

LAMELLA FORMATION AND EMIGRATION FROM THE WATER BY A LABORATORY COLONY OF *BIOMPHALARIA GLABRATA* (SAY) IN A FLOW-THROUGH SYSTEM

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Lamella formation and emigration from the water were investigated in juvenile Biomphalaria glabrata reared at two temperatures in aquaria with a constant water flow. Most snails (97.4%) reared at the lower temperature (21 °C) formed lamella at the shell aperture and emigrated from the water, whereas only 10.1% did so at 25 °C. Eighty percent of emigrations at 21 °C occurred within a period of 15 days, 70-85 days after hatching. A comparison of the studies done so far indicates that the phenomenon may be affected by the ageing of snail colonies kept in the laboratory and their geographic origin, rather than the rearing conditions. This hypothesis, however, requires experimental confirmation.

Key words: *Biomphalaria glabrata* – juvenile snails – emigration – lamella formation – effect of temperature

The knowledge of what factors play a role on lamella formation and emigration from the water by juvenile *Biomphalaria glabrata* snails is still scarce. An environmental manipulation with a proven effect on inducing this phenomenon is the rearing of snails at a temperature lower than usual (Pieri, 1985). However, the effectiveness of low temperature on triggering the phenomenon may vary a great deal, depending on factors not yet understood (Dannemann & Pieri, 1991).

Lamella formation followed by emigration from the water has been reported mainly from studies in closed systems (Paraense, 1957; Richards, 1963, 1967, 1968; Pieri & Thomas, 1986; Dannemann & Pieri, 1991), where the snails are particularly subjected to detrimental factors, such as depletion of resources and accumulation of toxic substances. No laboratory investigation has yet been done on the effects of temperature in open, flow-through systems, where such adverse factors are better prevented. Thus, the present study aims to evaluate the effect of two temperature conditions on la-

mella formation and emigration from the water in a flow-through system.

MATERIALS AND METHODS

Two 75 l aquaria were used in the experiment. On May 24, 1990 they were filled at the bottom with a 2 cm thick substrate layer consisting of 82% mud, 4% calcium carbonate, and 14% ground oyster shells. Filtered tap water was introduced in the aquaria by means of a drip-feed working continuously at a rate of 5 ml min⁻¹. In order to avoid water overflow, the aquaria were provided with an outlet. Both aquaria remained in a room with light intensity varying from 22 to 80 lux at daytime to less than 1 lux at night. Room temperature was kept permanently at 21 ± 1 °C by air conditioning. One of the aquaria was equipped with a water heater regulated by a thermostat at 25 ± 1 °C. For the sake of simplicity the aquaria will hereinafter be designated according to their recorded temperatures as 25 °C and 21 °C.

The test snails were obtained through cross-fertilization of snails which formed lamella and emigrated from the water at the juvenile stage in August, 1989. These parental snails belonged to the third generation of a laboratory colony of *B. glabrata* founded by snails collected from Pontezinha (34° 58'W, 8° 31'S), northeast Brazil, in April 1989 (PON-2 stock). Two

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batches of newly-hatched snails were introduced in each aquarium according to the following timetable: 110 snails on June 27 and 200 on July 5, 1990. From the first introduction of snails onwards, food was provided at two-day intervals as small amounts of oven-dried (40 °C), crushed, lettuce leaves, which were evenly scattered on the water surface of the aquaria. Care was taken that some lettuce always remained uneaten.

Emigration of snails from the water was recorded periodically along the experiment. The experiment ended when the resident snails had become adults (about 10 mm shell diameter). Snails were inspected for the presence of lamella as described by Pieri & Thomas (1986), and their maximum shell diameter was measured using a caliper. Results were statistically evaluated through Chi-square tests with a correction for continuity (Siegel, 1956).

RESULTS

Lamella formation followed by emigration from the water was observed in 97.4% of the snails in the 21 °C aquarium, as they reached the juvenile stage, whereas it occurred only in 10.1% of the snails in the 25 °C aquarium (Table I). All the emigrants in both aquaria were lamellate. Lamella formation was also observed among resident snails in both aquaria, with no significant difference between the two groups ($\chi^2 = 2.61$; $P > 0.05$).

The figure depicts the emigration of snails from the water in a temporal pattern, as well as the shell diameter of the emigrants. It reveals a massive concentration of emigrants from the 21 °C aquarium in a short span of time, as 80% of them were collected within a period of 15 days, 70-85 days after hatching. The shell diameter of emigrating snails at 21 °C was within the 3-5 mm range, averaging 3.9 ± 0.61 mm (mean and standard deviation). The seven lamellates which emigrated from the water at 25 °C did so 30-57 days after hatching, as they were 4.5 ± 0.61 mm of shell diameter.

The water temperature of the aquaria remained rather constant throughout the experiment: 24.7 ± 0.4 °C (mean and standard deviation) in the one provided with the water heater, and 20.9 ± 0.3 °C in the other aquarium. Maximum and minimum room air temperatures were 22.0 ± 0.7 °C and 19.9 ± 0.4 °C, respectively.

