Prevalence Of African Giant Snails For Parasites In A South-East Region Of Côte d'Ivoire

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Abstract

Two species of snails (*Achatina achatina* and *Archachatina ventricosa*), from surrounding forest of Azaguié's district, were collected at the market of the locality aforementioned in order to identify their various parasites. For that, 50 snails of each species were sacrificed. Parasites were searched on the level of the flesh, between the flesh and the shell, the stomach, the intestine, the liver and the reproductive apparatus. The collected parasites were *Balantidium spp,* the larvae of *Protostrongylus spp,* the larvae of *Dicrocoelium spp* and *Trichomonas spp*. Our results showed that 52% of *Achatina achatina* and 74% of *Archachatina ventricosa* were parasitized. Among the parasites collected in the snail *Achatina achatina*, 95.8% were nematodes and 4.1% were trematodes. Whereas in the snail *Archachatina ventricosa*, 97.7% were protozoa, 8.8% nematodes and 0.4% consisting with trematodes. The prevalence of *Protostrongylus spp* (48%) were higher than that of *Dicrocoelium spp* (4%) in *Achatina achatina*. For the snail *Archachatina ventricosa*, the prevalence of parasite were dominated by

Trichomonas spp (38%) and *Protostrongylus spp* (24%).

Keywords: African giant snail, parasites, prevalence

Introduction

Some terrestrial molluscs of the genus *Achatina* and *Archachatina* are sources of proteins that are appreciated by population in Africa. The consumed snails come from essentially collecting carried out in the forests during the rainy seasons (Otchoumou *et al.*, 1990). Therefore, it's not surprising that the preferential relative humidity of snail is between 75 and 95% and its optimal growth is 25 ^{o}C (Takeda and Ozaki, 1986). The annual consumption of this animal in Côte d'Ivoire reached 7800 tons in 1990. The consumption in Abidjan was around 1800 tons in 2008 (Kouassi *et al.*, 2008).

However, many terrestrial mollusc species are regarded as intermediate hosts of trematodes (Manga-gonzalez *et al.*, 2010). In addition the trematodes of *Dicrocoelium* genus would have as intermediaries' hosts *Helix aspersa* found in Turkey (Gurellï and Göçmen, 2007). Then, Shan *et al*. (2009) and Hu *et al.* (2011), announced a serious illness and sometimes fatal in mankind such as eosinophilic méningo-encephalopathy and the radiculomyélo-encéphalite whose *Achatina fulica* would be the vector. This snail is intermediate host of the nematode *Angiostrongylus cantonensis*. These studies related to only the species *Helix aspersa* and *Achatina fulica*. *Achatina achatina* and *Archachatina venticosa* not having been studied. However these two species are much consumed in Côte d'Ivoire (kouassi *et al*., 2008; Sika, 2015). So, would the species *Achatina achatina* and *Archachatina venticosa* are a potential reservoir of diseases? In other term do these two snails species are the reservoir of parasites?

In order to answer this question, we proposed this study which aims to identify parasites on two species of giant snail (*Achatina achatina* and *Archachatina ventricosa*) in the area of Azaguié.

Material

The biological material is composed of two snail species: *Achatina achatina, Archachatina ventricosa*.

The observation of the parasites was done with a binocular magnifying glass of mark CETI and a microscope of mark Carl Zeiss.

Methods

Study area

Snails used in this survey were collected in Azaguié, a locality of the forest belt of Côte d'Ivoire. Located at the South-east of the Côte d'Ivoire

between the latitudes 5°35' and 6°15' N and longitudes 3°55' and 4°40W (Figure 1), Azaguié is 40 km away from the north of Abidjan.

The choice of Azaguié is justified by the fact that this locality shelters a classified forest and several former studies showed that this area is snails' purveyor for the town of Abidjan.

Sampling

We did two missions to purchase snails. The first mission has been hold in 5 November 2011 and consisted in collecting randomly 50 snails of the species *Archachatina ventricosa*. The second mission took place in 4 December 2011 and it permitted us to collect also randomly 50 snails of the species *Achatina achatina*. The living and active snails of which shell was not damaged and without lesion on the flesh were taken. The dead snails or snails of which the shell was damaged, were not retained in this study.

The choice of 50 snails was made on the basis of formula suggested by Fosgate (2009):

$n = \log \alpha / [\log (1-p)]$

n = required sample size

p = prevalence expected (found in previous study, failing of previous study a prevalence of 50% was fixed)

α = 1- desired absolute precision (95%)

The collection was made early in the morning in order to avoid contaminations from surrounding traders. The tow species of snail are packaged separately in the bags before display on the market stand. We bought snails when there were packaged in bags before traders exposed them on the market table.

