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Research Note

Thymic Atrophy in Experimental Murine Schistosomiasis Japonica

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Key words: thymic atrophy, schistosomiasis, *Schistosoma japonicum*, mouse

Adult schistosomes reside in the portal and mesenteric venules of hosts and eggs released by the female worms cause granulomatous reactions in the liver and intestinal walls. Among three major human schistosome species *Schistosoma japonicum*, *S. mansoni*, and *S. haematobium*, *S. japonicum* causes the most severe disease. Chronic advanced schistosomiasis japonica is characterized by the liver fibrosis, hepatosplenomegaly, portal hypertension, esophageal varices and ascites with finally death (Warren, 1973). In the murine model of schistosomiasis japonica, mice also develop those symptoms and they occasionally die in early days of infection.

The most important histopathological changes of host tissues are granulomatous inflammations around parasite eggs. In the murine model, the granuloma size is maximum in acute stage and decreases in chronic stage (Warren *et al.*, 1978). In T-cell depleted or athymic nude mice, inflammatory granulomatous reactions around eggs are weak and those mice die in the acute stage of infection (Byram and Lichtenberg, 1977). On the other hand, in normal mice granuloma size is modulated by suppressor T lymphocytes at the chronic phase of infection (Colley, 1981).

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The relationship between inflammatory granulomas around schistosome eggs and hepatosplenomegaly has been long discussed. However there are only a few papers about the role of the thymus, which supplies T-cells to whole body (Wellhausen and Boros, 1982).

In the present study, we investigated the changes of thymocyte number, thymic weight and histopathology during infection.

Five weeks old female ddy mice (Schizuoka Agricultural Cooperative Association for Laboratory Animals, Schizuoka, Japan) were anesthetized by intraperitoneal injection of pentobarbital and 40 cercariae of *S. japonicum* (Yamanashi strain) were put on previously shaved abdominal skin by the coverglass method. Seven mice were killed by ether anesthesia at 2, 3, 6, and 9 weeks after infection respectively. The intact thymi were removed and weighed on an electronic reading balance (Libror 200, Schimazu Co.). In 5 mice removed thymi were respectively homogenized by a teflon planzer in plastic tube (Falcon 2057) with phosphate buffer solution and the number of thymocytes obtained was counted by the hemocytometer in Turk's solution. For the purpose of histopathological examination, thymi removed from 2 mice were fixed in 10% neutral formalin solution, dehydrated in ethanol, embedded in paraffin,

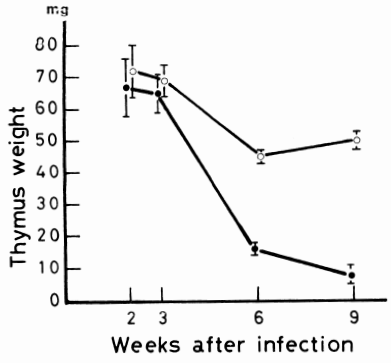

Fig. 1 Kinetic of thymic weight in mice infected with 40 cercariae of *S. japonicum*. (●) infected mice, and (○) control mice. Each point represents the arithmetic mean of the thymic weight obtained from 7 individual mice at indicated time after the infection. Each bar represents the limit of 1 standard error.

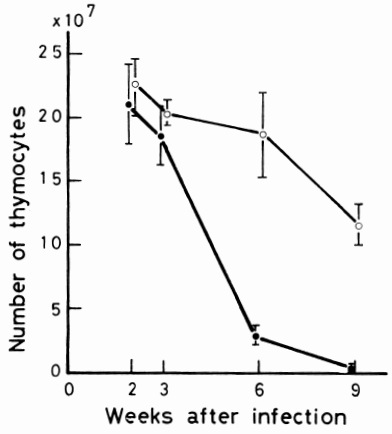

Fig. 2 Kinetic of the number of thymocytes in mice infected with 40 cercariae of *S. japonicum*. (●) infected mice, and (○) control mice. Each point represents the arithmetic mean of the thymocyte number obtained from 7 individual mice at indicated time after the infection. Each bar represents the limit of 1 standard error.

sectioned 3 to 4 μ in thickness, and stained with hematoxylin and eosin. Possible significant difference between infected and control animals was determined by using the parametric Student's *t*-test.

