A Study of the Paracapillary Nutrition Canals and their Possible Sympathetic Innervation.

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In a previous study of intravascular injections with colloidal solutions (3) the author attempted to show that the dispersion of these solutions is not solely confined to the vessels generally classified as belonging to the circulatory system. The intravascular injection with crystalline solution proved useless inasmuch as these diffused through the vessel walls and tissue membranes.

In order to demonstrate that tissue structures lying beyond the circulatory system are stained by colloidal solutions, the author's experimental procedure was as follows:

1. The animal is bled to death under ether narcosis.
2. The exsanguinated animal is immediately removed to an incubator heated to 37° C.
3. The residual blood in the blood vessels is washed out with saline to which 0.2 per cent sodium nitrate has been added.
4. The colloidal solution is injected intravascularly.
5. If the colloidal solution consists of gelatin, the animal is immersed in cold saline towards the end of the experiment. If the solution consists of India ink, the latter if precipitated by an acid fixation liquid, as soon as the injection is terminated.

The following injection solutions are used: gelatin and India ink; gelatin alone; India ink alone; trypan blue in 1 percent saline; 1 percent trypan blue prepared like India ink; gelatin and staphylococci; a sediment of staphylococci prepared like India ink; a sediment of staphylococci stained with methylene blue. All the injection substances are made homogeneous and free from sediment.

The principal experimental results are the following:

1. After injecting gelatin into the common carotid artery, the dental enamel in the dog was found to have become resistant to acid; com-

plete decalcification which otherwise takes place within 24 hours of ordinary teeth, was now prolonged to two weeks. It was observed, however, that the enamel in the injected tooth is not completely destroyed during the process of decalcification, which is otherwise the case when the enamel is exposed in the usual way to the influence of acid under atmospheric pressure. The enamel stroma in the gelatin injected tooth remains as a transparent cap over the dentine of the dental crown after decalcification. It is well-known that both the dentine and the enamel lack blood vessels. Bearing in mind the current opinion concerning the extent of the circulatory system, the results already mentioned must be considered as remarkable.

2. In the omentum of the rabbit it was observed that the finely grained injection substances did not throughout fill the lumen of the blood vessels. On the other hand, a certain part of the capillary wall, namely the argyrophile adventitia — *das Grundhäutchen* — was often clearly stained in a characteristic fashion which suggest the effect obtained by silver impregnation of the capillaries. The interior endothelial part of the capillary wall did not, however, appear to be stained by the injection substance.

3. The finely grained injection substances did not only stain the vessel walls in the rabbit's omentum, but also the connective tissue fibers between the vessels and the fairly large number of the tissue cells, the histiocytes.

In seeking an interpretation of these observed facts, the author found a valid explanation difficult, unless he takes into account the fact that the circulatory system does not solely consist of blood-carrying vessels commonly called arteries, veins and capillaries, but also a system of smaller vessels — *ultracapillaries* — through which the *blood plasma* is transported to all the parts of the tissue. According to the author's experience, he is inclined to believe that these ultracapillaries are identical with the reticular fibers, also called argyrophile fibers, *"Gitterfasern"*, or precollagenous fibers. In order to throw more light on this subject, the author has carried on a series of experiments which will be accounted for further in the text.

One tissue is probably more than any other suitable for the study of paracapillary nutrition, namely the *dental enamel*, which until quite recently was generally considered completely devoid of nutrition. The principal organic substance of the enamel consists of a system of reticular fibers. In fact, the enamel seems to be exclusively made up of reticular fibers, apart from *nerve fibers* and *nerve endings*, which shall be described later. Thus the enamel is the tissue where the argyrophile threads appear as it were to be produced in pure

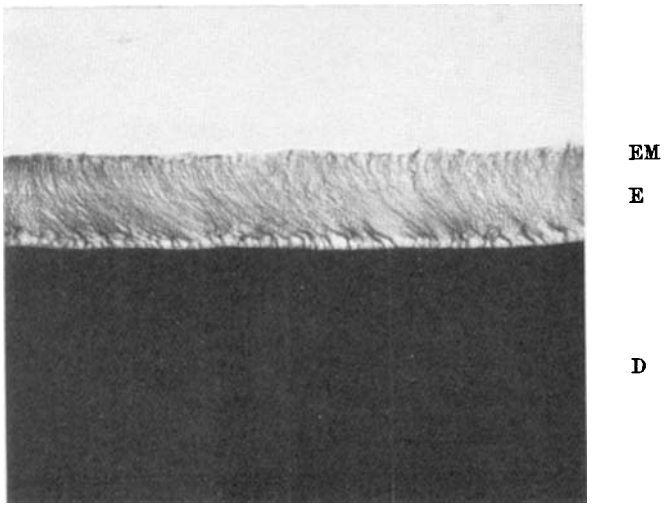

Fig. 1. Section from the incisor of a dog. The direction of the section is perpendicular to the longitudinal axis of the dental crown. Fixation: formalin. Decalcification under pressure. Embedding: celloidin. Staining: anilin blue. Magnification × 120. E. Enamel fibers. D. Dentine. EM. Enamel cuticle. The dento-enamel juntion membrane does not appear in this staining in contrast to the dentine.

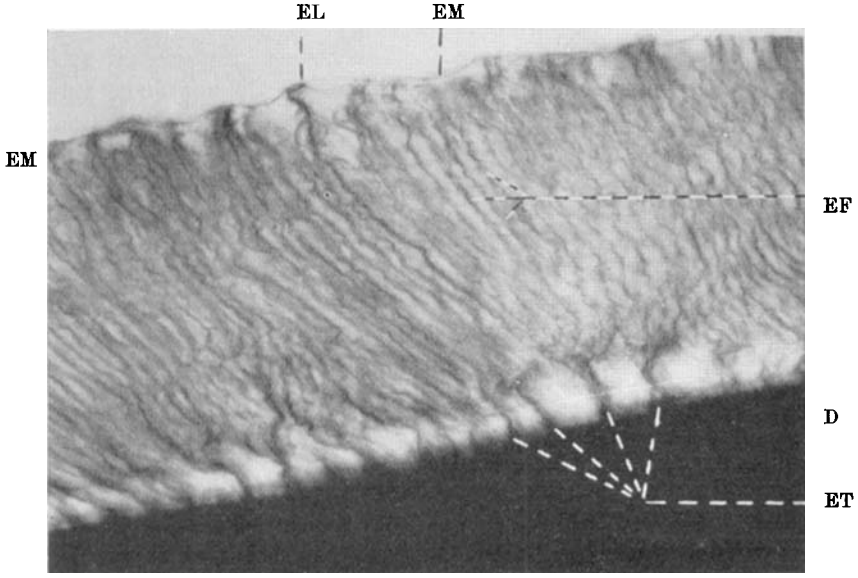

Fig. 2. Higher magnification, × 470, from Fig. 1. EM. Enamel cuticle. ET. Enamel tufts. EL. Enamel lamella. EF. Enamel fibers, and D. Dentine.

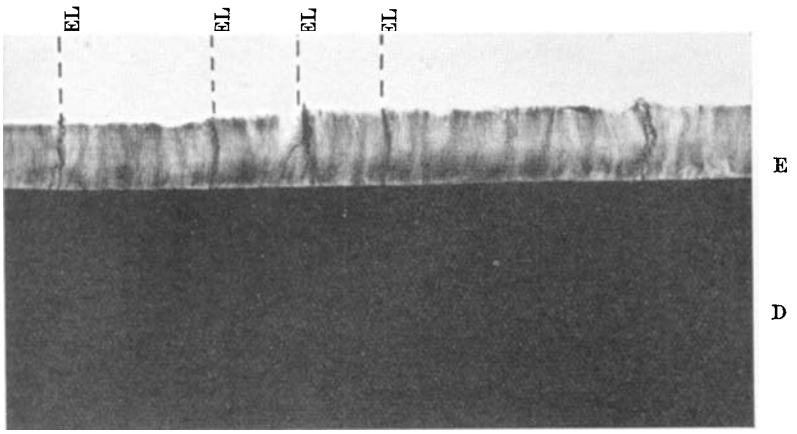

Fig. 3. Section from the incisor of a dog. The direction of the section is parallel to the longitudinal axis of the dental crown. Technique and magnification same in Fig. 1. EL. Enamel lamella. E. Enamel stroma, and D. Dentine.

culture. It is separated from the nearest blood vessels — in the dental pulp — by the dentine which is devoid of any blood vessels. The enamel being the hardest and least porous of any tissue in the body, is stained with greatest difficulty *in vitro*. Thus there is no risk that the injection susbstance might penetrate through vessel ruptures or other vessel injuries into the tissue and adhere to tissue elements which do not form nutrition canals. If, by injecting reliable colloidal dyes into a main artery, it is possible to stain the dental enamel in certain definite parts, then there can hardly be any doubt that these lines contain ramifications from the blood vessels, finer than the capillaries, and that these ultracapillaries are composed of reticular fibers.

The reticular fibers of the dental enamel are organized into a system whose structure is typical for the enamel (vide Figs. 1 and 2). Next to the dentine border there is an argyrophile, border membrane, the dento-enamel junction membrane. From this membrane, tufts of »threads» — the so-called *enamel tufts* — radiate into the enamel with even intervals. From the enamel tufts, as well as the from the dental-enamel junction membrane, run finer »threads», with constant intervals and mutually parallel, which finally are transformed into a border membrane on the enamel surface, the so-called enamel cuticle. This was once supposed, as was likewise the case with the enamel as whole, to be of epithelial nature and hence is called in the literature by the name *enamel cuticle*. Between these above mentioned »threads», which

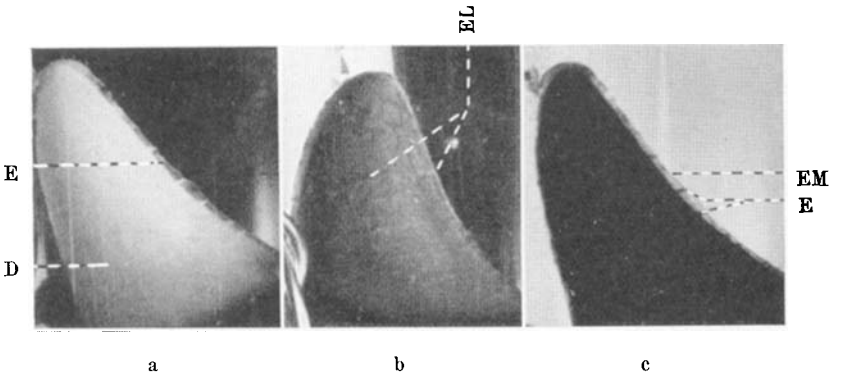

Fig. 4. Macrophotograph magnified of a decalcified incisor of a dog. a. labial side; b. lingual side, and c. labial side illuminated from behind. One observes in the picture b: the enamel lamellae surround the dentine like "Saturnian rings". E. Enamel stroma. EM. Enamel cuticle. EL. Enamel lamella, and D. Dentine.

the author called *enamel fibers* in a previous work (6), finer *membranes* run. Together with the enamel fibers these form the basis of the *prism sheaths*, described by RETZIUS, and which seem to surround every so-called *enamel prism*. These latter were considered to constitute the primary element and the corner-stones of the enamel in the days when the morphological studies of the enamel was done with thin ground sections of non-decalcified teeth and the enamel was considered as a dead mineral deposit, originating from special enamel-forming cells, the so-called ameloblasts. The above-mentioned prisms are *cross-striated*. To this cross-striation, which was formerly explained to be only a layer in the deposits of mineral salts in the prisms, correspond fine argyrophile membranes in the sections from decalcified enamel.

Moreover, argyrophile so-called *enamel-lamellae* are also present in the enamel (vide Fig. 3). They run, at least in the dog's enamel, as macroscopically visible lamellae through the entire enamel substance — like Saturnian rings — if the comparison is permitted (vide Fig. 4). There are besides these regularly running enamel lamellae, other bigger argyrophile bands in the enamel, irregularly situated and of varying intensity, which apparently form repairing phenomena after cracks in the enamel. In literature these scar-formations also bear the name of enamel lamellae.

All the reticular fibrous structures mentioned above have been included in the author's definition of *enamel stroma*. To the just mentioned facts regarding the enamel structure must be added another, which in the present connection does not seem irrelevant, namely that the enamel tufts have a *lumen*. In other words, *they look like tubes* (vide Figs. 5, 6, 7 and 8). That the enamel tufts appear like tubes or vessels is readily seen in each series of

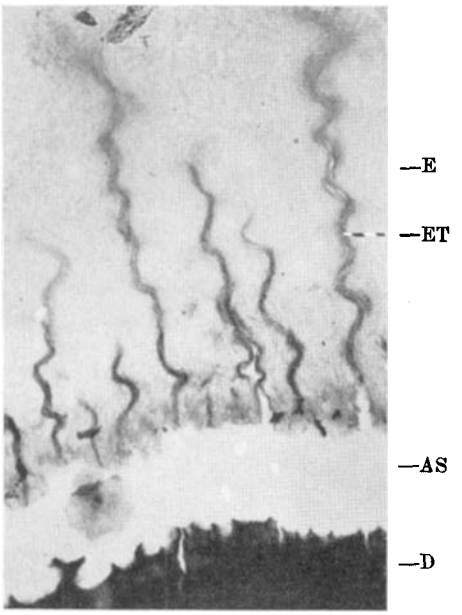

Fig. 5. Section from a premolar of a 12 year old boy. Fixation: formalin. Decalcification under pressure. Embedding: paraffin. Staining: Heidenhain's Azan. Magnification × 120. E. Enamel stroma ET. Enamel tufts. D. Dentine, and AS. Artificial mounting space between enamel and dentine.

sections — ca. 30 in all — of physiological enamel which the author has investigated. It is quite improbable that this is a question of merely artificial cracks in the sections (vide Fig. 7).

The connection between the dento-enamel junction membrane, from which the enamel tufts originate, and the blood vessels in the dental pulp, is formed by the reticular fibers of the dentine which lastly emanate from the adventitia of the capillaries in the dental pulp (vide Figs. 9 and 10).

In this connection the author likes to emphasize that BERGGREN (1) has clearly shown how methylene blue, which in substance was placed in drilled cavities of *vital* teeth, was transported relatively far into the enamel, whereby the enamel tufts appeared as the principal connecting canals of the dye.

When the author tried to impregnate the enamel in erupted teeth with the staining solutions mentioned at the beginning of this publication, no convincing results were obtained. It is true that the teeth after the injection got a color corresponding to that of the injection substance, but no evident objective proof

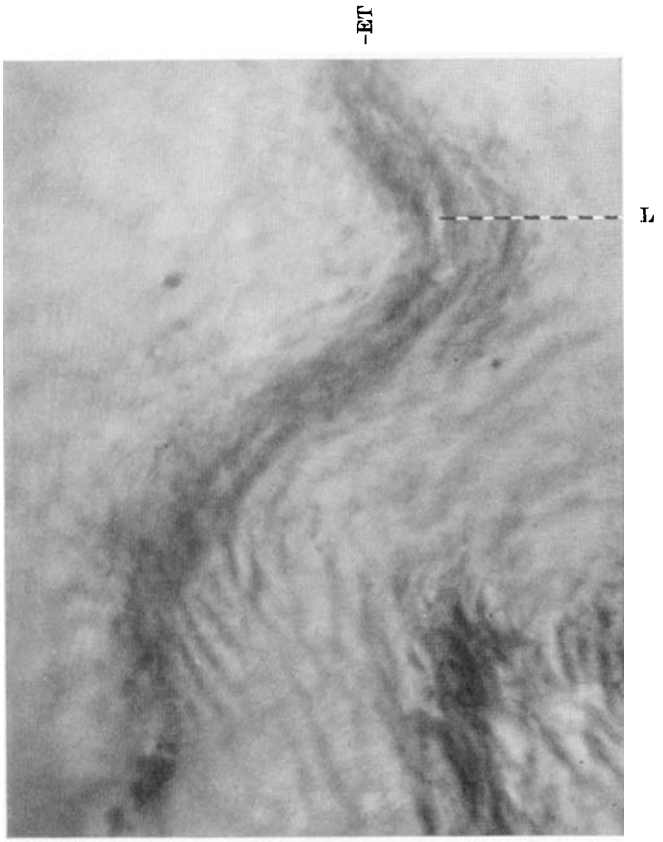

Fig. 6. Higher magnification of Fig. 5, × 1000. ET. Enamel tuft with L. Lumen.

was obtained that the enamel had been injected except by the indirect evidence, mentioned in the beginning of the article, after the injection of gelatin. When a dog's teeth, injected with India ink, were decalcified under pressure, the enamel lamellae were found to be sharply stained brownish-black. But when the sections were prepared, this tinctorial effect had unfortunately disappeared. Thus we have no absolute proof that the enamel lamellae were really filled with India ink.

