

Immunoparasitology series

IgE: a question of protective immunity in *Trichinella spiralis* infection

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The best-known function of immunoglobulin E (IgE) is the initiation of allergic reaction. However, it does not make sense that IgE exists only for allergy, which is harmful for the body and does not have a beneficial role. In this article, we focus on IgE in *Trichinella spiralis* infection in rodents and humans, and discuss the possible roles of IgE in protection and pathogenesis.

Functions of immunoglobulin E

Enhancement of immunoglobulin (Ig)E production is rarely seen in infections other than helminth infections. Therefore, it is likely that IgE provides protection against the helminth. However, there are not many cases in which helminth infection has proven IgE-dependent protection, especially in *in vivo* systems [1].

Epidemiological studies in humans have suggested that IgE production reflects the expression of resistance to reinfection by *Schistosoma* [2]. IgE-dependent protection in *Schistosoma* infection has been reported by comparing IgE-deficient mice with IgE-producing mice in terms of granuloma formation [3] and worm burdens [4] and, in rats, by measuring resistance to reinfection [5].

Trichinella spiralis infection worldwide is sporadic in humans and common in wild animals. Characteristic immune responses to *T. spiralis* are IgE production and eosinophilia. The IgE production has been explained by the induction of T helper (Th)2 cells that produce cytokines such as interleukin (IL)-4 and IL-13, which are key molecules for determining B-cell differentiation to IgE-producing cells [6]. Pathophysiology and immune responses to infection with *T. spiralis* have been reviewed previously [7–12].

Protective role of IgE in rodents

The sites of protective response to *T. spiralis* infection are the intestine and tissue (mainly muscle). The targets in the intestine are adult worms during primary infection and muscle larvae during reinfection. The targets in tissue are newborn and muscle larvae. Figure 1 shows muscle larvae surrounded by many inflammatory cells.

In rats, rapid expulsion from the intestine is considered to be IgE-mediated protection against *T. spiralis*; this is

the phenomenon whereby muscle larvae are expelled quickly from the intestine of immunized animals after oral challenge infection. Rapid expulsion in rats is induced by IgE antibodies and CD4⁺OX22[−] T cells [13]. An interesting finding is that, in this process, the IgE antibody is functional in the intestinal lumen and is transported from plasma. IL-4, presumably derived from CD4⁺OX22[−] T cells, activates enterocytes to concentrate the IgE antibody in the intestinal lumen [14]. The IgE antibody binds to *T. spiralis* to prevent invasion [11].

A selective suppression of IgE production was developed in rats by treatment with anti-IgE. The IgE-suppressed rats were more susceptible to primary infection with *T. spiralis* [15]. They harbored two to three times the amount of larvae harbored by the controls. The target of the IgE antibody for resistance seems to be the larval stage in muscle. Although the kinetics of expulsion of adult worms was not determined, the invasion of newborn larvae terminated at the same time in both IgE-suppressed and control rats. This report provides the first direct *in vivo* evidence that the IgE antibody is crucial for immune protection against helminth infection.

Biozzi mice, selected into high and low responder lines by antibody responsiveness to sheep erythrocyte, were

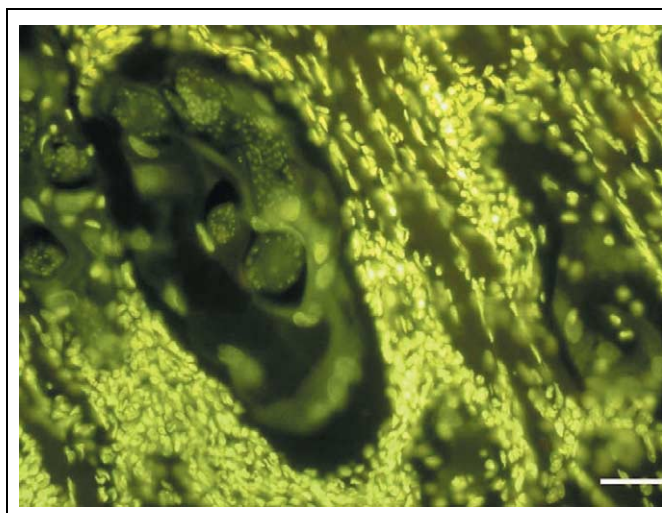


Figure 1. Muscle tissue section from mice experimentally infected with *Trichinella spiralis*, treated with Sytox® to stain cell nuclei. The nurse-cell–parasite complex is surrounded by inflammatory cells. Scale bar = 50 µm.

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infected with *T. spiralis*. Anti-*T. spiralis* IgE antibody was produced depending on the characteristics of antibody responsiveness of the mice. The protective immunity, evaluated by the number of muscle larvae present after primary infection, correlated inversely with IgE antibody responsiveness. High-responder-line mice produced higher levels of anti-*T. spiralis* IgE with lower worm burden [16].

