

MORPHOLOGICAL CHANGES IN VEINS AND LYMPHATICS OF THE DOG'S LIMB  
AFTER LIGATION OF THE MAIN VESSELS

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Experimental and clinical studies of the state of the venous and lymphatic systems of the limb after ligation of the main limb veins [1, 3, 5, 7] have shown the general principles governing the dynamics of the process and the interconnected and combined changes in veins and lymphatics. According to one opinion [6], because of the development of the collateral circulation at various times after the beginning of the experiment the normal microcirculation and the normal hemodynamics of the limb are completely restored (not later than 5-7 months after the operation). However, these observations do not fully correspond to the progressive clinical picture of chronic venous insufficiency and, consequently, they are not a sufficiently faithful model for extrapolation of the results to clinical practice. In addition, the lack of any accurate quantitative evaluation or mathematical analysis of the data leads to some ambiguity in the evaluation of the dynamics of the changes and their time course.

EXPERIMENTAL METHOD

Experimental venous insufficiency was studied in 38 male dogs weighing from 6 to 10 kg at different times after ligation of the main veins of the left hind limb at the level of the inguinal ligament. The right hind limb served as the control. Between 2-5 and 175-180 days after the operation the animals were killed (after preliminary amputation of the hind limbs). Fragments of thigh tissues (covering the whole area of its cross-section, namely  $3.0 \times 3.5$  cm) were embedded in celloidin-paraffin. Histological survey sections were prepared from them and stained with hematoxylin and eosin and with picrofuchsin by Van Gieson's method. The number of veins, venules, and capillaries per unit area and their diameter were calculated. To study the state of different components of the venous and lymphatic systems of the limbs small fragments of skin, subcutaneous tissue, and muscles were fixed in 10-12% neutral buffered formalin, after Lillie, and embedded in paraffin wax. Sections were stained with hematoxylin and eosin, with picrofuchsin by Van Gieson's method, and with fuchselin by Weigert's method. To detect accumulation of glycosamines, Steedman's method of staining with alcian blue was used. The state of the argyrophilic skeleton of the limb tissues of the venous and capillary walls was determined by impregnation by Foot's method. For a more exact assessment of the degree of vascularization of the tissues and development of the capillaries immediately before amputation of the limbs their vessels were filled with a solution of Direct black-3.

EXPERIMENTAL RESULTS

In the early stages (2nd-5th days) after ligation of the main veins acute insufficiency of the venous and capillary circulation was observed in the limb undergoing the operation. This was expressed as severe congestive manifestations and disturbances of capillary permeability with the development of edema and hemorrhages, mainly in the dermis and subcutaneous tissue. The congested and dilated capillaries, veins, and venules showed swelling of their endothelium and stretching of their muscular coats and elastic membranes. Meanwhile an associated response of the lymphatics and capillaries was observed, in the form of dilatation of the paravasal lymphatics and of the capillaries of the dermis and subcutaneous and inter-

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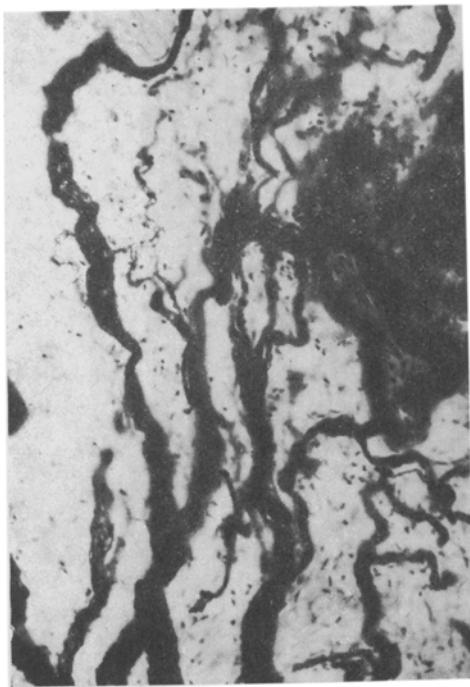


Fig. 1

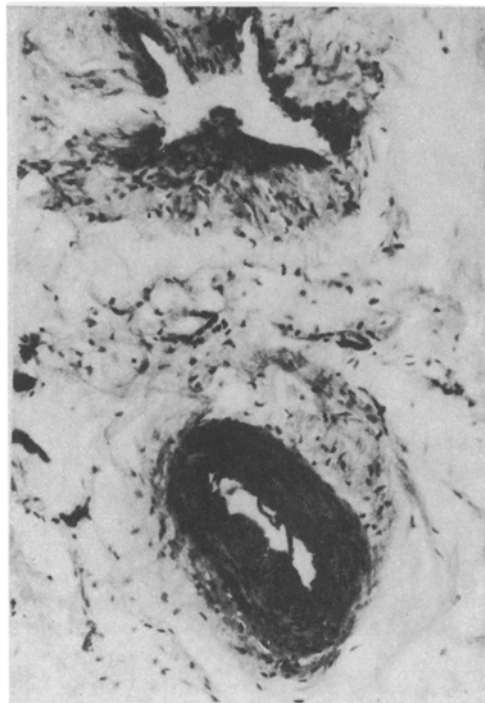


Fig. 2

Fig. 1. Increase in number of injected blood vessels and capillaries (30th day of experiment). Hematoxylin-eosin, 260  $\times$ .