This fact inspired a study of the possibility that the injection substance had not been suitable for giving tangible results of the injection of the dental enamel. The size of the particles in trypan blue for example is not bigger than that they might pass through the basement membranes and the enamel tufts. But the question

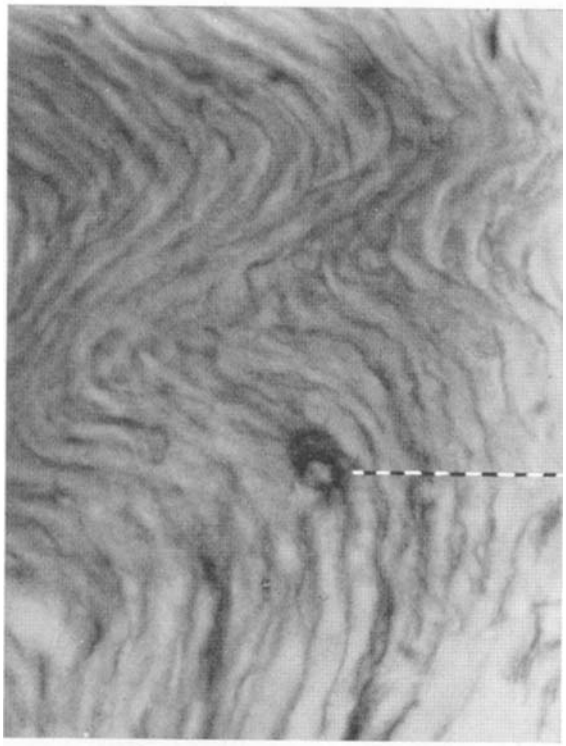

Fig. 7. Section from the same series as Fig. 5. Enamel fibers with cross-sectioned enamel-tuft ET, with visible lumen.

is whether or not it is sufficient that the injection substance should be finely grained. To answer that question one must elucidate the force which propels the circulation into the supposed ultracapillaries. *A priori* it seems rather obvious that it cannot reasonably be the hydrostatic pressure produced by the heart, which drives the blood plasma into the ultracapillaries. As motor for the circulation in the supposed ultracapillaries, one must reckon with the contraction of the capillaries — the so-called capillary pulse, discovered by STRICKER in 1865 and later confirmed by a large number of investigators. If this is the motive-power for the ultracapillary circulation, or let us say for the pumping of blood plasma into the principal stems of the ultracapillary system, then it becomes an essential prerequisite for a sucessful ultracapillary injection that the tissue survives during the time of injection. In the injection experiments mentioned

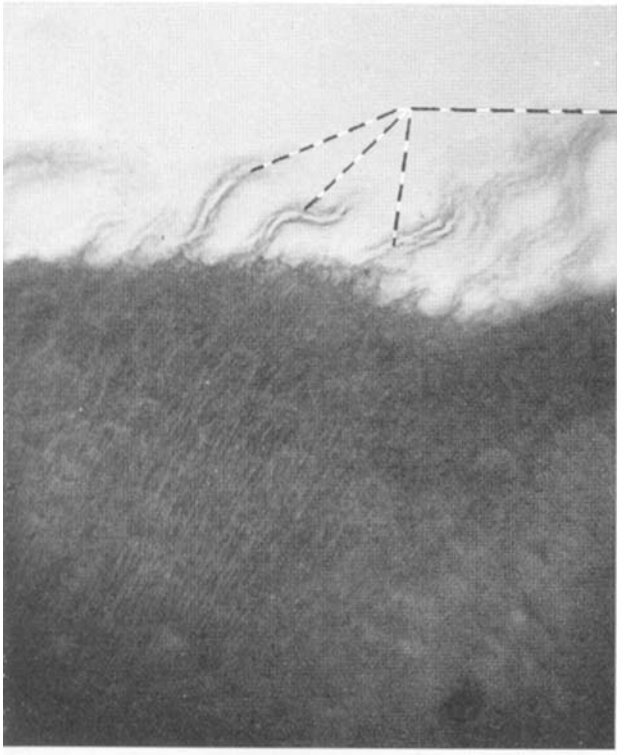

Fig. 8. Section from a premolar of a 15 year old girl. Fixation: formalin. Decalcification under pressure. Embedding: celloidin. Staining: Rio Hortega-Erasquin silver impregnation. Magnification × 120. The lumen of the enamel tufts is clearly visible. The stretched section does not give any information of the magnitude of this lumen. ET. Enamel tufts. D. Dentine, and E. Enamel.

here, this has probably generally been the case inasmuch as the injection of the substances always caused spasmodic jerks in the body musculature in question.

If we follow this argumentation that it must be the capillary pulse which feeds the ultracapillaries, then it seems doubtful that a finely grained homogeneous injection substance has a fair chance of being effectively pressed into the ultracapillaries. For the contraction of the respective blood capillary has probably no other effect on such an injection substance than that the circulatory speed is increased in the blood capillary. It is unlikely that any pressing out of the substance occurs to any great extent by the way of the ultracapillaries. However, when the blood

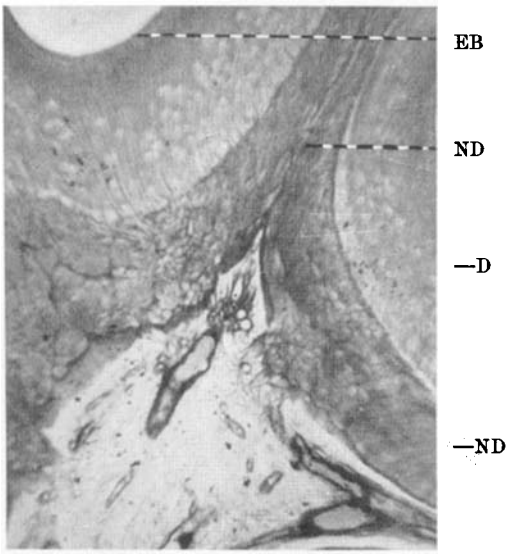

Fig. 9. Section from a molar of a rat, regenerative phase after operative trauma. Fixation: formalin, Decalcification under atmospheric pressure. Embedding: celloidin. Staining: Rio Hortega-Erasquin silver impregnation. Magnification × 120. EB. Enamel border. ND. Newly formed dentine. D. Ordinary dentine, and DP, Dental pulp.

capillary is filled with whole blood, the red blood cells probably prevent that the increase of the circulatory speed becomes the only consequence in this respect of capillary contraction. The red blood cells are likely to function as brakes on the circulatory speed in the capillaries or as valves. In agreement with a similar argumentation, an injection substance which is heterogeneous and consists of a mixture of extremely fine as well as large particles of similar size as red blood cells, would have a greater chance to fill the ultracapillaries than a homogeneous substance. The question whether or not this hypothesis is correct had better be left open to further discussion in this connection. However, by having made use of this hypothesis, the author has reached much more detailed results than in his earlier experiments with injection of the ultracapillaries. On the basis of his experimental data, the author feels inclined to make the following demands of a colloidal injection substance which is intended for injection of the supposed ultracapillaries:

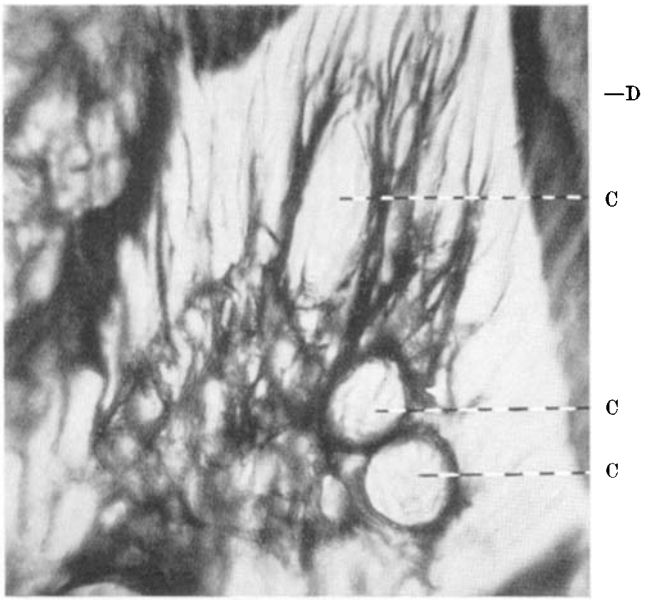

Fig. 10. Higher magnification of Fig. 9, × 1000. Argyrophile protrusions from the capillary adventitia — reticular fibres — radiate into the dentine, where they form argyrophile sheaths of the dentine canals — Neumann's sheath. D. Dentine, C. Capillary, and DP. Dental pulp.

1. The substance should consist of a finely grained and a coarsely dispersed component, the latter having the size of red blood cells.

2. The finely grained component should be quickly precipitable in order that it might remain in the place reached at the termination of the injection.

3. When a dye solution, such as the finely grained one, becomes so insignificant in the ultracapillaries that an eventual staining of the injected ultracapillaries is not directly observable, it should be possible to detect the presence of the injection substance by means of some specific reaction.

4. The injection substance should not be deteriorated beyond recognition by the material employed when preparing histological sections. Thus, an ideal substance for experiments with enamel injections ought to resist the liquid used for decalcifying the enamel.

5. The injection substance should be tolerated by the blood

of the test animal so that no agglutination or flocculation takes place. This demand is particularly important if the blood vessels are not washed out for residual blood before the injection. Naturally, the injection substance must be isotonic with the blood.

The author's endeavours to prepare such an ideal injection substance have unfortunately met with failure. However, he has found that a solution of *hemoglobin* containing the ruptured red blood cells — by hemolysis — fulfills demands 1, 2, 3 and 5. But the hemoglobin solution does not resist the nitric acid decalcification liquid used in these experiments.

Experimental Technique.

The preparation of the injection substance. — Stirred ox-blood is centrifuged in a usual milk separator and the red blood cells containing part of the centrifuged ox-blood — about half the volume of defibrinated whole blood — is hemolyzed with 2 parts distilled water added to one part of red blood cells liquid. One ml of 10 percent chinosol and 1.3 ml of 10 percent formalin are added to one liter of the prepared liquid in order to render it bacteriostatic. The pureness and isotonicity of the injection substance are controlled as well as the fact that no flocculation nor agglutination result from the mixture of the test animal's blood and the injection substance.

The injection procedure. — When the test animal is a dog — an animal especially suitable for dental enamel injection experiments — 5 to 10 ml of 1 percent morphine are given subcutaneously one hour before beginning the experiment.

The common carotid artery and the interior jugular vein are exposed bilaterally under ether narcosis. The animal is then placed in the incubator held at 37° C. The artery on one side is ligated and the vein on the same side is cut across. The artery on the other side is furnished with a cannula connected with the injection solution flask. The insertion of the cannula is made when the latter is filled with the injection solution in order to avoid air embolism. The injection is then carried on under 100—150 mm mercury pressure until 4—8 liters of hemoglobin solution are injected. The solution is held at 38—40° C. In the most success-

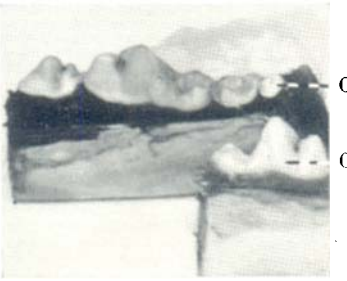

Fig. 11. Two mandibular fragments from a dog. One contains 3 molars, whose enamel is stained by hemoglobin which was injected into the common carotid artery. The color has not penetrated into the enamel of the fourth and smallest molar, OT, probably by mere coincidence. The other mandibular fragment contains a molar from a control dog, CT.

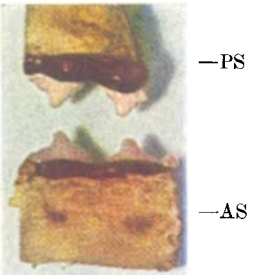

Fig. 12. Fragments from jaws, each containing two premolars from right and left lower jaw of a dog injected with hemoglobin solution. The lower maxillary fragment shows the injected side. The dental enamel of the other maxillary fragment is also stained red, albeit faintly. AS. Active side of injection, and PS. Passive side.

Fig. 13. Mandibular fragment from the injected side of a dog injected with hemoglobin solution. The red color appears very distinctly in the enamel of the front teeth — black in the photograph — the canine tooth is only slightly stained, probably by mere coincidence.

ful case 4 liters were injected during 1 hour and 20 minutes. The ligature on the artery on the opposite side was removed after 50 minutes. The animal breathed during 1 hour and 5 minutes kicked now and then rather forcefully — the animal was not tied down — and wagged its tail once or twice. The chewing muscles worked spasmodically during the first 10 minutes of the experiment. Other muscle reflexes were also observed.

When the test animal is a rabbit, the above mentioned procedure is mainly followed. The injection is made, however, in the descending aorta. The head and the upper part of the thorax are separated from the body and the injection cannula is inserted into the aorta just below the heart. In the rabbit the injection was continued as long as the animal was kicking or wagging its tail and as long as contractions of the toes and ventral musculature showed that the tissues were alive. During an injection time of 45 minutes ca. 1.5 liter hemoglobin solution was injected.

The fixation was generally done with a mixture of 3 parts of 20 percent formalin and 1 part of saturated picric acid which precipitates the hemoglobin. In the case represented by Figs. 11—14, the jaws were not fixed in a general sense; they were placed directly in 96 percent alcohol which also precipitates hemoglobin.

Experimental Results.

Teeth. — The dental enamel of a 14 months old dog became deeply pigmented during the injection. The staining was most marked on the side of the injection. But the enamel on the other side was likewise stained distinctly red (vide Figs. 11—14). A few days after the injection, the pigmentation of the enamel changed into a bluish-red deeper tinge. A few days later the pigmentation changed into indefinable shades of red-blue-brown. Two single teeth presented no pigmentation in the enamel. The injected teeth showed spots here and there which were not stained at all (vide Figs. 11 and 13).

The teeth were sectioned and examined under the dissection microscope. Distinct red stripes, running from the dentine border to the enamel surface, are visible in the enamel (vide Fig. 14). If the tooth is boiled, these stripes become brown-tinged in the typical hematin color. It is well-known that hematin is produced

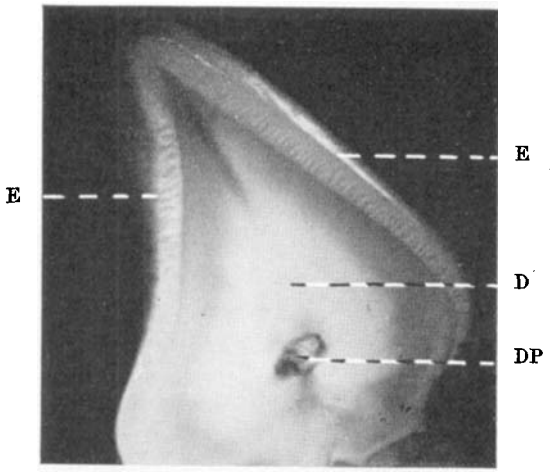

Fig. 14. Dissection microscope photograph of a sectioned premolar from a hemoglobin solution injected dog. The red staining follows the *Hunter-Schreger lines*. E. Enamel. D. Dentine, and DP. Dental pulp.

when hemoglobin is boiled. These pigmented lines coincide with the so-called *Hunter-Schreger lines*, which are visible also in the normal non-injected enamel. In these the darker stripes appear faintly grey when a sectioned tooth is examined under the dissection microscope. The stripes appear very clearly in the injected teeth, either as brown or red according to the teeth having been boiled or not after injection. The enamel parts between the strongly stained lines seem also to be pigmented, although very faintly.

