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Larval stages of *Brachylaima fuscatum* in the terrestrial snail *Limicolaria aurora* from southern Nigeria

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Abstract

Of 150 specimens of the gastropod snail *Limicolaria aurora* examined from the Edo and Delta states of Nigeria, 63.4% were infected with larval digeneans comprising mother sporocysts (12.1%) daughter sporocysts (20.4%) cercariae (43.1%) and metacercariae (24.5%). Attempts to experimentally infect three 14-day-old chicks (*Gallus domesticus*) and two laboratory-bred 4-month-old mice (*Mus musculus*) by oral feeding and peritoneal injection with cercariae were negative, although experimental infections of chicks via a cloacal drop yielded 62 immature and 37 mature worms from the intestinal caeca and ileum. The worms were identified as *Brachylaima fuscatum* (Trematoda: Brachylaimidae). The study also revealed that *L. aurora* acts as an intermediate host for *B. fuscatum*, in addition to *Eulota* sp., *Helix* sp., *Helicella* sp., *Oxychilus* sp. and *Agrolimax* sp.

Introduction

Limicolaria aurora is a small land snail, which commonly occurs in the high and fringing forests of the derived savannah regions of West Africa. These snails are delicacies and form an important source of animal protein where they occur. *Limicolaria* has been found to act as an intermediate host to the digenean, *Dicrocoelium hospes* in some parts of Africa (Odei, 1966; Bourgat *et al.*, 1975; Lucius & Frank, 1978; Lucius *et al.*, 1981). Adult *D. hospes* is known to infect man (King, 1971) and livestock (Kajubiri & Hohorst, 1977; Malek, 1980). Apart from *D. hospes* larvae, *Limicolaria* has not been shown to harbour any other digenean larvae and no previous studies have been undertaken in this regard in Nigeria.

The present paper reports on the occurrence of the larval stages of the digenean *Brachylaima fuscatum* from naturally infected *L. aurora* and attempts to experimentally infect avian and mammalian hosts with *B. fuscatum*.

Materials and methods

A total of 150 specimens of *Limicolaria aurora* (fig. 1) were collected by hand from bushes, fields and gardens in various parts of Edo and Delta States in southern Nigeria and maintained in the laboratory as described by Olsen (1974). Snails were dissected to isolate the head, foot, digestive gland, heart, ovotestis, hermaphrodite duct, stomach, oesophagus, crop, intestine and rectum. Each organ was teased open under a microscope in a Petri-dish containing normal saline (0.9% NaCl solution) and any parasites recovered were counted. Parasite identification was carried out at the former International Institute of Parasitology, St Albans, United Kingdom.

Six 4-month-old laboratory bred mice (*Mus musculus*) and eight 14-day-old chicks (*Gallus domesticus*) were used for experimental infections. Sixty metacercariae were fed orally by pipette to each of three mice while each of two mice were injected peritoneally with 60 metacercariae. The sixth mouse was used as a control. These methods have been used by Herman & Bacha (1978). All experimental mice were maintained separately and nourished on processed flour feed and clean water. In addition, 60 metacercariae were administered orally by pipette to each of three chicks. Three other chicks were anaesthetized with Nembutal (pentobarbitone sodium) by injection through the thigh muscles ($0.03\,mg\,g^{-1}$ of body weight) and were immediately administered each with 80 metacercariae using the cloacal drop infection method (Herman & Bacha, 1978). Two chicks were used as controls. All experimental chicks were maintained in separate cages and nourished daily on processed broiled feed and clean water. One mouse and one chick from those exposed to infection were each examined on days 7, 16 and 29 post-infection.

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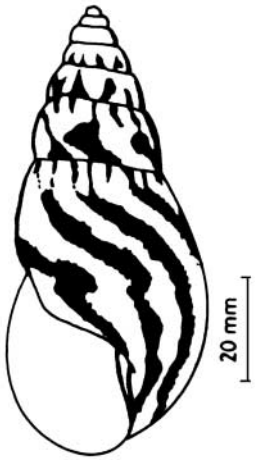

Fig. 1. The land snail *Limicolaria aurora*.

