

Trabecular Barriers (FRENULAE) of the Extracapsular Clefts (38563)

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Tissues tend to be organized into clusters of parenchymal cells, each such module being enclosed in a fibrous connective tissue capsule (1) (Fig. 1). The capsule is a barrier which can inhibit the emigration of cells from the cluster, and can exclude extraneous cells or materials from entering into its territory.

The services of the structure, including nerves, arteries, veins and lymphatics pass through the clefts between adjacent capsules. Histologically, these clefts (trabeculae) appear as relatively open avenues. An open avenue would be vulnerable to the intrusion of foreign materials or to the immigration of extraneous cells.

We have had extensive experience with the intraparenchymal injection of particulate materials and of liquid plastic (2-4). The fibrous capsules exclude these injected materials with the result that the injectate is limited to the extracapsular clefts.

The present study examines anatomical and functional arrangements of the capsules and the extracapsular clefts that may impede the spread of intraparenchymal injections, or that may determine the direction of flow of the injected material.

Methods and hypothesis. More than 100 tests were made on skeletal muscles of the hind limbs of 10 dogs. A two-injection technique was used.

1. The artery to the leg was cannulated and perfused with 100 ml of Ringer's solution containing 1-4% potassium ferrocyanide. Some of these ions filtered across the blood capillary endothelium into the extracapillary fluids, from which they could filter or diffuse across the capsular wall, ultimately to enter the extracapsular clefts.

2. Ringer's solution containing 1-4% ferric chloride was then injected intramuscularly into the parenchyma (not intravascularly). Previous studies have shown that except for the relatively few capsules impaled on the needle, such intramurally injected materials enter only the extracapsular

clefts, and into the lymphatic vessels that reside in these clefts.

Contact of ferric ions with ferrocyanide ions precipitates the highly insoluble Prussian blue.

Tissue specimens were fixed in 10% formalin, sectioned, and stained with nuclear-fast red. By use of this dye, the only blue stain in the section was the Prussian blue.

Results. Ferrocyanide studies. Histological sections of specimens which received only the intra-arterial infusion of ferrocyanide exhibited no blue coloration. Similarly, tissues which received only the intraparenchymal injection of ferric ion exhibited none of the blue precipitate.

In tissues receiving both ions, the blue precipitate was evident in extracapsular clefts, on the walls of the capsules, in the media of some of the arteries and veins, and sometimes in the lymphatic vessels that traversed the trabeculae.

The blue precipitate was restricted to local areas and did not extend freely throughout the trabeculae. A sharp margin of the blue precipitate demarcated the injected portion of the cleft from the immediately adjacent color-free portion of the cleft. Microscopic examination showed that collagenous sheets (frenulae) extended across the clefts to attach to the capsules on opposite sides of the cleft. (Figs. 2, 3) These frenulae limited the extent of deposition of the precipitate. Frenular sheets more proximal to the site of injection were apparently ruptured by the injection. Blue depositions were present on the sheets. Filling of the lymphatic vessels of the cleft was apparent as a set of long lines of blue coloration in the trabeculae of the tissues.

Discussion. In previous studies, we have called attention to the isolating role of the fibrous capsules which enclose each cluster of parenchymal cells. Thus, each cluster of parenchymal cells is enclosed in a tough cap-

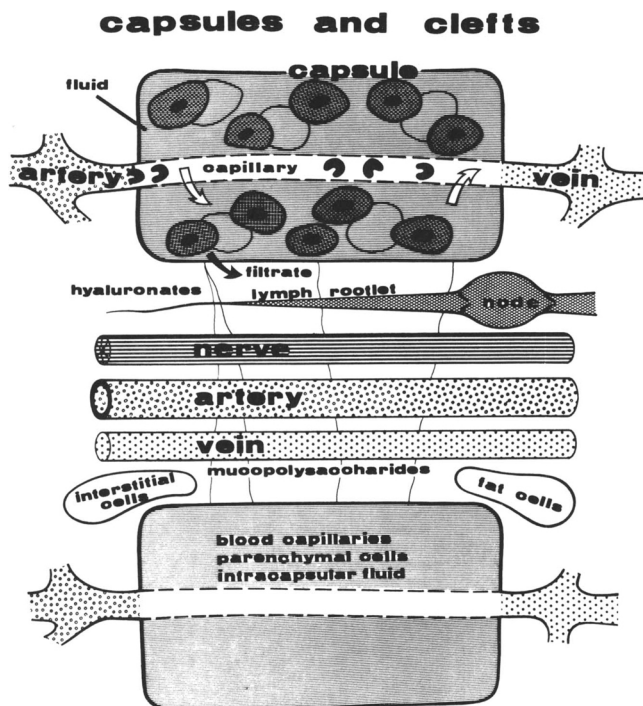


FIG. 1. Schema of tissue structure. A cluster of parenchymal cells (rounded bodies with nuclei, above) is perfused by a blood capillary. A capsule encloses each parenchymal cluster of perhaps 100 cells and many capillaries (only one is shown). Capillary ultrafiltrate fills the capsule. Some of this filtrate passes into and across the wall of the capsule to enter extracapsular clefts between adjacent capsules (middle portion of figure). The extracapsular cleft is the service route for blood vessels and nerves, and it is the site of origin of the rootlets of the lymphatic system. The cleft also is the locus of mast cells, interstitial cells, fat cells as well as ground substances (hyaluronates, mucopolysaccharides). Fine lines are shown between adjacent capsules to indicate the loci of frenalae.

