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Histochemical Studies of Liver of Mouse During Schistomiasis

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Abstract

Comparative histochemical studies on the liver damage following infections with *Schistosoma mansoni* and *Schistosoma margreborwiei* have been reported after 42, 49, 77, 91, 105 and 119 days post-infection in definitive host mouse. The most significant changes were the increase of protein in the hepatocytes present in the dark zones appeared during infections of the liver. However, normal hepatocytes were positive for glycogen and it was considerably reduced around the granuloma. Some areas of the liver exhibited magenta/purple red in colour and secretory cells of bile ducts were also stained for glycogen. Acid mucopolysaccharide was negative in the hepatocytes but increased amount of ribonucleic acid was stained in normal and infected liver. The ferric irons and lipofuscin pigments were stained negative in the hepatocytes but in few areas of the liver some positive granules of these substances were visible.

Introduction

Schistosomiasis is a disease caused by hypersensitivity reactions against parasite eggs trapped in the venules. Eggs release antigens that produce varying degrees of granulomatous response in the liver of the definitive host (Andrade and Andrade, 1965; Hirata *et al*., 1993; Von Lichtenebrg, 1962). The newly formed granulomas consisted either of one egg (single egg granuloma) or many eggs (multiple egg granulomas) in the centre surrounded by variable numbers of eosinophils, some neutrophils, epithelioid cells,s plasma cells and lymphocytes. In the later stages of infection a fibrotic tissue reaction occurred and the surrounding liver sinusoids were dilated. Most of the granulomas occupied portal veins of various sizes and provoked an intense perivascular infiltration of eosinopils, plasma cells ad lymphocytes were seen in the liver of mice with *S. mansoni* and *S. margrebowiei* infections (Soomro, 1996). The marked increase of glycogen, consistent changes in nuclear morphology have been noted in association with changes in the amount and distribution of certain histochemical constituents of the cell, which may ultimately prove beneficial in evaluating the functional state of the liver under the influence of schistosomiasis infections (Iwada *et al*., 1956). Numerous histopathological studies have been done on the liver of mouse following infections with *S. mansoni* and *S. margrebowiei*. The present paper describes the determination of protein, glycogen, acid mucopolysaccharide, ribonucleic acid, ferric iron and lipofuscin pigments during acute and chronic infections in the liver of the definitive host mouse.

Materials and Methods

Age-matched mice of the Bantim and Kingman Tylers original (BKTO) strain, weighed approximately 20-35g each were infected with 200 and 25 cercariae of either *S. mansoni* (Puerto Rican strain maintained in albino *Biomphalaria glabrata* snails and random-bred TO mice by method of Taylor *et al*. (1969) and *S. margrebowiei* (originally obtained from Lochinvar National Park, Zambia) and maintained in *Bulinus natalensis* intermediate host snails (the original stock was obtained from the Experimental Taxonomy Unit of the British Museum of Natural History, London). Before administering the cercaria, the experimental animals were anaesthetized with Sodium pentobarbitone (Nembutal) and the abdominal hair was clipped. The cercaria were applied to the abdominal skin by using ring. All mice infected with 200 cercariae were killed on day 42, while rest were killed at days 49, 77, 91, 105 and 119 p.i., Autopsies were performed immediately after the animals were killed by dislocation of neck region. The liver from each animals were fixed in Heidenhain's Susa fixative, washed and dehydrated in ethanol, infiltrated and embedded in historesin (Soomro, 1996). Selected 4 μm thick sections were stained in variety of histochemical methods; bromophenol blue for protein, Periodic acid and Schiff reaction for glycogen, alcian blue for acid mucopolysaccharide, ribonucleic acid for RNA, ferric iron and lipofuscin pigment demonstrations.

Results

Histochemical changes in the normal, *S. mansoni* and *S. margrebowiei* **infected livers of mice:** Normal hepatocytes showed light blue foamy cytoplasm, pale nuclei stained with bromophenol blue were positive for protein. The nucleus, cytoplasm of hepatocytes and connective tissues around the granuloma (termed as intermediate zone) were moderately and mildly stained for protein from 42 to 199 days p.i., However, the nucleus and cytoplasm of hepatocytes of dark zones were markedly and moderately stained for protein from 49 and 77 days p.i.. Normal endothelial cells were positively stained for protein and around granulomas these cells were weakly stained for protein from 42 to 119 days p.i. Normal and infected sinusoid stained mildly for protein from 42 to 119 days p.i. The normal epithelial cells bile ducts were positive for protein and around granulomas they were stained weakly

for protein.

