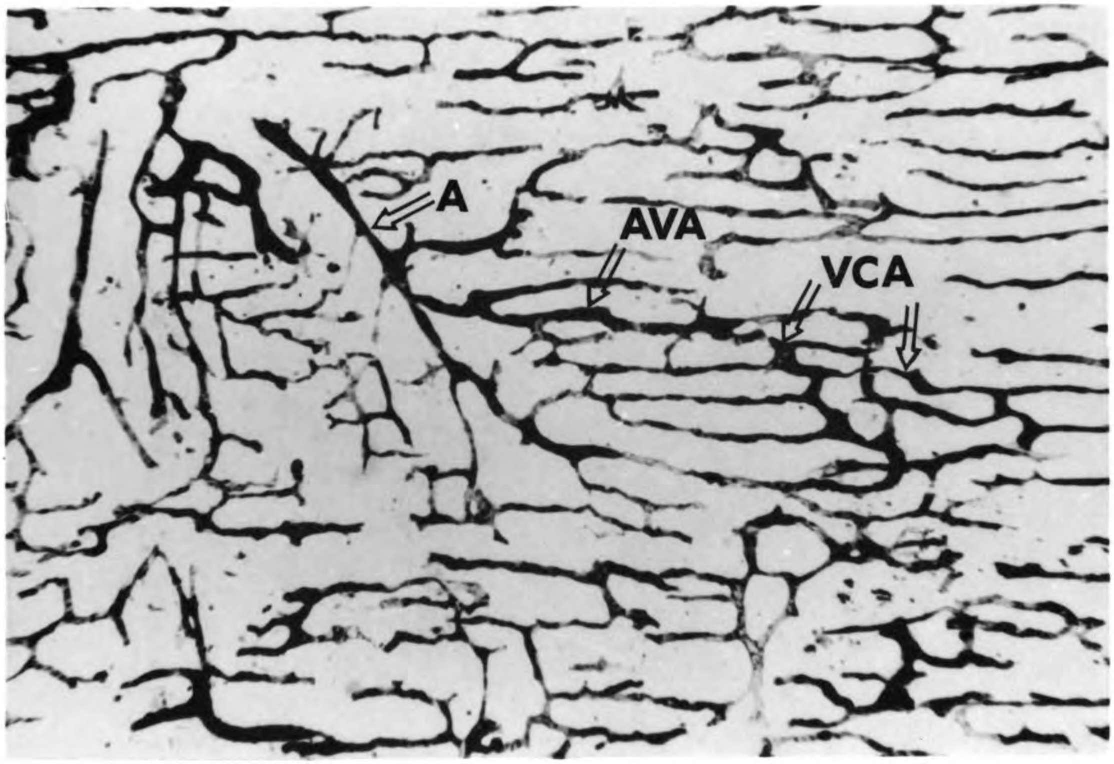


Figure 13. 1 week-old heart - Terminal vascular bed. Note sinusoidal nature of venous collection area. X175.
A - Arteriole
VCA - Venous Collection Area
AVA - Arteriovenous Anastomosis

Figure 14. 1 week-old heart - Terminal vascular bed. Note the short length and sinusoidal nature of the capillaries. X175.
A - Arteriole
VCA - Venous Collection Area


A
AVA
VCA

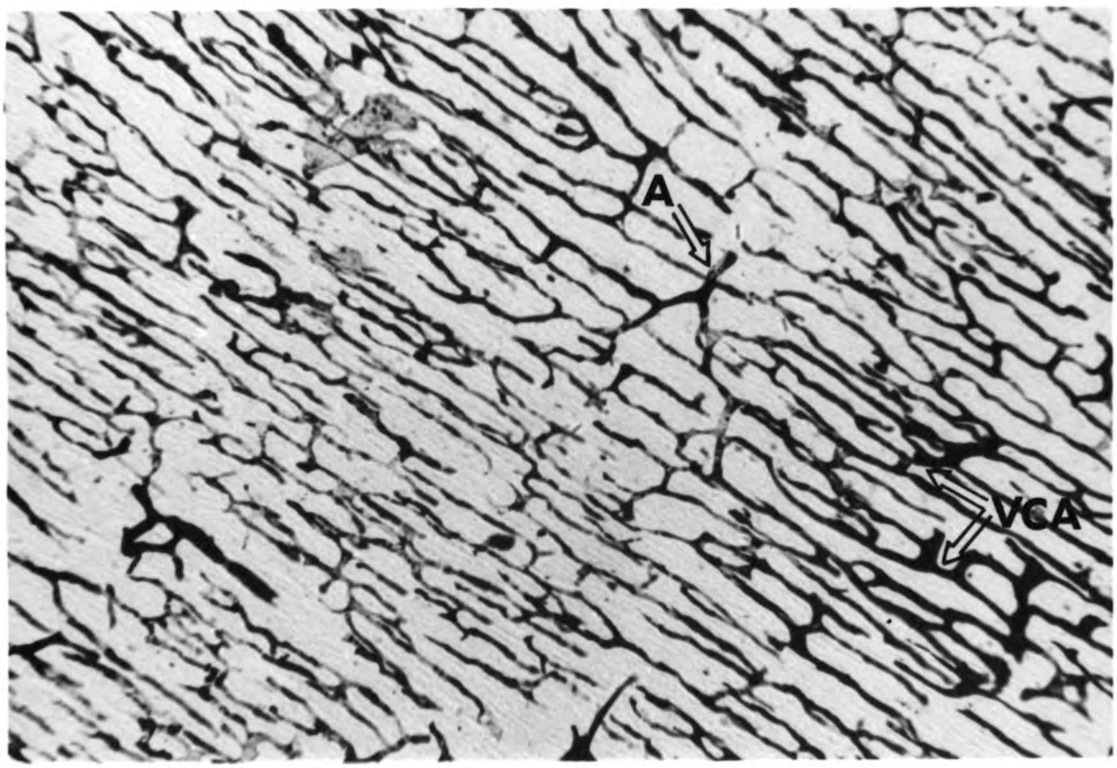

A
VCA

Figure 15. 1 week-old heart - Terminal vascular bed. Note multiple venous collection areas for single arteriole. X175.

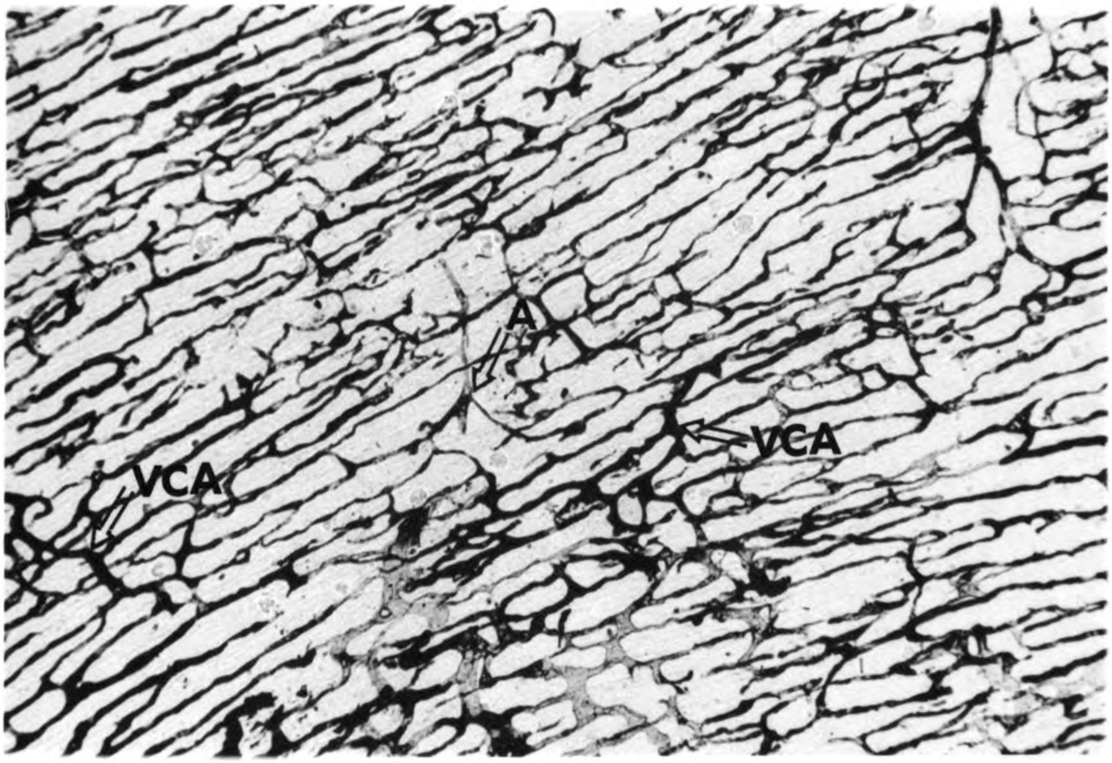
A - Arteriole

VCA - Venous Collection Area

Figure 16. 1 week-old heart - Terminal vascular bed. Note multiple arteriole supplies to single venous collection. X175.

