Review

Cellular and chemokine-mediated regulation in schistosome-induced hepatic pathology

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In hepatic schistosomiasis, pathology arises when schistosome eggs become lodged in the host liver, evoking an interleukin 4 (IL-4)- and IL-13-mediated dominant CD4$^+$ Th2 immune response. This response leads to the development of granulomas and fibrosis, with eosinophils, neutrophils, macrophages, hepatic stellate cells, and lymphocytes all identified as major cellular contributors to these events. This review outlines the cellular and molecular mechanisms of hepatic schistosomiasis, with an emphasis on the major cellular components and their release of chemokines. The differences between *Schistosoma mansoni*- and *Schistosoma japonicum*-induced hepatic granuloma are also discussed. This comprehensive overview of the processes associated with hepatic schistosomiasis may provide new insights into improved treatment for both schistosomiasis and other granulofibrotic diseases.

Schistosomiasis-induced pathology

Schistosomiasis is a chronic helminthic disease affecting approximately 200 million people in over 74 countries where it is endemic [1,2]. Human schistosomiasis is caused by infection with one of five species of schistosomes: *S. mansoni*, *S. japonicum*, *Schistosoma haematobium*, *Schistosoma intercalatum*, and *Schistosoma mekongi*, with the former three species being of the greatest clinical and socioeconomic importance. All species, with the exception of *S. haematobium*, cause hepatosplenic and intestinal schistosomiasis in which the parasitic worms inhabit the mesenteric veins. *S. haematobium*, by contrast, resides in the venous plexuses of the urinary bladder and causes urogenital schistosomiasis [2,3].

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The formation of granulomas (see Glossary) is a dynamic process that involves the accumulation of immune cells at the site of antigen release, leading to the confinement of the eliciting agent [4]. Granulomas are, in general, composed of a tightly clustered macrophage population, including epithelioid and multinucleate giant cells, and are often surrounded by a ring of lymphocytes [5]. Granulomatous disorders can arise both in infectious and non-infectious diseases, with gastrointestinal and hepatic granulomas being the most common clinical presentations. The

Glossary

Alternatively activated macrophages (AAMs): are activated by Th2 cytokines such as IL-4 and IL-13 to induce the production of proline, an amino acid essential for the synthesis of collagen. AAMs are thought to be involved in promoting collagen deposition and the onset of fibrosis.
Chemokines: a group of cytokines produced by cells present at the site of inflammation; they have the ability to induce chemotaxis in various leukocytes.
Chemokine-binding proteins: secreted proteins that selectively bind to and inhibit the action of specific chemokines.
Classically activated macrophages (CAM): activated by Th1 cytokines such as IFN-γ, TNF, and IL-12 to induce the production of nitric oxide that leads to inflammation and tissue damage.
Endothelial cells: a thin layer of cells lining the inner wall of the blood vessels and lymphatic vessels which form the endothelium.
Eosinophilia: an increase in the number of peripheral blood eosinophils. This condition is usually associated with allergic reaction or parasitic infection.
Fibrosis: excess deposition of fibrous tissue in an organ that normally occurs to replace tissue lost through injury or infection.
Granuloma: a small inflammatory lesion containing an organised collection of immune cells that is often produced in response to an infectious agent or a foreign substance.
Hepatic stellate cells (HSCs): also known as perisinusoidal cells, HSCs are located in the space of Disse between hepatocytes and sinusoidal endothelial cells of the liver. HSCs in normal liver are in a quiescent state, become activated when the liver is injured, and are the major cell type involved in liver fibrosis.
Kupffer cell: specialised macrophages lining the wall of the hepatic sinusoids that play a role in the breakdown of red blood cells.
Neutrophil extracellular trap (NET): a network of extracellular fibres composed of DNA and histones generated by activated neutrophils that bind pathogen.
Schistosomal myeloradiculopathy (SMR): known also as spinal cord schistosomiasis, SMR is the schistosomiasis of the brain and spinal cord caused by infection with the digenean trematodes of the genus *Schistosoma*.
Schistosomiasis japonica: helminth disease caused by the human blood fluke *Schistosoma japonicum*, primarily endemic in China and the Philippines.
Schistosomiasis mansoni: helminth disease caused by the human blood fluke *Schistosoma mansoni*, found mainly in regions of South America and the Caribbean, Africa, and the Middle East.

initiation, development, and regression of granuloma are mediated by the communication of cytokines and chemokines, leading to the recruitment of inflammatory cells towards the antigenic stimuli [5].

The pathology of schistosomiasis results from the eggs that inevitably become lodged in host tissue, particularly the liver and intestine. Because of the continuous antigenic stimulation from the trapped ova, inflammatory and immune cells are sequentially recruited to the sites of infection, leading to the formation of periovular granulomas and eventually chronic fibrosis in some infected individuals. This inflammatory response is associated with a dominant $CD4^+$ T cell-dependent immune response, with interleukin IL-4 and IL-13 being the main cytokines driving this reaction [6].

Granuloma arising from schistosomiasis

In schistosomiasis, egg-induced granulomatous lesions are found in the liver, intestines, lungs, bladder, brain, skin, adrenal glands, skeletal muscle, lymph nodes, spleen, kidney, and stomach [2,7]. The principal site of granuloma formation is around the eggs that are lodged in the pre-sinuisoidal capillary venules of the liver. These granulomas give rise to severe fibrosis which disrupts blood flow through the liver, causing portal hypertension and porta-caval shunting, which may lead to potentially fatal esophageal bleeding [1,2]. The intestines, another site for egg deposition, are characterised by a mucosal granulomatous response, leading to pseudopolyposis, microulceration, and superficial bleeding [1]. In schistosomiasis haematobia, egg deposition occurs primarily in the urinary bladder and results in urinary bladder calcification, which eventually leads to dysuria, pollakisuria, proteinuria, and haematuria [1], and subsequently to the development of bladder cancer or renal failure later in life [8]. The mortality rate due to under-performing kidney function due to *S. haematobium* infection exceeds that resulting from haematemesis in individuals infected with *S. mansoni* [8].

