Rev. Inst. Med. trop. S. Paulo
51(2):73-75, March-April, 2009
doi: 10.1590/S0036-46652009000200003

MUSCULAR STRENGTH DECREASE IN *Rattus norvegicus* EXPERIMENTALLY INFECTED BY *Toxocara canis*

Pedro P. CHIEFFI(1,2), Renata T.R. AQUINO(1), Maria A. PASCHOALOTTI(1), Manoel Carlos S.A. RIBEIRO(1) & Antonia G. NASELLO(1)

SUMMARY

The muscular strength of experimental infected *Rattus norvegicus* with 3rd. stage *Toxocara canis* larvae was investigated. Fifty Wistar rats, divided in three groups (G1 - 20 rats infected by 300 eggs of *T. canis*; G2 - 20 rats infected by 2,000 eggs of *T. canis* and G3 - 10 rats without infection) had been used. Ten and 30 days after infection the muscular strength in the fore-feet of the rats was checked; at the same time, the body weight was determined. No significative differences in the body weight were noted among the infected and control rats in both occasions. Otherwise, an impairment on the muscular strength was observed in rats infected with *T. canis* 30 days after inoculation.

KEYWORDS: *Toxocara canis*; *Rattus norvegicus*; Muscular strength.

INTRODUCTION

Toxocara canis is an ascarid nematode parasite of the small bowell of dogs, with wide geographical distribution. Human infection by *T. canis* larvae is a quite common event[2,12] and, sometimes, results in the occurrence of visceral and/or ocular larva migrans[14].

T. canis transmission to dogs depends on several mechanisms; one of them includes the transference of third stage larvae from paratenic host tissues to dogs by prey-predator relationships[12]. *Rattus norvegicus* and some other rodent species are known as possible paratenic hosts of *T. canis* [3,4,10], harboring live third stage larvae in their tissues and organs.

There are some evidences of behavioral changes in paratenic hosts infected by *T. canis* larvae resulting in facilitation of prey-predation transmission mechanisms[6], as a consequence of the presence of larvae in their central nervous system (CNS). However, before reaching the CNS *T. canis* larvae have to pass through other rodent tissues like the striated muscular system[1,10]. In natural conditions this kind of migration should impair muscular performance in infected animals, favoring their predation by carnivores.

In the present paper the effect of muscular migration of *T. canis* larvae on the muscular strength of infected *R. norvegicus* was investigated.

MATERIAL AND METHODS

Fifty Wistar rats, females, 6 - 8 weeks old, were divided in three groups; G1 - 20 rats infected by 300 *T. canis* eggs; G2 - 20 rats infected with 2,000 *T. canis* eggs and G3 - 10 rats without infection. The rats were infected *per os* with a solution of *T. canis* eggs, obtained after dissection of female worms and maintained at least 30 days in a 2% solution of formaline, at 28 ºC. Before inoculation the eggs were carefully washed with distilled water. Rats of the control group (G3) only received distilled water.

Ten and 30 days after infection, employing a special apparatus (Grip Strength Meter, made by Ugo Basile, cat. no. 47105/47106), the muscular strength in the fore-feet of any rat was checked. This apparatus was designed for measuring forelimb grip strength in rodents. The rat is placed over a base plate, in front of a T-shaped grasping bar fitted to a force transducer connected to a peak amplifier. When pulled by the tail the rat grasps at the bar, until the pulling force overcomes its grip strength (Fig. 1). After the rat loses its grip on the grasping bar the peak pull-force achieved by the forelimbs was shown on a liquid crystal display in grams, and transformed to Newtons (N).

In both occasions the muscular strength was determined three times successively for each rat. In the same occasion the forelimbs strength was performed, the body weight of all rats was recorded.

In the 60th day post-infection all rats had been sacrificed and *T. canis* larvae were recovered from liver, lungs, brain, kidneys and muscles after digestion with HCl 0.5% for 24 hours at 37 ºC[15]. During all the experiment rats were carried out in special cages submitted to light-dark cycles of 12 hours; food and water *ad libitum* were allowed.

(1) Faculdade de Ciências Médicas da Santa Casa de São Paulo, São Paulo, SP, Brasil.
(2) Instituto de Medicina Tropical de São Paulo (LIM 06), São Paulo, SP, Brasil.
Correspondence to: Pedro Paulo Chieffi, Instituto de Medicina Tropical de São Paulo, Av. Dr. Enéas de Carvalho Aguiar 470, 05403-000 São Paulo, SP, Brasil. E. mail: pchieffi@usp.br

Fig. 1 - Grip strength meter for determining muscular strength in rodents.

The data were analyzed by repeated measures analysis of variance applying an interaction factor between time and experimental groups. All data were analyzed by SPSS-15 and the level of significance was $p = 0.05$.

RESULTS

Ten days after infection no significant difference in the muscular strength determined in the fore-feet was observed among the three groups of rats. However, 30 days after infection a significant decrease in muscular strength was observed in rats infected only with 2,000 (G2) *T. canis* eggs, when compared to the control group (G3), as shown in Figure 2. In both occasions no differences in body weight had been noted when infected rats were compared to no infected animals.

