Africa Journal of Animal and Biomedical Sciences 7(1), 2012
ISSN: 1819-4214

Angiostrongylus cantonensis and other parasites infections of rodents of Budongo Forest Reserve, Uganda

L. Mugisha[1,2*], Okot Bwangamoi [3], M. R. Cranfield[4],
[1]Department of Wildlife and Aquatic Animal Resources, College of Veterinary Medicine, Animal Resources and Biosecurity, Makerere University, P.O.Box 7062, Kampala, Uganda, 256-772-566551
[2]Conservation & Ecosystem Health Alliance (CEHA), P.O.Box 34153, Kampala, Uganda
[3]Department of Pathology Faculty of Medicine, Gulu University, P.O.Box 166, Gulu, Uganda.
[4]Mountain Gorilla Veterinary Project, Department, Maryland Zoo, Druid Hill Park, Baltimore, Maryland21217. 410-396-0070
*Correspondence email: lmugisha@vetmed.mak.ac.ug.

Abstract

Angiostrongylus cantonensis has been documented for the first time in tissues of rodents from Budongo Forest Reserve in Uganda. Budongo Forest Reserve (BFR) is situated in Western Uganda in the Disricts of Masindi and Hoima. Thirty-three rodents were humanely captured, euthanized and tissues processed for histological examination. Six % (2/33) of rodents belonging to *Lophomurus, sikapusi* were infected with *Angiostrongylus cantonensis.* The parasite was found in the rodent lung tissues with lesions characterized by granulomatous pneumonia, periarteritis, and the presence of eosinophils, macrophages, plasma cells and degenerating adults, eggs and larvae.

Keys words: *Angiostrongylus cantonensis*, rodents, Budongo Forest Reserve,

Introduction

Angiostrongylus cantonensis is the most common infectious cause of eosinophilic meningitis worldwide (1). Although human infections with *A. cantonensis* are traditionally associated with South East Asia and the Pacific Basin, sporadic cases have been reported in several other countries outside this region (1,2). In the Caribbean, eosinophilic meningitis has not been commonly reported, although *A. cantonensis* has been found in rats from Cuba, Puerto Rico, and the Dominican Republic (3,4,5). Enzootic *Angiostrongylus cantonensis* was reported in rats and snails after an outbreak of human eosinophilic menengitis among tourists to Jamaica in 2000 (6). Beyond the Indo-pacific region, this worm has been found in rodents in Madagascar (ca 1963), Egypt (1977), New Orleans, Louisiana (1985), Port Harcourt, Nigeria (1989) (6)

Angiostrongylosis in humans may present as transient meningitis or a more severe disease involving the brain, spinal cord and nerve roots, with a characteristic eosinophilia of the peripheral blood and cerebral spinal fluids (1). The disease usually presents as non-specific symptoms such as headache, fever, paralysis and even coma which may be difficult to differentiate from other infections that involve the brain including hydatid disease, cysticercosis, strongylodosis and visceral larval migrans (7). However, a fatal case of infection with *Angiostrongylus cantonensis* was reported in a 14-month-old Jamaican boy recovering multiple sections of the worm in the boy's brain and lungs on autopsy (8).

Adult *A. cantonensis* worms are found in the branches of the pulmonary artery and sometimes in the right ventricle of rats, which are principal and definitive host for the parasite. The females produce eggs, which are carried to the lung capillaries where they hatch and first-stage larvae enter the alveoli of the respiratory tract. From here the first –stage larvae migrate up to the trachea, are swallowed and passed in the host's feces (9). This parasite requires an intermediate host to complete its life cycle, most often a slug or an aquatic or marine snail. The first-stage larvae are ingested by the intermediate host and molt twice into the infective third-stage larvae. Humans become infected by ingesting raw or undercooked snails or slug. In addition, freshwater prawns, crabs, and toads may act as sources of infection to humans (9). Third-stage larvae released from host tissues penetrate the intestinal tissues of humans and are carried by the blood to the liver, heart, lungs, and ultimately to the central nervous system (CNS) (9). While in the CNS, the larvae continue to develop to the fourth and fifth stage. In humans, there is rarely

migration to, and further development in, the lung (9). This study was initially undertaken to understand the role of rodents in the transmission of diseases in the current scenario of increased levels of human-wildlife interactions in Budongo Forest Reserve, Uganda.. It was during this study that *Angiostrongylus cantonensis* infection was discovered in rodents in this study area.