A - Arteriole

VCA - Venous Collection Area


A
VCA
VCA

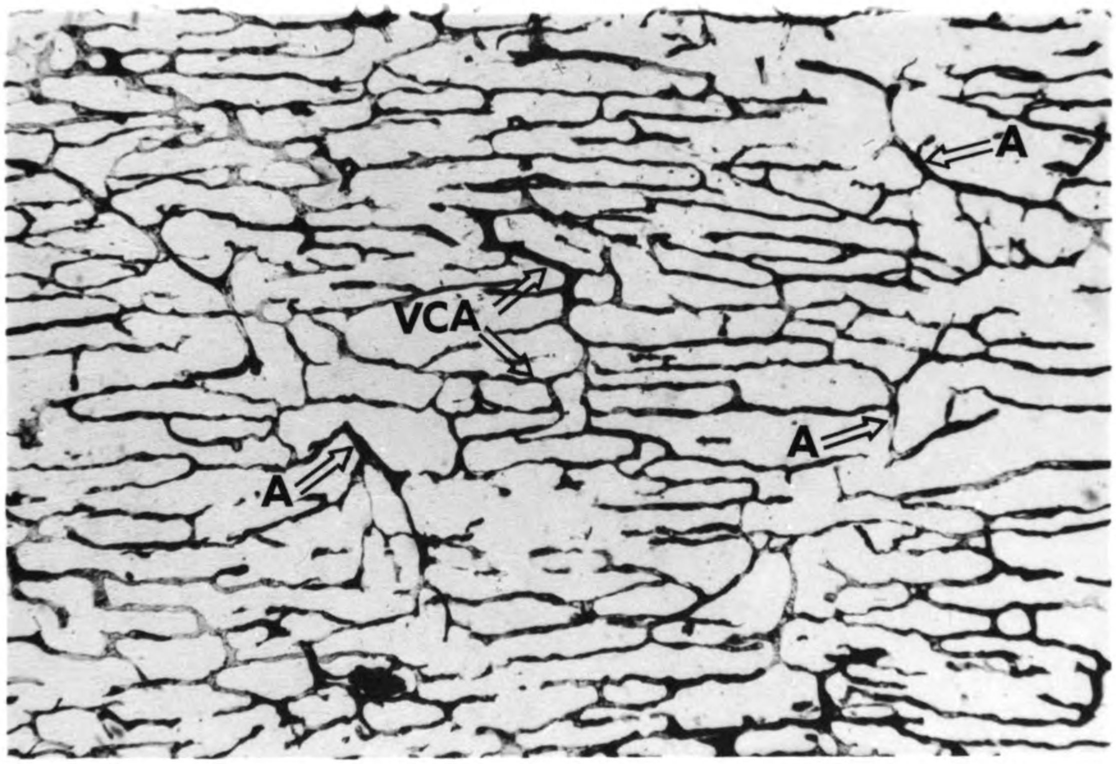
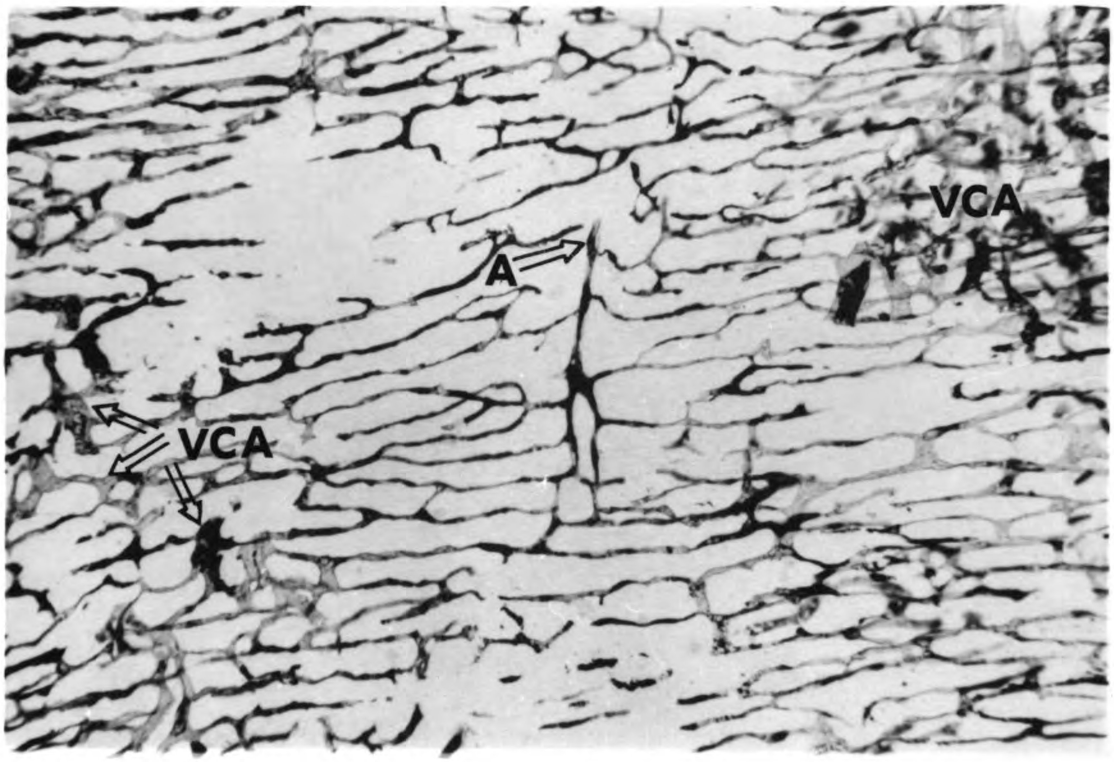

A
VCA
A
A

Figure 17. 2 week-old heart - Terminal vascular bed. Note that the capillary loops are becoming confined to the venous collection areas. X175.
A - Arteriole
VCA - Venous Collection Area

Figure 18. 2 week-old heart - Terminal vascular bed. Note sinusoidal nature of the expansive venous collection areas. X175.
A - Arteriole
VCA - Venous Collection Area


VCA
A
VCA

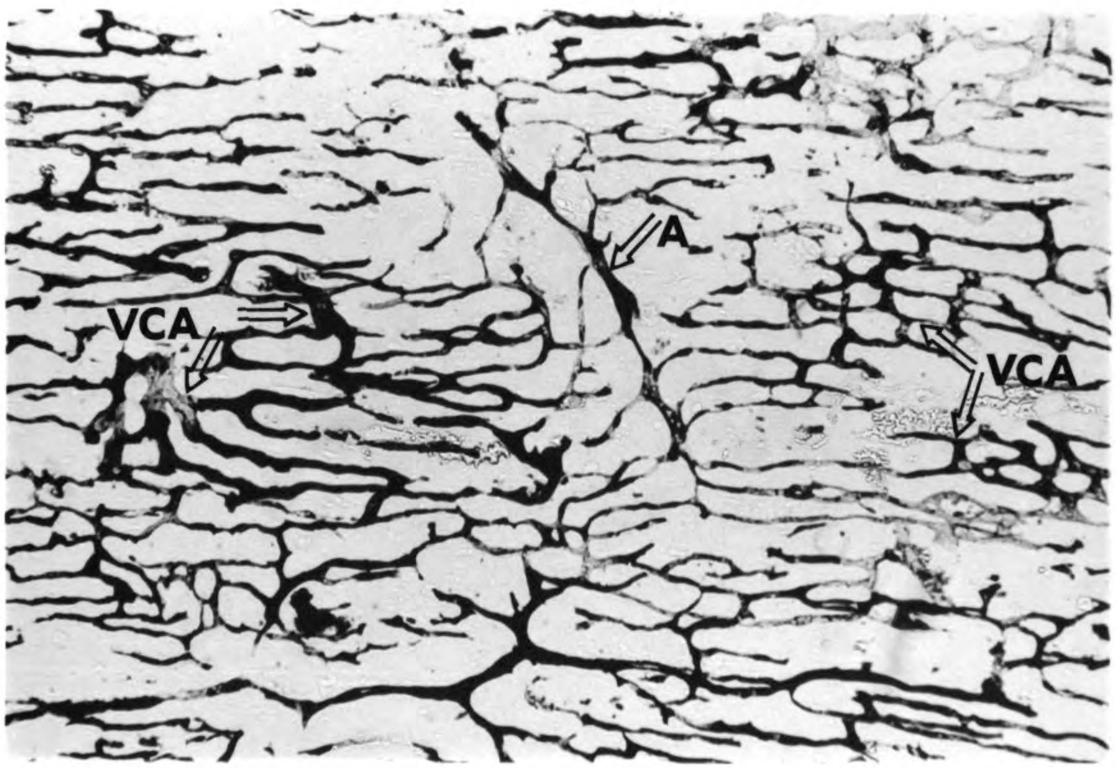
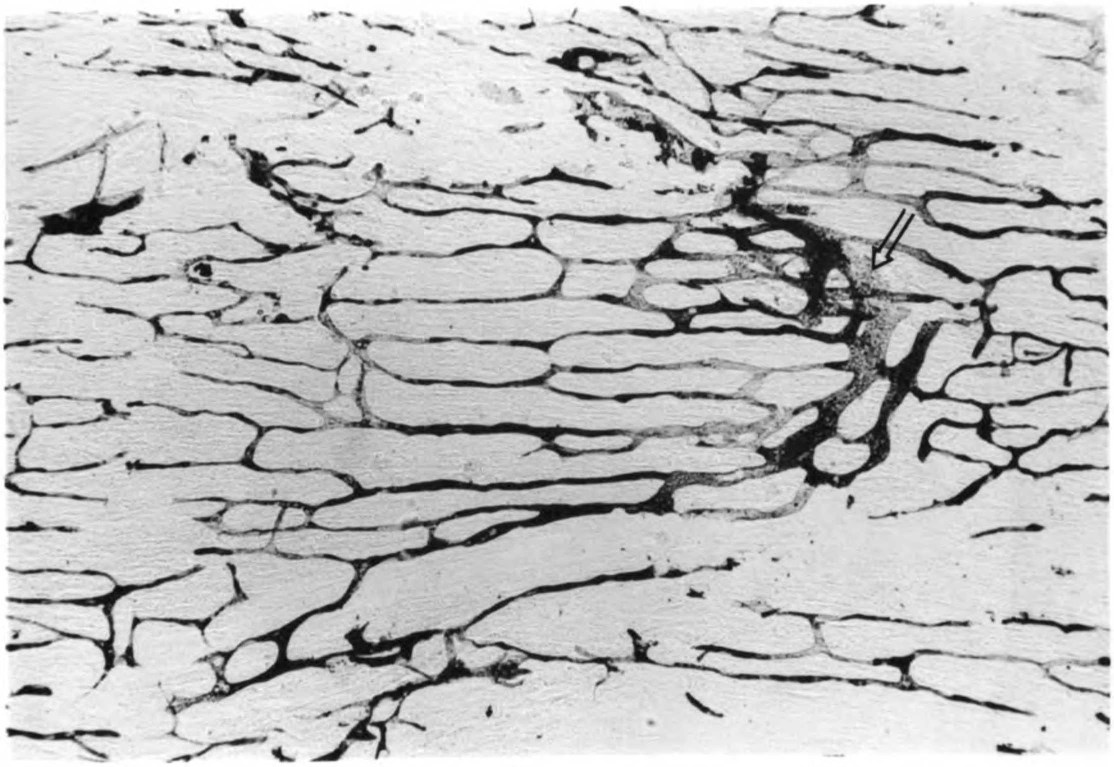

A
VCA
VCA

Figure 19. 2 week-old heart - Venous collection area at arrow. Note the anastomosing nature of the area. X175.