Transport

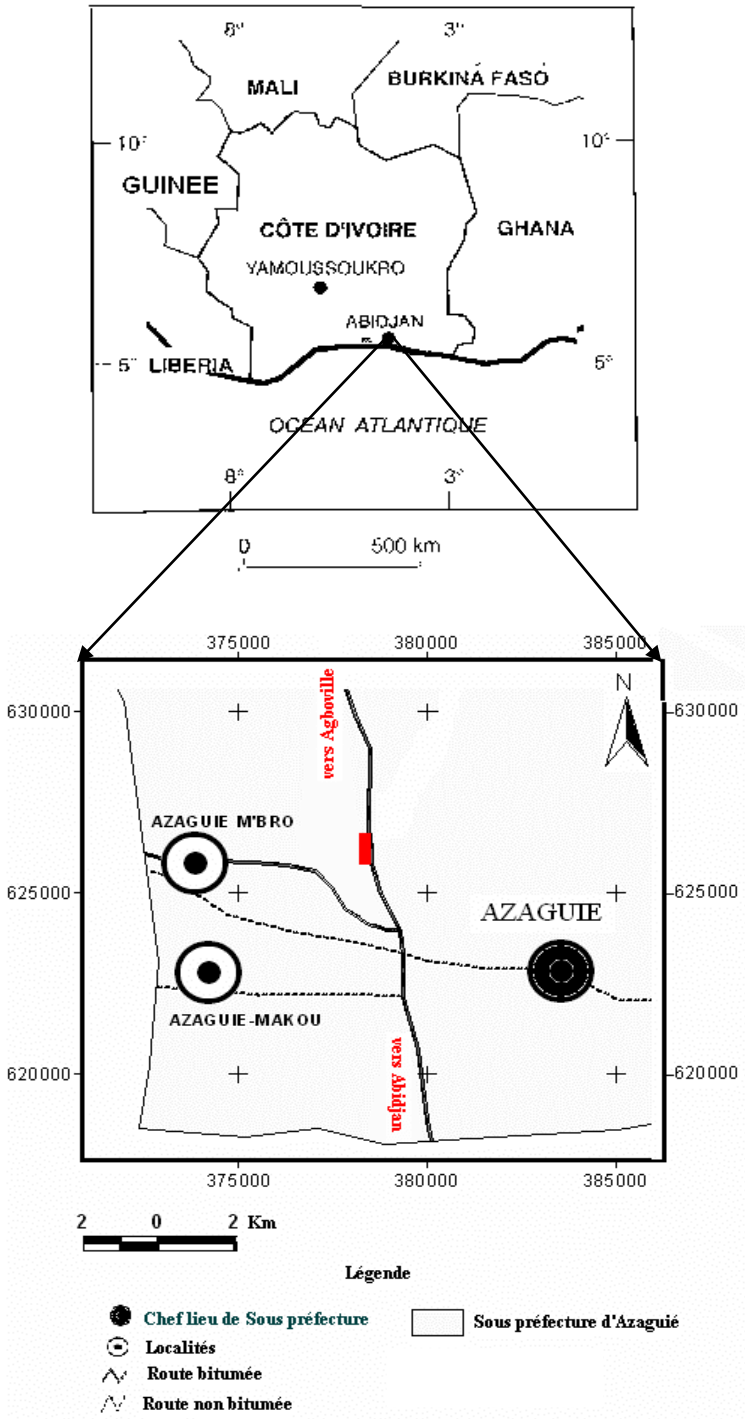
The snails were conditioned in individualized sterile sachets and conveyed to the laboratory in a cooler bag.

Macroscopic examination

Once at the laboratory, the shell and the flesh of snails were thoroughly examined in order to prevent the animals having from lesions and damage on their shell.

Preparation of the sample for parasitic analysis

Collection of the external parasites was done by scraping the pedal plate by using a sterile blade of lancet in a sterile plastic box of Petri containing 40 ml sterile distilled water. The shell of snails were broken with a stone cleaned beforehand (with liquid soap, rinsed with the



nFigure 1. Localization of the zone of study in the Southeast of the Côte d'Ivoire:
A: Map of Côte d'Ivoire, **B**: Situation of Sub-prefecture of Azaguié (Vroh et al., 2010).

tap water followed by an ethanol bath at 70^{o} during 10 minutes). This operation has been implemented for each snail.

The parasites located between the flesh and the shell were collected by rinsing the visceral mass and inside the shell with 40 ml sterile water distilled in a sterile plastic box of Petri.

After this operation, dissection was carried out with a sterilized pair of scissors and sterile pliers of dissection. The stomach, the intestine, the liver and the reproductive apparatus were taken. The organs were put individually in sterile plastic boxes of Petri.