Materials and methods

Thirty three rodents (n=33) were trapped using Sherman traps containing peanut butter as bait in the Budongo Forest Reserve. (BFR) is situated in Western Uganda in the Disricts of Masindi and Hoima located at latitude 1^0 35' and 1^0 55N and longitude 31^0 18'and 31^0 42'E. The traps were placed in typical rodent traveling paths under bushes or next to large objects in the environment such as buildings, trees or large rocks. The traps were set at the identified locations each evening and examined by 8: 30 am the next day. The captured rodents were euthanized using halothane and placed in a plastic bag for post-mortem. The rodents were dissected and the major organs; the heart, liver, lungs, kidneys, diaphragm, intestines as well as the stomach as well as intestinal contents were removed and preserved in 10% formalin for histological purposes.

Rodent tissue samples were processed for histological examination. The fixed tissues were sectioned and dehydrated in Isopropyl alcohol and wax impregnated using the automatic tissue processor (Tri-3-matic, model No. 2500, Lipsaw manufacturing Corporation). The tissue samples were embedded in paraffin wax mounted on wooden blocks and sectioned (3-4µm) using a rotary microtome (Baird and Tatlock London). The sections were placed on slides and dried in the oven overnight at 60ºC, stained using Haematoxylin and Eosin (H &E) and Masson's trichrome techniques and then mounted in DPX mountant. The slides were then examined and photographed using a camera mounted on Carl Zeiss microscope.Samples of the faecal droppings left in the traps were also collected. If necessary, additional feces were collected from rectum on postmortem. The samples were preserved in 10% buffered formalin. In the laboratory, faecal samples were subjected to flotation, sedimentation and (Immunofluorescent assay kit for *Cryptosporidium* and *Giardia* (IFA MERIFLUOR test kit, Meridian Diagnostic, Inc., Cincinnati, Oh).

Results

During this study, 33 rodents (60% males and 40% females) were captured which comprised of 7species (Table 1). *Praomys jacksoni* was the most abundant rodent caught in BFR comprising 33.3% of the trapped rodents. Examined rodents grossly and histopathologically, were infected with *Angiostrongylus cantonensis,* tapeworms and Sarcocyst (Table 2). The associated lesions were characterized by granulomatous pneumonia, periarteritis, and the presence of eosinophills, macrophages, plasma cells and degenerating adults, eggs and larvae of *Angiostrongylus* species. .Figure 1 and 2 shows cross sections of the lung tissues showing adults of *Angiostrongylus cantonensis* in the lung artery and lung granuloma containing larvae of *Angiostrongylus* species respectively. The faecal samples of rodents examined in this study identified *Strongyloides* species (6.45%), *Enterobius* species (6.45%) and Oocysts of *Cryptosoporidium* (6.45%) and *Giardia* species (12.9%). Unidentified mites were found infecting the skin of 27.2% (9/33) rodents and most of the infested rodents had associated alopecia

