Rev. Inst. Med. trop. São Paulo
19(2):109-112, *março-abril*, 1977

UDC 616.995.122.22

CONGENITAL MODIFICATION OF HOST RESPONSE IN MURINE SCHISTOSOMIASIS MANSONI

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SUMMARY

Pathology of liver and spleen in the acute phase of experimental murine schistosomiasis was studied in mice born to infected mothers, and compared to that of mice born to non-infected mothers. Hepato- and splenomegaly, characterized by hepatic inflammatory infiltration and intense reticulo-endothelial and lymphoid proliferation, were more pronounced in mice born to infected mothers. The transplacental transfer of schistosomal antigen, the consequent modification of the immunological response of the host, and the clinical evolution of the disease are discussed.

INTRODUCTION

Congenital modification of host response in murine schistosomiasis was first demonstrated by LEWERT & MANDLOWITZ [9]: offspring born to mothers heavilly infected by schistosomes were shown to form diminished granuloma around eggs embolised in the lungs. HANG, BOROS & WARREN [8] have obtained the same result, explaining it as an induced tolerance-like state, and postulating the transplacental transfer of antigen. WARREN [13], at the conclusion of a long series of studies, has established a hypothesis that tolerance to schistosomal antigen and diminished periovular granulomas should give a milder overt disease. The possibility of immunosuppressive treatment in humans was thus proposed (WARREN [13]). Nevertheless, FINE, BUCHANAN & COLLEY [5] and BUCHANAN, FINE & COLLEY [2] have shown that the suppression of granuloma formation in lymphocyte-depleted mice is followed by greater mortality and heavier lesions in the liver, thus demonstrating the protective character of the sequestring function of the granulomas, postulated earlier by VON LICHTENBERG [11].

The following work was done to study the effect of a congenitally induced modification of host response, described as a tolerance-like state by LEWERT & MANDLOWITZ [9] and HANG et al. [8], on the evolution and severity of murine schistosomiasis. Hepatic and splenic pathologies were studied in the acute phase of the disease.

MATERIALS AND METHODS

Three groups of mice were studied.

Group 1 — female C-3-H mice were infected with 50 cercariae of **Schistosoma mansoni** (FS-strain, Bahia). After 40 days, infection was monitored by stool examination and the mice were mated. Experimental Group-1 consisted of 30 mice, born to the infected mothers and exposed as sucklings (day 1) to transcutaneous penetration of 50 cercariae. They were sacrificed 8 to 10 weeks later, their livers and spleens weighed, fixed and treated by standard histological methods.

This work was supported by the «Service de la Coopération Scientifique et Techniques du Ministère des Affaires Etrangères» France, «Conselho Nacional de Pesquisas» Brasil, and «Centre National de la Recherche Scientifique» France.

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Group 2 — consisted of 30 mice, born to uninfected mothers and infected and treated by the same methods.

Control group of 20 adult mice (2-3 months) was exposed to 90 cercariae, so that the total number of adult worms in experimental and in control groups was equal, as previous research has shown that in experimental conditions used in our laboratory, 20-30% of cercariae develop to adult worms when infecting an adult mouse, whilst 50-60% of cercariae mature in the infection of newborn mice.

The statistical value of results was calculated by the Student's test.

RESULTS

Hepatomegaly and splenomegaly, estimated by the weight of the organs, were both most pronounced in **Group-1.** When weights of liver and spleen are expressed as a function of total body weight, their mean values show a statistically significant difference between **Group-1** and **Group-2,** with $0.001 < P < 0.01$. There was no significant difference observed between Group-2 and controls.

Histologically, in all groups livers and spleens showed the classical aspects of acute murine schistosomiasis (GOENNERT [6]). In the liver, the parenchymal injury was shown by local and focal cell degeneration and irregular regeneration with numerous binuclear cells and generalized anisokaryosis. The corresponding mesenchymal hypertrophy included endothelial and Kupffer-cell proliferation, periportal and diffuse mesenchymal infiltration composed of polymorphonuclear, mononuclear and lymphoid cells. Periovular granulomas were found to be similar in all groups, no qualitative or quantitative differences being detected in their morphology or composition. They were characterized by intense eosinophil and mononuclear infiltration, with fibrotic reaction in later stages. Their diameter was measured in all groups, but no significant difference was observed between the mean values.

TABLE I

Values of liver and spleen weights, expressed as a function of total body weight, compared in experimental and in control groups

	Mean value Body/Liver	Values Student's Test	Mean value Body/Spleen	Values Student's Test
Group 1	14.2	$t = 3.0$ $0.001 < P < 0.01$	79.1	$t = 3.1$ $0.001 < P < 0.01$
Group 2	16.4	$t = 0.16$ $0.50 < P$	102.6	$t = 0.09$ $0.90 < P$
Controls	16.6		105.8	

Massive reticular and lymphoid proliferation and myeloid metaplasia were the main characteristics of splenomegaly in all groups. Eggs and granulomas were rarely found in spleens, but local haemorrhages could often be seen. Fibrotic processes in spleen are not prominent at this stage of the disease.

