# STUDY OF A POPULATION OF *BIOMPHALARIA TENAGOPHILA* (ORBIGNY, 1835) AND OF SCHISTOSOMIASIS TRANSMISSION IN "ALTO DA BOA VISTA", RIO DE JANEIRO

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The present study was performed using data from a Biomphalaria tenagophila population located in a water cress garden in the Alto da Boa Vista region representing an isolated focal point of schistosomiasis in the city of Rio de Janeiro.

The density and age structure of this B. tenagophila population and its rate of infection by Schistosoma mansoni were studied for a period of 15 months.

The snail population showed seasonal variation in density, with a decrease in number of individuals at the beginning of the rainy season. At the end of this season, the population consisted mainly of adults (92.8% in May 1985 and 82.8% in April 1986). The population growth curve was logistic and of sigmoidal configuration.

Schistosoma mansoni cercariae were eliminated over a short period of time (March, April and May 1986). The release of cercariae of S. mansoni and of birds seems to depend on environmental temperature, which during certain months would show a daily variation of up to 13 °C, with the lower thermal limit approaching the limit value for sporocyte development.

Key words: Biomphalaria tenagophila - Schistosoma mansoni - age structure - ecology - water cress garden

In the states of Rio de Janeiro and São Paulo the transmission of schistosomiasis is partly associated with the practice of agriculture, which uses fresh-water springs for the irrigation and drainage systems. This is done for the water cress, rice and sugar cane cultures, which are excellent breeding sites for the snails acting as intermediate hosts of S. mansoni. In general, water cress gardens represent important focal points os schistosomiasis transmission (Pinto, 1945; Coutinho, 1950; Antunes, 1953; Andrade et al., 1955; Rey, 1958; Ruiz, 1957) and this is what occurs in the Alto da Boa Vista region, Rio de Janeiro, which has been found to represent an isolated focus of schistosomiasis (Coura et al., 1970; Paes et al., 1970). The continued presence of schistosomiasis in this region has been recently confirmed by Schall et al. (1985) and Silva et al. (1986).

Even though populational aspects have been extensively investigated in field studies on

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Biomphalaria glabrata no such studies are available on B. tenagophila.

The presente study was conducted to follow the populational fluctuation of *B. tenago*phila and the frequency of infection by *S. man*soni and by bird cercariae in the Alto da Boa Vista region. The possible causes that affect the distribution, density and age structure of this snail inside a water cress garden were analyzed.

## MATERIALS AND METHODS

Site and time of study — The study was carried out in a water cress in the Alto da Boa Vista region, RJ. The garden consists of 30 patches arranged in decreasing plateaus and interconnected by irrigation canals.

Three areas (patches) were selected for the study: (i) site of entry of water from the irrigation system (area 1 - A1), (ii) an intermediate point (area 2 - A2), (iii) site where the water leaves the system (area 3 - A3) R teng-

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Water samples for physicochemical analysis and snails were collected from A1, A2 and A3 twice a month from March 1985 to May 1986 for a total of 15 months of study.

Collection method — A 5  $m^2$  square frame subdivided into 80 portions, 40 of wich were picked at random using a table of aleatory numbers, was used to collect snails. The 40 portions corresponded to 2.5  $m^2$ , i. e. 5% of the total area of each plateau (50  $m^2$ ). In each of these 40 portions, all snails found on the surface, the sediment or on the water cress leaves were collected with tweezers. Since A1 and A3 were found to be free from B. tenagophila, snail collection was limited A2.

Quantitative evaluation of the animals and infection test — The snails were carried to the laboratory on wet filter paper placed on moistened petri dishes; they were them counted and measured for the populational study. To detect the presence of snails infected with cercariae of S. mansoni or of birds, the animals were placed 30 cm from a light source (an incandescent 100 W lamp) in glass containers with 20 ml of water (3 to 5 animals per container) for 2 to 3 hours. Infected snail were sacrificed.

Water collection and analysis — Water was collected in 1 liter plastic containers for physicochemical analysis, while water for pH and dissolved oxygen (DO) determination was collected in containers with specific preservers.

Control of variables — After examination in the laboratory, 90% of the snails were returned to the field within approximately 4 days. Meteorological data were obtained from the Alto da Boa Vista Station of the National Meteorology Institute.