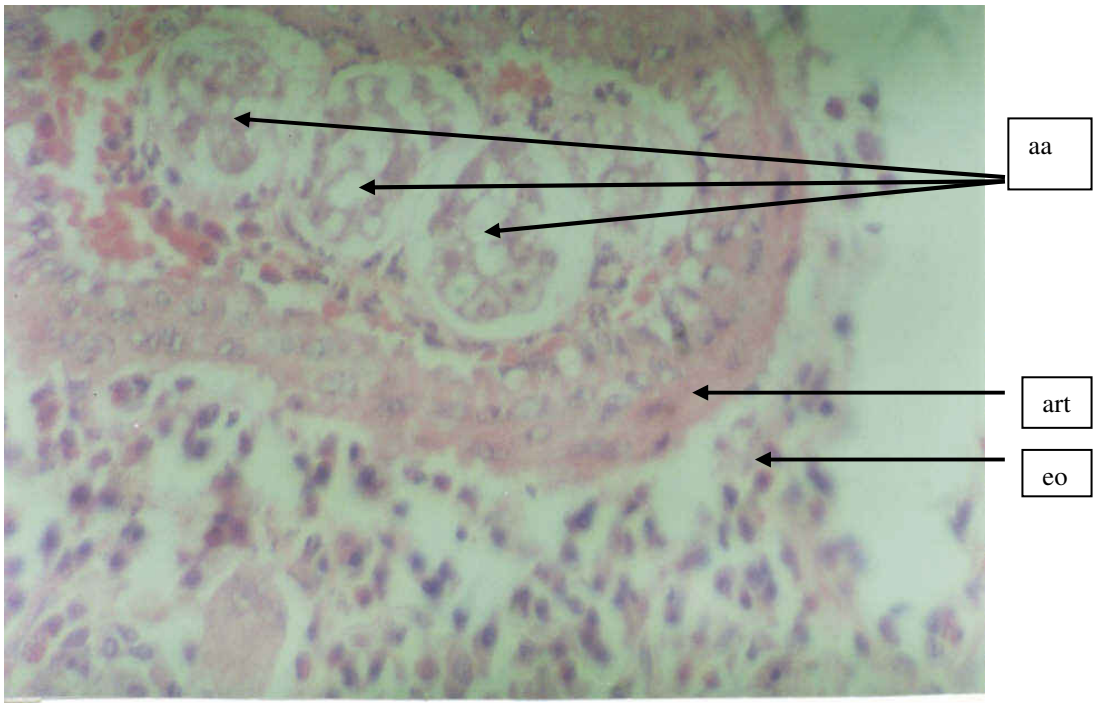
Table 1: Abundance of the rodent species in Budongo forest reserve

Species	Frequency (n)	Percentage
Aethomys hindei	1	3
Rattus rattus	3	9.1
Lophuromys stratus	3	9.1
Lophuromys flavopunctatus	4	12.1
Praomys stella	5	15.2
Lophuromys sikapusi	6	18.2
Praomys stella	5	15.2
Lophuromys sikapusi	6	18.2
Proamys jacksoni	11	33.3

Table 2: Parasitological findings on gross and histopathological examination of rodent tissues

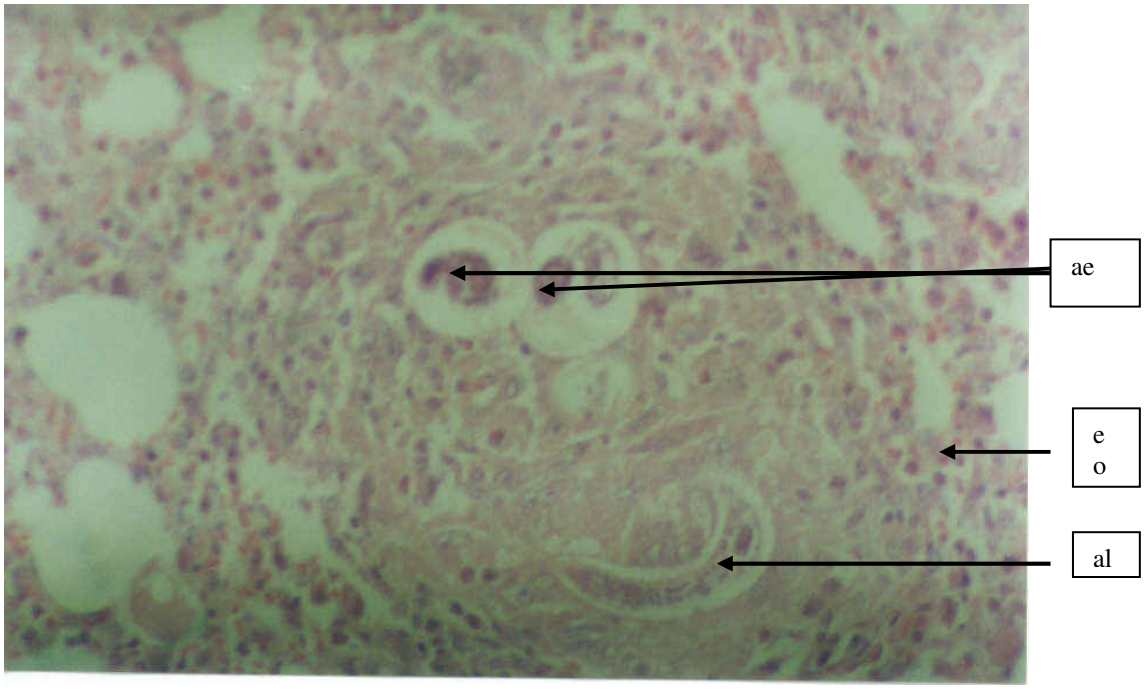
Rodent species	*C. hepatica*	*Angiostrongylus*	Tape worm	Mites	Sarcocyst
L. sikapusi	-	-	+	-	-
L. sikapusi	-	+	-	+	-
L. sikapusi	-	+	-	+	-
L. sikapusi	-	-	+	-	-
L .sikapusi	-	-	-	-	+
A. hindei	-	-	+	+	-
L. flavopanctus	-	-	-	+	-
L. flavopanctus	-	-	-	+	-
P. jacksoni	-	-	-	+	-
P. jacksoni	-	-	+	-	-
P. jacksoni	-	-	-	+	-
P. jacksoni	-	-	-	+	-
L. stratus	-	-	+	+	-
R. rattus	-	-	+	-	-
Total Numbers	0	2	6	9	1

