

BRIEF COMMUNICATION

BEHAVIORAL CHANGES IN *Rattus norvegicus* COINFECTED BY *Toxocara canis* AND *Toxoplasma gondii*

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SUMMARY

Using an elevated plus maze apparatus and an activity cage, behavioral changes in *Rattus norvegicus* concomitantly infected by *Toxocara canis* and *Toxoplasma gondii* were studied, during a period of 120 days. Rats infected by *Toxocara canis* or *Toxoplasma gondii* showed significant behavioral changes; however, in the group coinfecting by both parasites a behavioral pattern similar to that found in the group not infected was observed thirty days after infection, suggesting the occurrence of modulation in the behavioral response.

KEYWORDS: Behavioral changes; *Toxocara canis*; *Toxoplasma gondii*; Concomitant infections.

There are several references to the occurrence of behavioral changes in rodents infected with *Toxoplasma gondii*^{9,10,11,17,18}, as well as with *Toxocara canis*^{1,2,4,5,8}, parasites of animals and human beings which are there in order to facilitate their transmission either through predator-prey relationships or other mechanisms^{10,15}. COMBES in 1991³ coined the term “favorization” while studying these processes, to draw attention to this way of facilitating the transmission mechanisms found in some species of parasites.

Although the occurrence of either coinfections or multiple infections in the same host is common in natural conditions, this has not usually been considered in studies that seek to characterize behavioral changes in parasitized hosts. In the present study it was evaluated whether or not such changes occurred in *Rattus norvegicus* experimentally coinfecting with *Toxocara canis* and *Toxoplasma gondii*, since both parasites could affect the host’s central nervous system.

Female Wistar rats (n = 55), 6 - 8 weeks old, were divided into four groups:

- G1 – 15 rats infected with 400 embryonated eggs of *Toxocara canis*.
- G2 – 15 rats infected with 10 cysts of *Toxoplasma gondii*.
- G3 – 15 rats coinfecting with 400 embryonated eggs of *Toxocara canis* and 10 cysts of *Toxoplasma gondii*.
- G4 – 10 rats without infection (control group).

Rats were kept in standard cages, five per cage, with water and food *ad libitum*, and were maintained in a room equipped with an exhaust air device and alternating periods of light and darkness every 12 hours. Rats

were infected with *Toxocara canis* and/or *Toxoplasma gondii* by gavage and the animals of the control group (G4) received an equivalent amount of tap water by the same method.

All animals were submitted to the determination of behavioral variables (anxiety and motor activity) at 30, 60 and 120 days after infection.

Anxiety levels were determined using an elevated plus maze apparatus, in accordance with the technique described earlier¹⁴. The following variables were evaluated using this apparatus: head dipping, number of entries and time spent in the open and closed arms, during a period of five minutes.

Motor activity was evaluated using an activity cage and the following variables were recorded over a period of five minutes: grooming, vertical and horizontal motion, and immobility time.

After day 120 post-infection all rats were sacrificed and their brains were submitted to digestion with 0.5% HCl, for 24 hours at 37 °C, in accordance with the technique described by XI & JIN¹⁹ for recovering larvae and cysts of *Toxocara canis* and *Toxoplasma gondii* respectively.

The statistical analysis of the data obtained was carried out employing an analysis of variance (ANOVA) with the Tukey post-test, using the Prism 3.0 program. The experimental protocol was approved by the Ethic Committee in Animal Experimentation of the *Faculdade de Ciências Médicas da Santa Casa de São Paulo*.

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Only results with statistical significance ($p < 0.05$) will be commented upon in this paper.

Head dipping: At 60, but not 30 or 120 days after infection, the group infected only by *Toxoplasma gondii* (G2) showed a statistically significant difference when compared to the control group (Fig. 1).

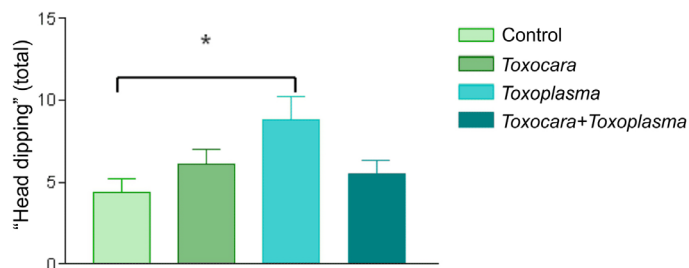


Fig. 1 - Head dipping observed in mice 60 days after infection by *Toxocara canis*, *Toxoplasma gondii* and concomitant infection by both parasites.

Grooming: No statistically significant difference was noted in the total grooming in all groups of rats at 30 and 120 days after infection, but significantly less grooming activity was found in the group infected only by *Toxocara canis* (G1) 60 days after infection when compared to the control (G4) (Fig. 2).

