

SPLENIC FIBROSIS IN PATIENTS WITH CHRONIC SCHISTOSOMIASIS

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In human schistosomiasis, chronic severe infections may lead to development of the hepato-splenic form of the disease, characterized by a great enlargement of the spleen. The consequent hypersplenism, with the associated hematological and endocrinological manifestations is one of the major aspects of the morbidity of this disease, particularly in younger patients.

The etiology of schistosomal splenomegaly is simultaneously related to various factors. Classical studies on human schistosomiasis mansoni have underlined the relationship between the establishment of portal hypertension and enlargement of the spleen (E. Koppisch, 1941, *Puerto Rico J. Publ. Health Trop. Med.*, 16: 395-455). Schistosomal splenomegaly is thus frequently classified as "fibro-congestive" (e. g. A. Bagshawe, 1970, *Brit. J. Haematol.*, 19: 729-737.). In a comparative study of patients from 5 to 21 years, A. Magalhães Filho and E. Coutinho Abath (1960, *Rev. Inst. Med. Trop. São Paulo*, 2: 251-259) concluded that the schistosomal splenomegaly may be initially related to lymphoid and reticular hyperplasia, while in later stages the congestion and fibrosis are more pronounced. In subsequent studies, Z. Andrade and S. G. Andrade (1965, *Rev. Inst. Med. Trop. São Paulo*, 7: 218-227) and A. E. Dumont et al. (1975, *Amer. J. Pathol.*, 78: 211-220) have stressed upon the proliferative character of schistosomal splenomegaly, and the former authors have indicated that the fibrosis of splenic cords was seldom seen. Recent studies have described the modification of extracellular matrix in splenic tissue of schistosomal patients (R. Borojevic & J. A. Grimaud, 1980, *Cell. Mol. Biol.*, 26: 247-250). Simultaneously, a secondary myeloid function of schistosomal spleen involving neutrophil granulocytogenesis and thrombocytogenesis was described (R. Borojevic & E. A. Carvalho, 1981, *Cell. Mol. Biol.*, 27: 671-676.; R. Borojevic et

al., 1983, *J. Infect. Dis.*, 148: 422-426.). A correlation between the hematologic manifestations of schistosomal hypersplenism and modifications of splenic extracellular matrix have been suggested (R. Borojevic & E. A. Carvalho, *loc. cit.*).

Although frequently mentioned, the fibrosis of splenic tissue in schistosomal splenomegaly has deserved little attention. In the present work we have analyzed a series of 15 spleens, obtained at splenectomy of patients with chronic severe schistosomiasis mansoni, ranging from 16 to 57 years. They were compared with 5 spleens obtained at autopsy of patients with pathologies unrelated with schistosomiasis. Tissues were fixed and studied in light and electron microscopy following treatment with standard histological and ultrastructural preparative methodology. A particular attention was given to modifications of connective tissue and its extracellular matrix.

The general organization of the splenic capsule and the trabecular framework was conserved in schistosomal spleens. However, both structures were thicker than in normal spleens, corresponding to the increased mechanical functions due to the splenomegaly and the increased intrasplenic blood pressure. Short silver-staining reticulin fibers extended perpendicularly towards the adjacent red pulp. Typical myofibroblasts with extensive perinuclear endoplasmic reticulum, indicating their high synthetic activity, were dispersed among thick collagen bundles and elastin deposits.

The extracellular matrix framework of the red pulp was strikingly modified in schistosomal spleens (Fig. 1A). In normal red pulp, the specific collagen stainings and silver impregnation demonstrated the extracellular matrix of blood vessel walls, and a delicate reticulin network. It corresponded mainly to fine reticulin fibers interspersed in splenic cords, most of which followed longitudinally the venous sinuses, and in lesser degree to basement

membrane circumferential ribs surrounding the myoendothelial cells lining the venous sinuses. In schistosomal spleens, the reticular network was remarkably dense. The circumferential ribs of splenic sinuses were heavily ornated with reticulin stains. They were frequently interconnected by perpendicular or oblique fibrils. They formed a continuous meshwork that substituted the regular ladder-like sequence of ribs and slits characteristic of basement membranes in normal splenic sinus walls. The reticular network of thickened splenic cords was dense and ramified (Figs. 1 B and D).