This experimental result proves that it is possible to stain the dental enamel by injecting the colloidal staining solution into a main artery. The enamel is not stained diffusely but in definite localities. One must not conclude, however, that the places where the hemoglobin gathers most compactly, are constituted of conductive canals while the areas only slightly stained should prove the reverse. It would appear from the following that the especially strongly stained parts in the hemoglobin injected tissue may be areas where the dye has been *retained*. Naturally such areas must contain nutrition canals of varying sizes. But other areas with less marked concentrations of the dye, ought not to be considered as lacking in nutrition canals on this ground alone. They may consist of *transitional parts*, although the absence of dye can

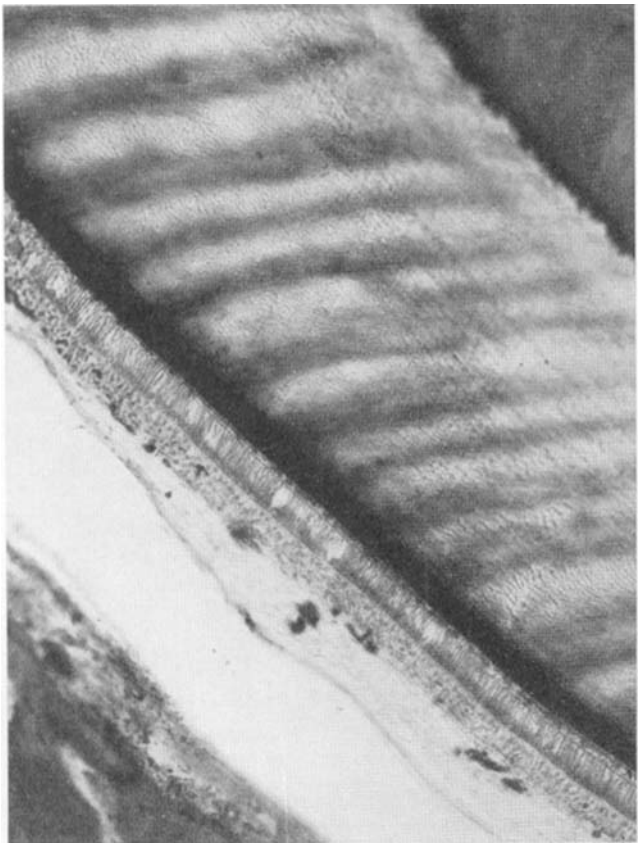

Fig. 15. Section from a not yet erupted molar of a pig. Magnification × 90. Fixation: formalin. Decalcification under atmospheric pressure. Embedding: celloidin. Staining: Heidenhain's azan. D. Dentine. AD. Adamantoid enamel. AM. Inactive interior enamel epithelium. The light and dark stripes in the enamel are the *Hunter-Schreger lines.*

also be due to lack of nutrition canals or lack of such canals which allow the passage of the dye. One should bear in mind the possibility of an obstacle being present in the blood capillaries which not permits the passage of the dye. In those parts of the enamel where the just mentioned lines have been seen at even intervals, it is quite likely that the last mentioned factor has not absolutely asserted itself.

Figures 15 and 16 are taken from a previous work (6) and show that in sections which illustrate the *formation of enamel*, one observes that every other stripe of the *Hunter-Schreger lines* is

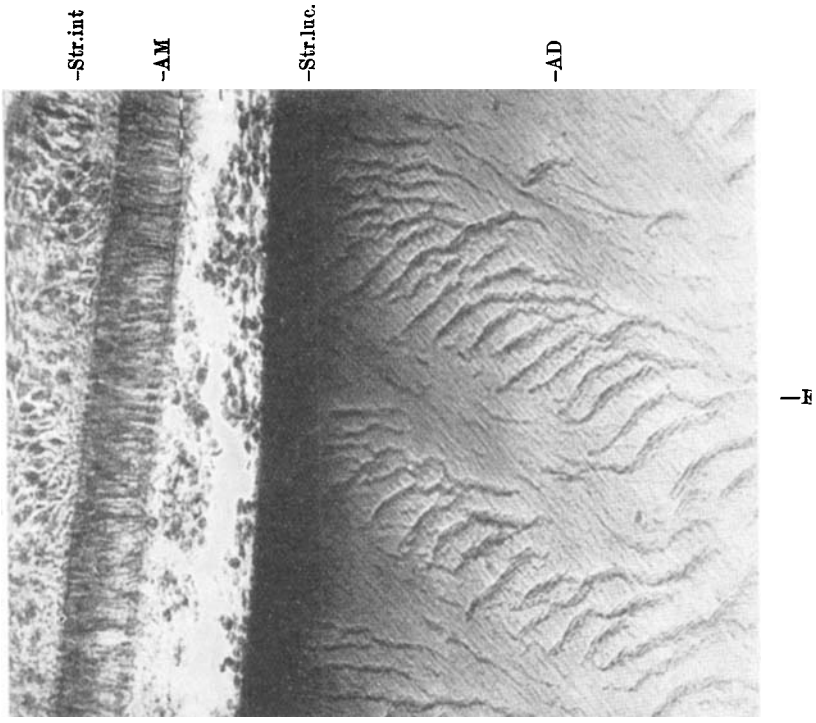

Fig. 16. Section from a non-erupted molar of a pig. Magnification × 240. Technique same as in Fig. 15. Str. int. Intermediary stratum. AM. Inactive interior enamel epithelieum. Str. luc. Elaidin enamel. AD. Adamantoid enamel. ES. Coarser network of reticular fibers of the enamel, which the author calls enamel stripes. They are identical with the enamel tufts. The lines in the enamel are the *Hunter-Schreger lines.*

dominated by still more marked bands of reticular fibers. Our investigation in this regard has not elucidated whether or not the more compact bands of reticular fibrous network are those which in Figure 14 appear as the most deeply injected. Still it seems quite plausible that such is the case.

It has previously been mentioned that hemoglobin does not resist the acid used in decalcifying teeth. Thus we cannot present any histological section of enamel injected with hemoglobin. Nevertheless, we have been able to prove that the enamel can be stained as a result of injecting the colloidal staining substance into a main artery. The stain is partly distributed along the *Hunter-Schreger lines.*

Other Investigated Tissues.

It is well-known that an abundance of reticular fibers occur in every tissue in the body. They originate in the capillary adventitia, and thence spread to the very smallest units in the tissue. The adventitia in the blood capillaries is the essential stable capillary wall which renders the blood circulation a closed system, according to HUZELLA. The endothelial wall does not constitute an impermeable wall. The reticular fibers are demonstrated by specific silver impregnation methods which stain these black. Figures 5—8 show that the coarser stems in the network of the enamel stroma have the appearance of tubes, even in the relatively coarse picture of the tissue structures seen in the usual histological section. One must not ignore the fact that the so-called reticular fibers, even in their finest ramifications, may appear not only like tubes or vessels, but they may also have the function of such. The irrefutable fact that the reticular fibers in the dental enamel can be injected from a main artery even as the blood vessels, makes it probable that the reticular fibers constitute in general, not only morphologically but also functionally, a direct continuation of the blood vessel system such as the latter is classically accepted. In other words, the reticular fibers form part of a more complete blood vessel system, with the object of bringing the nutrifying blood plasma to all parts of the tissue.

It will necessitate extensive investigations in order to elucidate the various dimensions of the reticular fibers — ultracapillaries — as well as their inter-connections and distribution. The dental enamel consists apparently of large "ultracapillary stems" (*i. e.* the enamel tufts) from which the finer ramifications emanate. It might be well worth investigating whether or not the same conditions prevail in the organism in other respects as well. Injections with colloidal solutions of various known molecular sizes should perhaps first of all be done. The first obstacle to be overcome is probably the difficulties of preparing suitable injection substances for such experiments. It would appear that hemoglobin, which is capable of penetrating into the finest structures discernible by histological technique, is not the ideal substance with which to illustrate the distribution and dimensions of ultracapillaries.

Thus it is not possible to render every separate transportpassage-

lumen visible by means of hemoglobin injections. Nevertheless, one is probably right in assuming that the intravascularly injected hemoglobin is most abundantly present in those tissue sections where the finer or coarser reticular fibers exist in largest numbers.

In order to continue the study of this problem, the author has examined various animal organs injected with hemoglobin, such as the liver, kidneys, spleen, muscles etc. To prove the presence of hemoglobin in the tissues, frozen sections were prepared and stained according to the method of SJÖSTRAND (9) for demonstration of red blood cells. This method is based on the specific color reaction of orthotolidine toward hemoglobin.

Kidneys. — Figures 17 and 18 show frozen sections of renal medulla from a control rabbit. The pictures give an outline of the capillary distribution. Figures 19 and 20 show frozen sections of renal medulla from a rabbit injected in the aorta with hemoglobin solution. SJÖSTRAND's histological technique was employed in both cases. The latter pictures demonstrate how the tissue between the capillaries is strongly impregnated with hemoglobin in spots. In comparing these pictures with Figures 21—23 of silver impregnated sections, we observe a denser network in spots of the argyrophile threads in this tissue. This may account for the earlier mentioned stronger hemoglobin impregnation which appear in spots in the medullary tissue.

Figures 24 and 25 present the renal bark from a control rabbit, stained by the SJÖSTRAND method in order to demonstrate the distribution of blood vessels. Figure 26 is taken from sections of the renal bark from a rabbit injected with hemoglobin. The dye is most markedly concentrated in definite localities, especially in the Malpighian corpuscles. Figures 27 and 28 illustrate the structure of the reticular fibers in the renal bark, which possibly correspond to the distribution of hemoglobin present in Figure 26. Figure 29 shows how the hemoglobin can be more compactly concentrated in the urinary pole in a Malpighian corpuscle and in the urinary canals than in the vascular pole.

Liver. — Except in the blood vessels, we have not found any special retention places for hemoglobin in the liver. All the paracapillary tissue seems to be rather evenly and compactly injected (vide Fig. 30). Since KUPFFER's time it is also known that the

Fig. 17. Magnification × 50.

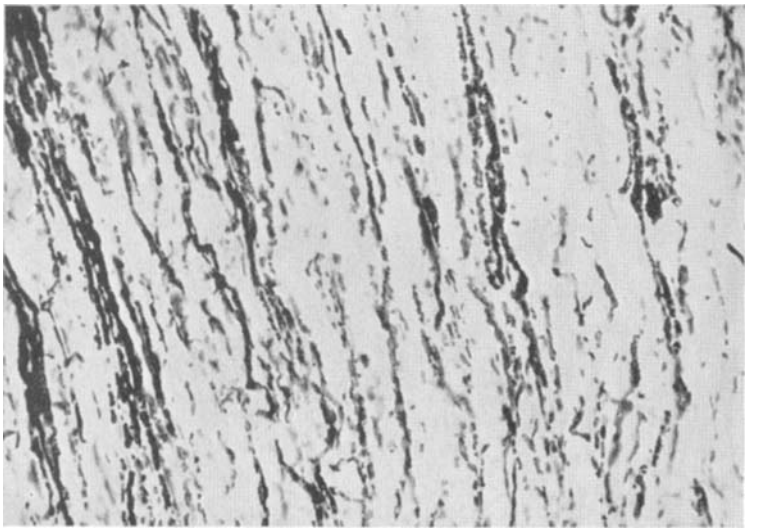

Fig. 18. Magnification × 140.

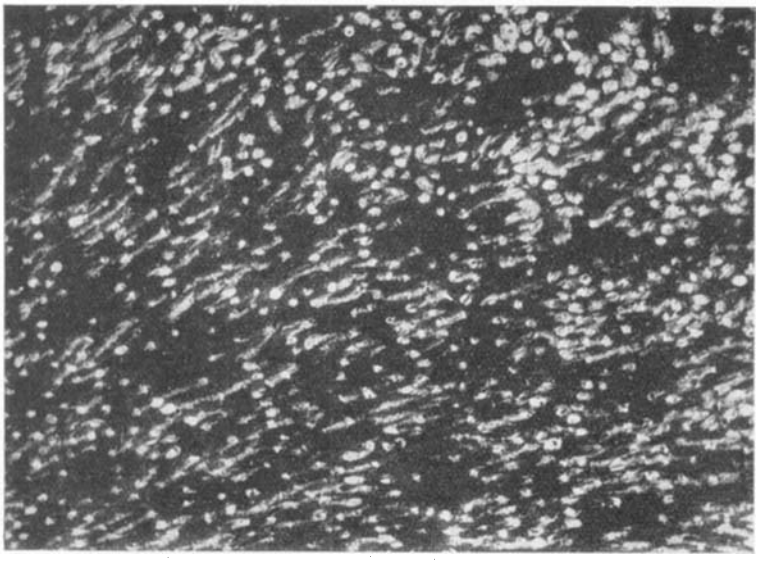

Fig. 19. Magnification × 50.

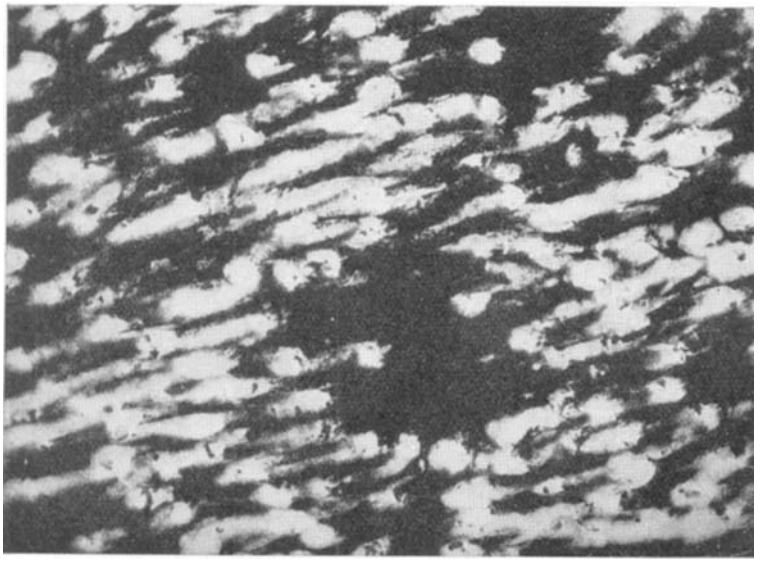

Fig. 20. Magnification × 140.

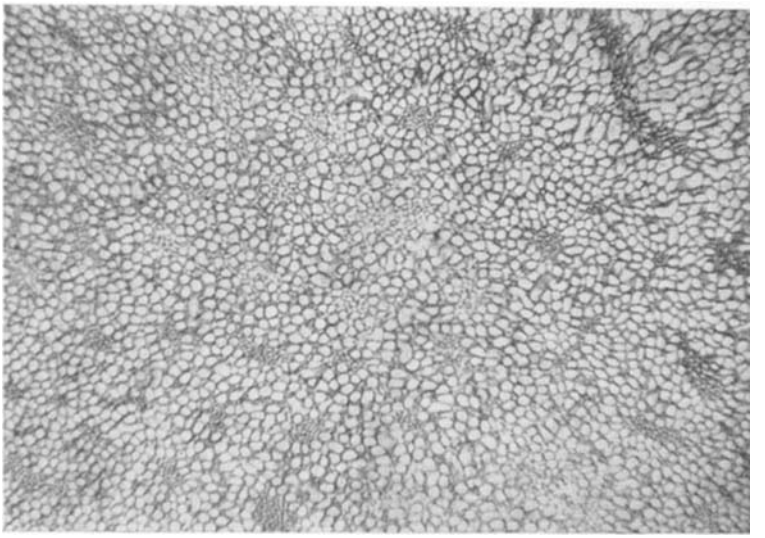

Fig. 21. Magnification × 50.

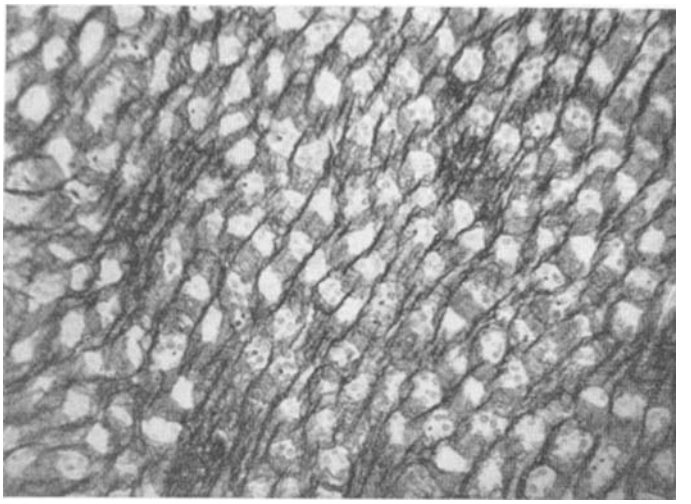

Fig. 22. Magnification × 140.

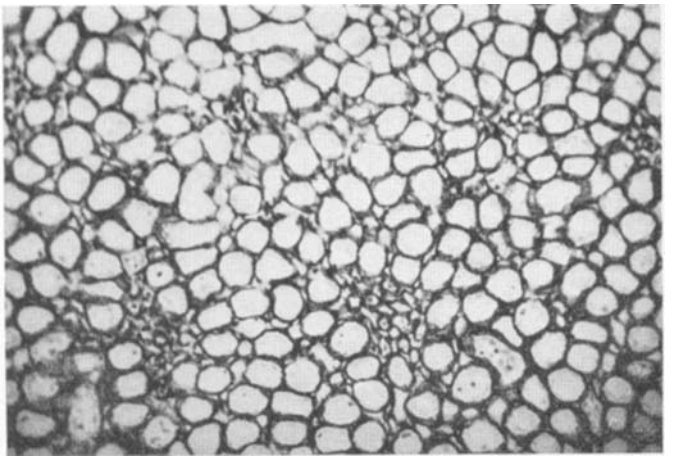

Fig. 23. Magnification × 140.

