

ANGIOSTRONGYLUS COSTARICENSIS NATURAL INFECTION IN VAGINULUS PLEBEIUS IN NICARAGUA

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SUMMARY

A study of 94 slugs, collected from urban and rural areas in and around León, Nicaragua, was carried out in order to confirm the role of *Vaginulus plebeius* as an intermediate host of *Angiostrongylus costaricensis*. Third-stage larvae of *A. costaricensis* were obtained from these molluscs. Some of these larvae were then orally inoculated into two laboratory-bred rats *Sigmodon hispidus*

and adult worms of *A. costaricensis* were recovered two months later. The infection rate of these slugs ranged from 4 % in urban areas to 85 % in rural areas. These data suggest that contamination to man is a major risk especially in rural areas and that abdominal angiostrongyliasis could be a health problem in Nicaragua.

RÉSUMÉ : Infection naturelle par *Angiostrongylus costaricensis* chez *Vaginulus plebeius* au Nicaragua.

Quatre-vingt-quatorze limaces collectées dans des zones urbaines et rurales de la province de León, ont été soumises à un processus de digestion artificielle. Des larves du troisième stade ont été isolées. Des rats du coton, *Sigmodon hispidus*, élevés au laboratoire, ont été infestés par voie orale à l'aide de ces larves. Les rongeurs ont été autopsiés deux mois après l'infestation et de nombreux vers adultes ont été retrouvés dans les artères mésentériques. Les

parasites ont été identifiés à *Angiostrongylus costaricensis*. Le taux d'infestation chez les limaces était de 4 % en zone urbaine à 85 % en zone rurale. Cette prévalence montre que la population rurale est très exposée à la contamination, ce qui permet de penser que l'angiostrongylose abdominale peut constituer un problème méconnu de santé publique au Nicaragua.

INTRODUCTION

Angiostrongylus costaricensis Morera and Céspedes, 1971 (= *Morerastrongylus costaricensis* Chabaud, 1972) is a nematode metastrongylid widely distributed throughout the american continent being found in humans and natural hosts from USA to northern Argentina (Loria *et al.*, 1980; Malek, 1981; Morera *et al.*, 1971 and 1983; Quintana, 1984; Sierra *et al.*, 1972; Ubelaker *et al.*, 1979; Zavala *et al.*, 1974; Zillioto *et al.*, 1975). Several studies on the life cycle of this parasite have shown that the gastropod *Vaginulus plebeius* is the most important natural interme-

mediate host (Morera *et al.*, 1970 and 1983; Morera, 1985) and the natural definitive hosts are at least twelve species of rodents, especially cotton rats *Sigmodon hispidus* (Morera, 1970 and 1985; Tesh *et al.*, 1973). Furthermore, marmosets, the coati-mundi (*Nasua narica*) and domestic dogs have been found naturally infected (Lewis *et al.*, 1982; Morera *et al.*, 1990). Man is an accidental host.

Infected molluscs release third-stage larvae with their mucus secretion, contaminating anything that comes into contact with them, including food and objects. Definitive and accidental hosts, including man, become infected by eating slugs, food or vegetables that have been contaminated with the mucus secretion of molluscs containing the infective third-stage larvae. Several witnesses have stated that children in Latin-American countries play with slugs, consequently contamination could be by molluscs—hand-objects—mouth contact. Slugs are infected by ingesting fecal material from rodents containing first-stage larvae.

In Nicaragua, several case of the human disease, abdominal angiostrongyliasis, have been reported (Duarte *et al.*, 1992). *V. plebeius* appears to be the main source of infection for humans in other countries (Morera, 1985). This mollusc is abundant and widespread in Nicaragua. The present study was carried-out to confirm the role of *V. plebeius* as the intermediate host of *A. costaricensis* in the country.

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MATERIALS AND METHODS.

SLUGS

Molluscs were collected from urban and rural areas in and around León, corresponding to the geographical locality of patients suffering from abdominal angiostrongyliasis, reported in an earlier study (Duarte *et al.*, 1992). The collection was done at night between May and June of 1986, the beginning of the rainy season.

GEOGRAPHICAL AREAS

León is the second most important town in Nicaragua, with a population of 320,000 inhabitants. It is situated about 90 km north of Managua, the capital city, on the pacific coast, close to the volcanic cordillera. The urban areas studied were three neighbourhoods, Fátima, San Francisco and Subtiava, located respectively in the north, centre and west of León. The rural area was a farm, San José del Tololar, located approximately 8 km north of León. Molluscs were collected in the backgardens of the patients and neighbouring dwellings in León and about 50 m from the farmhouse, in its banana garden.