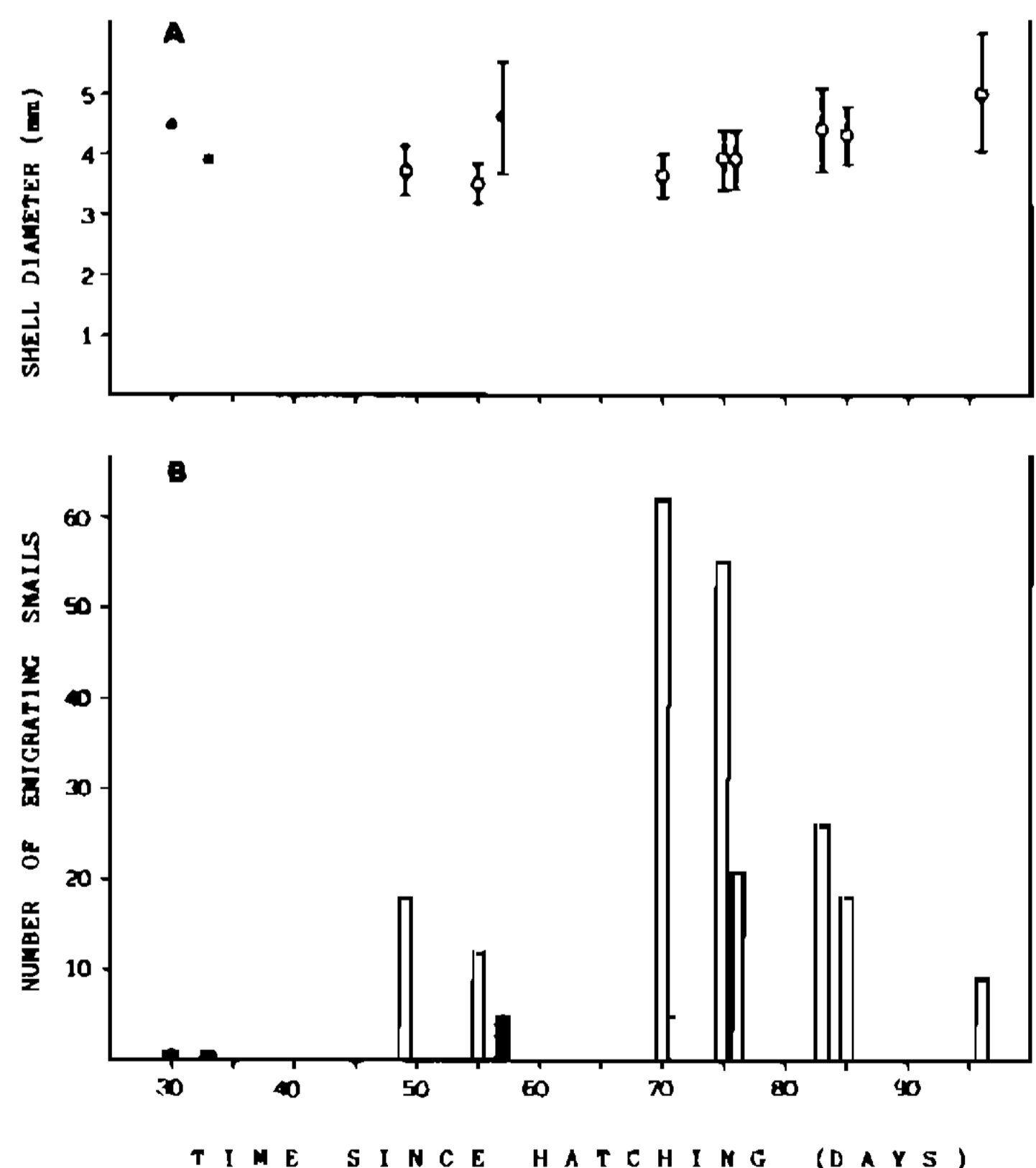
TABLE I

Number of juvenile *Biomphalaria glabrata* snails forming lamella at the shell aperture (A) and emigrating from the water (B).

A – Lamella formation				
Categories	Temperature conditions			
	21 °C	25 °C	Total	
Lamellates	224	16	240	$\chi^2 = 191.7$
Non-lamellates	3	53	56	
Total	227	69	296	$P < 0.001$

B – Emigration from the water				
Categories	Temperature conditions			
	21 °C	25 °C	Total	
Emigrants	221	7	228	$\chi^2 = 222.5$
Residents	6	62	68	
Total	227	69	296	$P < 0.001$

Egg-masses were obtained from a laboratory colony originated from Pontezinha, northeast Brazil. The test snails were reared since hatching in a flow-through system either at 25 °C or 21 °C. Chi-square values (χ^2) and corresponding probabilities (P) are also given.