Fig 1: Lung of a rat (*L. sikapusi*) showing cross section of three degenerating adults of *Angiostrongylus cantonensis* in an artery



Legends:
aa : adult *Angiostrongylus* cross-section; art: Arterial cross-section
eo: eosinophils

Fig 2: Lung of a rat showing cross section of granuloma containing larvae of *Angiostrongylus* species



Legends: ae: *Angiostrongylus* sporulated eggs; eo: eosinophils, al: larva of *Angiostrongylus*

Discussion

Angiostrongylus species were found in lung tissue of 6% of all rodent species and 40% of *Lophomurus sikasipusi* examined. Histologically, the lesions were characterized by granulomatous pneumonia, periarteritis, and the presence of eosinophills, macrophages, plasma cells and degenerating adults, eggs and larvae (Figs 1 and 2). The morphological features and histological lesions described here were compared with those in published reports (10) for identification confirmation. Presence of the degenerative gravid adult parasite in the lung tissue was definitive for *A.cantonensis*. Only *Angiostrongylus vasorum* has been reported in Dogs in Kampala, Uganda (11, 12) and the rats were involved in the life cycle only as paratenic hosts containing L_3 larvae. This finding was the first evidence of *Angiostrongylus cantonensis* infection in rodents in Uganda.

Angiostrongylus cantonensis was thought to be a parasite of rodents only until it was also found in the brain of a human teenager in Taiwan (7). Since then, it has been found in humans in Hawai, Tahiti, the Marshall Islands, New Caledonia, Thailand, Vanuatu, the Loyality Islands, Cuba, and even in Louisiana and Western Hemisphere (6,7,8). However, the true geographic distribution of this parasite remains unknown (7). Since 1961 it has been known that human infections are usually acquired by purposeful or accidental ingestion of infective larvae in terrestrial mollusks, planaria and fresh-water crustacean. The African land snail, *Achatina fulica* played an important role in the pan-Pacific dispersal of the organism which may be true for Africa. In Uganda, the intermediate host has not been yet established. Likewise, the human disease in Uganda has not been documented.

The use of mollusks and crustacean as famine foods, favored delicacies and medicines has resulted in numerous outbreaks and isolated infections (1, 2, 9). Economic and political instability, illicit trade, unsanitary peridomestic conditions and lack of health education promote the local occurrence and insidious global expansion of parasitic eosinophilic meningitis (1,2). Further studies should be done determine

the full distribution of the parasite and the species of snails involved in transmission process in Uganda.

Other parasites seen included unidentified tapeworm cysts in the liver, *Hymenolepsis* species in the intestines and sarcocyst in the diaphragm.