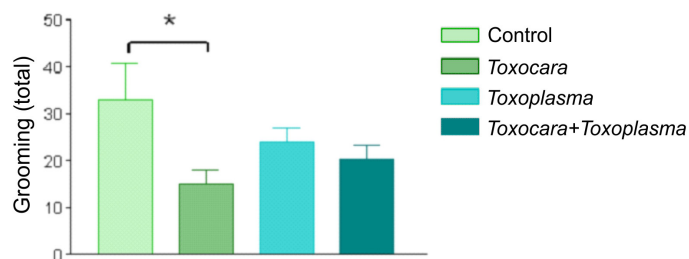


Fig. 2 - Grooming activity observed in mice 60 days after infection by *Toxocara canis*, *Toxoplasma gondii* and concomitant infection by both parasites.

Time of immobility: At thirty days after infection a decrease in immobility was observed in the coinfecting group (G3) when compared to rats with single infection (G1 and G2). The total time of immobility of the animals of G3 was similar to that observed in the uninfected control group (Fig. 3). On the other hand, 60 days after infection all three groups of infected rats showed a significant reduction in immobility time when compared to the control group (Fig. 4).

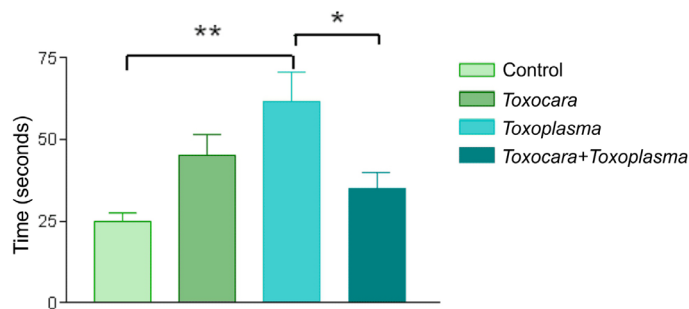


Fig. 3 - Time of immobility observed with activity cage in mice 30 days after infection by *Toxocara canis*, *Toxoplasma gondii* and concomitant infection by both parasites.

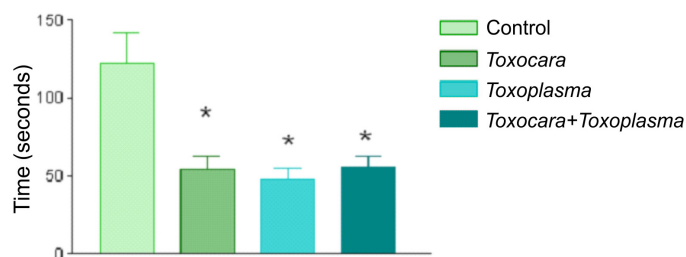


Fig. 4 - Time of immobility observed with activity cage in mice 60 days after infection by *Toxocara canis*, *Toxoplasma gondii* and concomitant infection by both parasites.

DISCUSSION

Host behavior manipulation is considered a strategy of transmission that enhances the chance of a parasite being passed from one host to another¹⁵. There are several examples of well-documented behavior changes in some rodents infected with *Toxoplasma gondii*^{7,16} or *Toxocara canis*^{1,2,8} that can be considered as host manipulation, aiming to facilitate the transmission rate of these parasites. The possibility of coinfection by both species is not usually considered in this kind of study, although such a situation is probably not rare in natural conditions, both in animals and in humans living in unsanitary conditions.

Recently LESCANO *et al.*¹³ showed the occurrence of some delay in the humoral immune response in mice coinfecting by *Toxocara canis* and *Toxoplasma gondii* when compared to animals infected only by *Toxocara canis*. On the other hand, in experimental models the interaction among *Toxoplasma gondii* and other parasites sometimes results in different frames than that observed in single infections^{6,11}.

In the present paper, despite being a pilot study which needs to be investigated further in order for a better understanding of the results observed, an apparently paradoxical finding was obtained, i.e. rats infected with *Toxocara canis* or *Toxoplasma gondii* showed significant behavioral changes, similar to the findings of other reports^{2,6,7,13}. However, when they were coinfecting with both parasites, they showed a behavioral pattern similar to the control group without infection (G4), suggesting a modulation in the behavioral response.

These results, which are still preliminary, are being studied in more depth in our laboratory using a large number of animals, in order to confirm these findings and try to clarify its mechanism.

RESUMO

Alterações comportamentais em *Rattus norvegicus* coinfectados por *Toxocara canis* e *Toxoplasma gondii*

Estudou-se a ocorrência de alterações comportamentais em *Rattus norvegicus* infectados de forma isolada e concomitante por *Toxocara canis* e *Toxoplasma gondii*, por período de 120 dias, utilizando labirinto em cruz elevado e actômetro. Ratos infectados apenas por *Toxocara canis* ou *Toxoplasma gondii* apresentaram alterações comportamentais em níveis significativos; todavia, no grupo infectado com ambos os parasitos observou-se padrão de comportamento semelhante ao do grupo não infectado no 30º dia após infecção, sugerindo ocorrência de modulação na resposta comportamental.

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