The liver was incised with a sterile blade of lancet. This blade of lancet has been used to take a small quantity of the hepatic liquid. It has been laid on a blade slide. A sterile distilled water drop was added on. The solution was homogenized with the edge of a plate which has been used as cover. This operation was repeated three times at tow extremity and at middle of the same liver.

The other organs were split with a sterilized pair of scissors and the inside is scrapped with a sterile blade of lancets. The contents of stomach and reproductive apparatus were rinsed with 20 ml and 30 ml of sterile distilled water respectively. For the search of parasite eggs, 3g of feces contained in the intestine were collected. We used flotation and sedimentation methods.

Parasites counting

For the search and the quantification of parasite eggs, Mac Master technique were used with saturated solution of chlorure of sodium (density 1.20). In an aim to facilitate the counting of parasites, 5 ml of each solution are taken and observed gradually until exhaust the total volume of each solution. The parasites observed in each time were identified and their manpower noted.

Microscopic examination

All solutions were watched with the binocular magnifying glass. The blades that stand the preparations have been observed in the microscope with magnitude of 100 and 400.

Identification of the parasites

The identification of the parasites has been facilitated by the keys proposed by Thiempon *et al.*, (1979), Troncy *et al.*, (1981) et Basson, (2010).

Prevalence

The prevalence of snails for parasites according to their class and

genus, were calculated with the following formula:

$$P = (ni/N) * 100$$

Where P : Prevalence of snails for parasite (%)
ni : Number of snails infested
N : Total number of snails analysed

Results

Distribution of the parasites according to the snail's organs

The parasites collected in the two snail's species were *Balantidium spp.*, the larvae of *Protostrongylus spp.*, the larvae of *Dicrocoelium spp.*, and *Trichomonas spp*.

No egg were found in feces. Table 1 shows parasites' distribution in the organs. While the other organs were not infected in *Achatina achatina*, three larvae of *Dicrocoelium spp* were found in the liver. *Protostrongylus spp* were present on the flesh (30 larvae), between the flesh and the shell (29 larvae), in the stomach (8 larvae) and in the intestine (3 larvae). The greatest values have been recorded on the flesh (30 larvae) and between the flesh and shell (29 larvae). The collected parasites in the snail *Archachatina ventricosa* were *Balantidium spp.*, *Protostrongylus spp.*, *Dicrocoelium spp.* and *Trichomonas spp*. *Balantidium spp.* were present on the flesh (9 parasites) and to a smaller extent in the reproductive apparatus (2 parasites). *Protostrongylus spp.* were collected in great number between the flesh and the shell (25 larvae) and on the flesh (14 larvae). *Dicrocoelium spp.* were only collected in the liver (2 larvae). *Trichomonas spp.*, except in the liver, has been observed in all the organs: 245 on the flesh, 171 between the flesh and the shell, 7 in the reproductive apparatus; 3 in the intestine, and 2 in the stomach.

Infested and no-infested snails rates

Our works showed that 52% of *Achatina achatina* and 74% of *Archachatina ventricosa* are parasitized (Table 2).

Prevalence level of snail for parasites' class

Among the parasites collected on the snail *Achatina achatina*, 95.8% were infested by the nematodes and 4.1% by trematodes. However none protozoans parasites were found on the snail *Achatina achatina*. Whereas on the other species *Archatina ventricosa*, 97.7% of them were infested by the protozoans, 8.8% by nematodes and 0.4% by trematodes (Table 3).

Table 1: Distribution of the parasites according to the organs of the snails *Achatina achatina* and *Archachatina ventricosa*

Species	*Achatina achatina*						*Archachatina ventricosa*					
Localization	Flesh	Flesh-shell	Stomach	Intestine	Liver	Reproductive Apparatus	Flesh	Flesh-shell	Stomach	Intestine	Liver	Reproductive Apparatus
Parasites												
Balantidium spp.	0	0	0	0	0	0	9	1	0	0	0	2
Protostrongylus spp.	30	29	8	3	0	0	14	25	2	2	0	0
Dicrocœlium spp.	0	0	0	0	3	0	0	0	0	0	2	0
Trichomonas spp.	0	0	0	0	0	0	245	171	2	3	0	7

Snail state	Infested	No infested	Total
Achatina achatina	52.00%	48.00%	100%
Archachatina ventricosa	74.00%	26.00%	100%

Table 2: Infested and not infested snail ratio in *Achatina achatina* and *Archachatina ventricosa*

Table 3: Parasites per class ratio and prevalence of parasitism in *Achatina achatina* and *Archachatina ventricosa*

Snail species	Class of parasites		
	Nematode	Trematode	Protozoan
Achatina achatina	95.80%	4.10%	0%
Archachatina ventricosa	8.80%	0.40%	90.70%

Snail species	Prevalence per genus			
	Protostrongylus spp.	*Dicrocœlium spp.*	*Balantidium spp.*	*Trichomonas spp.*
Achatina achatina	48.00%	4.00%	0%	0%
Archachatina ventricosa	24.00%	4.00%	8.00%	38.00%

Prevalence level of snail for parasites' genus

The prevalence in *Achatina achatina* is 4% for *Dicrocoelium spp*, 48% for *Protostrongylus spp*. On the contrary, zero for *Balantidium spp.* and *Trichomonas spp.* (table 3). In *Archachatina ventricosa*, it's 38% for *Trichomonas spp*, 24% for *Protostrongylus spp.*, 8% for *Balantidium spp.* and 4% for *Dicrocoelium spp*.