The normal hepatocytes cytoplasm showed mild amounts of glycogen. Negative stained with periodic acid-Schiff reaction nucleus of the hepatocytes around granulomas and cytoplasm were weakly stained for glycogen from 42 to 119 days p.i. The normal epithelial cells of bile ducts were positive for protein and around granulomas they were stained weakly for protein.

The normal hepatocytes cytoplasm showed mild amounts of glycogen. Negative stained with periodic acid-Schiff reaction nucleus of the hepatocytes around granulomas and cytoplasm were weakly stained for glycogen from 42 to 119 days p.i. The cytoplasm of hepatocytes in the some areas of the liver was stained positively from day to 42 to 19 and moderately on 105 and 119 days p.i. The cytoplasm of hepatocytes of dark zones stained weakly for glycogen. Normal endothelial cells were weakly stained for glycogen and around the granuloma these cells were negative. However, few areas in the infected liver exhibited magenta/purple red in colour. Sinusoid also shows positive and secretory cells of the bile ducts were stained purple red mildly stained for glycogen.

Acid mucopolysaccharide was absent in normal cells. However, a few positive granules were observed in sinusoids, endothelial cells and secretory cells of the bile ducts. From 42 to 119 days p.i. increased positively stained material in these structures. The ribonucleic acid was stained mildly in normal hepatocytes. But around the granulomas the nuclei and cytoplasm of hepatocytes of intermediate zones were showed reduced amount of the ribonucleic acid from 42 to 119 days p.i. The nucleus and cytoplasm of hepatocytes of dark zones were moderately and mildly stained for ribonucleic acid from 42 to 77 days p.i. The nuclei of normal endothelial and secretory cells were strained positively for ribonucleic acid. Around the granulomas nuclei of these cells were stained mildly for ribonucleic acids. Normal and infected sinusoids were positively stained for ribonucleic acid. The ferric irons were negative in the normal hepatocytes and around the granulomas. The normal endothelial cells, sinusoids and secretory cells were also remained unstained for ferric iron. In a few areas of the liver ferric irons were weakly stained it could be endothelial cells or sinusoids from 42 to 119 days p.i. The normal hepatocytes and secretory cells were negative from lipofuscin. Normal endothelial cells were stained weakly for the lipofuscin. The positive lipofuscin pigments were stained in some of the endothelial cells from 42 to 119 days p.i.. Sinusoids and connective tissue around granulomas were stained weakly for lipofuscin pigments.

Discussion

The hepatocytes present in the dark zones showed markedly amount during infection of the protein. Another study reported by Andrade and Barka, (1962) reported that the release of mucoprotein into the granulomatous tissue can be assumed although it is difficult to ascertain its presence histochemically because of dilutional factors its admixture with mucoprotein of the ground substan Nevertheless, further investigation of the character of t mucoprotein as possible antigens is warranted. Junquei *al.* (1992) regarding the hepatocyte has an abundant both smooth and rough. In the hepatocyte, rER f aggregates dispersed in the cytoplasm; these are c basophilic bodies. Several proteins (eg. blood pla fibrinogen) are synthesized on polyribosomes in t structures. In addition to synthesizing proteins for its maintenance, the liver produces various plasma protein export-among them albumin, prothrombin, fibrinoge lipoproteins. Lesson and Leeson, (1976) described pro as large molecules composed of a variety of amino monomers linked by peptide bonds in a definite seque Schalm, (1965) reported that the plasma protein, albu is synthesized by the liver and in chronic diseases o organ the synthesis of albumin is affected resulting reduction of total plasma proteins.

The cytoplasm of normal hepatocyte stained mildly glycogen, whereas, this amount was reduced aroun granuloma. In contrast, Andrde and Barka, (1962) reported that newly formed granulomas contained scat cells with diastase resistant PAS positive material in cytoplasm. Lesson and Lesson, (1976), reported glycogen, a highly branched polymers of d-glou constitutes a storage depot from which glucose, neede many chemical activities within cell, may be released r upon demand and is a common reserve substan animals.

Acid mucopolysaccharide was absent in normal infected hepatocytes. The nucleus and cytoplasm of n hepatocytes were mildly stained for ribonucleic However, around the granuloma the amount of ribon acid was reduced in the hepatocytes. Negative ferric i normal and infected hepatoytes, however endothelial or sinusoids were stained weakly for these substance normal hepatocytes, sinusoids, and secretory cells negative, however, lipofuscin pigments were stained w in the endothelial cells, sinusoids and connective ti around granulomas.

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