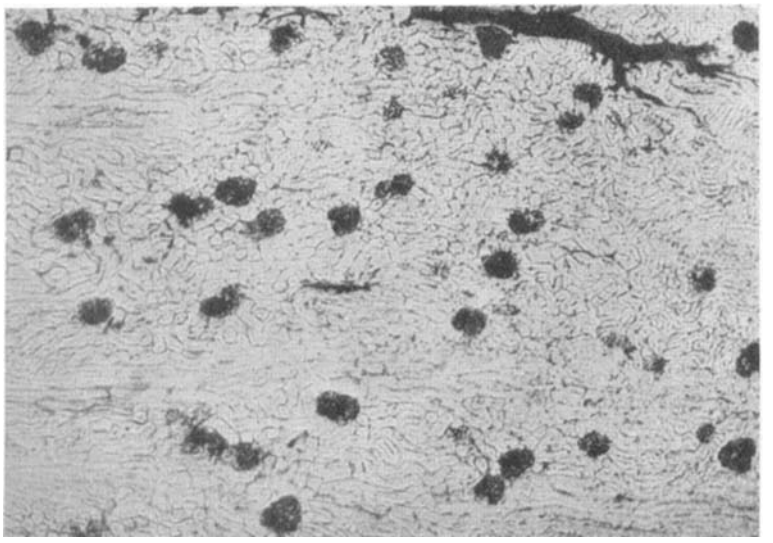

Fig. 24. Magnification × 50.

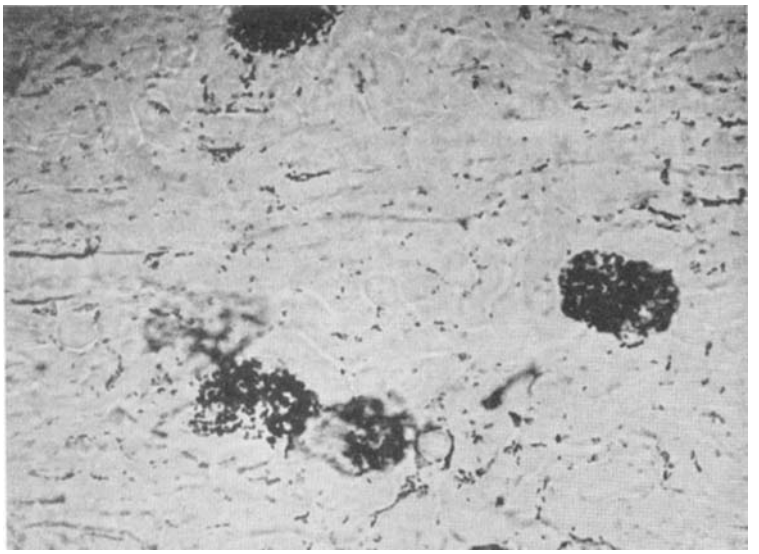

Fig. 25. Magnification × 140.

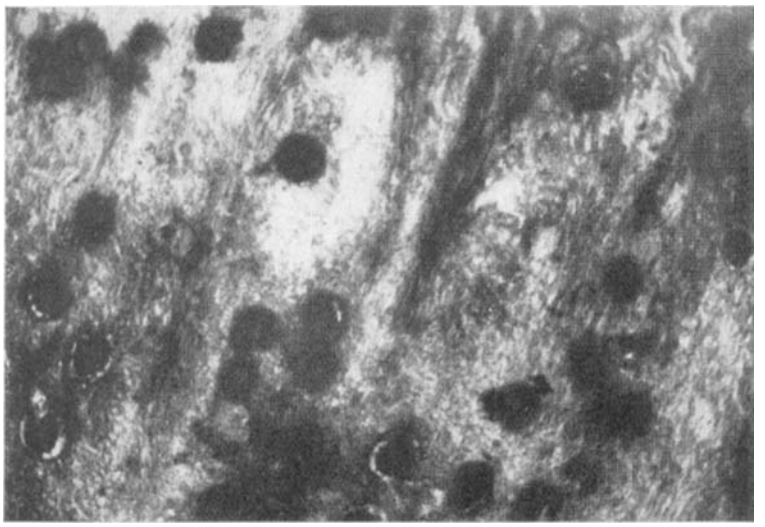

Fig. 26. Magnification × 50.

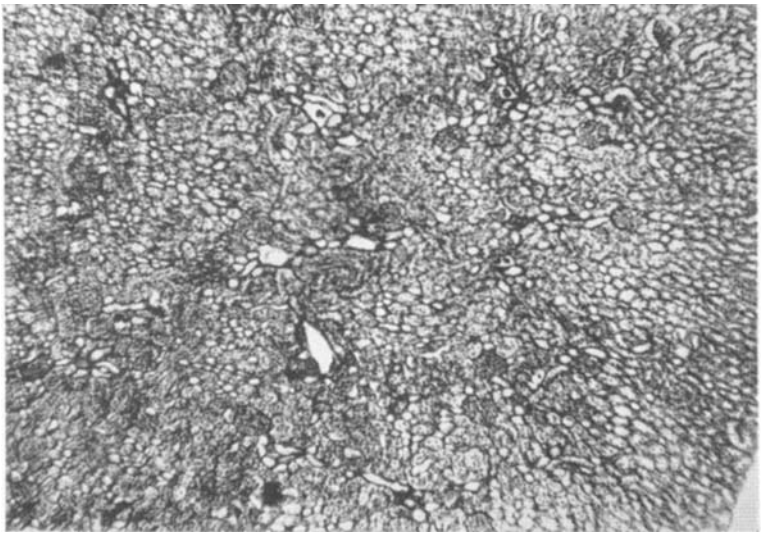

Fig. 27. Magnification × 50.

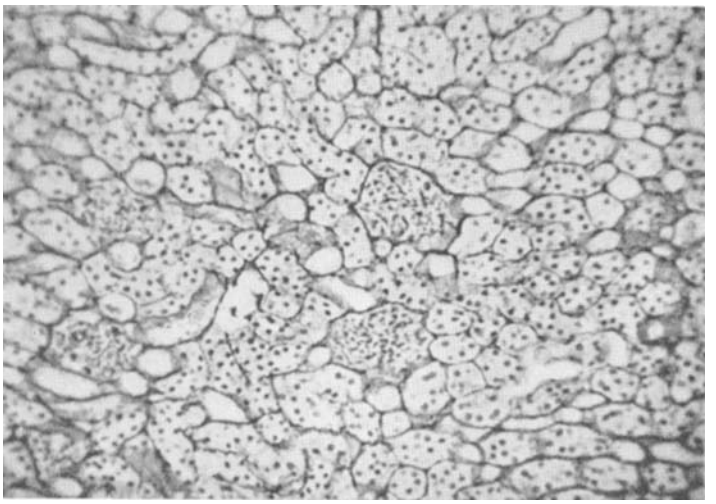

Fig. 28. Magnification × 140.

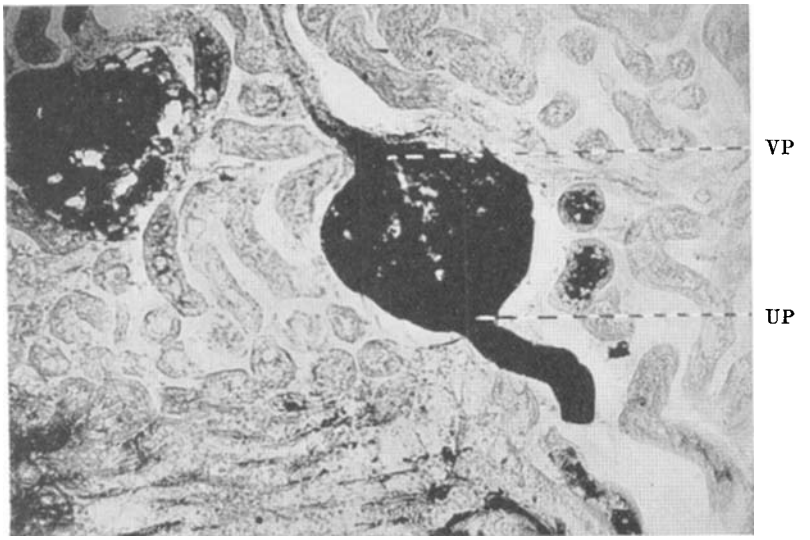

Fig. 29. Magnification × 140. VP. Vessel Pole, and UP. Urinary pole.

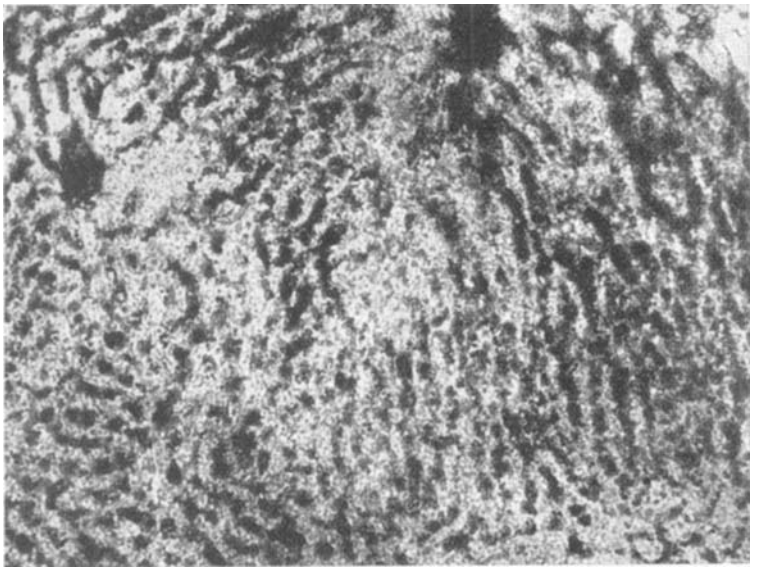

Fig. 30. Magnification × 140.

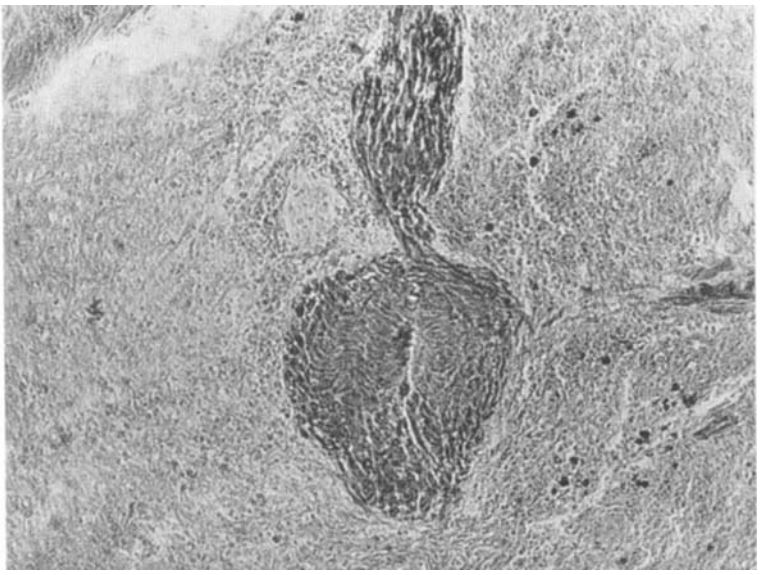

Fig. 31. Magnification × 140.

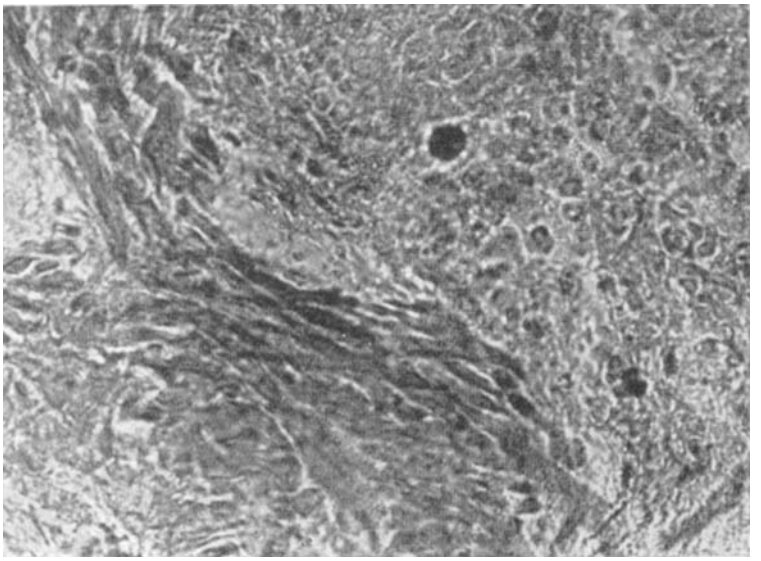

Fig. 32. Magnification × 580.

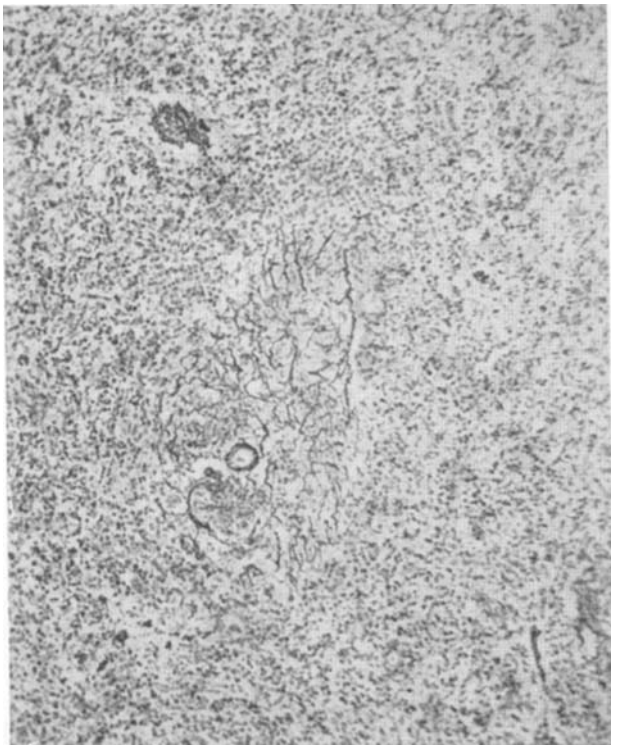

Fig. 33. Magnification × 140.

liver possesses a regularly arranged network of reticular fibers, emanating from the central veins and capillaries.

Spleen. — Although the red and white pulp of the spleen are impregnated with the injected hemoglobin solution, the color is especially concentrated in the white pulp. Figures 31 and 32 show sections from the spleen of an injected rabbit. Figures 33 and 34 show silver impregnated sections from the spleen of a control rabbit. The cause of the heavier impregnation with hemoglobin in the white pulp seems of be the well-known fact that the argyrophile fibrous network in the spleen is most strongly developed in the white pulp.

Parotid gland. — Figure 35 shows a section from the parotid gland of a dog injected with hemoglobin. Figure 36 is from a silver impregnated parotid gland from a dog. Here too we observe

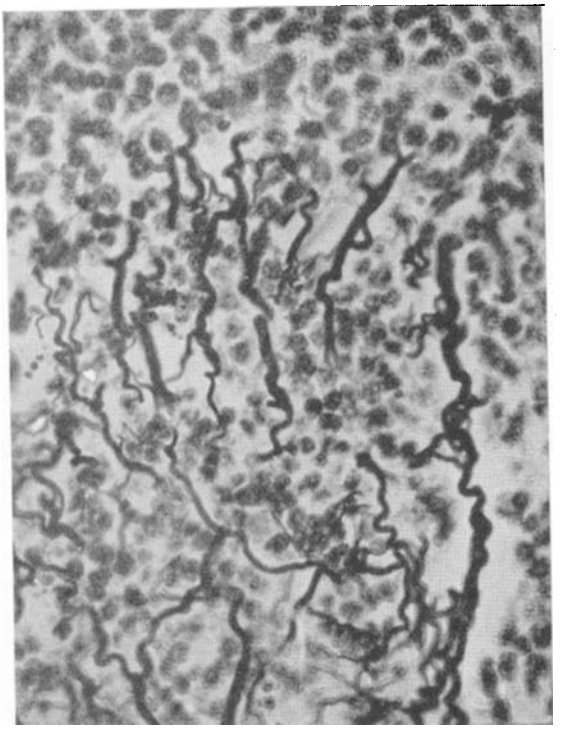

Fig. 34. Magnification × 580.

a certain parallel between the extension of the argyrophile fibrous network and the retention of hemoglobin in the paracapillary tissue.