Temporal pattern of emigration from the water by juvenile *Biomphalaria glabrata* snails reared since hatching in a flow-through system either at 25 °C (black circles and bars) or 21 °C (empty circles and bars). Egg-masses were obtained from a laboratory colony originated from Pontezinha, northeast Brazil. A: mean shell diameter at the time of emigration (vertical lines indicate standard deviations). B: frequency of emigration from the water.

TABLE II

Number of juvenile *Biomphalaria glabrata* snails emigrating from the water under different rearing conditions either at low (18-21 °C) or standard (24-25 °C) temperatures. Both laboratory colonies originated from Pontezinha, northeast Brazil, founded by snails collected in April 1982 (PON-1) and April 1989 (PON-2)

Conditions	Snail stock	PON-1		PON-2		PON-1		PON-2	
	Water system	Closed		Open		Closed		Open	
Temperature (°C)		18	20	21	24	25	25		
Authors		(a)	(b)	(c)	(a)	(b)	(c)		
Categories	Emigrants	37 (92.5)	51 (41.5)	221 (97.4)	12 (46.2)	7 (4.7)	7 (10.2)		
	Residents	3 (7.5)	72 (58.5)	6 (2.6)	14 (53.8)	141 (95.3)	62 (89.8)		
Total		40 (100.0)	123 (100.0)	227 (100.0)	26 (100.0)	148 (100.0)	69 (100.0)		

(a) Pieri (1985); (b) Dannemann & Pieri (1991); (c) present study. Percentages are given in parentheses.

A total of 284 snail deaths occurred in the 25 °C aquarium, 84.9% of the total being among hatchlings (< 3 mm shell diameter). Some deaths in that aquarium occurred among juveniles (12.3%) and adults (2.8%), probably due to a direct contact with the water heater. The survival of the snails in the 21 °C aquarium was 73.2%.

DISCUSSION

It is clear from the present results that lamella formation followed by emigration from the water in a flow-through system is significantly facilitated by rearing the snails at a lower temperature. This further confirms previous findings by Pieri (1985) and Dannemann & Pieri (1991) obtained in closed systems with a snail stock originated from the same locality in North-east Brazil.

Table II presents the data obtained by the above authors, as compared with those from the present study. It can be seen that in all three studies the proportion of emigrants was significantly greater at low (18-21 °C) than at standard (24-25 °C) laboratory temperatures. However, a comparative analysis of the three studies strongly suggests that at least one experimental condition may have substantially affected the outcome of the phenomenon. This will be briefly considered below.

The two studies with the PON-1 stock were carried out at different times since the founder snails were collected from the field. Pieri (1985) started his experiment in January 1984,

20 months after the colony was established in the laboratory. In contrast, Dannemann & Pieri (1991) made their experiment when the colony was 4 years 5 months old. As laboratory colonies of snails are known to lose some of their original features after many succeeding generations, this may in part explain why the proportion of emigrants was substantially greater in the former study than in the latter. It is interesting to note that the present study, which was carried out only 14 months after the PON-2 stock was established, also produced over 90% of emigrants at the low temperature condition (Table II).

A recent study by Barbosa (1989) also supports the hypothesis that newly established laboratory colonies of *B. glabrata* are more prone to form lamella and emigrate from the water than old ones. The snails which formed the parental generation had come directly from the field and were allowed to breed in a flow-through system at 22 °C. The data from that study enable one to estimate that 38.7% of the juvenile snails formed lamella and emigrated from the water one month after the colony was founded. Twelve months later, however, the phenomenon ceased to occur. Further experiments are required to confirm this hypothesis.

The study by Barbosa (1989) has some methodological similarities with the present one. Firstly, both studies used flow-through systems; secondly, the low-temperature condition in the present study was only 1 °C below the temperature used by that author; thirdly, the snails employed in both studies originated

from recently established laboratory colonies. However, a comparison of the results of the two studies shows that at low temperature the proportion of snails forming lamella and emigrating from the water, as well as the time elapsed until emigration, were substantially greater in the stock from Pontezinha than in that from Paulista. As these localities are situated in areas with different geological and vegetation characteristics (Pieri, 1985) it is possible that the expression of this phenomenon varies among different snail strains. This hypothesis could be evaluated by testing snail stocks originated from different regions.

The studies above clearly indicate that the phenomenon can occur at great proportions both in closed and open systems, being not significantly affected by depletion of resources or accumulation of toxic factors, nor by environmental shocks due to handling at the times of medium renewal. Moreover, it can be induced whether the hatchlings are fed on live *Oscillatoria* cyanobacteria or lettuce (either fresh or dried). It may occur both in small, standing water and large, continuously renewing water systems.

As stressed by Dannemann & Pieri (1989), the occurrence of this phenomenon in *B. glabrata* is of particular relevance for snail control in endemic areas of schistosomiasis. Further studies should focus on the elucidation of the mechanisms responsible for its causation.

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