IgE-dependent protection against muscle larvae was examined in mice by comparing IgE-deficient mice with IgE-producing controls. In congenitally selective IgE-deficient SJA/9 mice, the number of muscle larvae four and nine weeks after primary infection and four weeks after secondary infection with muscle larvae was not significantly different from that in IgE-producing SJL/J mice with a background gene of SJA/9, which indicates no IgE dependency in protection against adults and larvae [17].

Contrasting results were reported recently; IgE antibody was responsible for the protection against muscle larvae using *IgE* gene knockout mice with a BALB/c background. The number of muscle larvae in IgE-knockout mice 28 days post-infection was twice that in control mice. The percentage of worm cysts containing necrotic larvae in IgE-knockout mice was half that found in IgE-producing control mice. A deposition of IgE around the muscle larvae in IgE-producing mice suggested an IgE-dependent protection against the larvae [18].

The discrepancy between the results from SJA/9 and IgE-knockout BALB/c mice is likely to depend on the IgE responsiveness of the mice used. Previously, strains of mice were immunized with *Nippostrongylus brasiliensis* infection and the antigen, and IgE responsiveness was assessed by magnitude and persistence of IgE production. SJL/J and AKR mice were observed to be IgE low responders, with low and transient IgE production compared with IgE high responders such as BALB/c mice, which have a high and persistent IgE production [19]. This regulation of IgE production was isotype specific and nonspecific for immunizing antigens. A genetic study, based on crossing the low-responder SJL/J and the high-responder BALB/c, suggested that IgE production was controlled by a single autosomal gene that was not linked to major histocompatibility complex (MHC) genes (*H2* genes in mice) [19]. In accordance with these characteristics, the gene was called the IgE level regulatory gene because it determined the amount of IgE produced. This gene is also considered to be one of the atopy genes; it is hypothesized that it functions as a protection gene against *T. spiralis*. This suggests an important concept: that atopic diseases and protection against helminth parasites are controlled by this gene.

Anaphylactic antibody (IgE and IgG₁) formation in mice infected with *T. spiralis* and protection against *T. spiralis*, evaluated by recovery of muscle larvae, were reported to be under the control of the *H2* gene complex [20]. A larger number of muscle larvae were recovered from SJL/J and AKR mice compared with from other strains, suggesting an inverse relationship between reaginic (IgE) antibody titers and the number of larvae. The number of muscle larvae in SJL/J mice was three

times the number in high IgE responders. A similar difference in muscle larvae burden after primary infection by *T. spiralis* has been reported in mouse strains by others [21]. However, one might interpret these results by hypothesizing that protection was controlled by the IgE level regulatory gene, as mentioned, independently of the *H2* gene complex.

Because IgE is present in extremely small amounts in serum compared with other immunoglobulins and because it cannot activate complement, IgE is not suitable for neutralization, phagocytosis or complement-dependent lysis of pathogens. IgE requires cells bearing IgE receptors to function fully as an antibody. In fact, mast cells have high-affinity receptors for IgE (FcεR I). Eosinophils, macrophages and other cells have low-affinity receptors for IgE (FcεR II).

Mast cells have a protective role in the expulsion of adult worms from the intestine after primary infection with *T. spiralis*, irrespective of the participation of FcεR. In mice, mast-cell-deficient W/W^v mice exhibited delayed expulsion [22]. Mice treated with anti-c-kit antibody to inhibit differentiation of mast cells showed delayed expulsion and suppression of mastocytosis in the intestine [23]. The involvement of IgE in the activation of mast cells for expulsion was suggested in IgE-knockout mice from which adult worms were expelled slower than from controls [18]. Although the effector molecules for adult worm expulsion have not been defined clearly, serum levels of mast cell protease-1 in infected IgE-knockout mice did not increase and were less than half those in control mice 7–14 days post-infection [18]. Recently, mast cell protease-1 was reported to be an indicator of mast cell activation and to facilitate expulsion after primary infection with *T. spiralis* [24]. In rats, mast cells are not likely to be involved in the expulsion of adult worms. It has been suggested that IgE antibody, independent of mast cells, and CD4⁺ T cells stimulate enterocytes to prevent penetration of adult worms to the intestine [7]. Intestinal IgE, which is different from serum IgE, seems to be responsible for this process [25]. Strain difference in rats has indicated that larger amounts of intestinal IgE resulted in faster expulsion [26].

According to these findings, IgE antibody has protective roles against *T. spiralis* larvae and adults (Table 1). However, the effector mechanism of protection through IgE antibody varies among host species.

