Laboratory Breeding of *Lymnaea natalensis* (Krauss, 1848), Intermediate Host of *Fasciola gigantica* (Cobbold, 1856)

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Abstract: *Lymnaea natalensis* the intermediate host of the *Fasciola gigantica* (the causative agent of liver fluke disease) was collected from the zoological and botanical gardens, University of Ibadan, Nigeria and reared in our laboratory. The aim was to improve and standardize a rearing and maintenance technique for this snail. Two kinds of diets were compared: Blanched, Dried lettuce (A) and blanched, dried lettuce + 10% Calcium carbonate ($CaCO_3$) (B). The age at oviposition, growth rate using shell size as indices and the durations of incubation and hatching were determined. Age at beginning of ovipostion ranged from 39 to 60 days. The snails fed with $CaCO_3$ enhanced diet presented an increased growth rate, however, the difference was not significantly different ($p>0.05$). The maximum size attained by snails fed with diet A was 18mm length and 8mm height and 23mm length and 14mm height for diet B. The maximum duration of incubation of the eggs for the diets are 12 days (B) and 11 days (A) while the duration of hatching for diet B and A are minimum of 2 days and 6 days respectively. The method of mass breeding and maintenance of *Lymnaea natalensis* using $CaCO_3$ as supplement to blanched dried lettuce was found to be suitable in our laboratory.
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Running title: Rearing of *Lymnaea natalensis*

1. Introduction

Snails are a group of animals which have a diverse habitat ranging from freshwater, marine and terrestrial environments (Brown, 1994). Pulmonate freshwater snails have been studied and shown to be intermediate hosts of some parasitic diseases which are of veterinary and public health importance (Ndifon and Ukoli, 1989). *Lymnaea* snails are a group of freshwater snails distributed worldwide (Godan, 1983)). In Nigeria, the most important genus is *Lymnaea natalensis*, which is the intermediate host of fascioliasis, a disease which has been observed to be of public and veterinary health importance. This organism is widely distributed geographically in Nigeria (Ukoli and Asamu, 1979; Thomas and Tait, 1984).

In a malacology survey of 11 local governments of Ibadan, *Lymnaea natalensis* was found in 6 without restriction of spread (Adedokun, 2005). Specimens collected from various locations were found to be susceptible to natural and experimental infections with *Fasciola gigantica* (Guobadia, et al, 1996).The main type of feeding regime used for the snails brought into our laboratory has been dried blanched lettuce without calcium carbonate ($CaCo_3$).

The study was initiated to compare two feeding regimes for these snails, collecting data on growth, oviposition, and productivity with the objective of improving and optimizing the breeding methods.

2. Materials and methods

Collection and identification of samples

Snails used in this study were collected from breeding colonies in streams running through the Zoological and Botanical gardens of the University of Ibadan, southwest Nigeria. After identification, the snails were divided in two groups of 20 and placed in two 60 x 30 x 30 cm glass aquaria filled with dechlorinated water and the aquaria were exposed to natural light supply in the laboratory. Eggs were collected daily on transparent plastic sheets placed into the aquaria.

Experiments

Aquaria A and B were set up in ten replicate each containing 3 newly hatched snails. The snails in A group were fed with only blanched dried lettuce which has been the staple for rearing *Lymnaea natalensis* in our laboratory (400mg every 72hours) while B group were fed with the blanched dried lettuce + 10% $CaCO_3$,(360mg lettuce + 40mg ground chicken egg shell). Snail development was evaluated by measurement of shell length and height. Egg masses and egg were collected and counted daily. Longevity was observed in each group.

Statistical Analysis

Average lengths of the snails were compared using Student t test ($p \leq 0.05$).

3. Results

The age at first oviposition ranged from 39 days for diet B to 60 days for diet A. The number of egg per egg mass in aquaria A and B ranged from 0 to 73 for A and 0 to 75 for B. Snail growth, incubation and hatching rate and average number of egg masses in both experiment are shown in table 1 and 2 respectively. The snails in aquaria B developed faster than those in aquaria A although the difference was not statistically significant ($p<0.05$). Water turbidity and colour was visibly higher in B than A although this was not measured.

Table 1: Age and Growth Rate of Snails

Group	No of snails	Age (days	Average shell length(mm	Average shell height(mm
A	30	14	4.1	2.2
B	30	14	4.8	2.5
A	30	28	8.5	4.3
B	30	28	8.9	4.8
A	30	42	10.8	4.8
B	30	42	11.5	5.7
A	30	56	11	5.8
B	30	56	11.7	6.83
A	25	70	12.2	7.9
B	30	70	14	8.0

Table 2: Age and Productivity Rate of Snails

Group	Age (days)	Average No Egg mass / snail	Average No of Eggs / Snail	Average Duration of Incubation (days)	Average Duration of Hatching (days)
B	39	1	67	10	5
B	40	2	45	12	5
B	41	2	45	9	4
B	43	2	57	10	5
B	45	1	42	9	2
B	46	2	52	8	3
A	60	3	50	11	6

4. Discussion

Several techniques have been developed by various authors for the laboratory rearing of the lymnaea snails (Madsen and Monrad, 1981 Sachez et al, 1998, Souza de Perieira and Magalhaes, 2000).The techniques, maintenance of the aquaria and diets however vary widely. The need to improve on our rearing methods which led to the search for an effective, cheap and more productive rearing method for our laboratory was achieved to a great extent.

The methodology in the present study used to rear *Lymnaea natalensis* was similar to that used to rear *Lymnaea columnella* by Souza de Pereira and Magalhaes, 2000 and satisfactory results were obtained.

The number of egg masses laid per snail was at a maximum of 3 egg masses which is in contrast to Islam et al, 2001 who said that freshwater snails generally have a high fecundity. This low fecundity could however be due to a change of environment (Callow, 1970 and Skoog, 1978) since they were offspring of snails brought in from the wild. There is a significant relationship between age at point of lay, number of egg masses per snail, duration of hatching and incubation. However, in contrast with the reports of Garaerts and Muhammed(1981) there is a lack of significance between the relationships of age at point of lay, height and length of shell.

There is also an indication that both indices of growth (height and length of snail shell) increased at same rate and according to the works of Russell-Hunter (1961 a and b) the onset of reproduction is determined by genetic and environmental factors. Both indices of growth (height and length of snail shell) increased simultaneously which is as reported by Madsen, 1982. The enhancing effect of calcium carbonate on snail size and productivity rate as seen in this study corroborates the works of Madsen, 1982 who reported that calcium enhances snail egg production.

This study shows that *Lymnaea natalensis* gets to point of lay between 39 days and 60 days and the older the snail, the more egg masses they produce. Also, dietary supplementation with calcium carbonate from egg shell (95%, $CaCo_3$) has a positive influence on growth and snail productivity rate.

However our research is continued for a detailed understanding of *Lymnaea natalensis* to ensure a comprehensive understanding of the factors that surround and influence the biology of this snail with the possibility of obtaining an environmentally friendly means of control.

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