Results

Ninety-five (63.4%) of one hundred and fifty *L. aurora* examined were infected with larval stages of digenetic trematodes. A total of 7265 larvae were isolated from infected snails and comprised 877 (12.1%) mother sporocysts, 1480 (20.4%) daughter sporocysts, 3130 (43.1%) cercariae and 1778 (24.5%) metacercariae, (table 1). The highest infection (29.7%) was recorded in the hepatopancreas, with the lowest (1.1%) in the hermaphrodite duct. Daughter sporocysts were highly branched and tubular and cercariae were of the microcercous type. The larvae (figs 2 and 3) are identified as those of *B. fuscatum* as described by Sumenkova (1902).

In experimental infections, no adult worms or larvae were recovered from mice infected orally and peritoneally nor from chicks infected orally. Each of three chicks infected via the cloaca yielded two juvenile worms, 60 immature worms and 37 sexually mature worms in the intestinal caeca on days 7, 16 and 29 post-infection, respectively (fig. 4). Features of the trematode stages recovered from experimentally infected chicks are listed in table 2. Characteristics of the adult digenean *B. fuscatum* include an elongated tongue-shaped body, with the acetabulum located near the midbody rather than towards the anterior extremity. Caeca are straight or somewhat sinuous with testes arranged in tandem at the posterior third of the body. Vitellaria commence at the level of the acetabulum and extend to the anterior testis.

Table 1. The prevalence, (%) total worm burden and distribution of brachylaimid larval stages in 150 specimens of *Limicolaria aurora*.

	Primary (mother) sporocyst	Daughter sporocyst	Cercaria	Metacercaria	Total	Prevalence (%)
Hepatopancreas	125	638	981	413	2157	29.69
Ovotestis region	579	480	360	244	1663	22.89
Other parts of digestive tract	81	267	646	234	1228	16.90
Heart	63	–	774	642	1479	20.36
Hermaphrodite duct	25	32	20	5	82	1.13
Head	4	18	139	85	246	3.39
Stomach	–	30	171	121	322	4.43
Foot	–	15	39	34	88	1.21
Total	877	1480	3130	1778	7265	
Prevalence (%)	12.0	20.37	43.08	24.48		

Table 2. Characteristics and measurements of juvenile, immature and adult worms of *Brachylaima fuscatum* from experimental infections in chicks.

	Measurements of worms (μm)		
Characteristics	7-day-old juvenile	16-day-old immature adult	29-day-old sexually matured adult
Body length	990	3420	4140
Body width	450	690	1770
Oral sucker	190 × 220	510 × 510	720 × 810
Ventral sucker	150 × 150	300 × 300	510 × 510
Pharynx	120 × 150	240 × 240	300 × 300
Distance between the two suckers	150	630	650
Right intestinal caeca	760	2760	3840
Left intestinal caeca	740	2760	3820
Upper testis	–	–	310 × 360
Lower testis	–	–	300 × 350
Ovary	–	–	190 × 290

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The ovary is located between the testes, and the uterus extends to the intestinal bifurcation.

Discussion

A 63.3% prevalence value recorded in *L. aurora* in the present study is high when compared with 3.2% and 37.1% values for larval infections of *D. hospes* in the same snail species from the Ivory Coast and other areas (Lucius *et al.*, 1981). The larval characteristics presented are clearly those of trematodes in the family Brachylaimidae and adult worms retrieved from experimentally infected chicks were identified as *B. fuscatum* from characteristics described by Yamaguti (1971).

Brachylaima fuscatum has been reported from several countries to occur in the caeca of various definitive bird hosts, e.g. *Tetrao coturnix, Corvus* sp., *Francolinus* sp., *Strix* sp., *Columba* sp., *Gallus* sp., *Passer* sp., *Coturnix* sp. and *Turdus* sp. (Dollfus, 1954; Yamaguti, 1971). Sporocysts, cercariae and metacercariae have been reported in some terrestrial snails, e.g. *Eulota duplicinita* (Sumenkova, 1902) *Helix pisana*, *Helix aspersa*, *Helicella scitnla*, *Oxychilus cellarius* and *Agrolimax agrestis* (Yamaguti, 1971) and more recently in the snail, *Phyllocaulis variegates* (Thiengo & Amato, 1995). Apart from birds, *Brachylaima* sp. infection has been reported in rodents. Recently, Cribb (1990) reported a *Brachylaima* sp. infecting three species of Australian rodents (*Mus domesticus, Rattus fuscipes* and