Muscle. — When studying the muscle tissue from hemoglobin-injected animals, we were surprised to find how the muscle bundles were heavily stained by the hemoglobin. Figures 37 and 38 illustrate this fact. In cases of incomplete injection, one gets the impression that the hemoglobin is retained primarily in the dark striae, the A-disks (vide Fig. 39). It is known that the muscle fibers are surrounded by a strongly developed network of argyrophile threads. The A-disks appear as deep black lines in silver impregnated sections (vide Figs. 40 and 41). Figure 40 is taken from a dog's tongue. Figure 41 is reproduced from MÖLLENDORFF and presents a drawing of a silver impregnated muscle. Thus the muscle tissue seems to present a clear parallel

Fig. 35. Magnification × 50.

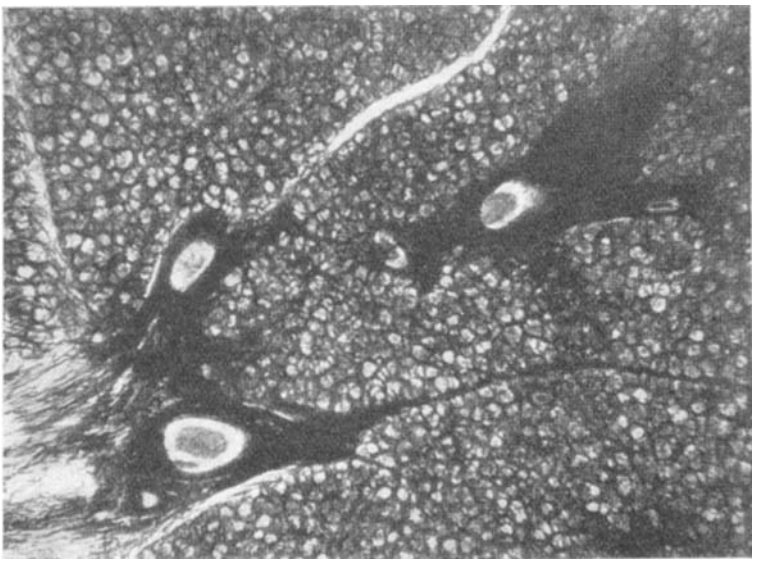

Fig. 36. Magnification × 50.

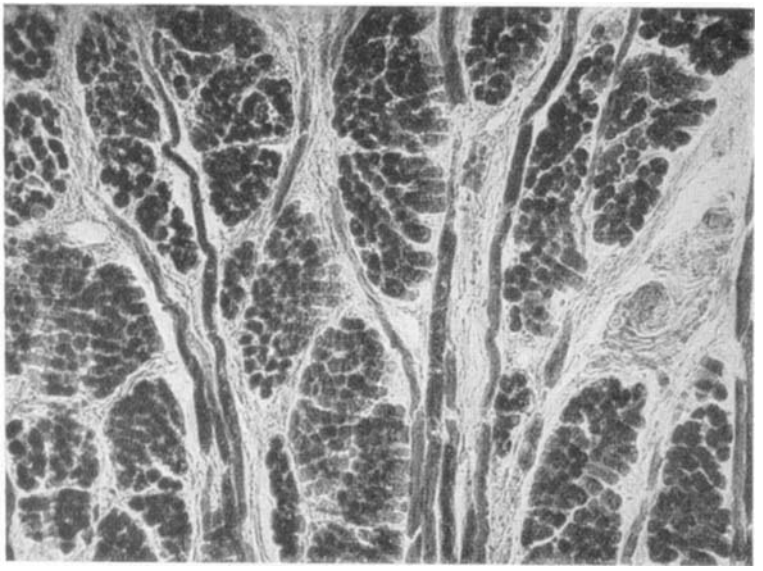

Fig. 37. Magnification × 70.

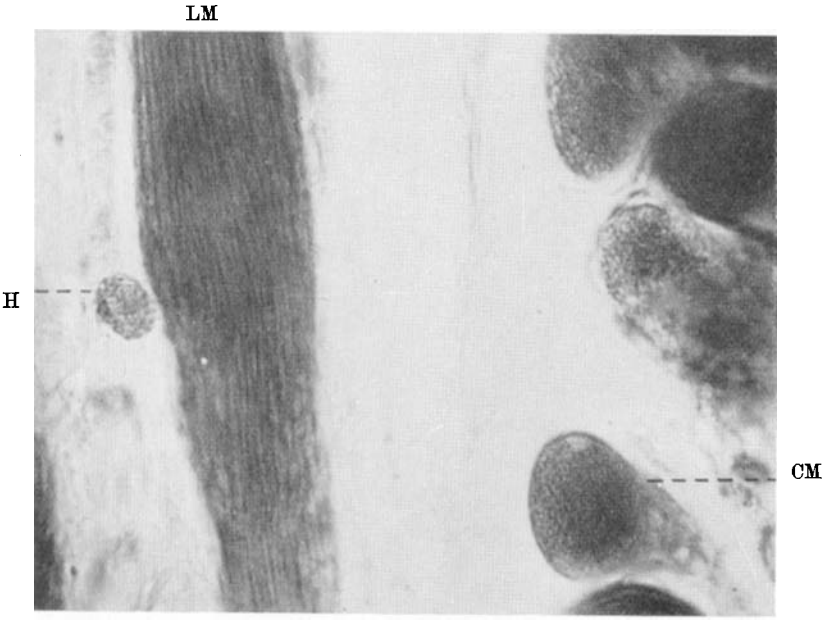

Fig. 38. Magnification × 900. CM. Cross sectioned muscle bundle. LM Longitudinally sectioned muscle bundle. H. Histiocyte.

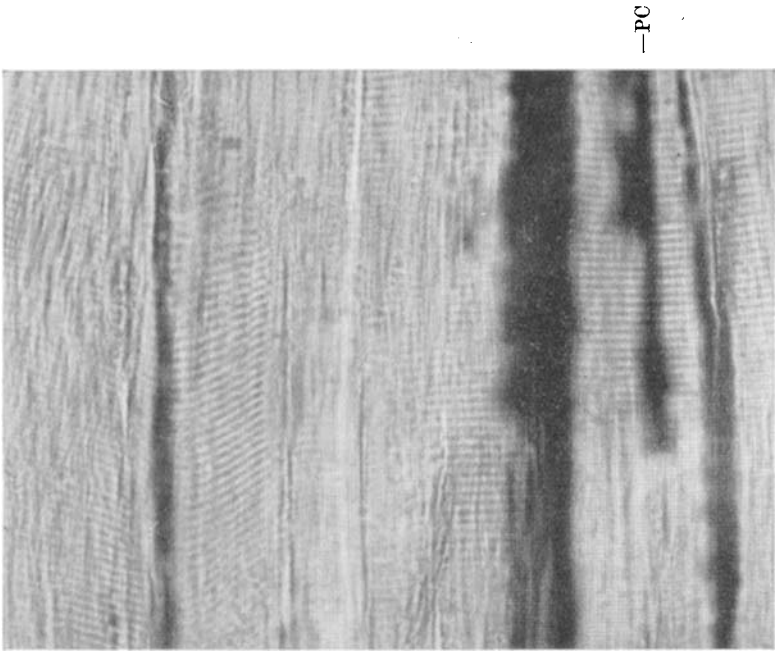

Fig. 39. Magnification × 580. PC. Capillary not injected in its entire length. Just above the point where the hemoglobin ceased to enter the capillary, the cross striation of the nearest muscle fibers is no longer visible.

between a more frequent presence of argyrophile elements and a more compact retention of the injected hemoglobin beyond the blood vessels.

Figure 42 shows a border area between tendon and muscle in a hemoglobin injected rabbit. The tendon seems much less compactly injected than the muscle bundles. The distribution of the reticular fibers is not the same in tendon and muscle. In the tendon, the collagenous fibers dominate. Silver impregnated sections show the difference between the distribution of reticular fibers in tendon and muscle (vide Figs. 43 and 44).

Interstitial connective tissue. — The interstitial connective tissue in muscular tissues is obviously less stained by the injected hemoglobin than the muscle bundles (vide Figs. 37, 38 and 45). The argyrophile fibers have not such a compact network in the interstitial connective tissue as in the muscle tissue. In the former tissues one notices, however, that some cells — histiocytes — have

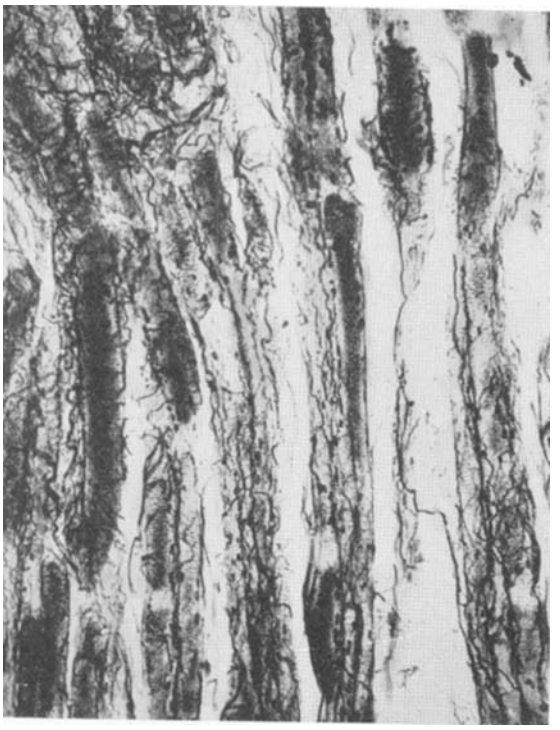

Fig. 40. Magnification × 140.

been filled with hemoglobin (vide Figs. 38, 45 and 46). Silver impregnated sections demonstrate how these injected cells are connected with fine argyrophile threads. Thus Figure 47 shows a section of tongue from an injected dog, stained according to SJÖSTRAND and impregnated with silver.

Adipose tissue. — Figure 48 is a section of adipose tissue from the kidney of a rabbit injected with hemoglobin. The section is stained by Sjöstrand's method. Figure 49 is a drawing reproduced from MÖLLENDORFF which shows the reticular envelope surrounding each fat cell.

Thus we find a common feature in all the presented investigation results, namely that the colloidal hemoglobin solution injected into the general circulation through a main artery, as a rule is unevenly distributed throughout the *paracapillary* part of the tissue, but is often concentrated in definite areas. The silver

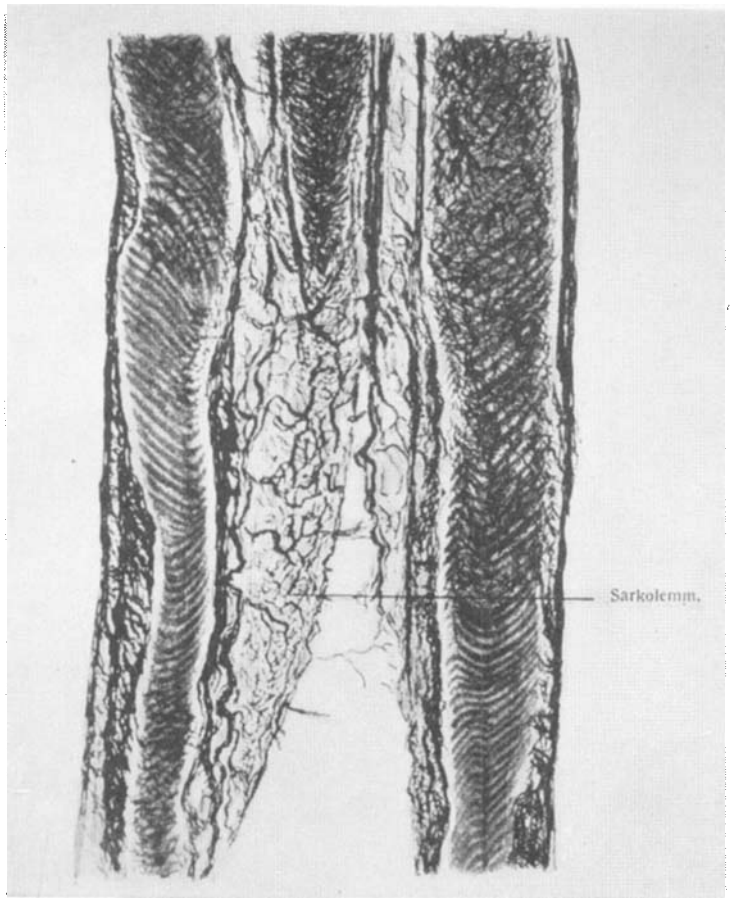

Fig. 41. Magnification × 700. Drawing reproduced from Möllendorff (8).

impregnation, employed to make the argyrophile connective tissue fibers visible, makes it apparent that the presence of paracapillary reticular fibers is unevenly distributed in the tissues but concentrated in certain parts. These parts seem identical with those which are strongly stained by the injected hemoglobin solution.

It is quite apparent, however, from looking at the figures mentioned, and especially obvious in Figure 39 in comparison with Figure 44, that the variations in concentration of the injected hemoglobin solution in the tissues is not dependable on a corresponding concentration in the capillaries.

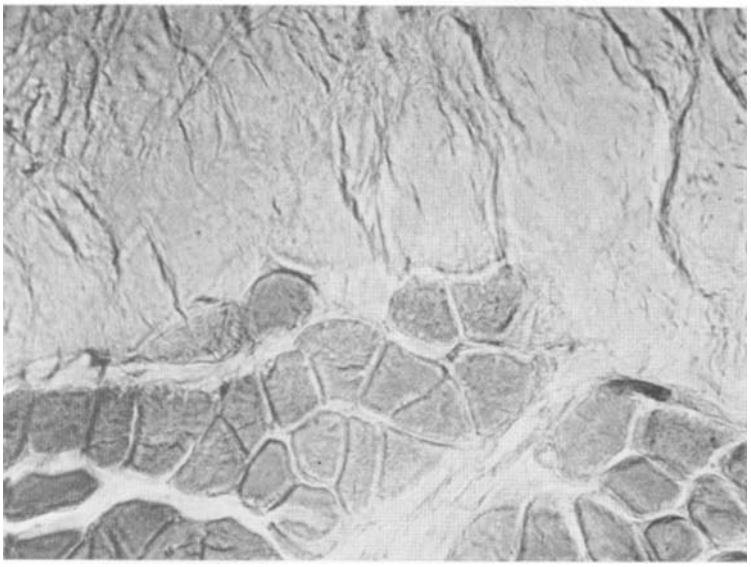

Fig. 42. Magnification × 140. T. Tendon, and M. Muscle.

Fig. 43. Magnification × 140.

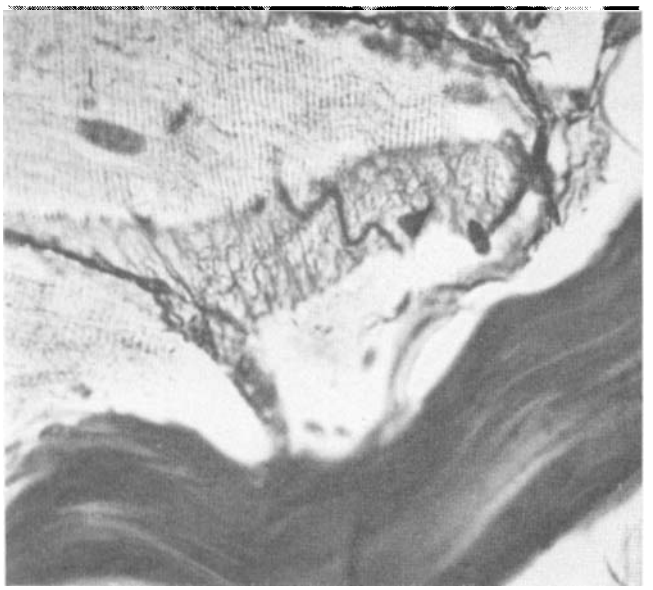

Fig. 44. Magnification × 580.

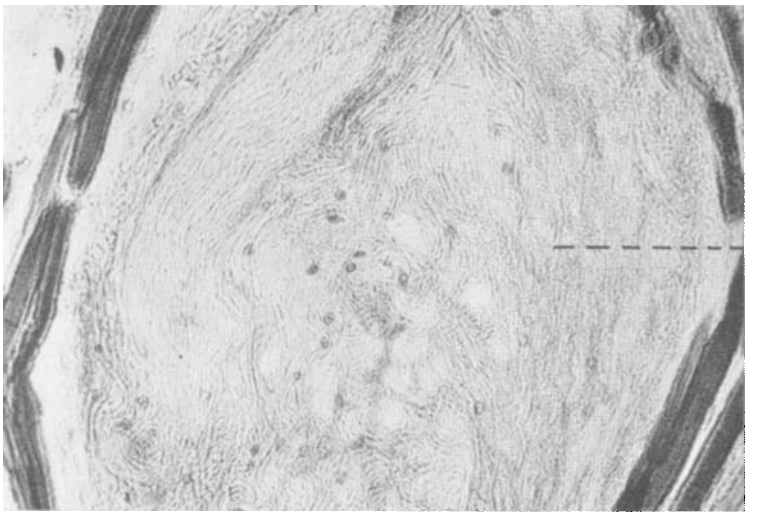

Fig. 45. Magnification × 95. IC. Interstitial connective tissue, and MB. Muscle bundle.