All the blood vessels of the red pulp were surrounded by thickened extracellular matrix. The adjacent sinuses were surrounded by continuous dense layers of matrix that obliterated completely the passage between sinuses and splenic cords. In these regions splenic cords were reduced to compact sinus walls so that an alveolated sponge-like fibrotic structure was formed, totally replacing the normal organization of the red pulp (Figs. 1 A and D). Around the major vessels, this fibrosed tissue occupied a considerable space corresponding in

thickness to several venous sinuses. The constitution of these perivascular fibrosed regions was progressive. In the younger patients of the studied series, the increased diffuse reticulin network of the red pulp was predominant, while in the older ones, in addition to diffuse fibrosis of splenic cords, the perivascular fibrosis became more pronounced. The electron microscopy depicted collagen deposits, associated most frequently to perivascular reticular cells and basement membranes of perisinusoidal myoendothelial cells. Initially, fibrous collagen deposits were seen associated with amorphous substance of basement membranes. Frequently, dense collagen bundles composed of irregular twisted fibers were interspersed in splenic cords. A clear gradient of collagen deposits was seen around blood vessels, the densest and the most organized deposits being always in close association with vascular structures. Plasma cells were frequently associated with collagen deposits, but their normal distribution being perivascular, this association was probably only topological.

Elastin modifications in the red pulp of

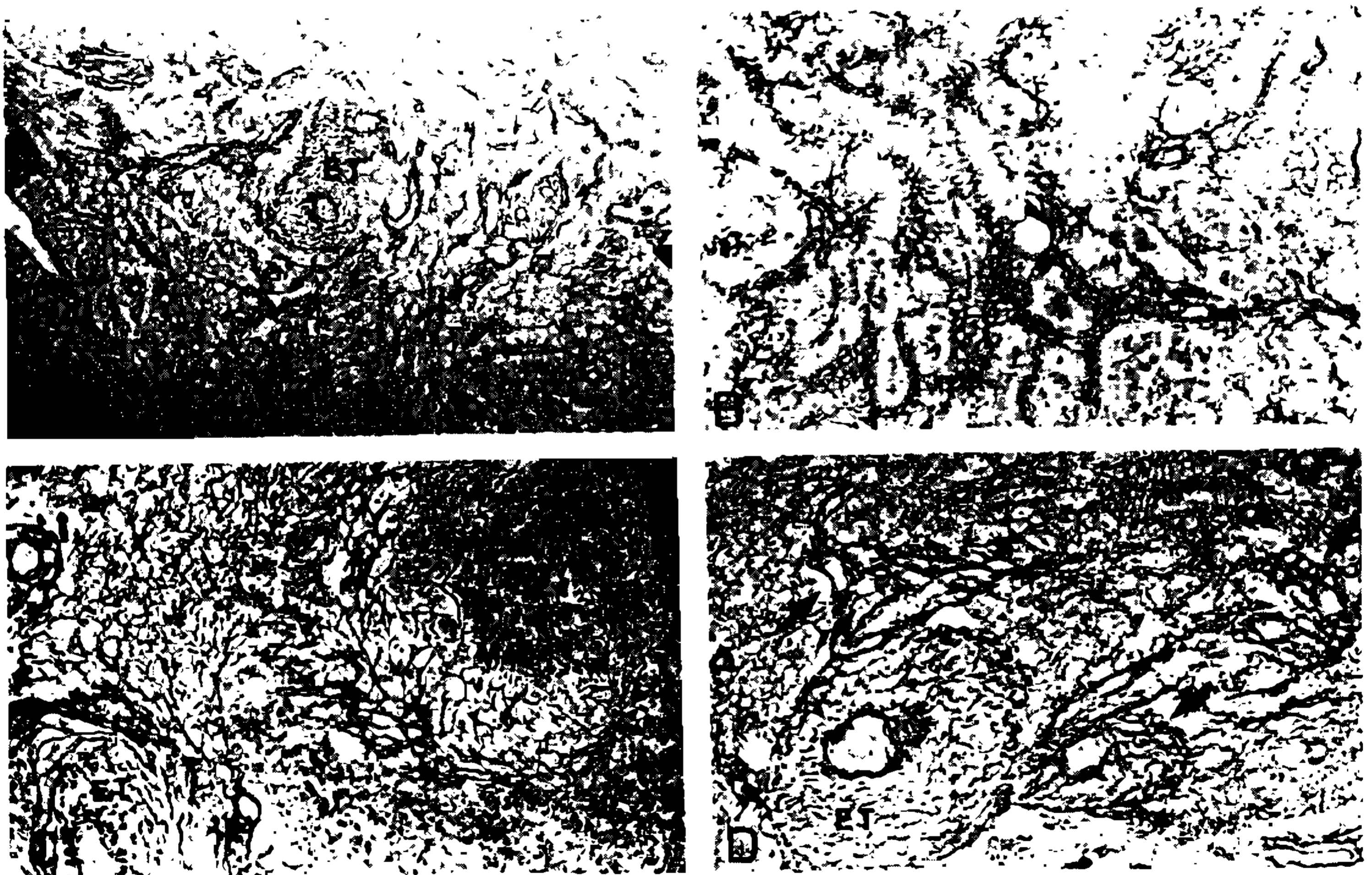


Fig. 1: chronic human schistosomiasis-silver impregnation of "reticulin fibers" in splenic tissue. A.) Low power view of the red pulp. ET = thickened elastic trabecules with large blood vessels. Arrows = smaller vessels with adjacent fibrotic reaction. B.) Red pulp. Note the reticulin meshwork surrounding the venous sinuses. C.) ET = elastic trabecule, MB = mapighian body: a tangential cut showing fine reticulin fibers penetrating into the peripheric part of the body. D.) ET = elastic trabecule, and adjacent blood vessels surrounded by fibrotic reaction (arrows). Note the fibrotic meshwork surrounding the venous sinuses.