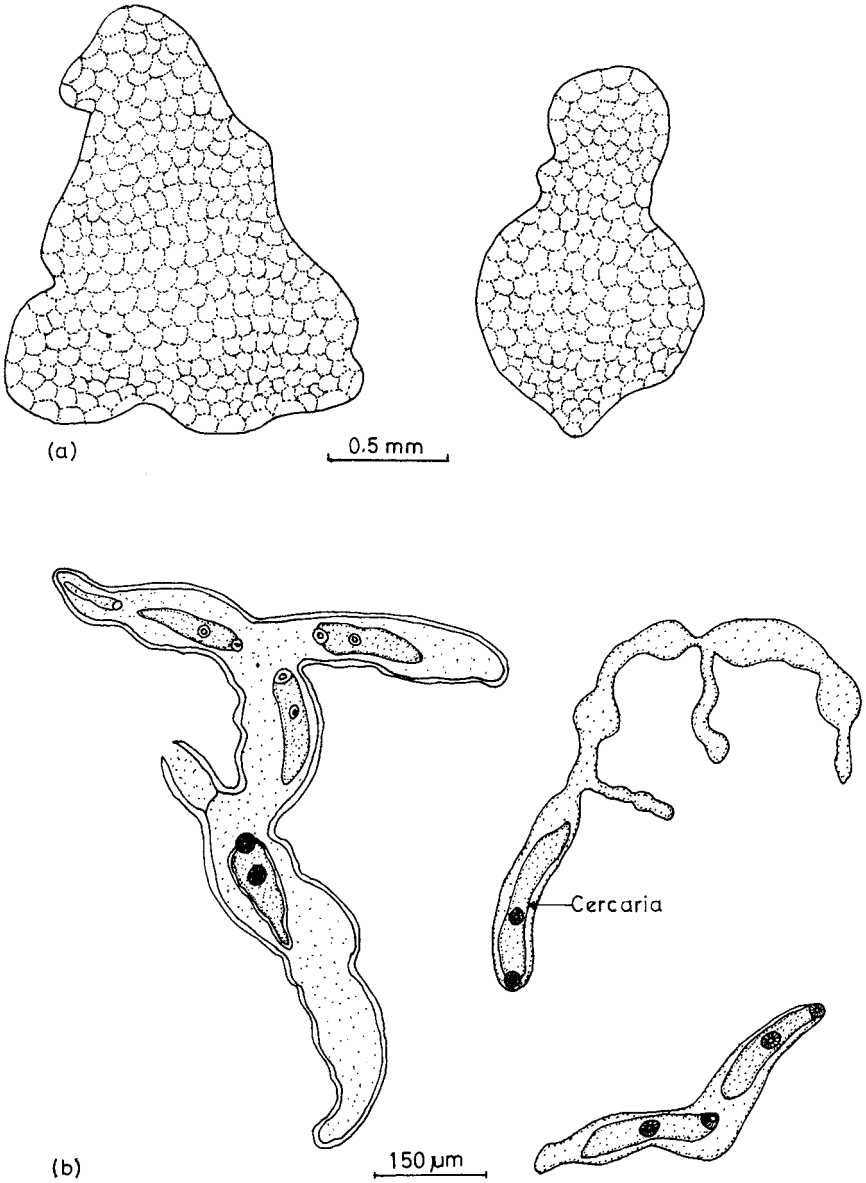

Fig. 2. *Brachylaima fuscatum* primary (mother) sporocysts (a) and branched secondary (daughter) sporocysts containing cercariae (b) from naturally infected *Limicolaria aurora*.