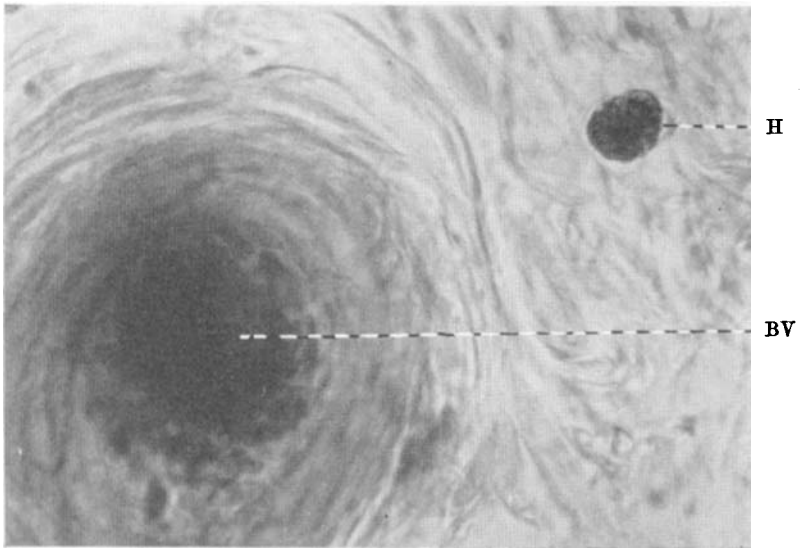

Fig. 46. Magnification × 900. H. Histiocytes, and BV. Blood vessel.

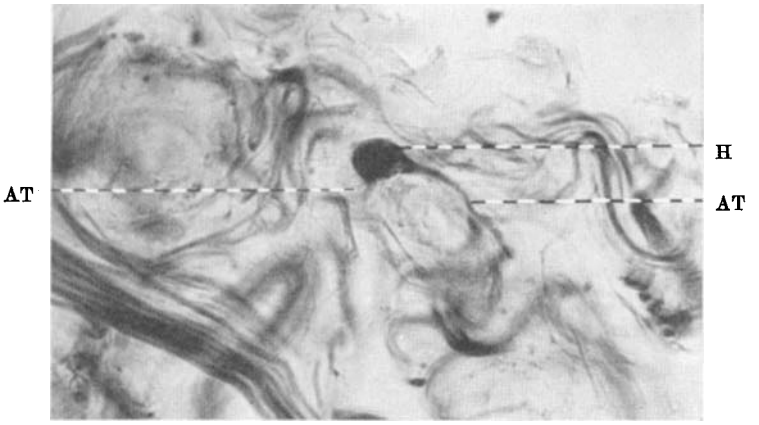

Fig. 47. Magnification × 580. H. Histiocyte, and AT. Argyrophile thread.

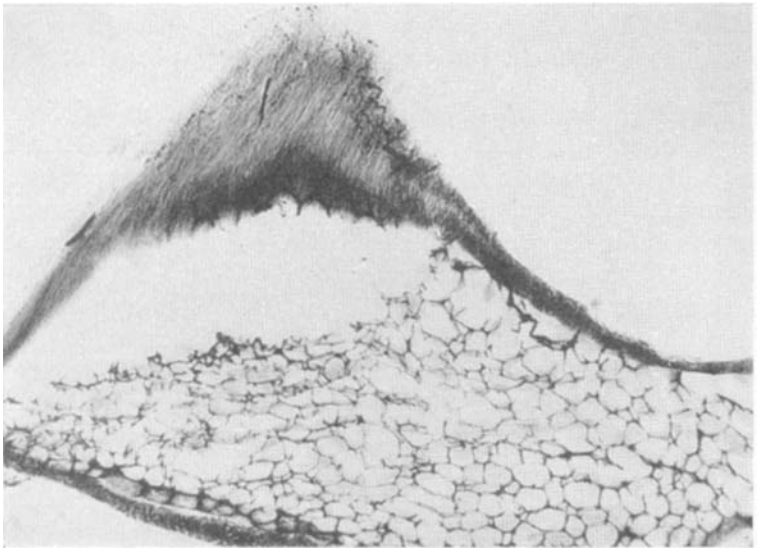

Fig. 48. Magnification × 50. F. Adipose tissue.

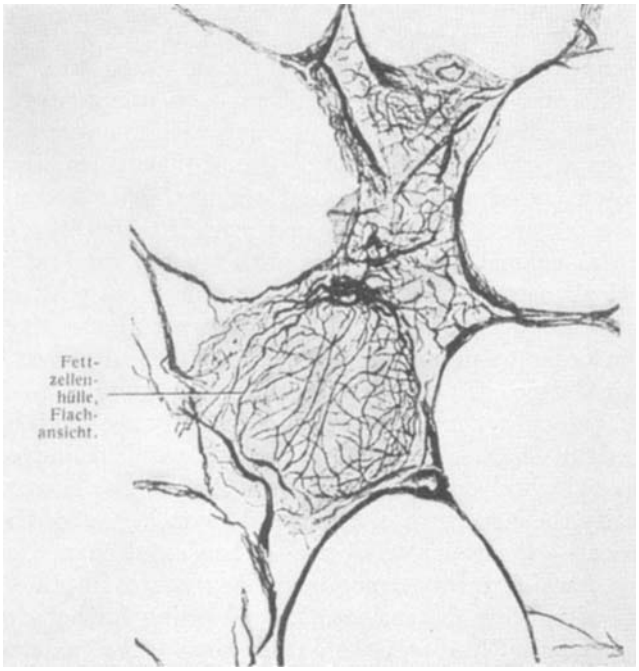

Fig. 49. Magnification × 900. Drawing reproduced from Möllendorff (8).

Sympathetic Innervation of Ultracapillaries.

The dental enamel. — It is well-known that the general circulatory system in its classical extent is richly innervated by the vegetative nervous system, sympathetic and parasympathetic, whose task is to regulate the blood supply in the respective organs. It remains to become elucidated if the ultracapillaries are possessed of sympathetic ramfications.

The ultracapillaries — the reticular fibers — which emanate from the blood vessels and surround the smallest units of the tissues, form an exceedingly fine network. But available experimental technique seems, in the hands of the author, utterly inadequate for the investigation of the special ultracapillary — sympathetic innervation throughout the tissues in general.

On the other hand it may prove to be an exceedingly difficult task to decide whether an observed nerve ending belongs to the reticular fiber or to some other part of the tissue. But among the various tissues it is most fortunate that the dental enamel is wholly made up of reticular fibers. Hence it is clear that if the dental enamel is possessed of nerve fibers, there should probably be no cause for doubting their functional connection with the ultracapillaries — the reticular fibers — in the dental enamel.

It is not known to the author that nerve fibers or nerve endings have been described in the dental enamel. This is perhaps due to the present unsuitable histological technique for disclosing the organic structure of this particular tissue. Hitherto, the study of the dental enamel has been done with mineralogical rather than histological methods because the enamel does not resist the decalcification processes which the hard tissues of the body must stand in order to prepare histological sections of these. But by an original decalcification process under a pressure of 3—4 atmospheres, the author has succeeded to decalcify the dental enamel and has thus been able to prepare both paraffin and celloidin sections of dental enamel. By this method it has been possible to identify the structures in the dental enamel. Besides the ultracapillaries — the reticular fibers — the dental enamel contains structures which may be non-medullary nerve fiber or nerve endings of varying dimensions. Before giving further details of these histological fir dings, the author should like to describe his technical procedure to prove this contention.

Technical Procedure.

Before sections can be prepared of the dental enamel, the tooth must necessarily be decalcified in such a way that the enamel is not destroyed during the decalcification process. The author has described this procedure in a previous work (4—5).

When the enamel has been decalcified, it is important that its fragile fibers are not destroyed during the process of embedding. It is necessary, therefore, to make the proper arrangements for the embedding even before decalcification has taken place. In order to achieve this aim of preserving the enamel fibers intact, the tooth is first of all tied to a frame made of rustless wire. This frame is screwed into a piece of cork to which is attached a weight of lead to keep the cork submerged (vide Fig. 50). A glass cylinder is now placed over the wire frame which stands free within the cylinder. With the aid of this contraption, one may pass the tooth through the various solutions required for the embedding. Thus the enamel stroma is always suspended in a liquid which protects the enamel fibers from injury during the necessary technical manipulations.

When the decalcification process is finished, the pressure in the glass jar, containing the glass cylinder with the suspended tooth, is slowly lowered from the high pressure which prevailed during decalcification. The glass jar is opened.

The glass cylinder is now pressed down over the cork, which has shrinked in the pressure jar. The tooth protective contraption is lifted out of the decalcification vessel and placed in a big vessel containing the decalcifying liquid, usually 5 percent nitric acid. The water of this liquid is previously boiled in order to drive off all air bubbles which might damage the enamel fibers. When the tooth protective contraption is carefully placed in the big vessel, the glass cylinder is removed while the cork is being held down with a pair of forceps.

The tooth is allowed to remain in this vessel during the time required for decalcification of the dentine, usually from one to six weeks. It is then transported to 10 percent formalin for several days. The glass cylinder must of course be put on during the transport. If formalin is a part of the decalcifying liquid then one may omit the transport in the 10 percent formalin alone. Thereafter, the tooth is rinsed in boiled water. It is then placed in a smaller cylinder in which it is passed through the various concentrations of dehydrating alcohols and then a mixture of alcohol and ether if it is destined for embedding in celloidin. At every change, the protective glass cylinder is replaced and the whole contraption is lifted out of the vessel while the liquid is being changed.

For the celloidin embedding one uses a cylinder of larger diameter than that in the original contraption (vide Fig. 51). Here the protective tube remains over the tooth during the whole first and second embeddings. After several days in 1 percent celloidin, with a cork closing the upper end of the contraption, the celloidin above the tooth

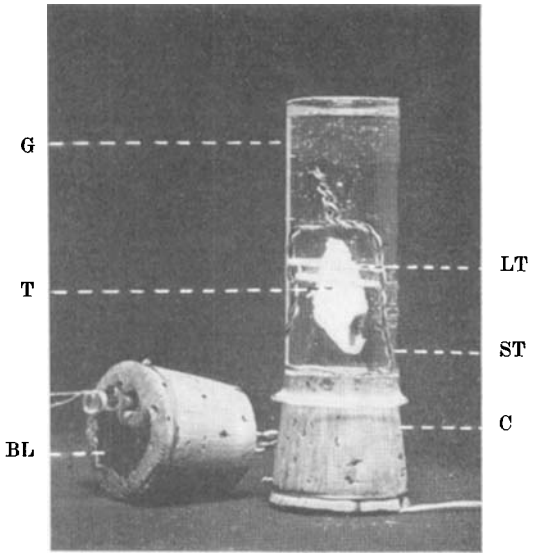

Fig. 50. Protective contraption for the embedding of teeth whose enamel is to be examined in decalcified sections. G. Glass tube. T. Tooth. BL. Ballast of lead. LT. Silk thread with which the tooth is tied to the frame of rustless wire ST, and C. Cork.

is sucked off and the tube is refilled with 4 percent celloidin. This procedure is repeated with one or two days intervals, using first 4 percent celloidin once or twice and then 8 and 12 percent. Thereafter, small sticks of dried celloidin are placed around the tooth in order to get the celloidin as thick as possible and thus diminish the shrinking which takes place on evaporation.

After homogeneization, a slow evaporation takes place. In the meantime, the celloidin is evaporated in the outer vessel and the risk is thereby diminished of air bubbles entering through the leakage around the cork in the lower part of the cylinder.

The now accomplished embedding is not useful for direct sectioning. In spite of all precautions, the celloidin becomes blistered and cracked. This is undoubtedly connected with the use of he rustless steel. Re-embedding is therefore indicated.

This is done by cutting a block, containing the tooth, free from the metal wire frame and removing the silk-threads from the block. A hole is now bored through the root of the tooth and through this a silk-thread is drawn. By means of the latter the block is suspended in a suitable evaporating dish containing 12 percent celloidin. The vessel remains closed until complete homogeneization has taken place. Then evaporation follows.

The tooth destined for paraffin embedding is passed from the absolute alcohol bath to methylbenzocelloidin and then into benzol or

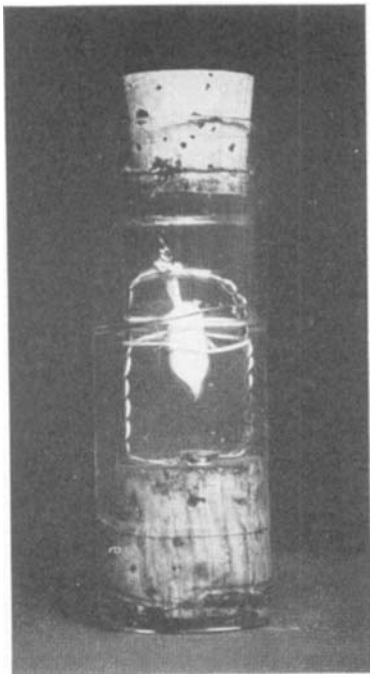

Fig. 51. The protective contraption for the enamel stroma is placed in a cylindrical glass vessel containing celloidin. The cylinder of the contraption is now filled with celloidin and a cork is stoppered even in its upper portion.

some other paraffin solvents. Thereafter it is placed in molten paraffin which is changed twice or thrice. At every transport the protection cylinder is put on. The tube is then cooled slowly from the lower end upward in ice water.

The paraffin also gets blistery as long as the metal wire is attached. It is convenient, therefore, to cut a block, containing the tooth, loose from the frame and to re-embed the block.

One may also make frozen sections of decalcified enamel. Taking the same precautions as mentioned above, the author has first embedded in gelatin in the usual manner for microtome sectioning. These attempts have not been very successful. This method should be further improved inasmuch as so many differential staining methods are based on the frozen section technique.

The following standard staining methods have been used:

1. VAN GIESON staining with WEIGERT's hematoxylin for general orientation.

2. MARTINOTTI's keratin staining with aniline blue which gives the basement membranes of the dentine contrast color (bluish violet) against the main substance of the dentine (yellowish red).

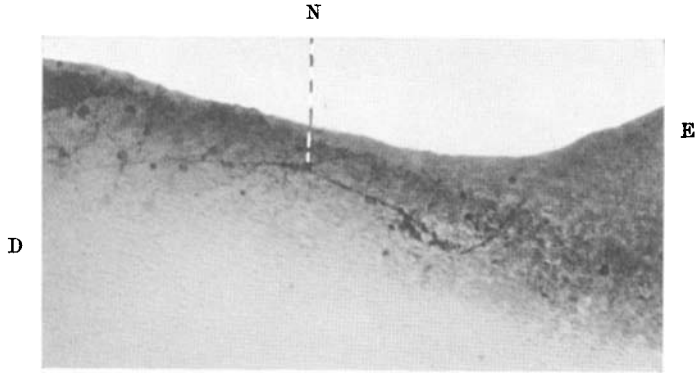

Fig. 52. Celloidin section from the dento-enamel junction in a maxillary front tooth removed from a 33 year old man. Staining: Martinotti. Magnification × 140. E. Enamel. D. Dentine, and N. Bundle of non-medullary nerve fibers with nerve endings (?).

3. ERASQUIN's modification of RIO HORTEGA's silver impregnation method for staining reticular fibers.

4. BODIAN's method for staining nerve fibers and nerve endings. This method differs from other nerve silver staining methods inasmuch as it is suitable for paraffin and celloidin sections.

Other staining methods, without particular interest for this present work, have also been used whenever necessary.

In examining enamel sections prepared by the above mentioned methods, the author was unexpectedly struck by the rich presence of reticular fibers in the dental enamel (vide Figs. 1 and 2). With regular intervals the enamel tufts radiate into the enamel. They taper off into exceedingly fine threads which more properly might be called vessels. Between the regularly parallel enamel fibers are observed fine membranes. Thick enamel lamellae run through the entire enamel without reduction in size. The cross stripes, which of old have been observed in the ground sections, are recognized as thin membranes, more distinctly stainable and easier to follow than in the ground sections of old.