schistosomal spleens were much less prominent than those observed for collagen. However, when large and continuous fibrotic deposits were formed in perivascular sinusoids, elastin was observed associated with collagen, participating in formation of alveolar matrix deposits around blood vessels.

In young patients the white pulp was hypertrophic, while in older ones it was frequently reduced. Malpighian bodies were surrounded by a prominent thickened system of marginal venous sinuses, with continuous meshwork of reticulin fibers. However, the matrix of marginal sinuses was not different from other red pulp sinuses. Fine reticulin fibers were occasionally seen arising perpendicularly from marginal sinuses, penetrating radially into the lymphoid corpuscle. In spleens attained by intense generalized fibrosis, the perpheric region of malpighian corpuscles contained a diffuse network of fine reticulin fibrils (Fig. 1C). Germinative centers contained a loose network of fine, tortuous but rarely branching fibers.

Frequently, hemorrhagic regions were observed in schistosomal spleens, surrounded by fibrotic reactions that subsequently formed extensive fibrotic scars.

In conclusion, two modes of fibrosis could be distinguished in schistosomal spleens: a) a diffuse fibrosis of the red pulp, with formation of continuous meshwork of silver-staining fibers in splenic cords and around venous sinuses, and

b) an intense perivascular fibrotic reaction, modifying considerably the normal organization of the perivascular tissue. The first modification has as its direct consequence an alteration of the blood cell passage through splenic cords. This is consistent with hematological aspects of the schistosomal hypersplenism, including retention of less actively moving cells as thrombocytes (R. Borojevic & E. A. Carvalho, 1981, *loc. cit.*). The second modification alters the general blood circulation pathway in the red pulp creating perivascular regions with considerably decreased permability. Hemorrhagic patches frequent in schistosomal spleens may be consequent to this modification.

A. Magalhães Filho and E. Coutinho Abath (1960, *loc. cit.*) have indicated that gross fibrotic involvement of the spleen is found in later stages of the disease. This is consistent with our observation of the gross perivascular fibrosis. The difuse fibrosis of the splenic red pulp is an earlier event, and together with hyperplasia of macrophagic and lymphoid splenic cell populations, it represents a fundamental characteristic of splenic pathology in schistosomiasis.

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