Fig. 2. Sclerosis of wall of a small vein with signs of hyperplasia of intima and endothelium and sclerosis of wall of a lymphatic (60th day of experiment). Picrofuchsin by Van Gieson's method, 90  $\times$ .

muscular tissue, which merged in certain places to give distinctive "coils" (Fig. 3). Meanwhile, in the subcutaneous tissue and dermis, avascular zones of sclerosis were found, and here and there islets of fatty areolar tissue were immured in the midst of scar tissue, often infiltrated by chronic inflammatory cells (Fig. 4).

From the 100th to the 180th day the defined stage of reorganization of the vascular venous and lymphatic system apparently is completed. In the subcutaneous, intermuscular, and deep connective tissue, major venous collaterals occur: large thick-walled veins with hyperplastic intima, hypertrophy of the muscular sheath and hyperelastosis and sclerosis of the walls by which used blood is unloaded. A similar unloading somewhat decreases venous congestion, although not sufficiently to completely eliminate it. This observation is supported by conservation in the subcutaneous and intermuscular tissue of a significant quantity of dilated, long, coiled, and (in some places) "knotted" varicosoid deformation of superficial veins, and by capillaries filled by Direct black-3 (which indicates their function); other evidence is the moderate interstitial edema.

Doubtlessly, venous-lymphatic congestion is in the final analysis the important factor underlying the existence of the pathological process.

In conclusion it must be pointed out that the basic principles of the dynamics of the venous and lymphatic systems of the limb after ligation of its main veins could be successfully established by the use of this experimental model. They include the development not only of manifestations of venous and lymphatic stasis, but also of compensatory and adaptive processes, namely the formation of a collateral venous circulation and structural changes in the vessel wall of the "collateral" veins.

The time course of the structural changes in the vascular network can be subdivided into three stages. In the acute stage I (the first 30-35 days of the experiment) acute insufficiency of the venous and lymphatic drainage is observed without any evidence of compensatory reactions. In stage II, starting with the 50th-60th day, the gradual formation of compensatory and adaptive processes is observed, with the formation of collaterals of



Fig. 3

Fig. 3. Numerous injected vessels and capillaries in subcutaneous tissues (80th day of experiment). Hematoxylin-eosin, 90  $\times$ .

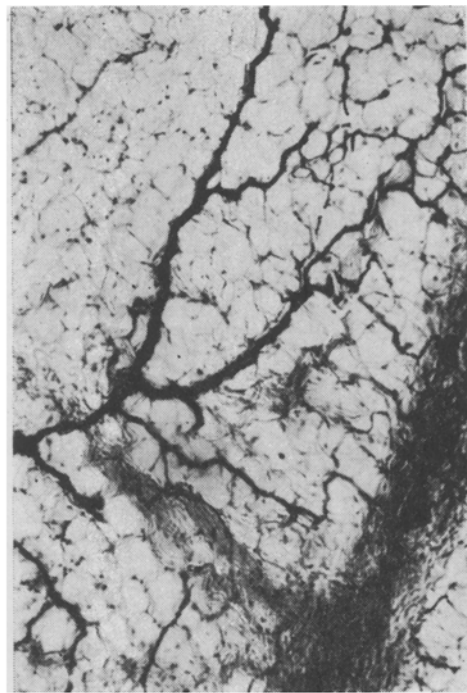


Fig. 4

Fig. 4. Capillaries injected with Direct black in subcutaneous tissue (99th day of experiment). Hematoxylin-eosin, 90  $\times$ .

the main veins. In stage III, starting from the 99th-100th day, a definite equilibrium is observed between compensatory processes and venous stasis.

When disturbances of venous and lymphatic drainage are simulated by the method described above, typical signs of chronic venous insufficiency such as long-lasting disturbances of the microcirculation and permeability of the lymphatic wall, chronic dilatation of the capillaries, hyperplasia of the intima, hyperelastosis of the venous wall, with its sclerosis and varicose deformation in some places, the development of chronic inflammation, and dystrophy and atrophy of muscle fibers arise in the limb. These developments are evidence of some degree of insufficiency of compensatory process, and in the opinion of some workers [3, 4], this lies at the basis of progression of chronic venous insufficiency.

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