sule of connective tissue which prevents the intrusion of injected foreign materials or the immigration of extraneous cells into the territory of the capsule. The capsule can also impede the emigration of the enclosed cells out of the capsule.

In studies utilizing the intraparenchymal injection of India ink, tantalum powder, barium sulphate paste, radiopaque fluids, or liquid plastic, we found that each of these materials tended to be limited to trabeculae near the site of injection. The extent of spread of the injected materials in the trabeculae increased with the volume of the injection and with the force used to introduce these materials. Some of the injected materials drained via the lymphatic vessels at the site, to lymph nodes usually at a distance from the sites of injection. The frenal barriers in the trabeculae therefore appeared to impede the widespread infiltration of

extraneous materials into the tissues. This effect was also demonstrated in the tissues injected with liquid (nonhardening) plastic containing pigment particulates. The particles were intruded along the trabeculae. At some point, the frenal sheets were of sufficient strength to impede the further extension of the injectate.

The two-phase injection demonstrates that sheets of connective tissue that extend across the trabeculae limit the movement not only of the particles, but also of the flow of fluids and solutes. Thus, some of the ferrocyanide ion infused through the artery filtered to the extracapillary fluid of each perfused capsule. The intraparenchymally injected Ringer's solution containing the ferric ion was injected into the trabeculae and this spread through the connecting trabeculae.

The results show that fibrous connective

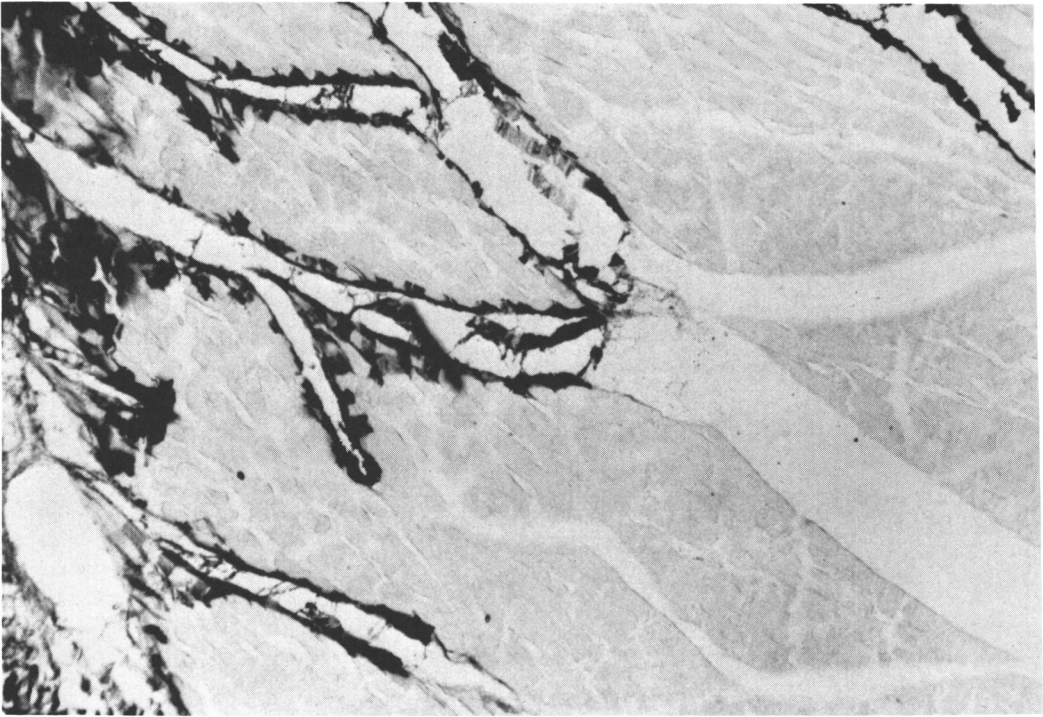


FIG. 2. Prussian blue precipitate on capsular walls bordering the trabeculae. The ferric ion, injected into muscle at upper left penetrated through the trabecular to the frenulae in the middle of the figure. Obstructions to the flow of the intraparenchymal injection are shown at several sites. 25A Red Filter 40X.