At 2 and 3 weeks after infection, mean thymic weight of infected mice was analogous to that in control mice. At 6 weeks after infection, however, thymic weight significantly decreased by 64.4% as compared with age matched control (mean ± S.E.; infected group : 16 ± 2 mg., control group : 45 ± 2 mg., $P<0.001$, Fig. 1). At 9 weeks after infection, it was difficult to recognize the thymi in infected mice. Thymic weight dramatically decreased by 90% as compared with control mice (infected group : 8 ± 3 mg, control group : 50 ± 3 mg, $P<0.001$, Fig. 1) and likewise thymocyte number was only 4.1% of those control mice (mean ± S.E./10^{-7}; infected group : 0.48 ± 0.36, control group : 11.6 ± 1.6, $P<0.001$, Fig. 2). Infected mice began to die at 10 weeks after infection and most died at 11 weeks after infection.

At 2 and 3 weeks after infection, no difference in thymic histopathology was observed between infected and control mice. At 6 weeks after infection, thymi of infected mice showed clear corticomedullary distinction with progressive depletion of cortex. At 9 weeks after infection, however, the corticomedullary distinction became unclear and most of small cells with dark stained nuclei disappeared (Photos. 1, 2). Finally lymphoid cells disappeared from subcapsular cortical (Photo. 3) and medullary sinus (Photo. 4). There were abundant cells with enriched cytoplasma and clearly stained nucleus in the medulla.

In murine schistosomiasis, hepatosplenomegaly and the enlargement of mesenteric lymph nodes are considered to be caused by inflammatory granulomas around parasite eggs. Thymus-derived lymphocytes serve as both effector and suppresor cells of granulomatous response in murine schistosomiasis mansoni (Chensue and Boros, 1979). The balance between these populations tilts increasingly towards the suppressive mode during chronic infection, resulting in diminished granulomatous formation (Wellhausen and Boros, 1982).