In sections where the enamel fibers have been damaged or loosened in the embedding or the mounting, certain structure appear which have not been described earlier. Deeply staining threads are seen running straight across the enamel fibers, often in a wide curve. These threads branch off dichotomously and are often provided with small dilatations at the branching points. On the threads themselves are seen swellings which sit very close. Here and there these threads are joined into larger bundles in

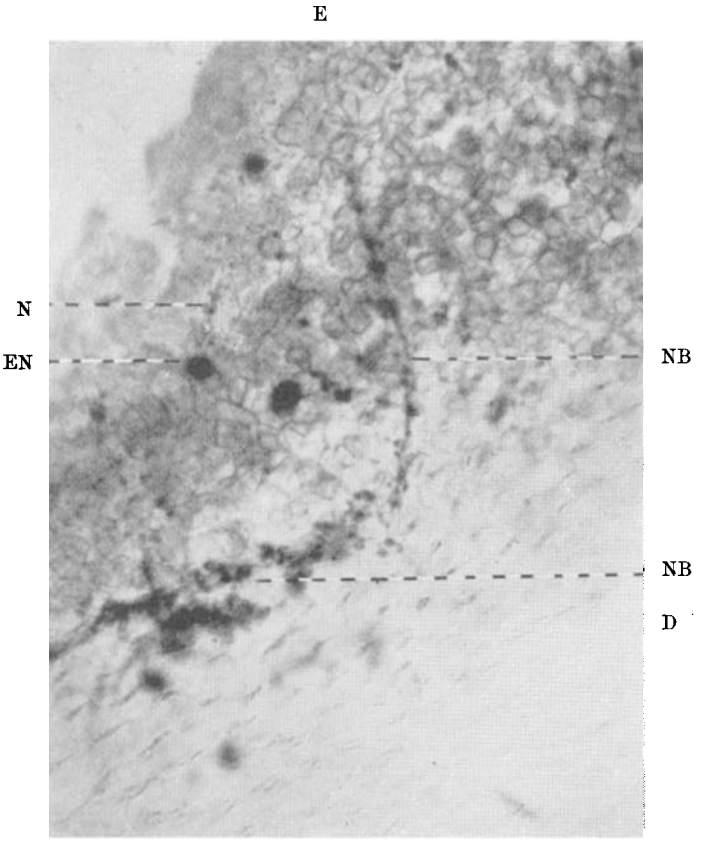

Fig. 53. Higher magnification of Fig. 52, × 580. E. Enamel. NB. Nerve bundle of non-medullary fibers with swellings at the branching points and endings (?). D. Dentine, EN. Nerve ending (?), and N. Nerve fibers (?).

which the separate threads are distinguishable. These threads seem to end in lump-like formations of various appearance and seemingly of different sizes.

Although these above mentioned structures are encountered in the whole enamel substance, they appear to be more abundantly present in the surface areas in the enamel and the parts adjoining the dentine border. It is interesting that through these areas run the two basement membranes of the enamel, namely the dento-enamel junction membrane and the enamel membrane. Figures 52—60 illustrate this point. If one compares the pictures of the enamel shown here, with the descriptions and illustrations in

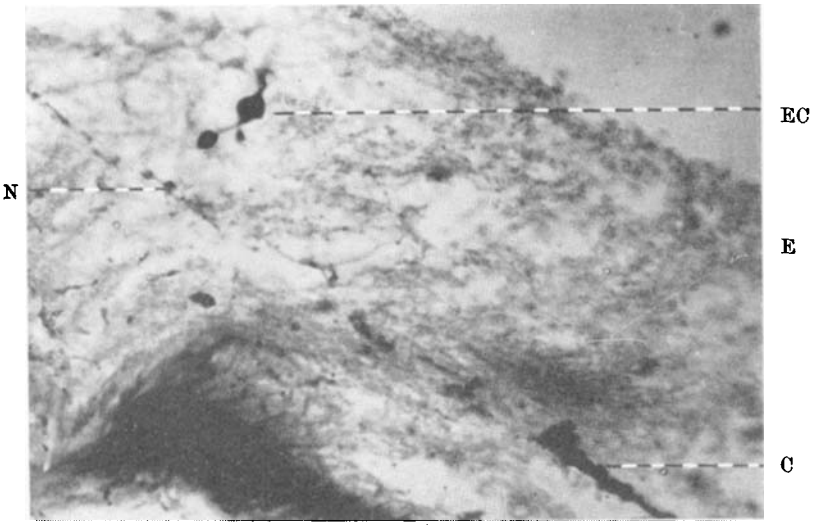

Fig. 54. Paraffin section from premolar of a 12 year old boy. Staining: Martinotti. Magnification × 580. EC. Three nerve endings (?) communicating with each other. E. Enamel. C. Conglomerate of nerve fibers and nerve endings, and N. Nerve fiber with swellings (?).

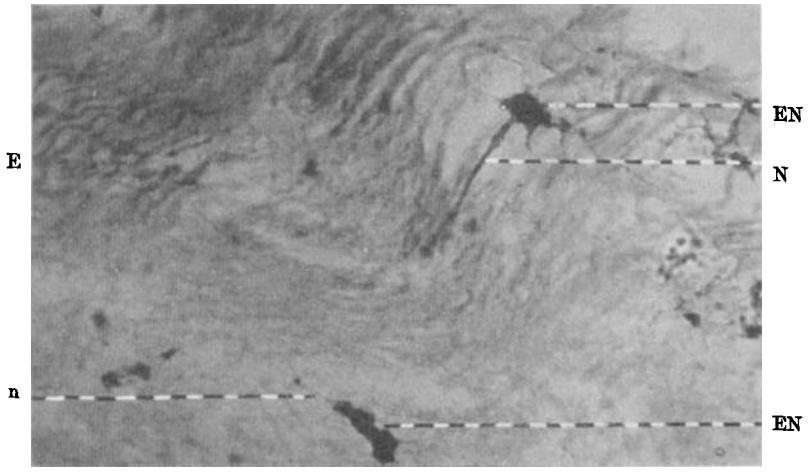

Fig. 55. Section same as Fig. 54. Same magnification. EN. Nerve ending (?). N. Nerve fiber bundle (?), n. nerve fiber (?), and E. Enamel.

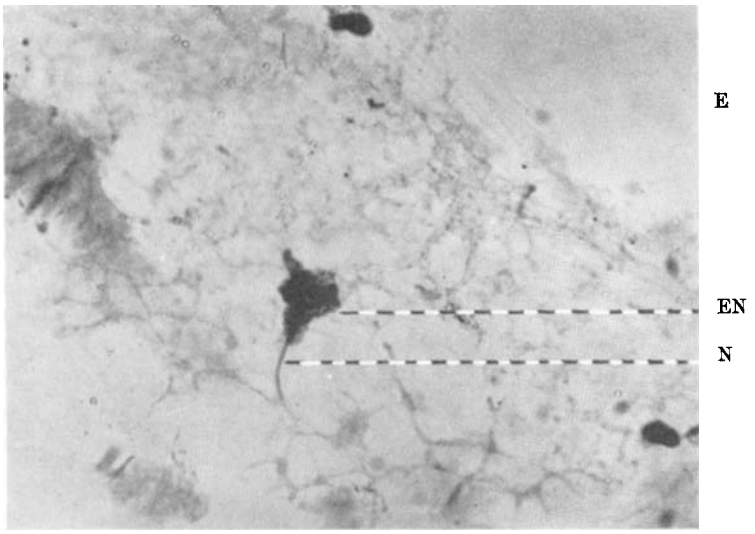

Fig. 56. Section same as Fig. 54. Same magnification. EN. Nerve ending(?). N. Nerve fiber(?), and E. Enamel.

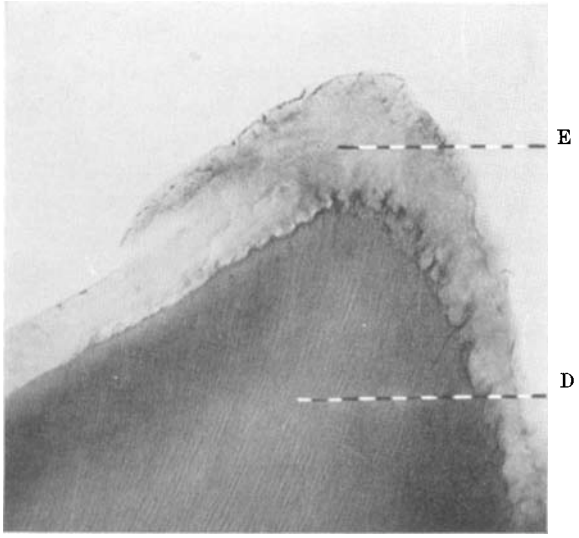

Fig. 57. Celloidin section of premolar from a 13 year old boy. Staining: Martinotti. Magnification × 140. E. Enamel, and D. Dentine.

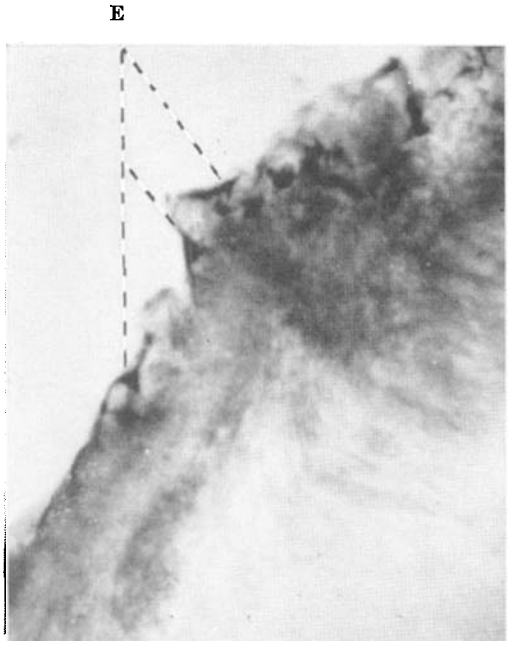

Fig. 58. Section same as Fig. 57. Magnification × 580. EN. Nerve endings(?) in the enamel surface — the enamel cuticle.

literature about the end ramif.cations of the vegetative nervous system, and adds the author's observation that the fibers and endings described by him are stained positive with staining methods considered to be extremely reliable, then one may be permitted to suppose that the afore-mentioned enamel structures and their terminations are non-medullary nerve fibers. Their likeness is especially striking with the sympathetic nerve endings in the blood vessel adventitia, such as these are described in literature (vide Figs. 61—64).

It seems rather apparent that these newly discovered structures in the dental enamel could be nerve elements. One knows that it is impossible on morphological basis to decide if they belong to the cerebrospinal or the vegetative nervous system. If one takes into consideration the familiar fact that no sensation has been ascribed to the dental enamel, then it becomes difficult to overlook the circumstance that the just mentioned nerve elements in the dental enamel could be referred to the vegetative nervous system.

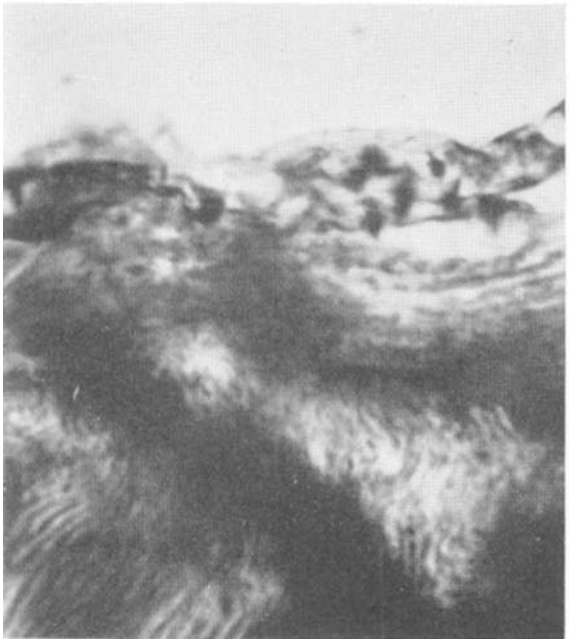

Fig. 59. Section from the same series as Fig. 57. Staining: Weigert's hematoxylin and v. Gieson. Magnification × 580. EC. Enamel cuticle with numerous nerve endings(?); connected with each other by means of fine nerve fibers and E. Enamel.

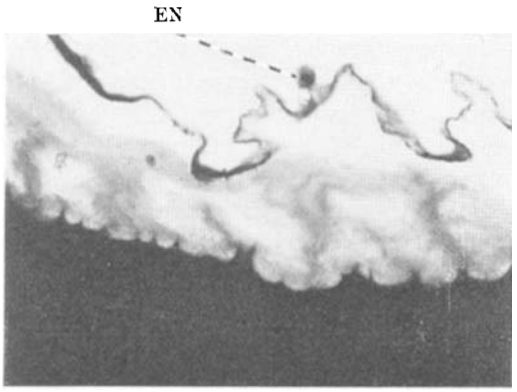

Fig. 60. Section from the same series as Fig. 57. Staining: Weigert's hematoxylin and v. Gieson. Magnification × 140. EN. Nerve ending(?), which, when the section was mounted turned out from the enamel cuticle. E. Enamel, and D. Dentine.

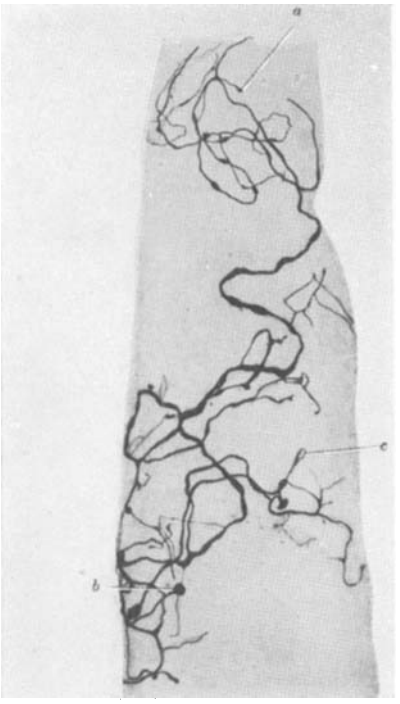

Fig. 61. Drawing reproduced from Stöhr (10). Vegetative nerve ramifications in the wall of a vein from human pia mater. Schultze's NaOH silver method. a. and b. nerve endings, and c. nerve ending with loop-like formation. Magnification × 200.

Since these supposed nerve elements are found in a tissue wherein no other organic substance than reticular elements heretofore have been described, it is not improbable that the vegetative nervous nutrient supply of the dental enamel is functionally connected with the reticular fibers, whose function it is to regulate the circulation in the ultracapillaries in the enamel.

From what has been said above, one may be permitted to draw the following conclusions:

The argyrophile connective tissue fibers — the reticular fibers — have for some time been known to form a morphological unit with the circulatory system. They have in their larger stems a lumen which is visible under the ordinary microscope. In other words, these stems look like tubes or vessels. They can be injected with colloidal injection substance by the technique used of old

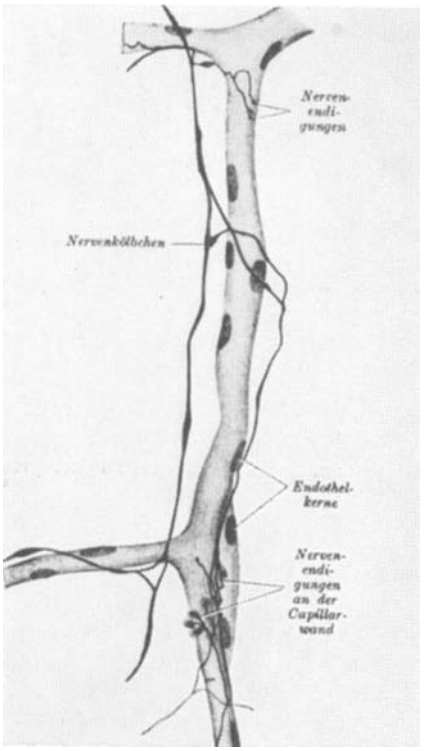

Fig. 62. Drawing reproduced from Stöhr (10). Capillary nerves from human pia mater. Schultze's NaOH silver method.
Magnification × 800.

to confirm the extent of the blood vessel system. They are connected in the enamel with fibers and other structures which possibly might belong to the vegetative nervous system and function as regulators of the circulation.

The question is now if blood plasma can be demonstrated in the ultracapillaries formed by the argyrophile connective tissue fibers? It might be well to reflect on certain phenomena which suggest the existence of the ultracapillaries through which blood plasma circulates, such as the formation of blebs after burns and trauma, as well as other non-sanguinous extravasations.