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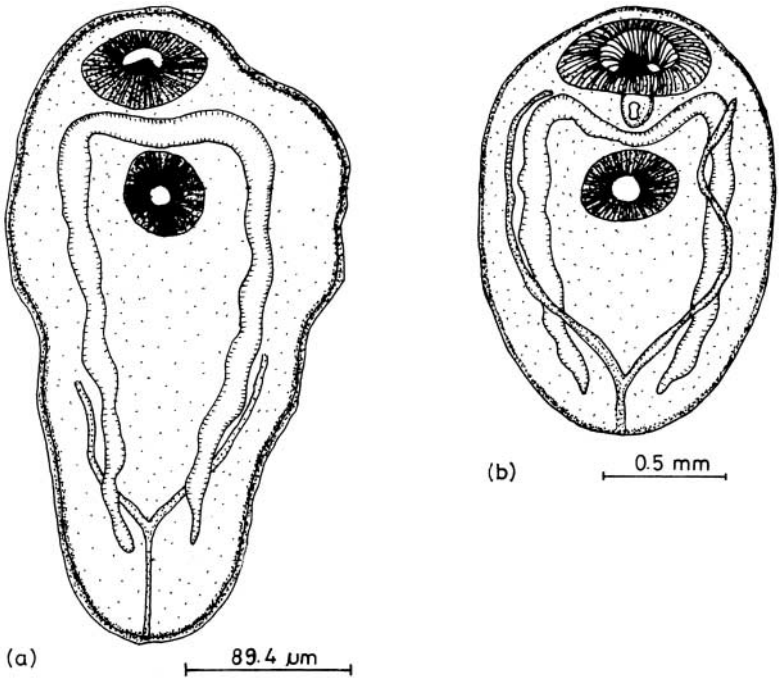

Fig. 3. Cercaria (a) and metacercaria (b) of *Brachylaima fuscatum* from naturally infected *Limicolaria aurora*.

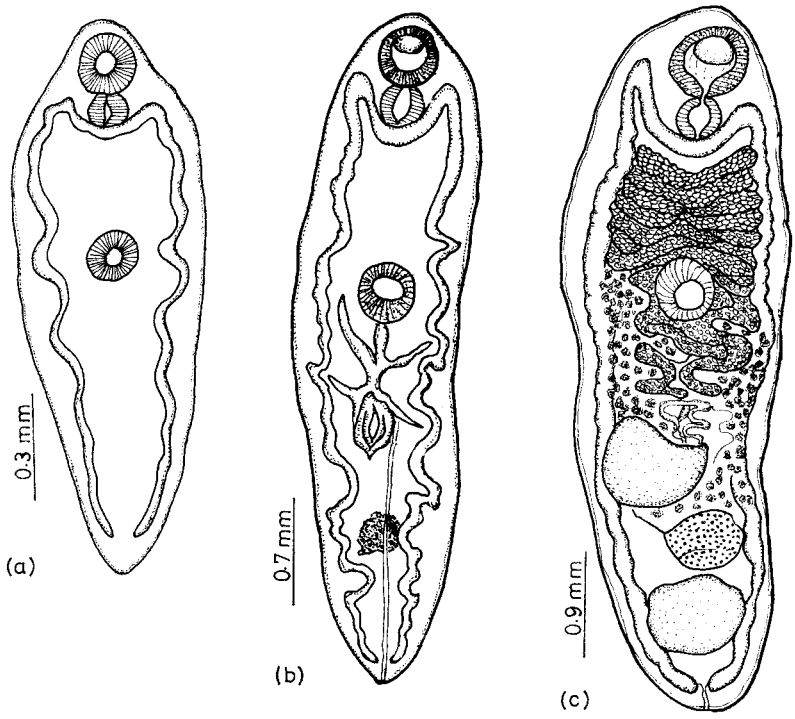

Fig. 4. Immature *Brachylaima fuscatum* recovered from (a) 7-day-old and (b) 16-day-old infections and (c) mature worm from 29-day-old infection in the chick.

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Leporillus conditor) and utilizing the snails *Cernuella virgata*, *Cochlicella barbara* and *Theba pisana* as intermediate hosts.

Soboleva *et al.* (1995) reported that some microelements of calcium, magnesium, copper, zinc, lead, iron, etc. which are assimilated into the body of snails from the soil and plants on which snails feed, influence populations of trematode infections in snails. They reported that trematode infections with sporocysts in the liver and metacercariae in the kidney of the snails require high levels of these microelements while non-infected snails show corresponding low levels. In the present work, it is possible that high levels of microelements in the surrounding environment might explain the prevalence value of 63.3% of *B. fuscatum* in *L. aurora*.

In the West African sub-region, few studies have been undertaken on the Brachylaimidae except on *Postharmostomum notowi* from the domestic fowl in Ghana by Hodasi (1967). The present finding is the first report of the occurrence of *B. fuscatum* in *L. aurora* in Nigeria, and thus this would form the basis of further experimental studies on the biology of this brachylaimid digenean.

Acknowledgements

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