When rats infected with 300 *T. canis* eggs were compared to controls it was also noted, 30 days after infection, a significant decrease in the muscular strength among the three successive determinations carried on (Table 1).

On average 59 and 348 larvae had been recovered in the muscles of rats infected, respectively, with 300 and 2,000 *T. canis* eggs.

DISCUSSION

A wide range of small animals, mainly rodents, may transfer encysted larvae of *T. canis* to carnivores[2]. *R. norvegicus* has been pointed out as *T. canis* paratenic host[3,4].

Recently it was observed the presence of 3rd stage larvae of *T. canis* in muscles of *R. norvegicus* from the 5th at least the 60th day after experimental infection with embryonated eggs of this ascarid and predominantly between the 10th and the 30th days after infection[10], showing a migration larvae pattern similar of that found in other rodent species[1,8]; corroborating the possibility of *T. canis* larval transference to carnivore animals by prey-predator relationship. On the other hand, there are some indications of muscle changes in humans infected by *T. canis*, resulting in the emergence of antistriational antibodies, suggesting the occurrence of anti-muscle autoimmune response[11]. Experimentally working with mice infected by *Trichinella spiralis* RAU & PUTTER (1984)[13] found a dose-dependent impairment on mice running responses, denoting that those hosts should be more susceptible to predation.

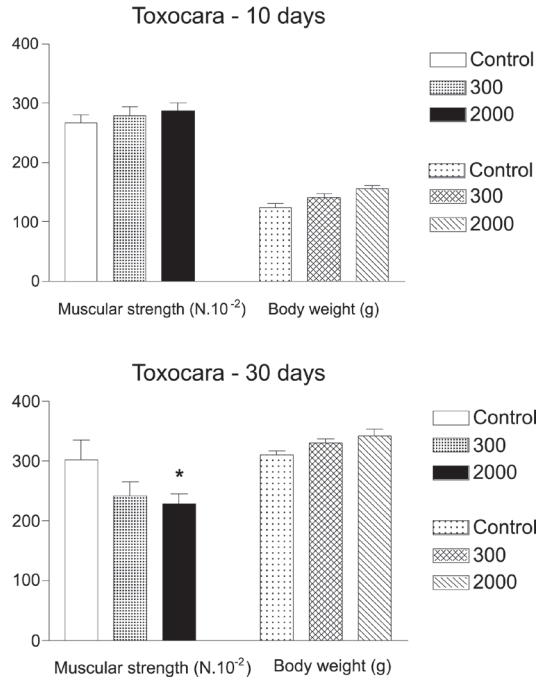

Fig. 2 - Muscular strength and body weight in *Rattus norvegicus* 10 and 30 days after infected by *Toxocara canis* eggs.

Table 1
Muscular strength in *Rattus norvegicus* infected with 300 *Toxocara canis* eggs, 30 days after inoculation, according to the three measures performed

Measures	Control Am (sd)	Infected Am (sd)
First	312.1 (194.6)	408.1 (251.5)*
Second	213.3 (112.5)	250.6 (178.7)*
Third	295.5 (163.4)	194.8 (108.7)*

*$p < 0.05$; Am = arithmetic mean; sd = standard deviation.

Otherwise, ZOHAR & RAU (1986)[16] observed behavioral alterations in mice harboring encysted muscle larvae of *T. spiralis*, prevented by specific chemotherapy.

In the present experiment it was noted similar growth pattern in rats infected with 300 and 2,000 *T. canis* eggs 10 and 30 days after infection, when compared with control group. Our data suggest that *T. canis* infection has no influence on body development (Fig. 1). Otherwise, as shown in Figure 1, a decrease on muscular strength was detected 30, but not 10 days after infection, mainly when rats received an inoculum of 2,000 *T. canis* eggs, being in accordance with previous data of others[7,9] testing behavioral parameters in mice infected by this ascarid.

It is worth-while to call attention on the decrease observed among the three measures of the muscular strength performed in rats infected with 300 eggs of *T. canis*, suggesting that those animals showed more fatigue than controls, when submitted to successive struggles. Curiously rats infected with 2,000 eggs did not show the same performance. However,

COX & HOLLAND (1998)[5] observed similar paradoxical results analyzing the behavior of mice experimentally infected by *Toxocara canis* larvae: those harboring the lower brain infection displayed a greater level of behavior alterations.

Our results reinforce the hypothesis that infection by *T. canis*, inducing a decrease of muscular performance, facilitates *T. canis* transmission by prey-predator relationship. Further studies are been developed in our laboratory to test the influence of these results on behavioral parameters of *T. canis* infected rats.