This problem can probably only be cleared up by proving the existence of blood plasma in and around the ultracapillaries in the dental enamel, since this tissue in all respects is most suitable

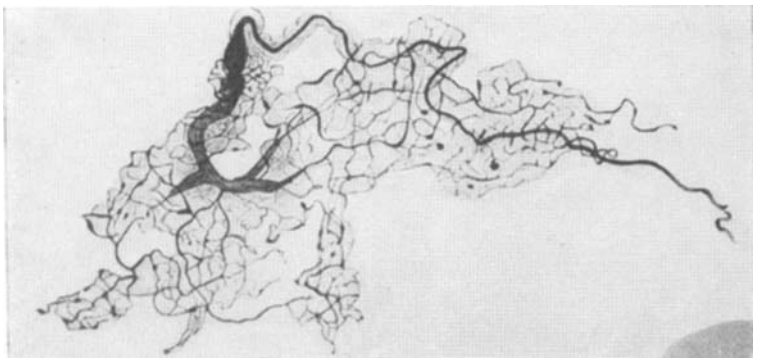

Fig. 63. Drawing reproduced from Stöhr (10). Bundle shaped nerve ending from human pericardium. Staining: Marynoff's methylene blue.

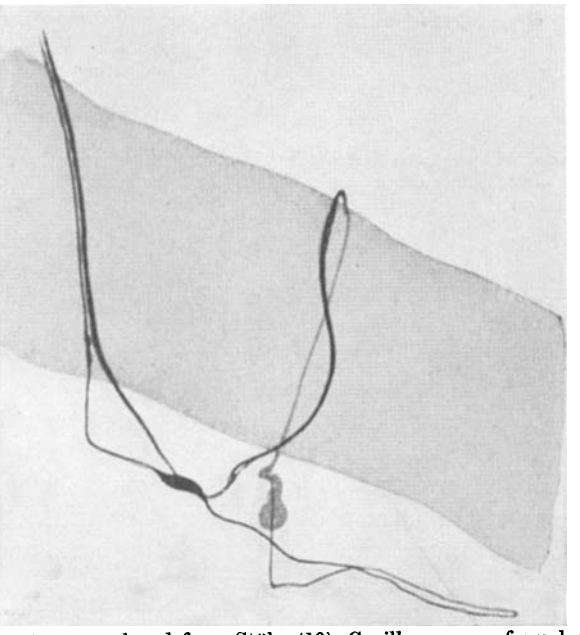

Fig. 64. Drawing reproduced from Stöhr (10). Capillary nerve from human pia mater. Schultze's NaOH silver method. Magnification × 1000.

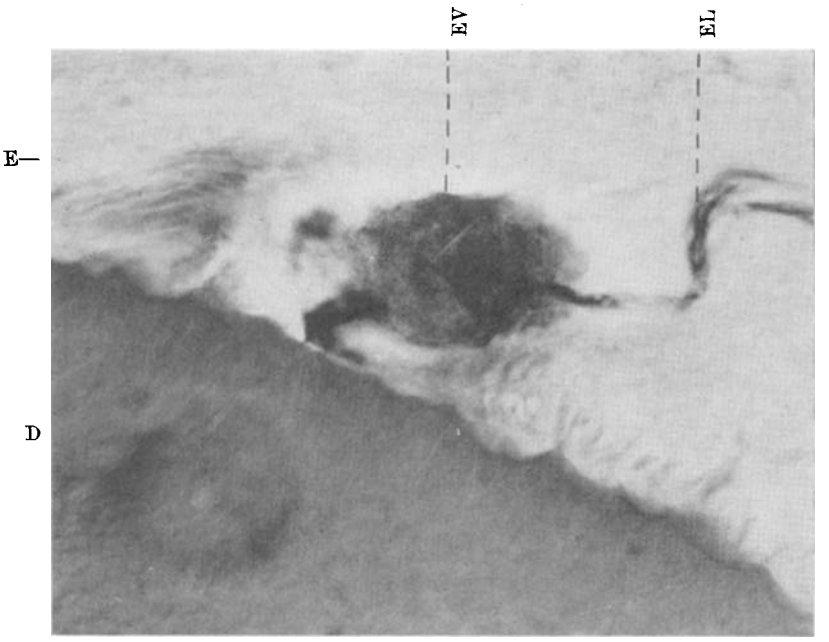

Fig. 65. Celloidin section from clinically intact surface of incisor from a 23 year old man. Staining: v. Gieson, Magnification × 580. D. Dentine. E. Enamel. EV. Extravasation in the enamel, and EL. Enamel lamella.

for a primary study of the ultracapillaries. All direct attempts to prove the existence of blood plasma in the dental enamel will meet with technical difficulties which cannot be mastered at present. The plasma derivative, fibrin, is, however, directly demonstrable with histological methods. The question is if fibrin can be demonstrated in the dental enamel?

The author has started an investigation of the pathogenesis of dental caries, in co-operation with BIRGER ÖSTMAN and FILIP PÅLSSON. With their permission he likes to show two illustrations obtained in these investigations.

Figure 65 shows a *van Gieson* stained section from an incisor of a 23 year old man. The dental portion under discussion is clinically intact. An extravasation is seen around an enamel lamella, which in the section has the typical yellowish-grey color for fibrous extravasation. Figure 66 shows an adjoining section of the same tooth, but stained with *Weigert's* fibrin stain. This

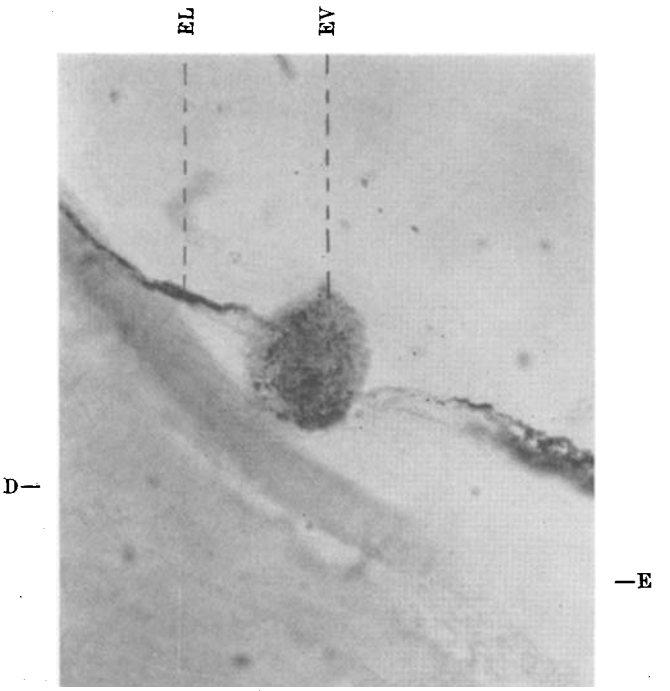

Fig. 66. Adjacent section to Fig. 72. Staining: Weigert's fibrin stain. Magnification × 580. D. Dentine. E. Enamel. EL. Thrombosed enamel lamella, and EV. Extravasation in the enamel, with fibrous threads. The enamel lamella is dislocated in mounting the section.

too gives a positive result and fibrin threads are even seen in the enamel lamellae.

These findings of fibrin in the enamel would seem to answer the author's question in the positive, that the ultracapillaries in the dental enamel represent vessels containing blood plasma. It is on this account that the author feels convinced that the circulatory system is incompletely investigated unless one takes into consideration the ultracapillaries of argyrophile substance. These would seem to emanate from the principal stems of the circulatory system — arteries, veins and capillaries — for the purpose of bringing the nutrifying blood plasma to the smallest units of the tissues.

Summary.

In an earlier series of investigations, the author has advanced the supposition that the circulatory system is not solely comprised of the heart, arteries, veins and capillaries. Experimental evidence has led him to believe that the circulatory system also should comprise the reticular fibers which he feels constitute a closed system of ultracapillaries.

The present study extends the experimental basis for this supposition of the existence of ultracapillaries. It deals mainly with the dental enamel which is essentially made up of reticular fibers. Proofs are advanced that these fibers originate in the adventitia of the capillaries of the dental pulp. That the reticular fibers of the tissues in general have their roots in the adventitia of the capillaries has previously been pointed out be several authors. Microscopic sections are presented by the author in which the largest fibers of the dental enamel stroma — the enamel tufts — appear like tubes having a distinct lumen. By saturating the enamel with a colloidal dye, injected directly into a main artery, the author has attempted to prove that the network of reticular fibers in the system consists of ultracapillaries through which the so-called paracapillary nutrition is brought about to the very smallest units of the tissues.

The principal experimental handicap in demonstrating the ultracapillary circulation in the dental enamel, seems to be the preparation of such colloidal dyes which might be pressed into the ultracapillaries by means of the capillary pulse. The author has succeeded in finding such a colloidal dye. This also contains a coarse-dispersive component which, by its occluding effect on the capillaries, presses the colloidal fluid into the ultracapillaries by means of the capillary pulse. The liquid contains hemoglobin and hemolyzed red blood cells. After being injected intravascularly, the dye stains the dental enamel deeply and is especially localized to the so-called *Hunter-Schreger lines*, the bands of which are connected with a greater concentration in every other band of coarser network of reticular fibers.

The intravascular injection of this colloidal dye penetrates extensively the kidneys, liver, spleen, muscle, tendon, adipose tissues and the interstitial connective tissues where it concentrates in definite localities. Silver impregnation control sections show that these localities contain strongly developed reticular

fibers. The presence of the dispersed hemoglobin is proved by the ortho-tolidin method devised by SJÖSTRAND.

By means of an original decalcification method, the author demonstrates that the enamel stroma contains besides reticular fibers, certain structures which might belong to the vegetative nervous system.

Finally, the author shows the presence of fibrin in an extravasation around an enamel lamella in clinically intact enamel.

Résumé.

A l'occasion d'une série de recherches antérieures l'auteur a émis l'hypothèse que le système circulatoire n'est pas constitué seulement par le cœur, les artères, les veines et les capillaires. Ses expériences l'ont conduit à la conception que le système circulatoire doit comprendre également les fibres grillagées, qui constitueraient un systeme fermé d'ultracapillaires.

L'étude présentée ici élargit la base expérimentale sur laquelle se fonde cette idée de l'existence des ultracapillaires. Elle s'occupe avant tout de l'émail dentaire, qui est constitué principalement par des fibres grillagées. L'auteur apporte des preuves que ces fibres proviennent de l'adventice des capillaires de la pulpe dentaire. Déjà précédemment il a été relevé par plusieurs auteurs que les fibres grillagées des tissus en général ont leur origine dans l'adventice des capillaires. L'auteur présente des coupes microscopiques où les fibres les plus grossières du stroma de l'émail — les fuseaux d'émail — apparaissent comme des tubes avec une lumière nettement visible. En saturant l'émail avec une substance colorante de nature colloïdale, injectée directement dans une artère principale, il a essayé de prouver que le réseau de fibres grillagées du système, forme des ultracapillaires, par lesquels est rendue possible la nutrition dite paracapillaire des unités infinitésimales des tissus.

La difficulté expérimentale la plus grande quand il s'agit de mettre en évidence la circulation ultracapillaire au sein de l'émail semble résider dans la préparation de colorants colloïdaux aptes pour qu'ils puissent être chassés par le pouls capillaire dans les ultracapillaires. L'auteur a réussi à trouver un colorant colloïdal répondant à ces exigences. Ceci contient aussi une composante à grains plus gros, qui, par son action occlusive sur les capillaires, a pour effet d'amener le pouls capillaire à pousser le liquide

colloïdal dans les ultracapillaires. Le liquide contient de l'hémoglobine et des hématies hémolysées. Après l'injection dans les vaisseaux le colorant colore profondément l'émail, et se dépose spécialement dans ce qu'on appelle *les stries de Schreger*.

Après l'injection dans les vaisseaux ce colorant colloïdal pénètre dans le tissu paracapillaire des reins, du foie, de la rate, des muscles, des tendons, du tissu adipeux et du tissu conjonctif lache, où il se concentre en des endroits déterminés. Des coupes de contrôle imprégnées par la méthode à l'argent montrent que ces endroits sont en majeure partie ceux où existent des fibres grillagées fortement développées. La présence de l'hémoglobine dispersée est mise en évidence par la méthode de l'orthotolidine indiquée par SJÖSTRAND.

Grâce à un procédé spécial de décalcification l'auteur démontre que l'émail dentaire, en plus des fibres grillagées, contient certaines structures qui pourraient appartenir au système nerveux végétatif.

Finalement l'auteur décèle la présence de fibrine dans une extravasation entourant une lamelle d'émail, dans un cas où celui-ci était cliniquement intact.

Zusammenfassung.

In einer früheren Arbeit hat Verf. die Vermutung aufgeworfen, dass das Kreislaufssystem nicht nur aus Herz, Arterien, Venen und Kapillaren besteht. Experimentelle Erfahrung hat ihn zu der Auffassung geführt, dass das Kreislaufssystem auch die Retikulinfasern umfassen muss, die ein geschlossenes System von Ultrakapillaren darstellen.

Die vorliegende Untersuchung erweitert die experimentelle Basis dieser Annahme der Existenz von Ultrakapillaren. Sie behandelt vorwiegend den Zahnschmelz, der hauptsächlich aus Retikulinfasern aufgebaut ist. Es werden Beweise dafür vorgelegt, dass diese Fasern von der Adventitia der Kapillaren der Zahnpulpa herstammen. Dass die Retikulinfasern der Gewebe im allgemeinen der Adventitia der Kapillaren entstammen, ist schon früher von mehreren Autoren betont worden. Verf. gibt mikroskopische Schnitte, in denen die gröbsten Fasern des Schmelzstromas — die Schmelzbüschel — als Röhren mit deutlicher Lichtung erscheinen. Durch Sättigung des Zahnschmelzes mit einem kolloidalen Farbstoff, der direkt in eine Hauptarterie ein-

gespritzt wird, hat Verf. versucht zu beweisen, dass das Netzwerk von Retikulinfasern in dem System aus Ultrakapillaren besteht, durch welche die sog. parakapillare Ernährung der allerkleinsten Einheiten der Gewebe ermöglicht wird.

Die grösste experimentelle Schwierigkeit der Darstellung des ultrakapillaren Kreislaufs im Zahnschmelz scheint in der Herstellung solcher kolloidalen Farben zu liegen, dass sie durch den Kapillarenpuls in die Ultrakapillaren hineingepresst werden können. Es ist Verf. gelungen, solch eine kolloidale Farblösung zu finden. Diese enthält auch eine grobdisperse Komponente, die durch ihre Verschlusswirkung auf die Kapillaren so wirkt, dass der Kapillarenpuls die kolloidale Flüssigkeit in die Ultrakapillaren treibt. Die Flüssigkeit enthält Hämoglobin und hämolysierte rote Blutkörperchen. Nach Einspritzung in die Gefässe färbt der Farbstoff den Zahnschmelz tief und schlägt sich besonders nach den sog. *Hunter-Schreger'schen Linien* nieder.

Nach Einspritzung in die Gefässe dringt dieser kolloidale Farbstoff in grosser Ausdehnung in die parakapillären Gewebe in Nieren, Leber, Milz, Muskeln, Sehnen, Fettgewebe und interstitielle Bindegewebe ein, wo er sich an bestimmten Stellen konzentriert ansiedelt. Silberimprägnierte Kontrollschnitte zeigen, dass diese Stellen kräftig entwickelte Retikulinfasern enthalten. Die Gegenwart des Hämoglobins in der Gewebe wird durch die von SJÖSTRAND angegebene Orthotolidin-Methode bewiesen.

Mittels einer besonderen Entkalkungsmethode weist Verf. nach, dass das Zahnschmelzstroma ausser Retikulinfasern gewisse Strukturen enthält, die dem vegetativen Nervensystem angehören können.

Schliesslich zeigt Verf. die Gegenwart von Fibrin in einem eine Schmelzlamelle umgebenden Extravasat in klinisch intaktem Schmelz.

The author wishes to express his appreciation to Dr. Anders Wassén, director of the Bacteriological Laboratories, and Dr. Olof Forselius, director of the Pathological Laboratories at the Sahlgren Hospital in Gothenburg, for the privilege of carrying on these investigations in their respective departments and for their always willing counsel and guidance.

He is likewise indebted to Hospital Director Hugo Höglund for his kind permission to work in these laboratories.

The microphotographs have been taken at the Cerebral Laboratory at the Lillhagen Hospital by the kind permission of the hospital superintendent, Dr. Sven Hedenberg.

The histological specimens have been prepared by my technical assistant, Miss Brita Wikblad.

When preparing the colloidal injection substances and in making the injections of the same into animals, the author has been ably assisted by the skilful laboratory nurse, Miss Pyrola Modig.

The translation has been done by Miss Barbro Dyhlén, B. A. and kindly revised by Professor Dr. Konrad Birkhaug.

To all these persons the author extends his deepfelt appreciation.

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