LIFE CYCLE

Each slug after evisceration was submitted to a pepsin artificial digestion, using the method described by Wallace and Rosen, 1969. The resulting product was placed in Baermann funnels. Five hours later the sediment was examined under a dissecting microscope and the larvae obtained from each slug were counted.

Two ten-week old laboratory-bred cotton rats, *S. hispidus*, raised in the Laboratory of Parasitology, University of Costa Rica, were each orally inoculated with 50 of these larvae. Two months later the rats were sacrificed with chloroform and examined for adult parasites.

The classification of the larva and adult parasitic stages as *A. costaricensis* was based on their morphological features.

RESULTS

A total of 553 slugs were collected, 487 from the urban areas and 66 from the rural area. However only 74 of the urban slugs (19 %) and 20 of the rural slugs (31 %) were used.

NATURAL INFECTION IN THE MOLLUSCS

Twenty of the 94 slugs studied were found to be infected with third-stage larvae, corresponding to a global infection rate of 21 % (Table I). However, the number of molluscs infected varied considerably between urban and rural areas. In the urban area, only 3 of the 74 slugs studied (4 %) were found to be infected. All three infected slugs were from the neighbourhood Fátima, corresponding to an infection rate of 15 % (3 out of 20 slugs). No slugs from the other two neighbourhoods were found to be infected. In the rural area 17 of the 20 molluscs were found to be positive for third-stage larvae, corresponding to an infection rate of 85 %. Also there was a marked difference between urban and rural areas in the number of larvae obtained per mollusc. In the urban areas 1, 3 and 9 larvae were obtained from each mollusc respectively but the

Table I. — Infection rate of molluscs by *Angiostrongylus costaricensis*.

Locality	No. of slugs studied	Infected slugs	
		No.	%
<i>Urban</i>			
León City	74	3	4
<i>Rural</i>			
San José del Tololar	20	17	85
<i>Total</i>	94	20	21

number of larvae per mollusc from the rural areas ranged from 200 to 4,600.

EXPERIMENTAL INFECTION IN THE RODENTS

The two infected rats began to discharge first-stage larvae in their feces 25 days after inoculation. One rat died spontaneously and the advanced state of decomposition did not permit a necropsy to be carried out. However, the autopsy of the surviving rat, performed 60 days after inoculation, showed abundant adult female and male parasite within the ileocecal arteries.

PARASITES

First and third-stage larvae and adult parasites were studied microscopically. The morphological features permitted their identification as *A. costaricensis* (Morera, 1973).

DISCUSSION

In rodents the adult parasites of *A. costaricensis* live in the mesenteric arteries and their branches, which supply the small intestine and cecum. The mature female deposits eggs, which embryonate in the intestinal wall and these larvae then migrate through it to the lumen and are eliminated with the feces. In humans, the eggs and larvae are trapped in the intestinal tissues, where they cause an important inflammatory reaction, associated with thrombosis and extensive zone of necrosis. These lesions are responsible for a severe clinical syndrome, potentially grave or fatal, if a surgical intervention is not performed. In areas where there is a shortage of information on the disease and the serological test are unavailable, clinical and parasitological diagnosis are very difficult (Morera, 1985; Morera *et al.*, 1990) as the disease can resemble many other inflammatory or neoplastic diseases and first-stage larvae are not found in human stool.

Vaginulus plebeius was first identified as a natural intermediate host of *A. costaricensis* in Costa Rica (Morera *et al.*, 1970). Later it was confirmed in Ecuador (Morera

et al., 1983) and now in Nicaragua. The infection rate of molluscs in Léon was similar to that reported in Costa Rica for the whole country, which ranged from 28 % to 75 % (Morera, 1985).

There was a marked variation in the infection rate of the molluscs between urban and rural areas of Léon. The causes for this are unclear. It is possible that there could be another species of slug or rodent acting as intermediate and definitive host in urban areas. Another possibility is that molluscs from urban areas were young and had a much smaller chance of coming into contact with feces of rats contaminated with first-stage larvae.

The high infection rate observed in rural slugs suggests that human contamination is a major risk in this area. However six of the seven human cases (85 %) reported from Nicaragua were observed in urban inhabitants (Duarte et al., 1992). There is no obvious explanation for this. It may be that there are more opportunities for contact between man and molluscs in urban areas or that there are differences in host responsiveness or in socio-economic and behavioural factors. Another consideration is that a number of the urban cases could have been infected in rural areas. Further studies are required to clarify the situation.

The data presented here shows that naturally-infected molluscs are widely distributed and that there are numerous opportunities for man to become infected. The results suggest that abdominal angiostrongyliasis could be a potential public health problem in Nicaragua.

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