#### RESULTS

Physicochemical analysis of the water — In view of the fact that A2 differed from A1 and A3 by presenting B. tenagophila, A1, A2 and A3 were compared in terms of physical (temperature) and chemical (pH, conductivity, chloride, total hardness in CaCO<sub>3</sub>, total Ca hardness in CaCO<sub>3</sub>, DO, Mg and Ca concentrations) water parameters. The results were

differences were detected among areas in water pH, conductivity and chloride content.

Climatic factors — The garden in question, which is located at an altitude of approximately 300 meters in the Alto da Boa Vista region, is not subject to periods of drought (Fig. 1). The highest rainfall indices were recorded between November and February, wich was the time when the snail population suffered a reduction and the frequency of adults increased.

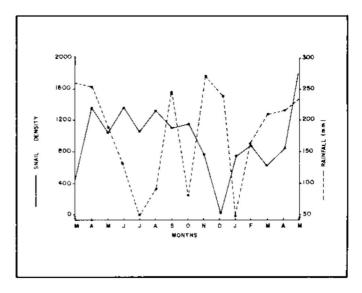


Fig. 1: month variation of snail density (number of snail per 2.5 m<sup>2</sup> of sampled area) and rainfall (total in mm) from March 85 to May 86 in the Alto da Boa Vista region, RJ.

Temperature is characterized by wide annual variations, with lows of 18 °C and highs of 35.6 °C recorded for maximum temperature, and highs of 24 °C and lows of 10.8 °C recorded for minimum temperature. Daily variations up to 13 °C were observed during the colder months (June and July).

Even under the artificial environmental conditions created for a vegetable, in which the impact of the strong rains was reduced by the partial deviation of water flow out of the patches and constant flooding was present, we detected a sharp inverse relationship between density and rainfall (Fig. 2).

Indices of infected snails — Monthly recording of the frequency of snails infected with S. mansoni showed that the release of S. mansoni cercariae approximately occurred in March, April and May. The release of bird carcariae occurred in all months except lune.

TABLE I	
Analysis of the physicochemical parameters of water from a watercress garden in the Alto da Boa Vist	region

Parameters			Ar	ea			Varia anal		,	A	В		
	A1 A		A	2	A	3	(F)	<b>(P</b> )	\r.		) (r		
	x	s	$\bar{\mathbf{x}}$	s	X	s	( <b>F</b> )	(P)	Min.	Max.	Min.	Max.	
pH	6.63	0.42	6.36	0.40	6.47	0.24	3.73	0.027	5.6	9.1	4.8	9.0	
Conductivity													
(micronhos cm <sup>-1</sup> )	96.6	25.8	121.8	26.0	117.0	26.0	5.84	0.005	117.0	2000.0		6000.0	
Chloride (mg/l <sup>-1</sup> )	12.50	1.54	15.20	3.94	14.80	3.46	6.60	0.003	1.0	3500.0	$\bar{\mathbf{x}} = 86$	2562.0	
Dissolved oxygen													
$(mg/l^{-1})$	7.10	0.89	5.74	1.41	5.38	1.15	15.90	0.000	_	_	_	_	
Ca conc. $(mg/l^{-1})$	5.90	2.37	5.80	2.28	5.32	1.38	0.48	0.623	2.4	129.0	$\bar{\mathbf{x}} = 1.9$	-	
Mg conc. $(mg/l^{-1})$	2.05	1.22	2.17	1.24	2.20	1.78	0.07	0.929	_		$\bar{\mathbf{x}} = 1.0$	_	
Ca total hardness													
in CaCO <sub>3</sub>	14.90	6.00	13.50	2.82	13.30	3.64	0.957	0.623	2.8	730.0	_	_	
Total hardness													
of CaCO <sub>3</sub>	23.20	8.50	22.90	9.30	20.70	7.03	0.59	0.559	14.0	980.0	_	_	
Temp. water OC	22.00	2.90	23.00	3.91	23.00	3.91	0.66	0.522	18.0	41.0	_	_	

A - Value from other places (B. glabrata) referred by Pieri (1985).

B - Value from other places (B. tenagophila).