Twenty seven point three percent of the studied rodents were infested with unidentified mites with characteristic loss of hair in some cases. The rodents infested with mites were likely to serve as carriers of viral, reckettsial and bacterial diseases infectious to humans and other wildlife species. It was therefore important to identify rodent species, estimate their population, habitats and range in addition to establishing parasites including mites infesting them.

The findings of *Angiostrongylus cantonensis*, *Cryptosporidium* and *Giardia* species and other multi host helminths parasites in the faecal and tissue samples of rodents in Budongo Forest Reserve with the potential for zoonotic transmission presents a public health concern requiring further investigation and expanding the scope to include the non-human endangered primate species especially chimpanzees in that region.

Acknowledgements

I wish to thank Uganda wildlife Authority, Uganda National Council of Science and Technology and Budongo Forest Project for the permission to carry out this research. This study was part of a MSc thesis (Mugisha) which was supported by Compton Foundation Institute, (USA), and DAAD In-country Scholarship Mountain Gorilla Veterinary Project of the Morris Animal Foundation.

References

1. **Kliks, M.M., Palumbo, N.E. 1999.** Eosinophilic meningitis beyond the Pacific Basin: the global `dispersal of a peridomestic zoonosis caused by *Angiostrongylus cantonensis,* the nematode lungworm or rats. *Soc Sci Med* 34:199-212.
2. **New, D., Little, M.D., Cross, J. 1995.** *Angiostrongylus cantonensis* infection from eating raw snails. *N Engl J Med Hyg* **332:**1105-1106.
3. **Aguair, P.H., Morera, P., Pasqual, J. 1981.** First record of *Angiostrongylus cantonensis* in Cuba. *Am J Tropical Med Hyg* **30:**963-965.
4. **Andersen, E., Gubler, D.J., Sorensen, K., Beddard, J., Ash, L.R. 1985.** First report of *Angiostrongylus cantonensis* in Puerto Rico. *Am J Tropical Med Hyg.,* **35:**319-322.
5. **Vargas, M., Gomes Perez, J.D., Malek, E.A. 1992.** First record of *Angiostrongylus cantonensis* in the Dominican Republic. *Trop Med Parasitol.,* **43:**253-255
6. **Lindo, J.F., Waugh, C., Hall, J., Cunningham-Myrie, C., Ashley, D., Eberhard, M.L., Sullivan, J.J., Bishop, H.S., Robinison, D.G., Holtz, T., Robinson, R.D. 2002.** Enzootic *Angiostrongylus cantonensis* in rats and snails after an outbreak of human eosinophilic meningitis, Jamaica. *Emerg Infect Dis*.**8:**324–326.
7. **Roberts, L.S. & J. Janovy, J. 1996.** In: Foundations of Parasitology, 5th ed. Wm.C. Brown, Publishers.
8. **Lindo, J.F., Escoffery, C.T., Reid, B., Codrington, G., Cunningham-Myrie, C., Eberhard, M.L. 2004.** Fatal autochthonous eosinophilic meningitis in a Jamaican child caused by *Angiostrongylus cantonensis*. *Am J Trop Med Hyg*. **70:**425–428.
9. **Cross, J. 1997.** Angiostrongyliasis. In: *Pathology of Infectious Diseases.*(Ed. Connor DH, Chandler FW, Schwartz DA, Manz HJ, Lack EE). Stanford, CT: Appleton & Lange, 1307–1314p.
10. **Ash, L.R. 1970.** Diagnostic morphology of the third-stage larvae of *Angiostrongylus cantonensis*, *Angiostrongylus vasorum, Aelurostrongylus abstrusus* and *Anafilaroides rostratus* (Nematoda: Metastrongyloidea*). J .Parasitol*.**56:**249
11. **Bwangamoi, O. 1972.** *Angiostrongylus vasorum* and other worms in dogs in Uganda. *The Vet Rec* **91:** 267.
12. **Bwangamoi, O. 1973.** Renal lymphoid and pulmonary lesions in naturally acquired canine angiostrongylosis in Uganda. *Bull Epiz Dis Afr*. **22:** 55-68.