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Achatina fulica infected by Angiostrongylus cantonensis on beaches, in the west zone of Rio de Janeiro, Brazil

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ABSTRACT

Angiostrongylus cantonensis is considered the main etiological agent of eosinophilic meningitis in humans. At present, this zoonosis is considered an emerging disease mainly in the Americas. The aim of this study was to determine the prevalence of Achatina fulica infected by Angiostrongylus cantonensis in restinga areas along beaches in the west zone of Rio de Janeiro city, Brazil. The study areas included the following beaches: Barra da Tijuca, Recreio dos Bandeirantes, Reserva, Prainha and Grumari. Ninety specimens of Achatina fulica were collected. Positive molluscs were found only in Barra da Tijuca. Infection prevalence was 5.5%. The presence of this parasite in the beachfront areas, in the west zone of Rio de Janeiro city demonstrates the potential risk of infection for visitors and the expansion of this helminth in the State of Rio de Janeiro.

KEYWORDS: Emerging parasitosis. Eosinophilic meningoencephalitis. Giant African snail Nematode.

The nematode *Angiostrongylus cantonensis* (Chen, 1935) is a zoonotic parasite that causes eosinophilic meningitis, the main clinical manifestation of human angiostrongyliasis^{1,2}. Synanthropic rats of the genus *Rattus* are the main definitive host^{3,4} and the African snail *Achatina fulica* the most important intermediate host⁵⁻⁸. Adult worms parasitize pulmonary arteries of the definitive host, where females lay eggs that develop into first-stage (L_1) larvae^{3,9}. These larvae move to the alveoli and pharynx and are swallowed, then eliminated in feces. Snails or slugs are infected by L_1 larvae through ingestion or penetration, after which the two molts occur and the larvae become infective to definitive hosts and humans. Humans become infected by eating undercooked or raw intermediate or paratenic hosts³.

In Brazil, the first record of *A. fulica* naturally infected by *A. cantonensis* was in the municipality of Vila Velha, *Espírito Santo State*, and *São Vicente*, in *São Paulo State*⁵. Since then, it has been reported in the municipalities of *São Gonçalo*, Barra do Pirai and Angra dos Reis in Rio de Janeiro State^{10,11}, Joinville and Navegantes in Santa Catarina State^{10,11}, *Paranaguá* in *Paraná State*¹¹, Escada in Pernambuco State⁷ and *Belém*, in *Pará State*¹². According to Morassutti *et al.*¹³, in Brazil, there are intermediate and definitive hosts infected with *A. cantonensis*, however, few cases of infection have been reported. This fact could be justified by the lack of knowledge about the parasite by physicians and professionals prepared to make the diagnosis for angiostrongyliasis. The aim of this study was to determine the presence and prevalence of *A. fulica* naturally infected by *A. cantonensis* on beaches in the west zone of Rio de Janeiro city.

Between October and November 2015, A. fulica specimens were collected in

nine points of beachfront areas in the west zone of Rio de Janeiro. Collection points coordinates were: Barra da Tijuca (23°00'39"S 43°21'48"W, 23°00'39"S 43°21'59"W, 23°00'41"S 43°22'15"W), Recreio dos Bandeirantes (23°10'22"S 43°26'56"W, 23°10'44"S 43°28'13"W), Prainha (23°20'06"S 43°30'16"W), Grumari (23°20'53"S 43°31'40"W, 23°20'55"S 43°31'38"W) and Reserva (23°10'20"S 43°25'09"W). A total of 90 snails were collected, ten per collection point. The specimens were collected manually, preferably at night or on rainy mornings. The collection was done using gloves, after which the snails were placed in plastic containers and transported in biological material boxes to the Laboratory of Biology and Parasitology of Wild Mammal Reservoirs, Oswaldo Cruz Foundation, Rio de Janeiro. The cephalopodal masses of molluses were individually fragmented and L, larvae were obtained according to Garcia et al. 14. The number of larvae recovered was expressed as mean \pm standard deviation.

After counting of L₃ larvae, three Wistar rats were infected with 60 larvae each through an orogastric probe (Medsonda, 5 mm) and kept in a vivarium to confirm the infection by *A. cantonensis*. All procedures were approved by the Animal Experimentation Ethics Committee of the Oswaldo Cruz Foundation (LW-47/14). Adult nematodes were recovered 40 days after infection in the pulmonary arteries of the rodents and their morphology was identified according to Anderson *et al.* ¹⁵ and Maldonado *et al.* ¹⁰.

Five A. fulica were infected by A. cantonensis in Barra da Tijuca with a prevalence of 5.5% (5/90). The mean intensity of L_3 recovery was 183 (\pm 238) per infected snail. Twenty-one, 31 and 39 adult worms of A. cantonenis with a total of 58 males and 33 females were recovered in the three infected rats, respectively. In males, specific morphological characteristics as the caudal bursa and the spicule length were observed confirming the identification of helminth specimens as A. cantonensis.

Human angiostrongyliasis is an acute disease caused by *A. cantonensis* that affects the central nervous system. This infection was previously restricted to Asia and the Pacific islands mainly due to food and cultural habits of these populations¹⁶. However, the rapid spread of *A. fulica* in different countries around the world has contributed to the dispersion of the nematode and consequently the zoonosis^{11,17}. In Brazil, intermediate and definitive hosts infected with *A. cantonensis* have been identified in urban areas of several Brazilian states, including those in the North, Northeast, Southeast and South regions¹³. Recent studies carried out in the municipality of *São Gonçalo*, Rio de Janeiro State, showed that *R. norvegicus*, *A. fulica* and *B. similaris* are the main definitive and intermediate hosts of *A. cantonensis*^{18,19}, playing important roles in

the transmission dynamics of this nematode, since they presented high abundance and high infection rates in various studies.

Infected molluscs were found exactly in the most anthropic areas of the study, where presence of trash left by beachgoers, garbage from food kiosks and rats was observed. This plentiful food availability contributes to the presence of both intermediate and definitive hosts, thus favoring the life cycle of the parasite, as also observed by Simões *et al.*⁴. The prevalence of the nematode in the intermediate host was low, differing from the results reported by Oliveira *et al.*¹⁹ that observed a prevalence of 78.7% in *A. fulica* infected by *A. cantonensis*. The rate of helminth recovery after the experimental infection corroborates with the results obtained by Garcia *et al.*¹⁴, who observed a variation of 25-51%.

From these results, it is possible to conclude that the presence of *A. fulica* naturally infected by *A. cantonensis* along the beachfront areas of Barra da Tijuca should serve as an alert to public health authorities of the potential risk of infection to visitors, especially children. Moreover, the presence of this helminth along the beaches of the west zone of the Rio de Janeiro demonstrates the expansion of this nematode in the State of Rio de Janeiro.

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