Figure 20. 2 week-old heart - Junction of ventricular wall and septum. Note the extreme meshwork of sinusoidal capillaries. X175.



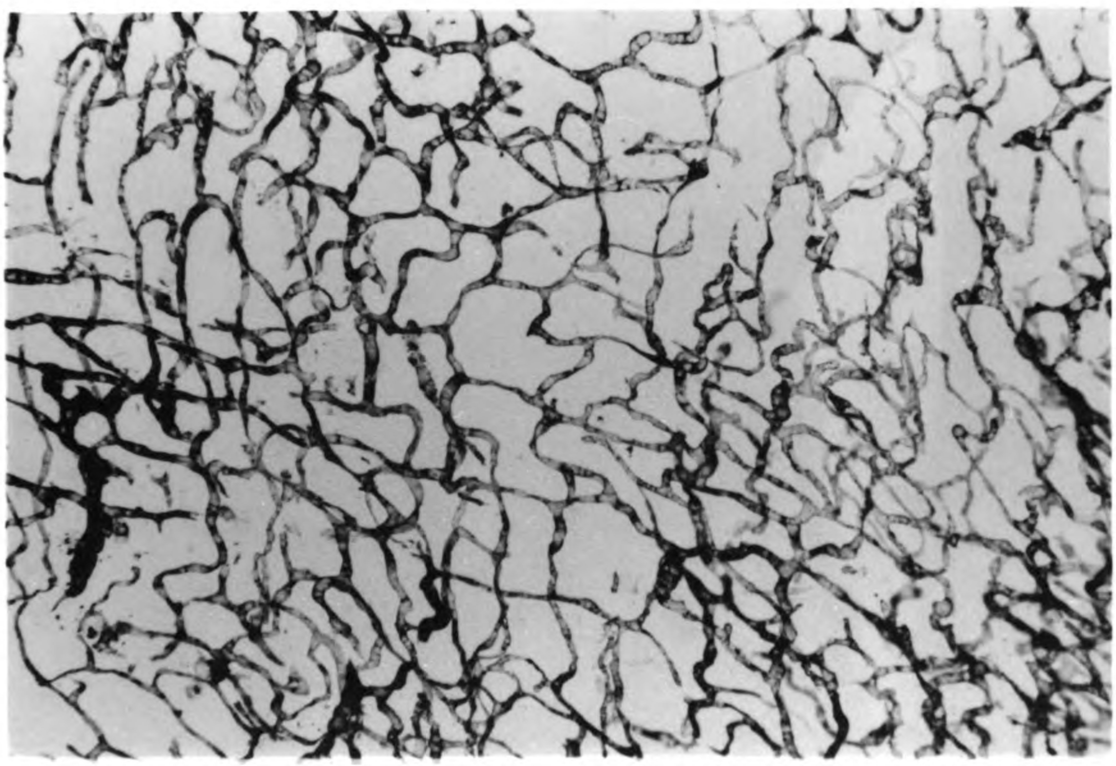


Figure 21. 3 week-old heart - Terminal vascular bed. Note the increased length of the capillaries and the decrease in anastomotic loops. X175.
A - Arteriole
V - Vein

Figure 22. 3 week-old heart - Terminal vascular bed. Note that the capillaries are losing their sinusoidal nature. (Compare with Figure 11). X175.
A - Arteriole
VCA - Venous Collection Area


A
V

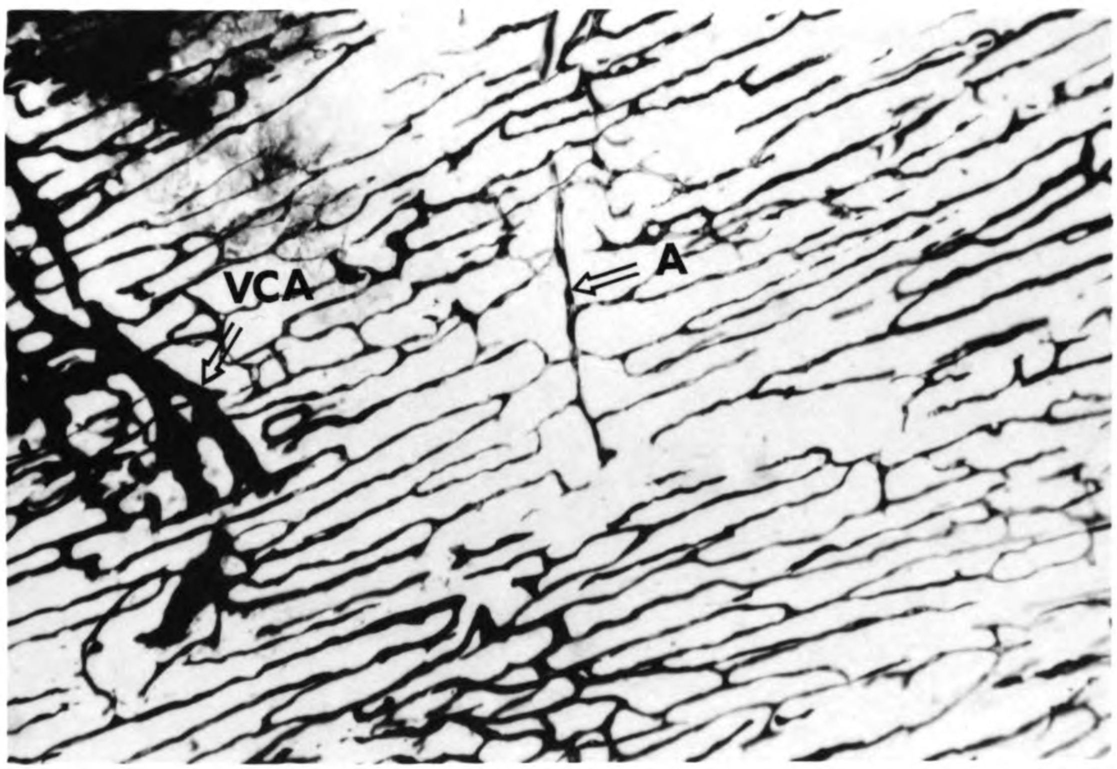
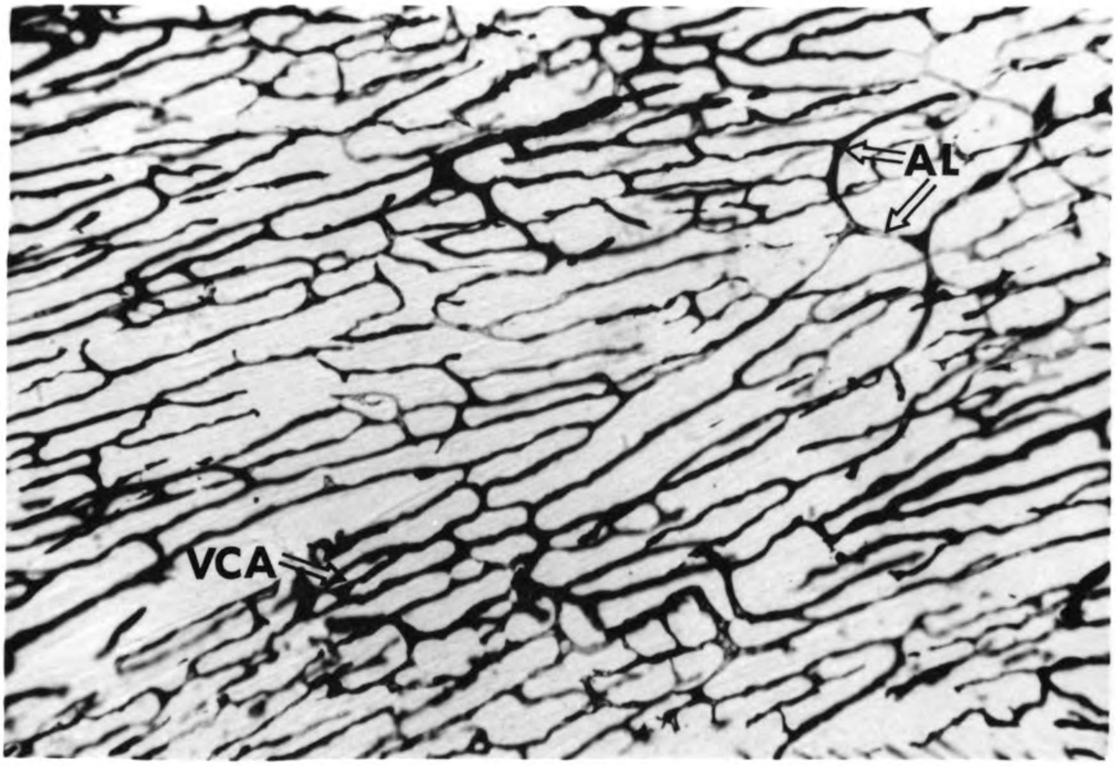