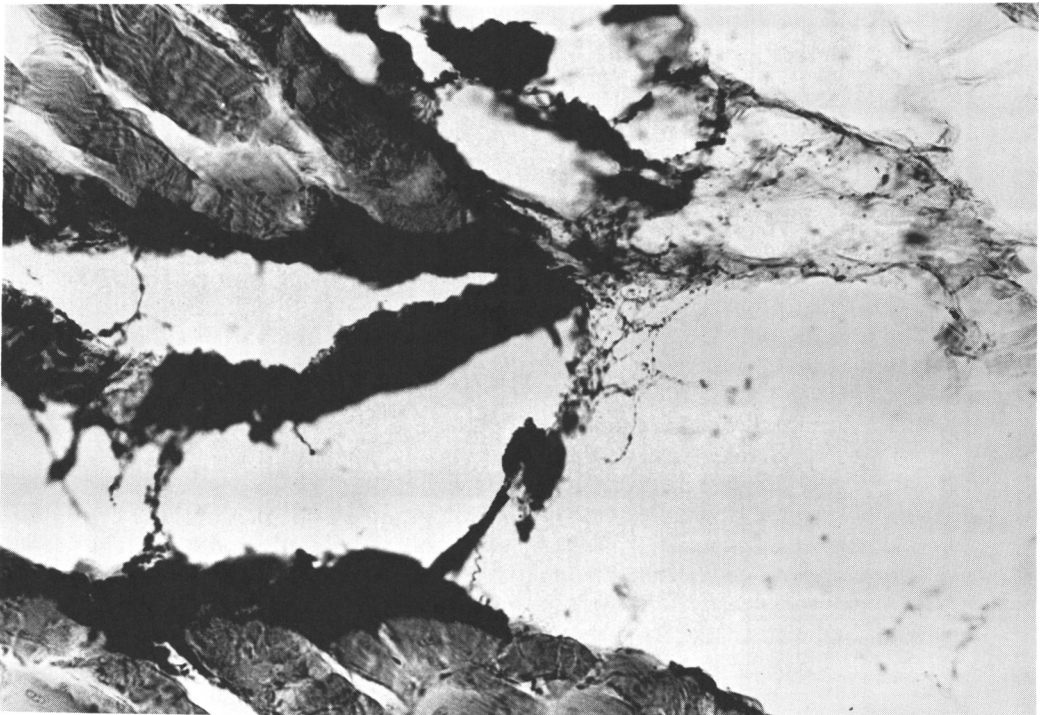


FIG. 3. Higher power of central portion of Fig. 2 showing two frenulae (below) which obstruct free flow into the trabeculae. Frenulae at the right beyond the locus of pigmentation cannot be seen. Lymphatic vessels appear at the upper center. Capsules up to the point of limitation of permeation of ferric ions are deeply stained with Prussian blue. 25A Red Filter 40X.

tissue sheets form gate-like barriers in the trabeculae. Despite their apparent delicacy, these remarkable fibrous sheets are sufficiently strong to impede the spread of extraneous materials or cells. The frenulae may thereby serve important localizing functions which prevent bacteria or other foreign materials from spreading in the tissue.

The foregoing properties may clarify certain questions concerning the characteristics of the casts which we have been producing by the injection of liquid plastic. The capsules prevent introduction of the injected material into the domain of the enclosed parenchymal cells. The frenulae impede the intrusion of the plastic into the extracapsular clefts. The injected material stretches and ruptures some of these frenulae. The extent of penetration of the injectate into the trabeculae probably varies with the force applied to the plunger of the syringe, and with the volume introduced. Injection of plastic into a compartment introduces this material only into the trabeculae inside the fibrous supercapsule which serve as the boundary of the compartment.

The frenular sheets extend across the trabeculae and mechanically bind adjacent capsules together. The extent of the movement affects the length and strength of the sheets and probably thereby determines the relative freedom of movement of adjacent

capsules with respect to each other. Thus, short sheets will limit the movement of the adjacent capsules. Looser tissues probably have longer frenulae between adjacent capsules.

Summary. Ferrocyanide was perfused through the vascular system of the isolated legs of 10 dogs. Ferric ion was then injected intramuscularly (not intravascularly) into the leg. The injected materials spread along the trabeculae of the tissues, rupturing the sheets of connective tissues which bind adjacent capsules together.

Precipitation of ferri-ferrocyanide on specific barriers in the extracapsular clefts in positions showed that sheets of fibrous connective tissue (frenulae) impeded the movement of materials in the trabeculae. The barriers consist of sheets of connective tissue which insert on adjacent fibrous capsules. The capsules enclose each cluster of muscle, glandular or other cells. These barriers appear to function in conjunction with the lymphatic drainage system.

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1. Rodbard, S., *Curr. Mod. Biol.* **3**, 27 (1969).
 2. Rodbard, S., and Taller, S., *Med. Exp.* **19**, 65 (1969).
 3. Rodbard, S., McMahon, N. J., and Denk, M., *Obstet. Gynecol.* **38**, 171 (1971).
 4. Rodbard, S., *Amer. J. Cardiol.* **32**, 877 (1973).
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