Discussion

The parasites collected in the two snail's species were *Balantidium spp*, the larvae of *Protostrongylus spp.*, the larvae of *Dicrocoelium spp*, and *Trichomonas spp*. The presence of these parasite could be justified by the fact that these species of snails would present a favourable environment to their development. The harvest of *Protostrongylus spp.* and *Dicrocoelium spp.* larvae corroborates the results of Dreyfuss and Rondelaud (2011) who observed that the terrestrial molluscs constitute intermediate hosts for most of nematode and trematode species.

The distribution of the parasites according to the organs revealed that *Archachatina ventricosa* was more parasitized than *Achatina achatina* as well in a number of parasites as in species. This species would displays more favorable for the survival of parasites. The larvae of *Protostrongylus spp* were very frequent on the flesh and between the shell and the flesh. *Protostrongylus spp* contaminates snail in its larval form (L1) by penetrating in the flesh of mollusk then leave it at the L3 stage (Dreyfuss and Rondelaud, 2011). *Dicrocoelium spp* was only localized in the liver. This finding seems to confirm the fact that it is a parasite of liver (Ducommun and Pfister, 1991; Gurellï and Göçmen, 2007).

Achatina achatina was infested at 52% and *Archachatina ventricosa* at 74%. Indeed Dreyfuss and Rondelaud (2011) maintain that the majority of the species of parasites would have closely specificity for the mollusc hosts species. Our works showed that *Archachatina ventricosa* is infested at 97.7% by the protozoa, 8.8% by the nematodes and at 0.4% by the trematodes. This preponderance of protozoa would be due to the conditioning. Because a flora and a fauna are associated to snails. This flora and this fauna could become explosive when the environment allows it (Pirame, 2003).

The prevalence of *Dicrocroelium spp.* were 4% for *Achatina achatina* and also for *Archachatina ventricosa*. This prevalence were higher than the result obtained by Gurellï and Göçmen (2007) in *Helix aspersa* (0.97%). However it were lower than the result obtained by Fashuyi·and Adeoye, (1986) in *Limicolaria flammea* (30%), *Limicolaria striatula* (20%) and *Lamellaxis gracilis* (20%). Additionally, we noticed that Fashuyi·and Adeoye (1986) observed nothing in 25 Achatinidae. This weak prevalence would be related to the mollusc species used in our work (*Achatina achatina* and

Archachatina ventricosa).

The prevalence of *Protostrongylus spp.* were 24% and 48% respectively in *Archachatina ventricosa* and *Achatina achatina*. Our results were largely higher than those of Sher *et al.* (2006) obtained in goats (1.05%). According to Dreyfuss and Rondelaud (2011) these parasites require the terrestrial mollusc intervention to ensure their transmission to the herbivorous. Moreover, the high-risk period includes November and December for molluscs (Cabaret *et al.*, 1980). Our samplings have been carried out in November and December 2011.

The prevalence of *Trichomonas spp* (38%) and *Balantidium spp* (8%) in *Archachatina ventricosa* seem to be new for us. Since *Balantidium* is a parasite of monoxene characteristic. It means that its evolution proceeds on the same host or partially in the external environment. Pig, man, dog, wild monkey and rodents are final hosts (Nanfah, 2008). The snails would probably contaminate in their origin environment. Did these parasites contaminate *Archachatina ventricosa*? Is *Archachatina ventricosa* an intermediate host for these parasites? Lastly, are these parasites pathogenic for this mollusc? Many interrogations which deserve further studies.

Conclusion

At the end of this study, we can retain that *Achatina achatina* and *Archachatina ventricosa* were colonized by larvae of nematodes, *Protostrongylus spp*, and trematodes, *Dicrocroelium spp*. The species *Archachatina ventricosa* is also infested by protozoa such as *Trichomonas spp* and *Balantidium spp*. *Archachatina ventricosa* were more infested than *Achatina achatina.* Another strong idea would be a prospective survey over the year in order to analyze evolution and dynamic of infestation and infection rates.

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Statement of conflicting interest

The authors attest that there is no conflict of interest with regard to the authorship and publication of this manuscript.

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