VCA
A

Figure 23. 3 week-old heart - Terminal vascular bed. Note the arteriole loop with arteriole radiations. X175.
AL - Arteriole Loop
VCA - Venous Collection Area

Figure 24. 3 week-old heart - Terminal vascular bed. Note the short length and anastomotic looping of the capillaries. X175.
A - Arteriole
VCA - Venous Collection Area


AL
VCA

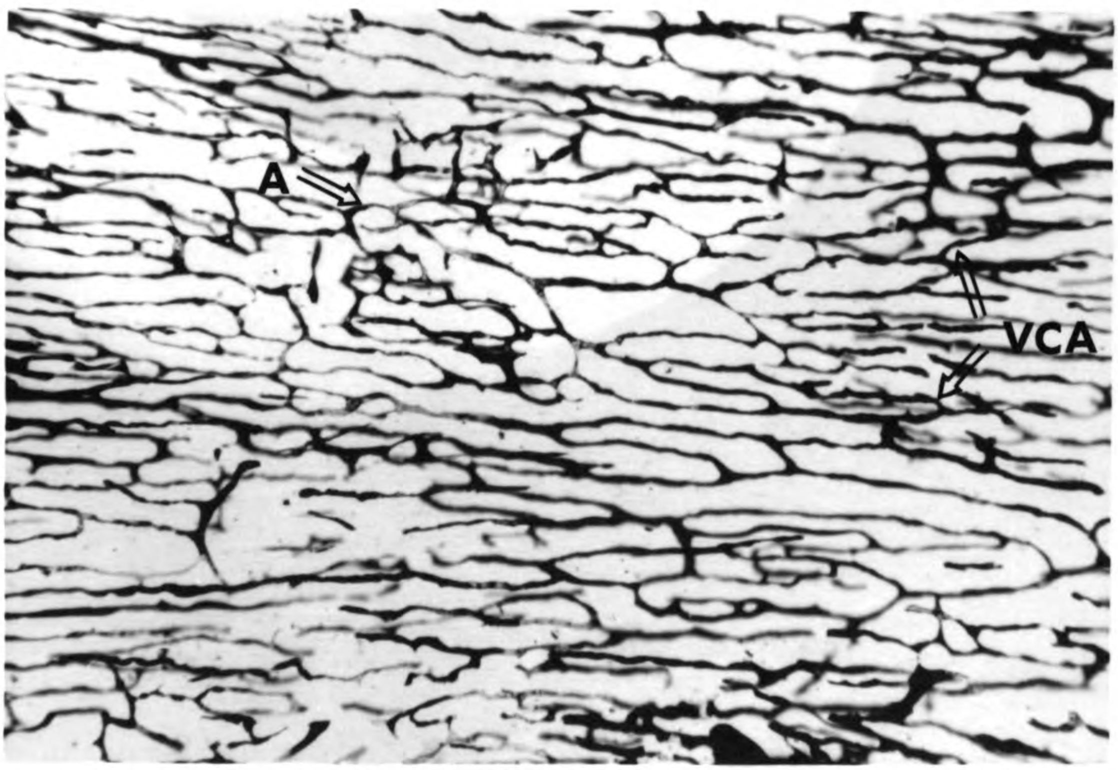
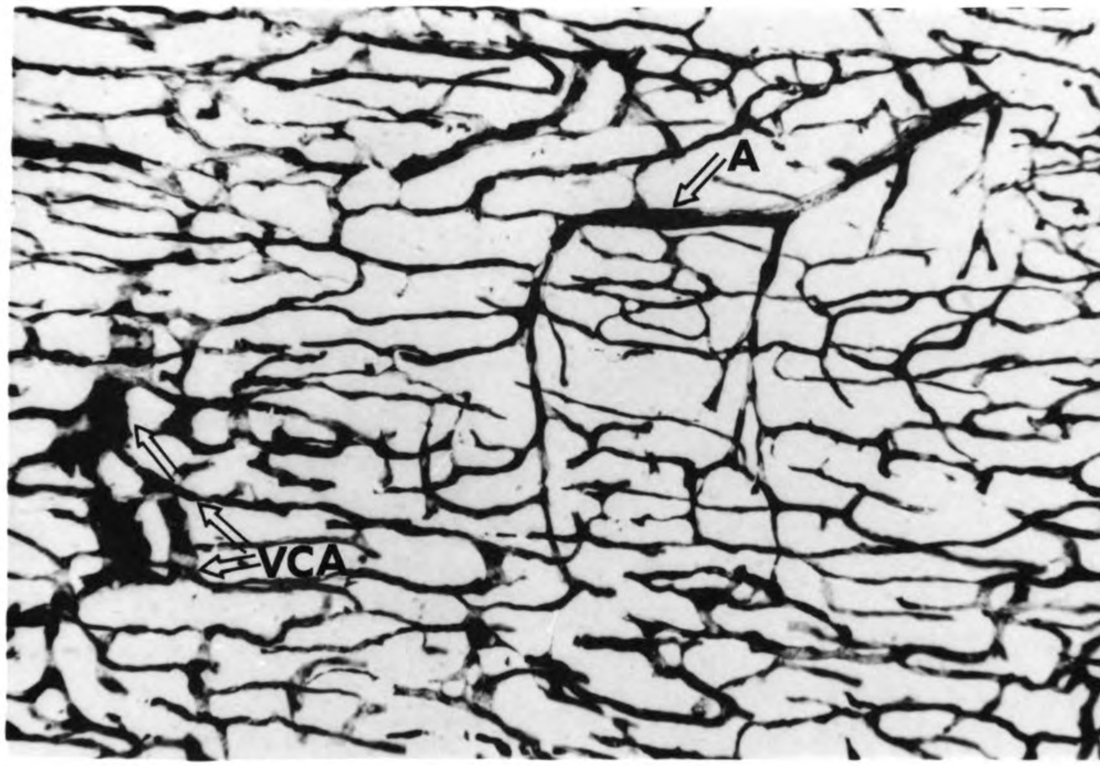

A
VCA

Figure 25. 4 week-old heart - Vascular arcade peculiar to the interventricular septum. Note the proximity of the parallel daughter arterioles. X175.
A - Arteriole
VCA - Venous Collection Area

Figure 26. 4 week-old heart - Junction of the ventricular wall and septum. Note the extreme meshwork of the capillaries. X175.


A
VCA

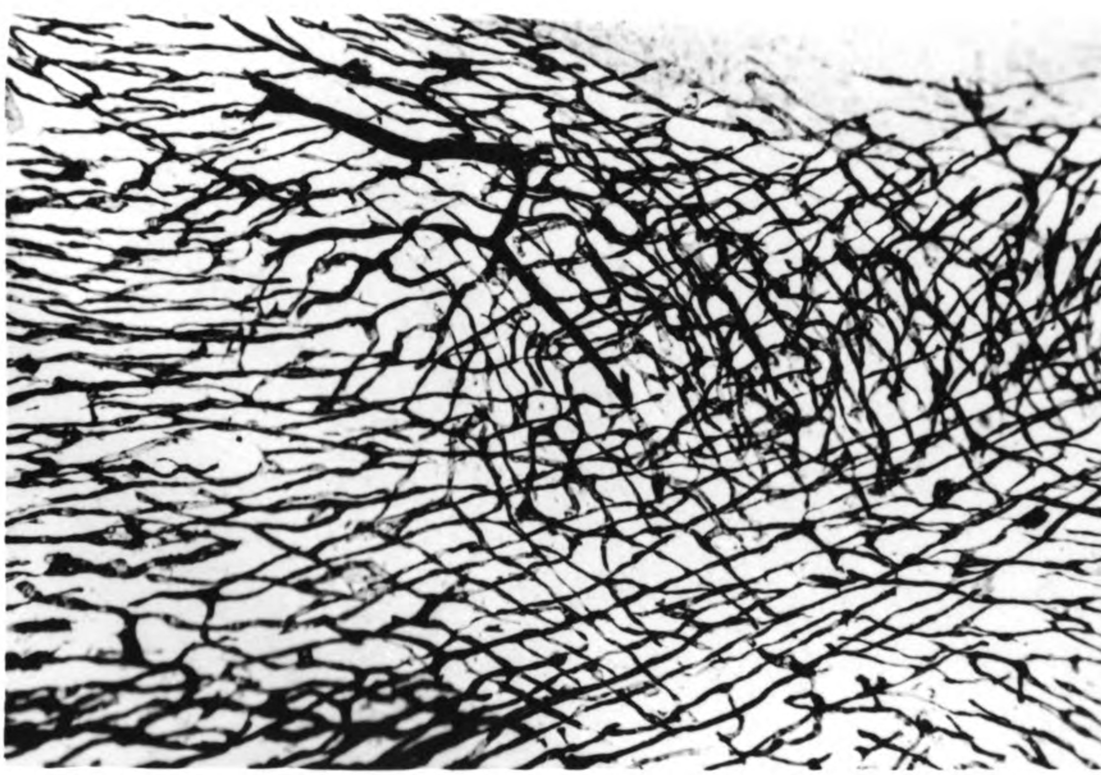


Figure 27. 4 week-old heart - Terminal vascular bed. Note the presence of both a long and a short capillary bed. X175.

AR - Artery
V - Vein
A - Arteriole
VCA - Venous Collection Area
LB - Long Capillary Bed
SB - Short Capillary Bed

Figure 28. 4 week-old heart - Terminal vascular bed. Note the drainage of the short capillaries directly into the large vein. X175.