Weights of livers and spleens partially overlapped in the experimental groups. Similarly their histology is not clearly different. Nevertheless, most of the livers in **Group-2** were characterized by well delimited granulomas and relatively little diffuse and perivascular infiltration. In **Group-1** an intense perivascular infiltration, consisting mainly of eosinophil granulocytes, mixed with neutrophils and mononuclears, formed continuous sheets around intrahepatic blood vessels. Diffuse parenchymal infiltration was prominent in **Group-1,** with formation of granulocytic folliculi described earlier (GRIMAUD & BOROJEVIC [7]). These differences were constant

and permitted recognition, in the "blind test", of histological preparations of mice with body/liver ratios inferior or superior to 15.

The spleen histology was similar in all groups, and the weight differences seem to be quantitative ones only.

We found no histological evidence of an associated non-schistosomal pathology which might have been incriminated for observed differences between the experimental groups.

DISCUSSION

Earlier reports have indicated the formation of smaller granulomas in lungs of mice born to infected mothers. The fact that no significant difference could be detected between the liver granulomas in our experimental groups is consistent with the observed fact that the model of granulomas around eggs injected into the lung, developed by VON LICHTENBERG [12], is more sensitive than analysis of granulomas around eggs deposited by worms in liver (DOMINGO & WARREN [3,4]). The eggs are deposited in liver at different times, and as each egg has its own cycle with consequent changes in the size of granulomas, only very prominent differences in the granuloma sizes could give a generally significant result.

Our results differ from those obtained by HANG et al. [8] in several aspects. Moderate infection of mice with 60 cercariae did not modify the responsiveness in offspring born in their experimental conditions, while it did in ours. This might be due to the fact that we have challenged newborn mice whereas those Authors have studied eight-week-old offspring, or related to the higher morbidity of schistosomes in our experimental model: 90 cercariae kill most of mice by the 8th week of infection in our laboratory, while 110 cercariae kill mice by 10 - 12th week in their experimental conditions, moderate infections here corresponding to heavy infections there.

HANG et al. [8] state that no significant differences were observed between spleen and liver weights of control animals and those treated with hyposensitizing doses of antigen administered prenatally. The notion of hyporesponsiveness is here ambiguous, as hepatosplenic disease was studied 8 weeks after infection, in mice sensitized prenatally by three 300 μg injections of schistosome egg antigen (SEA). Although this treatment induced diminished granulomas at 8 and 16 days after challenge, at 32 days "the granulomas in the lungs of treated mice were actually larger than in those of the control group", and the Authors concluded that this treatment "resulted in an enhanced granulomatous response". Accordingly, at the 8th week the responsiveness was probably enhanced.

The size of granulomas alone is a useful parameter in studies of cellular reaction in the lung. It is difficult to correlate it with a general response to schistosome eggs and to schistosomal antigens, as smaller granulomas around eggs in the acute phase of a primary infection (non-sensitized animals) are qualitatively different from smaller granulomas in a chronic phase of infection (so-called "endogenous desensitization"). The first type of granuloma correspond to more severe destructive lesions in liver, exemplified by necrosis, inflammatory and exudative infiltration, whilst the second corresponds to an equilibrated and compensated general state of liver, with nearly normal parenchyma, where only the progressive periportal fibrosis with consequent complications will eventually provoke severe liver disease.

Although the granulomatous reaction to schistosome eggs is known to be diminished in mice born to infected mothers, the hepatomegaly has shown to be more prominent in our experiments. Hepatomegaly in murine schistosomiasis is a result of inflammatory infiltration of liver tissues and of progressive fibrosis caused mainly by periovular granulomas in later stages of the disease. Our results are consistent with comparative observations reported by VON LICHTENBERG et al. [12] in hamsters, in which an inverse correlation was found between granulomatous and non-granulomatous pathology in liver. When the degree of destructiveness of liver lesions was considered, smaller granulomas were associated with more intense inflammatory injury throughout the liver. BUCHANAN et al.[2] have observed a similar relationship in mice depleted of thymus-dependent lymphocytes.

Splenomegaly in the acute phase of murine schistosomiasis results from reticular proliferation, lymphoid hyperplasia, myeloid me-

taplasia and only in lesser degree from a fibrotic and congestive reactions (ANDRADE [1]). The elevated splenomegaly thus corresponds to the elevated stimulation of poliferative reactions in spleen, which might be related to the higher levels of diffused antigenic and other substances in the circulation, due to lesser sequestration of egg antigens in granulomas (VON LICHTENBERG [12]).

It is to be concluded that congenitally induced modifications of host response, described earlier by LEWERT & MANDLOWITZ [9] and HANG et al. [8] have, in the acute murine schistosomiasis, an adverse effect on the evolution of the overt disease. The periovular granulomas, although causing lesions in venous circulation in the liver, with all the complications consequent to the progressive portal obliteration, have a protective function, preventing the diffusion of antigenic and toxic substances, known to diffuse of the eggs to the surrounding liver parenchyma.

RESUMO

Modificação congênita da resposta do hospedeiro na esquistossomose mansônica do camundongo

A patologia do fígado e do baço na fase aguda da esquistossomose experimental foi estudada em camundongos nascidos de mães infestadas, e comparada com a patologia dos camundongos nascidos de mães não-infestadas. Os maiores índices de hepatesplenomegalia, caracterizada pela infiltração inflamatória hepática e pela proliferação intensa retículo-endotelial e linfóide esplênicas, foram observadas nos camundongos provenientes de mães infestadas. A passagem transplacentária do antígeno esquistossomótico, a modificação da resposta imunológica do hospedeiro e a evolução clínica da doença são discutidas.

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Recebido para publicação em 27/8/1975.