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BRIEF COMMUNICATION

Schistosoma mansoni: EXACERBATION OF INFLAMMATORY GRANULOMATOUS RESPONSE IN MICE CHRONICALLY INFECTED AND SUBMITTED TO REINFECTION

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In schistosomiasis, the acute phase of the disease is histologically characterized by an exuberant inflammatory response against parasite eggs. During the chronic phase, the granuloma size decreases as a result of immunomodulation of the inflammatory response. The typical granuloma of the chronic phase can be considered a beneficial inflammatory response to the host, in spite of schistosomiasis being classified as immunopathologic. So, in animals immunosuppressed by drugs or thymectomized and with inhibition of inflammatory granulomatous responses, an increase of tissue damage around the eggs has been observed. This was probably due to toxic substances and potent enzymes produced by miracidial spreading over surrounding tissues, thus resulting in an extensive area damaged by coliquative necrosis[3,6,7,9,10,11,14,15]. In this way, the granulomas of the chronic phase abrogate the spreading of these substances preventing a more severe tissue damage.

By using a larger number of animals, the present study intended to corroborate the results obtained by COELHO *et al.*[5] and RASO *et al.*[16] who have demonstrated the phenomenon of exacerbation of the granulomatous response in mice submitted to reinfection, during the chronic phase of the disease.

Albino Swiss mice (outbred females) were infected with the LE strain (Belo Horizonte, MG, Brazil) of *Schistosoma mansoni* isolated from a patient and kept at the Schistosomiasis Research Unit – Prof. José Pellegrino Laboratory (Federal University of Minas Gerais) by passage in laboratory reared *Biomphalaria glabrata* and hamsters, for more than 30 years. The animals were transcutaneously infected with 20 cercariae through the abdominal skin, according to the technique described by BARBOSA *et al.*[1].

On day 120 after infection, one group of mice was reinfected with the same number of cercariae by using the same technique previously described[1], and the remaining mice were maintained as controls. On day 70 after reinfection the animals of both groups (Reinfected n = 49, Control n = 42) were sacrificed by sulphuric ether, the livers removed and preserved in 10% formalin buffered by addition of NaH_2PO_4 and NaOH, pH 7.2. The livers were cut into small pieces 3.0 mm thick each, and embedded in paraffin. The sections 5.5 μm thick so obtained were stained with hematoxylin and eosin.

Only granulomas in necrotic-exudative, exudative and productive phases were considered for diameter

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measurements, whereas granulomas in process of healing by fibrosis were not taken into account. The average of the diameter size was determined by the mean between the smallest and largest measurements of diameter passing through the egg in the centre of the inflammatory reaction. In this way, only the recent granulomas were examined with a split eyepiece (10X, Ernest-Leitz, Wetzlar, Germany) adapted to a Zeiss microscope.

The Student's t test was used to detect differences between the two groups.

As can be seen in Table 1, the mean of granuloma diameter of the reinfected group was statistically higher than the one of the control group with only primary infection ($p < 0.0001$).

The process of immune regulation of inflammatory granulomatous response is governed by a complex balance of lymphokines produced by $CD4^+$ cells especifically from subsets Th1 and Th2[17]. The process of immunomodulation is transient, starting around day 90 after infection and ending around day 120 after infection. In this occasion, the histopathology defines the granulomas as typical of the chronic phase[2,4,8,12]. PERRIN & PHILLIPS[13] demonstrated that $CD8^+$ (suppressor cells) also play a role in the declining of immunomodulation of the inflammatory granulomatous response.

The present study reveals that there must occur alteration of the immunomodulation mechanism in mice chronically infected and compared to reinfected mice. This result corroborates the findings of COELHO *et al.*[5] and RASO *et al.*[16], who demonstrated an exacerbation in the granulomatous process in mice submitted to reinfection, in the chronic phase. The presence of granulomas in reinfected animals could be due to changes of the balance of interleukines from Th1 and Th2. The skin, pulmonary and portal system phases of migration of the parasites derived from reinfection, as well as the increase of parasites and eggs in tissue due to the increased worm burden, could explain in part the exacerbation of the granulomatous response. Further studies will be conducted to shed light on the mechanisms involved in this intriguing immunopathologic phenomenon, mainly on the evaluation of interleukines from Th1 and Th2 lymphocytes.

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TABLE 1

Mean granuloma diameter in livers of mice chronically infected with *Schistosoma mansoni* and submitted to reinfection

	Sacrifice day	Mean granuloma diameter M ± SD	n	p <
Mice chronically infected	190	210.07 ± 11.39	42	0.0001
*Mice chronically infected and submitted to reinfection	190	260.00 ± 20.38	49	

Mice were reinfected on day 120 after primary infection and sacrificed 70 days later.

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