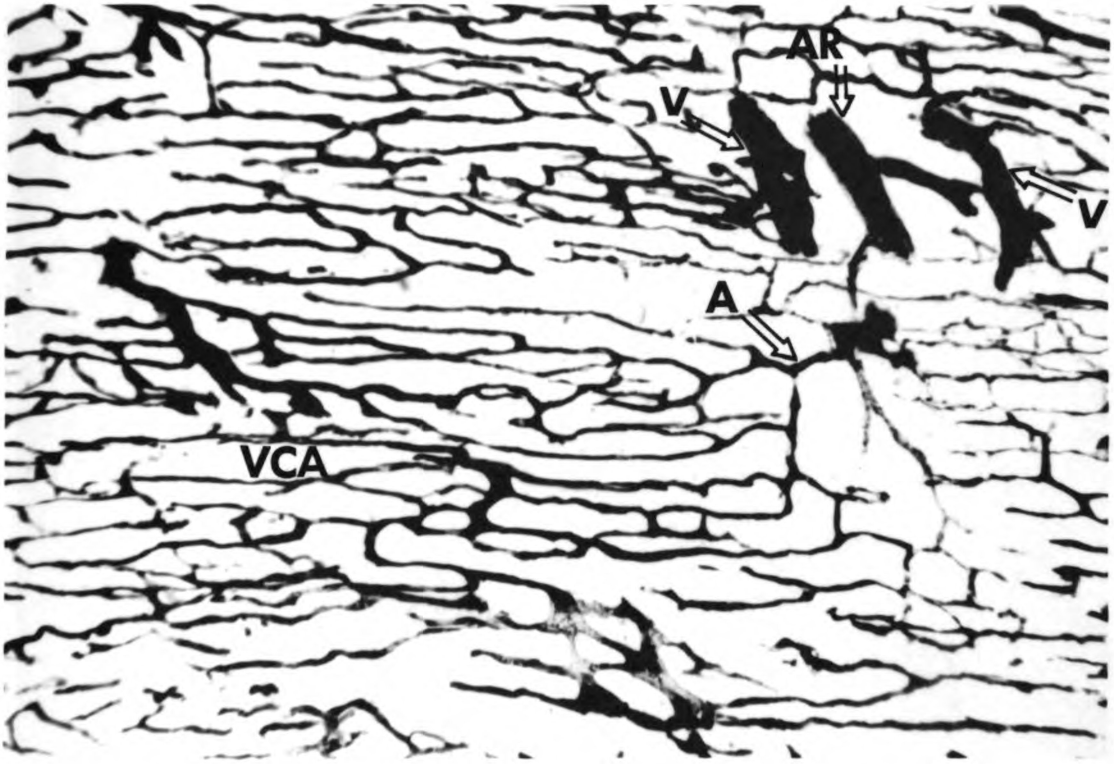
A - Arteriole
V - Vein


AR
V
V
V
A
SB
LB
VCA


V
A

Figure 29. 4 week-old heart - Terminal vascular bed. Note the arteriole from the artery of the vein-artery-vein triad. X175.
AR - Artery
V - Vein
A - Arteriole
VCA - Venous Collection Area

Figure 30. 4 week-old heart - Terminal vascular bed. Note the expansive, anastomotic nature of the venous collection area. X175.
A - Arteriole
V - Vein
VCA - Venous Collection Area


AR
V
V
A
VCA

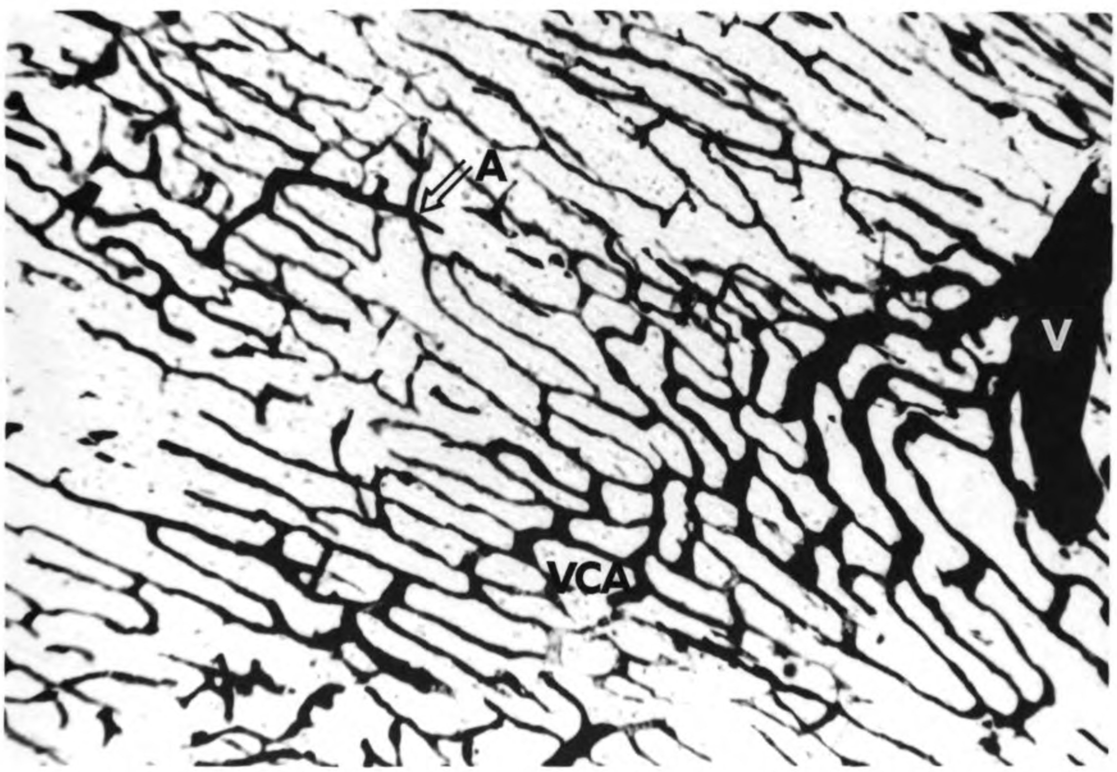
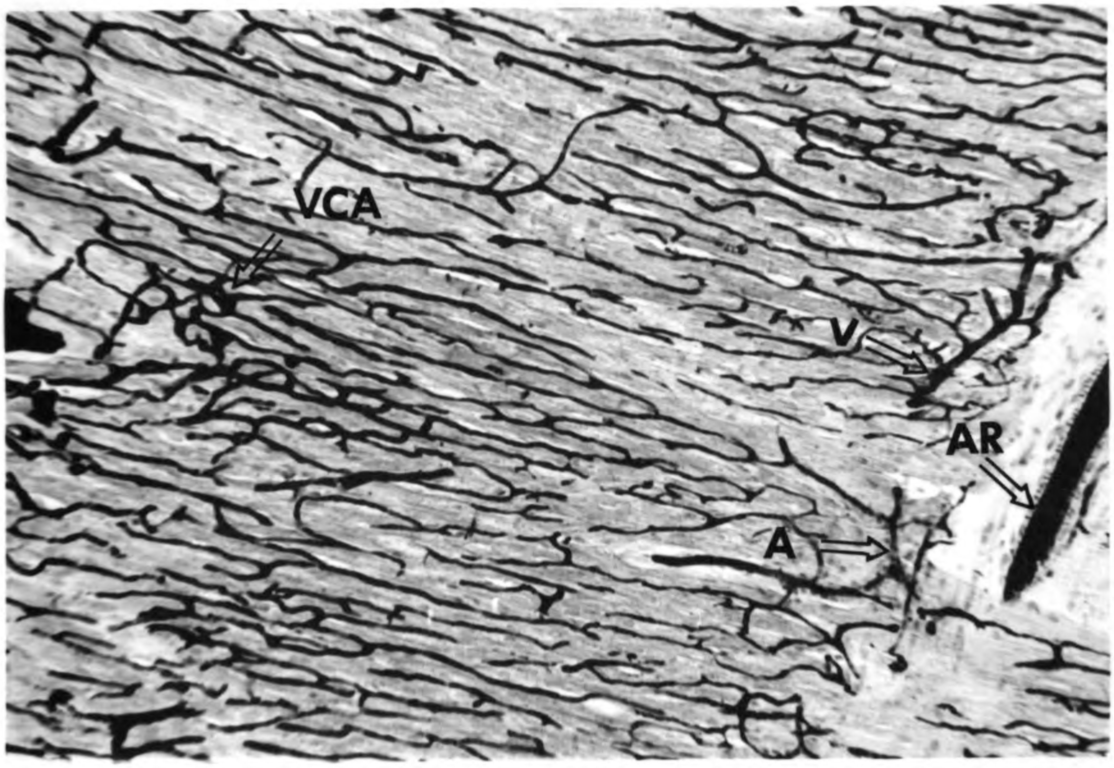

A
V
VCA

Figure 31. 5 week-old heart - Terminal vascular bed. Hematoxylin-Eosin stain. X175.
AR - Artery
V - Vein
A - Arteriole
VCA - Venous Collection Area

Figure 32. 5 week-old heart - High power of Figure 31. Note the layer of muscle tissue around the arteriole and the lack of muscle tissue around the vein. Hematoxylin-Eosin stain. X340.
A - Arteriole
V - Vein
M - Muscle Wall of Arteriole