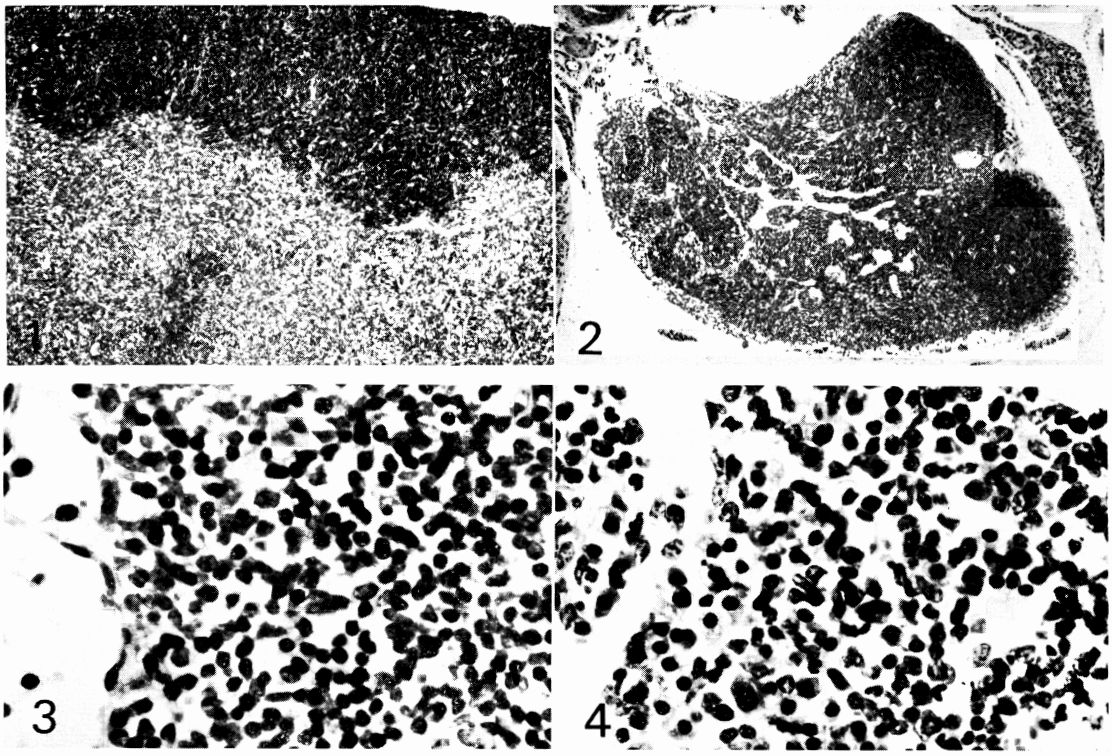

Photo. 1 In age mached control mice, corticomedullary distinction of the thymus is clear and lymphoid cells in cortex are very rich. (×40)

Photo. 2 At 9 weeks after infection, atrophy of the thymus is significant and corticomedullary distinction is lost. (×40)

Photo. 3 At 9 weeks after infection, the number of small, darked nucleated cells decreases in the cortex and thymocytes disappear in the sinus under the capsule. (×200)

Photo. 4 At 9 weeks after infection, the cells with enriched cytoplasma and clear large nucleus are significant and the number of thymocytes decreases in the medulla. (×200)

In chronic schistosomiasis mansoni, the spleen cells of infected mice have a function to diminish the granuloma (Colley, 1981). Also, in the freshly infected mice to which are transfered the spleen or lymph node cells from mice infected with the chronic schistosomiasis japonica and mansoni, the degree of granuloma around parasite eggs is suppressed (Olds *et al.*, 1982; Colley, 1976).

Little attention has been paid to the thymic atrophy during the course of murine schistosomiasis. Wellhausen and Boros (1982) have recently reported the occurrence of atrophy of thymic cortex in CBA/J mice infected with 200 cercariae of *S. mansoni*. In the present study we infected ddy mice with 40 cercariae of *S. japonicum* but recognized more severe thymic atrophy than those reported by Wellhausen and Boros (1982). Thymic atrophy due to parasite infections has been also observed during the course of acute infection of mice with *Trypanosoma rhodesienses* (Mansfield and Bagasra, 1978), *T. brucei* (Murray *et al.*, 1974) and *Plasmodium berghei* (Igarashi and Waki, 1983). It is very interesting to note that thymic

atrophy occurs in mice heavily infected with *S. japonicum* and *S. mansoni*. Thymic atrophy, observed during the course of murine schistosomiasis, may be due to the suppression of production of precursor thymic cells, the destruction of thymocytes in thymic cortex by thymocytotoxic autoantibody (Kawabata *et al.*, 1981) and the marked exhaustion of T cells from the thymus (Wellhausen and Boros, 1982).

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短　　報

日本住血吸虫感染マウスの胸腺萎縮

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日本住血吸虫セルカリア40隻を ddy マウスに経皮感染させ，感染後 2，3，6，9 週の胸腺変化を観察した．

胸腺重量と胸腺細胞数は，感染 6 週以後急激に減少した．特に，感染 9 週目には，胸腺重量は対照群の約10%に，胸腺細胞数は対照群の約 4% にまで減少した．感染 6 週目の組織像では，皮質内細胞の減少と皮質の萎縮が著明であった．感染 9 週目には髄質内のリンパ球様細胞の減少が著るしく，胸腺全体の萎縮が目立った．また，感染10週以後，大部分の感染マウスは死亡し，実験を中止せざるを得なかった．