RESUMO

Diminuição da força muscular em *Rattus norvegicus* experimentalmente infectados por *Toxocara canis*

Roedores são reconhecidos como hospedeiros paratênicos de *Toxocara canis*. Um dos mecanismos de transmissão desse ascarídeo para cães, seus hospedeiros habituais, consiste na predação de hospedeiros paratênicos, que albergam larvas de terceiro estágio em seus órgãos e tecidos, entre os quais músculos estriados. No presente trabalho estudou-se se a infecção por larvas de *Toxocara canis* provoca alterações na força muscular de exemplares de *Rattus norvegicus* experimentalmente infectados. Cinqüenta *Rattus norvegicus* foram divididos em três grupos: G1, com 20 ratos infectados com 300 ovos de *Toxocara canis*; G2, com 20 ratos infectados com 2.000 ovos do ascarídeo e G3, com 10 ratos sem infecção. Dez e 30 dias após a infecção determinou-se a força muscular nas patas dianteiras dos roedores; ao mesmo tempo, avaliou-se seu peso corporal. Não foram observadas diferenças no peso dos animais dos três grupos; contudo, verificou-se decréscimo na força muscular dos ratos infectados com 2.000 ovos após o 30º dia de infecção.

ACKNOWLEDGMENTS

This investigation was supported by a FAP/FCMSC grant.

REFERENCES

1. BARDÓN, R.; CUÉLLAR, C. & GUILLÉN, J.L. - Larval distribution of *Toxocara canis* in BALB/c mice at nine weeks and one year post-inoculation. **J Helminth., 68:** 359-360, 1994.

2. BARRIGA, O.O. - A critical look at the importance, prevalence and control of toxocariasis and the possibilities of immunological control. **Vet. Parasit., 29:** 195-234, 1988.

3. BURREN, C.H. - The distribution of *Toxocara canis* larvae in the central nervous system of rodents. **Trans. roy. Soc. trop. Med. Hyg., 66:** 937-942, 1972.

4. CHIEFFI, P.P.; DEL GUÉRCIO, V.M.F.; UEDA, M. & MELLO, L.B. - Importância de *Rattus norvegicus* capturados no município de São Paulo, SP, Brasil, como hospedeiros paratênicos de *Toxocara* canis (Ascaroidea, Nematoda). **Rev. Inst. Adolfo Lutz., 41:** 89-91, 1981.

5. COX, D.M. & HOLLAND, C.V. - The relationship between numbers of larvae recovered from the brain of *Toxocara canis*-infected mice and social behaviour and anxiety in the host. **Parasitology, 116:** 579-594, 1998.

6. COX, D.M. & HOLLAND, C.V. - The influence of mouse strain, infective dose and larval burden in the brain on activity in *Toxocara*-infected mice**. J. Helminth., 75:** 23-32, 2001.

7. COX, D.M. & HOLLAND, C.V. - Relationship between three intensity levels of *Toxocara canis* larvae in the brain and effects on exploration, anxiety, learning and memory in the murine host. **J. Helminth., 75:** 33-41, 2001.

8. EPE, C.; SABEL, T.; SCHNIEDER, T. & STOYE, M. - The behavior and pathogenicity of *Toxocara canis* larvae in mice of different strains. **Parasit. Res., 80:** 691-695, 1994.

9. HAMILTON, C.M.; STAFFORD, P.; PINELLI, E. & HOLLAND, C.V. - A murine model for cerebral toxocariasis: characterization of host susceptibility and behaviour. **Parasitology, 132:** 791-801, 2006.

10. LESCANO, S.A.Z.; QUEIROZ, M.L. & CHIEFFI, P.P. - Larval recovery of *Toxocara canis* in organs and tissues of experimentally infected *Rattus norvegicus*. **Mem. Inst. Oswaldo Cruz, 99:** 627-628, 2004.

11. MACURA-BIEGUN, A.; PITUCH-NOWOROLSKA, A.; REWICKA, M.; MROZEWICZ, B. & NOWOROLSKI, J. - Antistriational antibodies during *Toxocara canis, Trichinella spiralis* infections. **Comp. Immunol. Microbiol. infect. Dis., 21**: 101-106, 1998.

12. OVERGAAUW, P.A.M. - Aspects of *Toxocara* epidemiology: toxocarosis in dogs and cats. **Crit. Rev. Microbiol., 23**: 233-251, 1997.

13. RAU, M.E. & PUTTER, L. - Running responses of *Trichinella spiralis*-infected CD-1 mice. **Parasitology, 89:** 579-583, 1984.

14. SCHANTZ, P.M. - *Toxocara* larva migrans now. **Amer. J. trop. Med. Hyg., 41**(3 suppl.): 21-34, 1989.

15. XI, W.G. & JIN, L.Z. - A novel method for the recovery of *Toxocara canis* in mice. **J. Helminth., 72:** 183-184, 1998.

16. ZOHAR, A.S. & RAU, M.E. - The role of muscle larvae of *Trichinella spiralis* in the behavioral alterations of the mouse host. **J. Parasit., 72:** 464-466, 1986.

Received: 13 December 2008

Accepted: 13 February 2009