VCA
V
AR
A

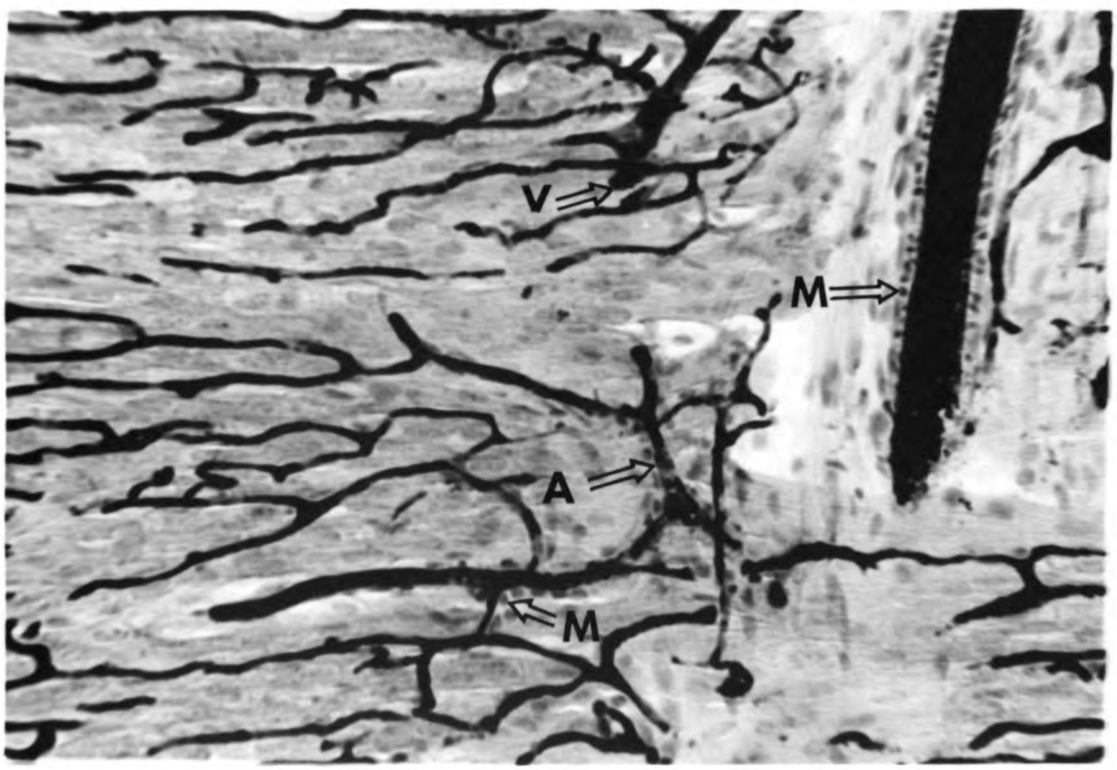
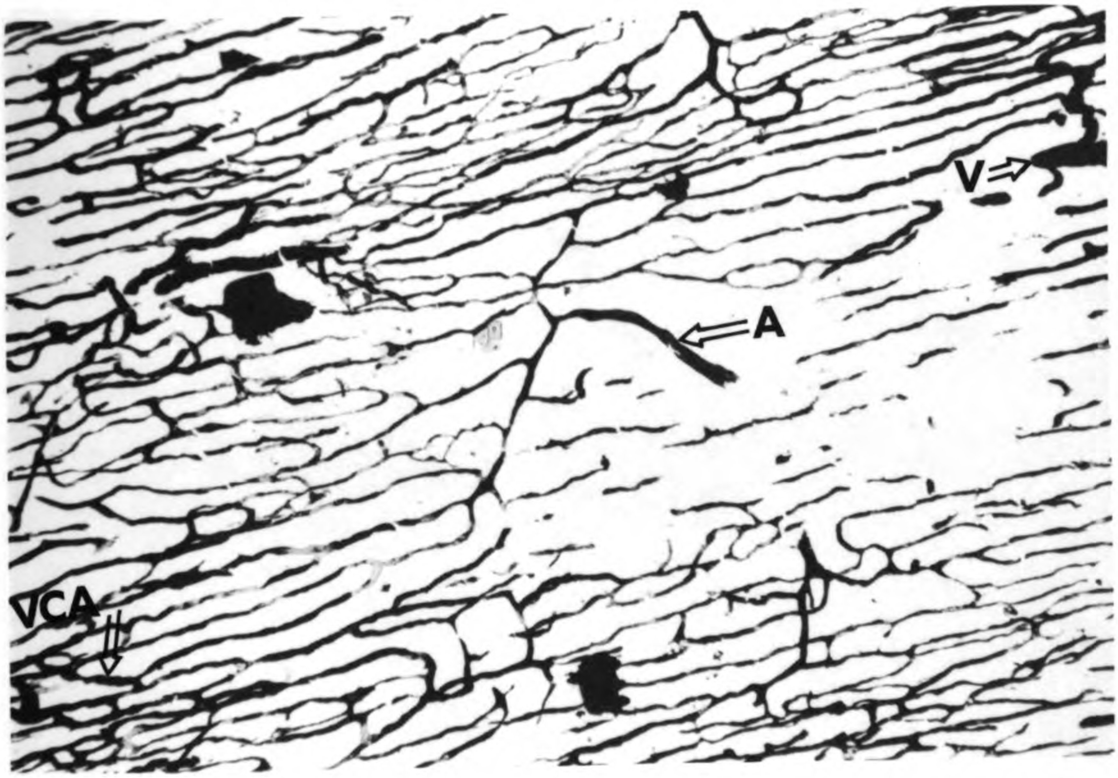

V
M
A
M

Figure 33. 5 week-old heart - Terminal vascular bed. Note the increased length of the capillary and the nearly complete absence of the sinusoidal nature seen previously. X175.

A - Arteriole
V - Vein
VCA - Venous Collection Area

Figure 34. 5 week-old heart - Terminal vascular bed. Note the increased distance between the arteriole and vein. X175.

A - Arteriole
V - Vein


V
A
VCA

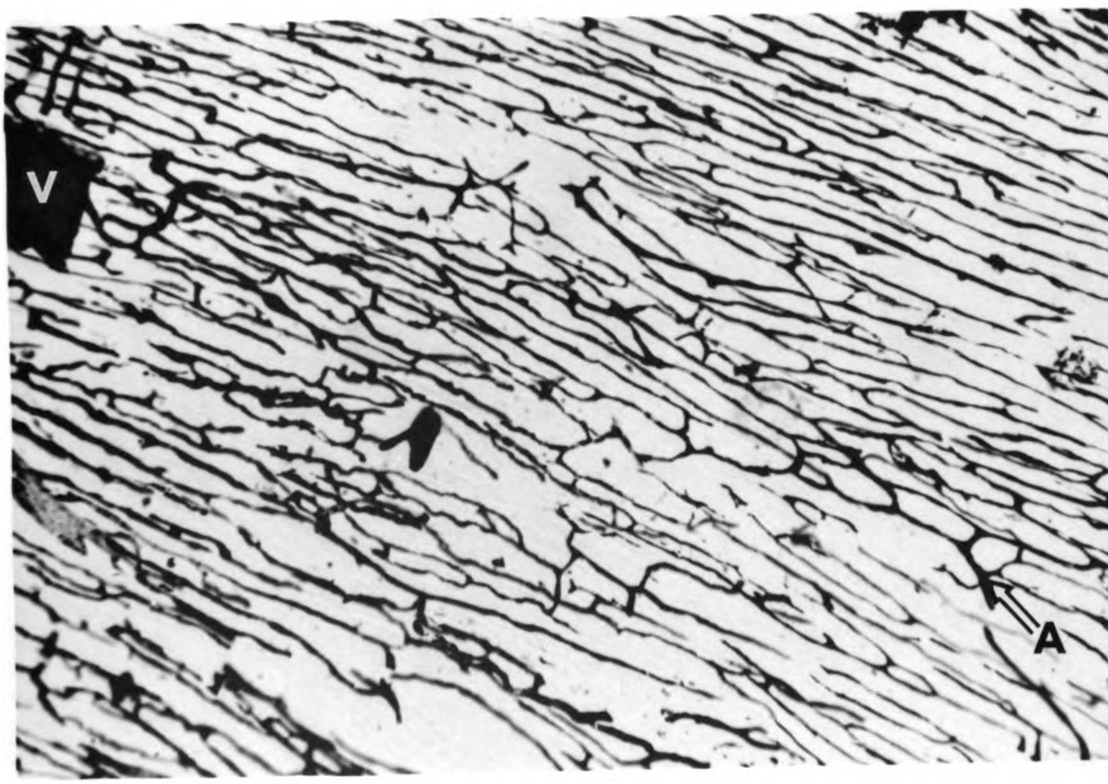
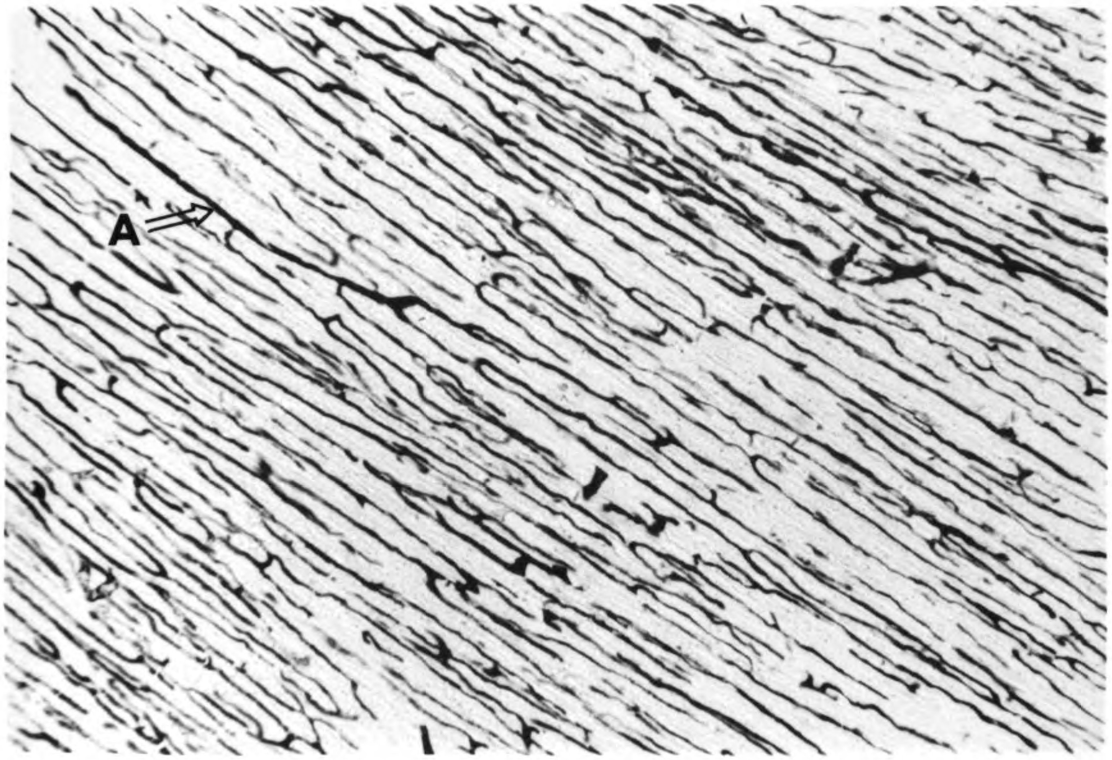