Schistosome egg-induced granulomas develop through two major stages, the pre-granulomatous and granulomatous stages [9,10]. The pre-granulomatous stage is characterised by the disorganised aggregation of cells, whereas the granulomatous stages are associated with a more clearly delineated structure. Lesion development starts with the infiltration of monocytes and eosinophils around the eggs. This early phase is followed by the recruitment of fibroblasts, leading to the production of extracellular matrix and collagen fibres that form an additional layer at the outer zone of the granuloma. Following the death of the miracidium within the egg, the granuloma structure disintegrates and calcifies, and this is associated with a reduction in the inflammatory response. During the final stages of the response, the granuloma is significantly reduced in size and contains only pigmented macrophages [10–12]. The continuous deposition of eggs at this later stage nevertheless triggers the formation of new granulomas. Representative images of hepatic granulomas at different developmental stages in murine *S. japonicum* infection are shown in Figure 1.

Although their formation is deleterious to the infected host, these schistosome-egg driven granulomas also play

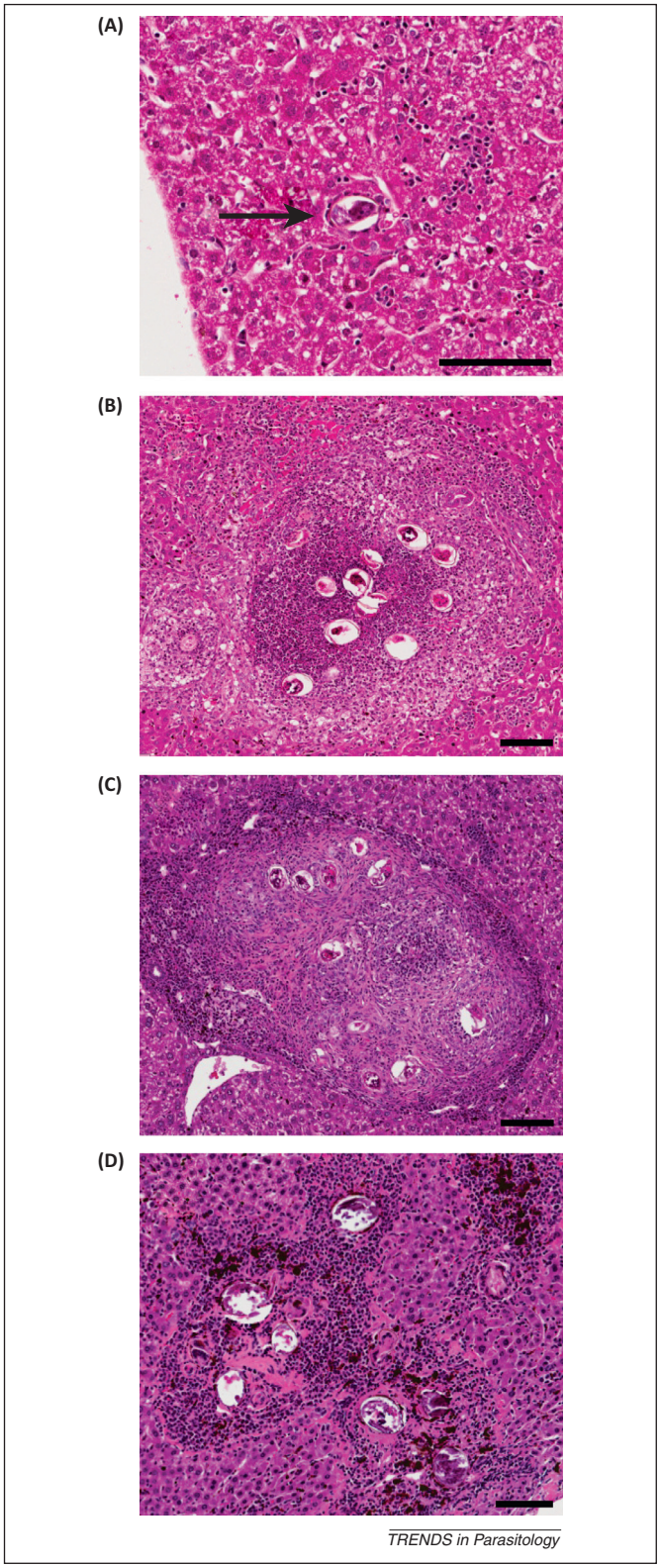

Figure 1. Representative histology of *Schistosoma japonicum*-induced granuloma in C57BL/6 mice at increasing time-points. **(A)** Eggs (arrow) begin to lodge in the host liver at 4 weeks post-infection (p.i.) with only minimal cellular infiltration. **(B)** At 6 weeks p.i. the eggs are surrounded by a dense population of immune cells, followed by a band of fibrovascular tissue leading to the formation of a mature granuloma. **(C)** Granulomas continue to increase in volume at 10 weeks p.i., delineated by a dense population of lymphocytes at the outer rim. **(D)** During the later stages (13 weeks p.i.) granulomas reduce in volume, often accompanied by the presence of dark-brown pigmented macrophages and calcified parasite eggs. Formalin-fixed, paraffin-embedded liver sections stained with hematoxylin and eosin. Scale bars, 100 μm.