Total IgE levels after infection were found to be 100 times those in uninfected mice [27]. The majority of IgE in helminth-infected hosts consists of potentiated IgE. Potentiated IgE production is the result of polyclonal IgE-B-cell activation by infection, and the IgE produced does not exhibit any binding activity to antigens of the infecting helminth. The biological importance of potentiated IgE production remains obscure. One possibility is that it is beneficial for the host because of the inhibition of severe anaphylactic reactions by hyper-IgE. Another possibility is that it is beneficial for the parasite: potentiated IgE interferes with defense mechanisms by means of the helminth-specific IgE antibody and, thus, leads to an easier establishment of infection. In other words, potentiated IgE could be interpreted as being an

Table 1. IgE-dependent protection against *Trichinella spiralis* in rats and mice, evaluated by modification of immune status

Immune status (host)	Target		Refs
	Muscle larva	Adult worm	
Anti-IgE treatment (rat)	Increased (primary infection)	ND ^a	[15]
IgE and T-cell transfer (rat)	Decreased (secondary infection: intestine)	ND	[11,13]
IgE deficient (mouse)	Unmodified (primary and secondary infection)	ND	[17]
IgE knockout (mouse)	Increased (primary infection)	Delayed expulsion (primary infection)	[18]

^aAbbreviation: ND, not determined.

escape mechanism from attack by the host immune system. In nature, it is not rare to find a single host infected by multiple species of helminth. In this situation, infection by the first species of helminth might lead to an easier establishment of infection in the host by the subsequent helminth species. Therefore, the presence of hyper-IgE as a general feature in helminth-infected hosts might be the result of survival by different helminth species.

IgE in humans infected with *Trichinella*

In human trichinellosis, as in many other helminthic infections [28], total IgE levels are increased; however, this increase is not a consistent phenomenon and trichinellosis cannot be excluded in its absence [29]. The issue that must be addressed is whether or not increased IgE levels are parasite specific.

Using a radioallergosorbent test, specific IgE was not detected in a group of 11 patients [30]. Using an indirect enzyme-linked immunosorbent assay (ELISA), *T. spiralis*-specific IgE was found in 7% of patients after one month of infection and in none after 11 months, when 17% of patients still had increased total IgE levels [31]. Using the same method, patients with specific IgE were found in other studies, in percentages ranging from 13 to 46% [32]. Using an amplified ELISA, 80–100% of patients in early infection were observed to have specific IgE that persisted for more than one year [32]. A low correlation between total and specific IgE has been observed for both *T. spiralis* and *Trichinella britovi* infections [32].

The human antibody response to affinity-purified antigens belonging to the *T. spiralis* larvae 1 (TSL-1) group was reported in 22 trichinellosis patients over a period of sixty weeks [33]. TSL-1 is a group of antigens with a molecular mass ranging from 45 to 100 kDa that is present among the excretory–secretory (ES) antigens of L₁ larvae [34], and glycosylated with the glycan β -tyvelose (a configuration that is unique to the *Trichinella* genus) [35]. Using an amplified ELISA, Salinas-Tobon *et al.* found TSL-1-specific IgE to be present after only three weeks of infection in ~60% of patients and, after six weeks and up to one year, all patients had specific IgE in their sera [33]. The IgE specific for the glycan moiety of TSL-1 antigens was evaluated in 50 trichinellosis patients with an amplified ELISA, using synthetic β -tyvelose as antigen. All patients were observed to have β -tyvelose-specific IgE in their sera, whereas IgE specific for ES antigens was detected in 78% of patients [36].

IgE antibodies against *T. spiralis* newborn larvae were detected after only three weeks in ~44% of patients, and specific IgE was still detected in 8% of them after one year [37]. A similar study was performed on IgE specific for

adult worms; of 22 individuals with trichinellosis, specific IgE appeared in 62% of patients after only three weeks of infection [38].

Owing to the different factors responsible for the severity of the clinical course of human trichinellosis, a correlation between specific IgE levels and clinical course severity is difficult to establish and, to our knowledge, there is no report focusing on this aspect. Certainly, elevated IgE levels are involved in the pathogenesis of some typical clinical signs observed in trichinellosis, such as edema – particularly periorbital, cutaneous rash [39]. In an outbreak in Italy caused by *T. britovi* involving ~300 people in 1986, specific IgE was significantly more frequent and present in higher levels in patients with periorbital edema (87%) than in patients who did not present this sign (62%), suggesting an allergic mechanism for the edema [40].

A significant correlation has been observed between the presence of specific IgE in patient sera during the first period of infection and during the blood eosinophilia period. 73% of patients with blood eosinophilia four months post-infection were found to be positive for the specific IgE, whereas only 28% of patients with normal blood-eosinophil levels had previously been found to be positive [41].

Clinical observations suggest that the *Trichinella*-specific IgE antibody is responsible for allergic manifestations that are typical of the clinical picture of trichinellosis but, at present, it is difficult to ascribe a protective role to this antibody in humans.

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