V
A

Figure 35. 6 week-old heart - Terminal arteriole. Note the delicate capillary ramifications. X175.
A - Arteriole

Figure 36. 6 week-old heart - Terminal vascular bed. Note the absence of distinct anastomotic loops in the venous collection area. X175.
A - Arteriole
VCA - Venous Collection Area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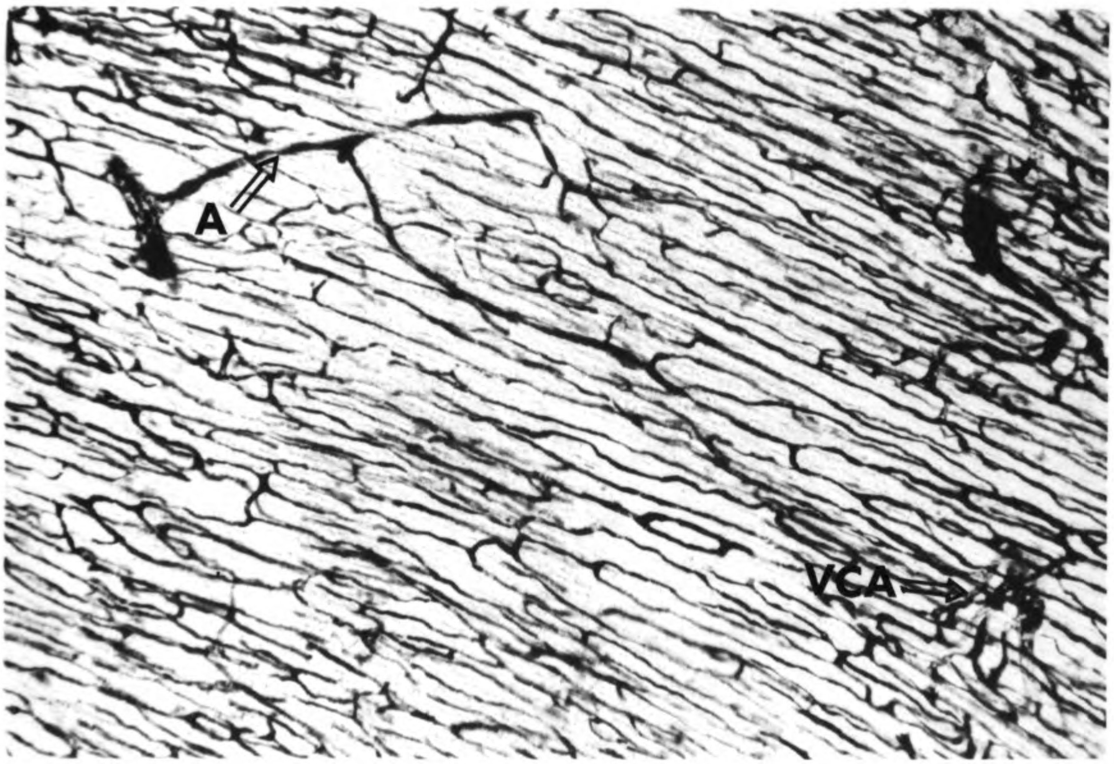
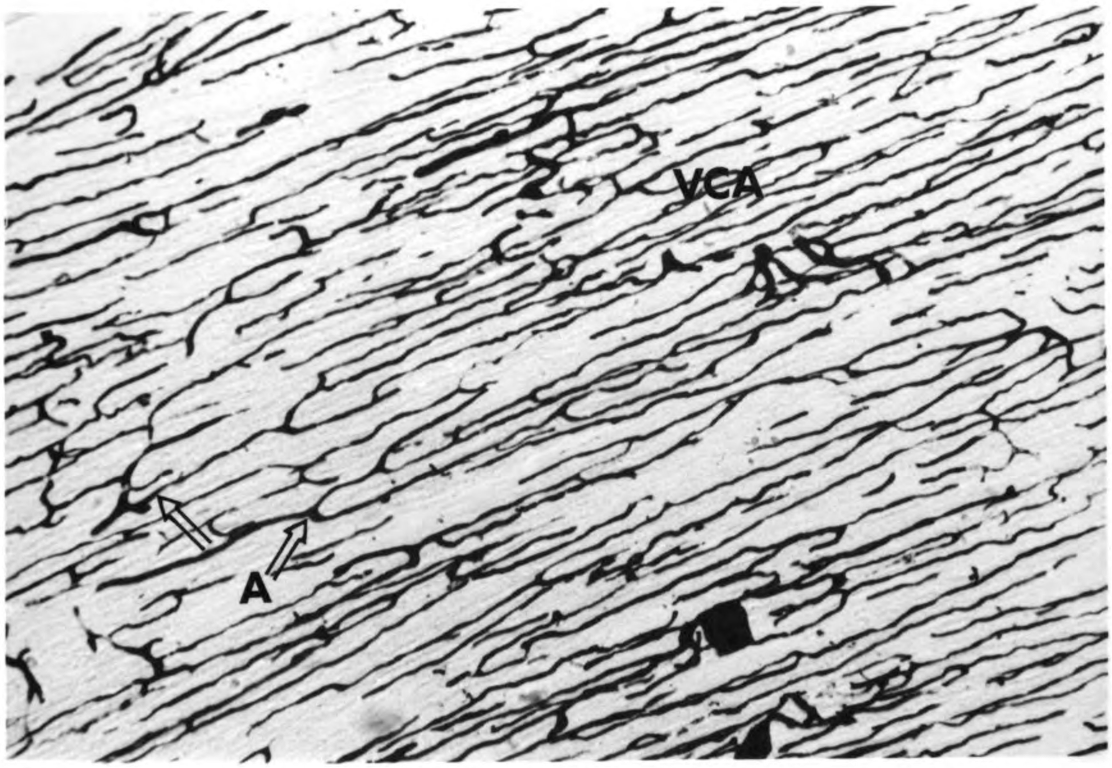

A
VCA

Figure 37. 6 week-old heart - Terminal vascular bed. Note the narrow areas of venous collection when compared with Figure 17. X175.
A - Arteriole
VCA - Venous Collection Area

Figure 38. 6 week-old heart - Terminal vascular bed. Note the 2 arterioles drained by a single narrow venous collection. X175.
A - Arteriole
VCA - Venous Collection Area


VCA
A

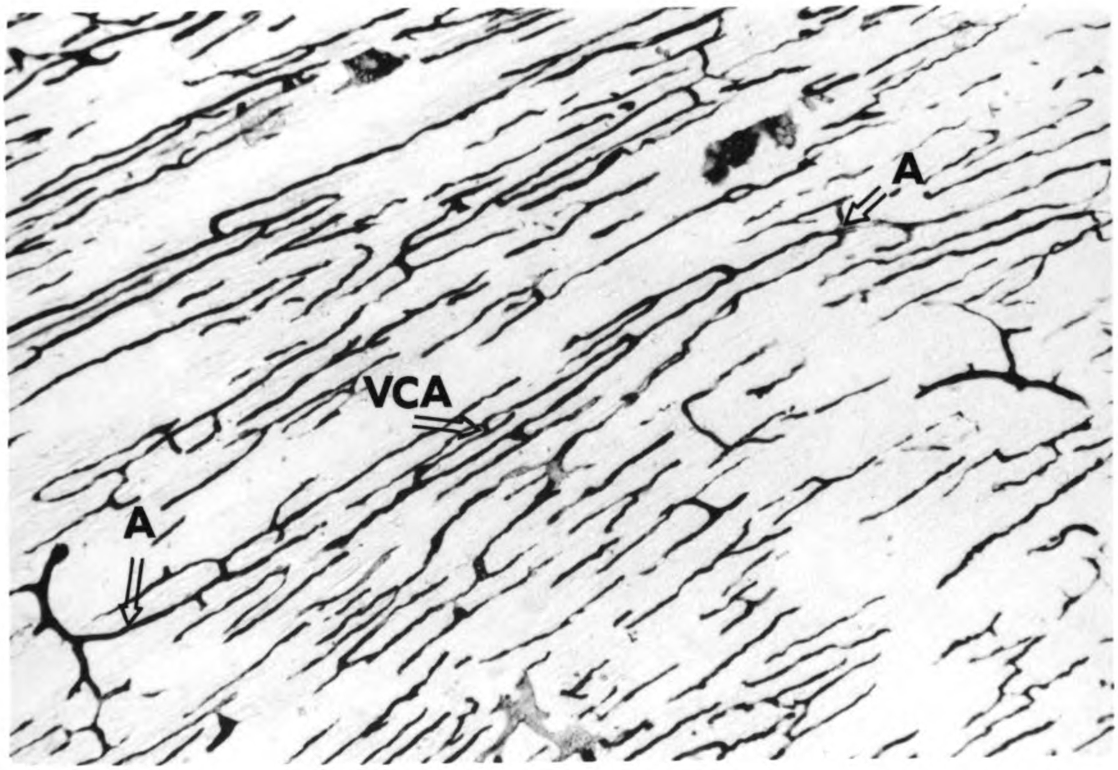

A
VCA
A

Figure 39. 7 week-old heart - Vascular arcade peculiar to the septum. Note the diverse ramifications of the terminal arteriole. X175.
A - Arteriole
V - Vein
VCA - Venous Collection Area

Figure 40. 7 week-old heart - Terminal vascular bed. Note the "turnip root" vein. X175.
A - Arteriole
TRV - Turnip Root Vein


VCA
A
V

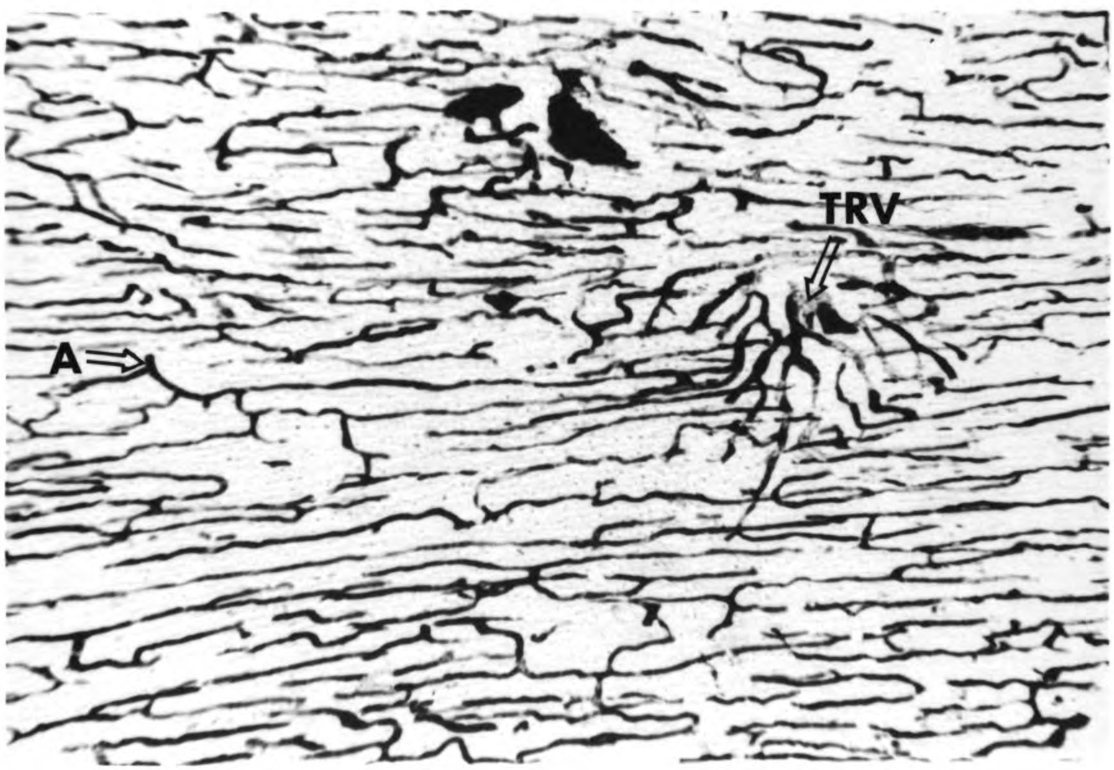
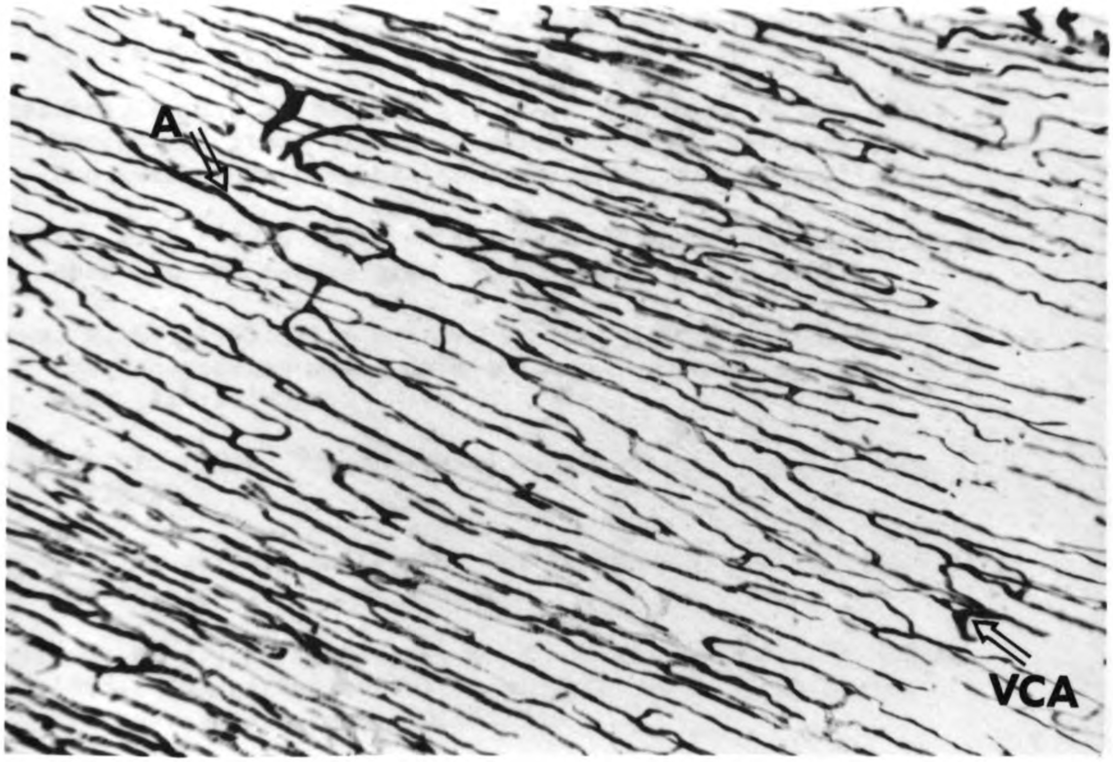
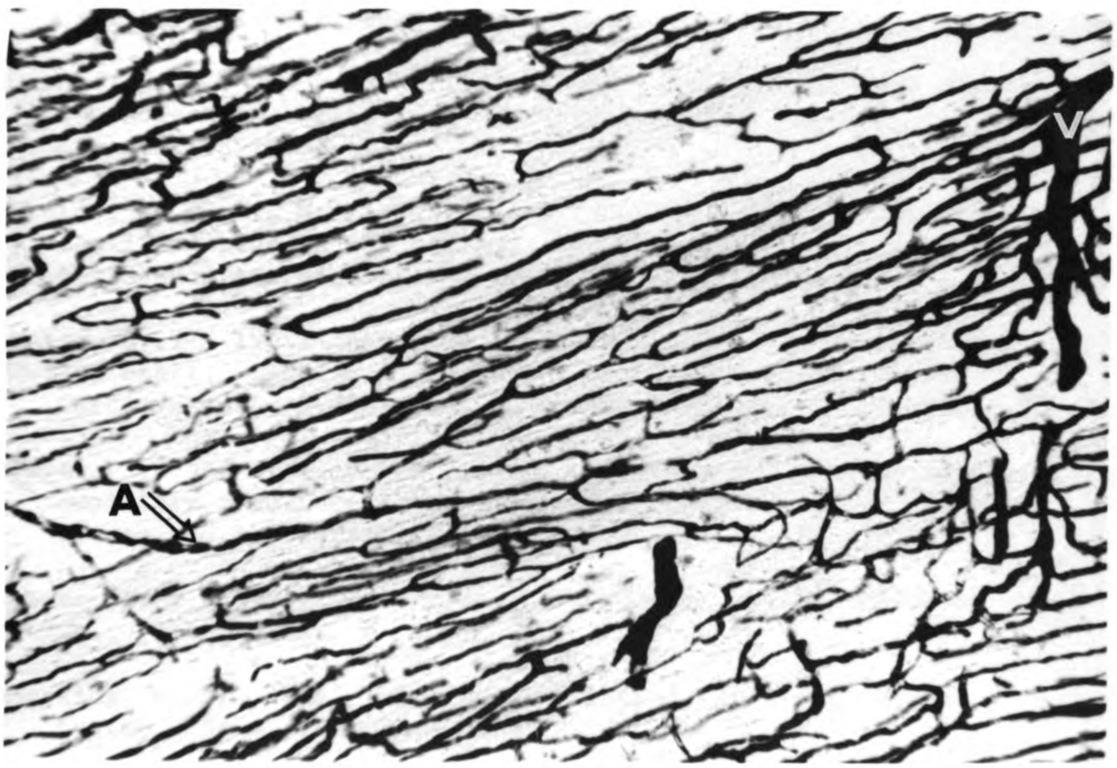

TRV
A

Figure 41. 7 week-old heart - Terminal vascular bed. Note the lack of sinusoidal capillaries and capillary loops and the increased distance between the arteriole and venous collection. X175.
A - Arteriole
VCA - Venous Collection Area

Figure 42. 7 week-old heart - Terminal vascular bed. Note the lack of sinusoidal capillaries and capillary loops and the narrow area of venous collection. X175.
A - Arteriole
V - Vein


A
VCA


V
A

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