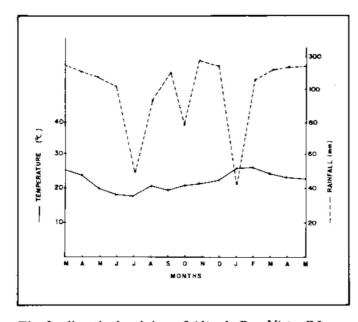


Fig. 2: climatic dominion of Alto da Boa Vista, RJ.

Population structure — Considering that the mean size of B. tenagophila at first spawning is about 7.5 mm (Rey, 1956), individuals were defined as young when they measured up to 8 mm and as adults when they exceeded this diameter. Occasionally, however, the individuals were divided into classes only for a clearer analysis of age structure. Monthly samples correspond to the sum of two byweekly samples.

As the study progressed, there was a sharp change in population age structure. In March 1985, mean population diameter was 11.19 ± 1.88 mm and the composition consisted mainly

TABLE II

Month distribution (%) of infected in relation to the total of snail colected

Month	Total	otal Inf. snail S. mansoni		Inf. snail bird cerc.	%	
March	448	1	0.22	_	-	
April	1172	6	0.51	2	0.17	
May	1046	4	0.38	1	0.10	
June	1382	_	_	_	_	
July	1066	_	_	-	_	
August	1331	_	-	1—	_	
September	1111	_	_	_	_	
October	1174	_	_	1	0.09	
November	762	-	_	_	_	
December	52	_	_	1	1.92	
January	725	_	_	3	0.41	
February	832	_	_	9	1.08	
March	617	-	_	2	0.32	
April	829	2	0.24	18	2.17	
May	1805	_	_	39	2.16	

Table III presents the frequency distribution by shell diameter, clearly indicating that the population tended to grow from April on, continuing to be high until October.

In April 1985, changes started in age structure owing to the appearance of young individual whose percentages increased from 7.14 to 32.2% in relation to the previous month. This change was more marked in May, with young individuals making up 80.1% of the population, and continued over the subsequent months,

	TAB	LE III		
Month sna	il frequence	by shell	diameter	(mm)

Month			·					Shell	diam	eter	(mm)						
	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	Total
March	1	2	3	1	6	19	31	67	132	89	62	18	12	2	3	_	448
April	16	26	45	77	71	143	196	246	142	90	64	41	12	2	_	1	1172
May	18	61	104	109	161	167	188	113	59	26	19	6	6	_	_	_	1046
June	23	69	156	245	331	204	144	105	59	24	14	6	1	_	1	_	1382
July	3	25	61	139	262	226	140	109	58	27	14	2	_	_	_	_	1066
August	34	40	82	209	342	331	162	87	25	10	8	1	-	-	_		1331
September	14	32	55	149	247	252	190	113	39	14	5	1	_	_	_	_	1111
October	25	42	144	220	286	224	105	85	35	6	2	-	_	_		-	1174
November	_		5	49	107	185	159	124	98	20	13	6	2	2	1	_	762
December	_	_		5	7	17	9	5	2	4	2	1	_	-	_	_	52
January	_	6	31	29	64	118	112	135	117	65	37	9	9	2	_	_	725
February	1	15	89	57	90	152	100	83	75	64	91	50	13	2	_	1-	832
March	_	4	21	24	42	105	131	105	83	33	33	24	8	2	1	1	617
April	_	1	6	15	27	93	124	106	165	102	115	48	20	5	1	1	829
May	5	36	106	109	89	158	224	221	303	154	219	122	36	8	2	_	1805

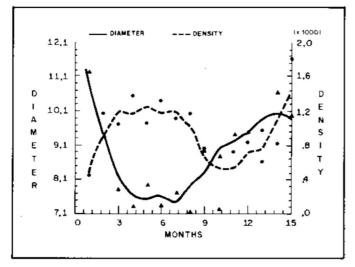


Fig. 3: adjusted curve of *Biomphalaria tenagophila's* populational density and shell medium diameter.

Even though many young individuals entered the population (within the diameter limit considered here) between June and October, the concentration of individuals measuring 7 to 8 mm (being 45 to 50% of the population the frequency of adults) remained constant, except for September when it increased and the frequency of young individuals decreased (Table III).

November was marked by a change in this general picture, with young individuals decreasing from 80.1% to 44.2% from October to November, and adults increasing from 19.9% to 55.8%. Adults then continued to increase in the population, reaching 82.9% in April 1986.

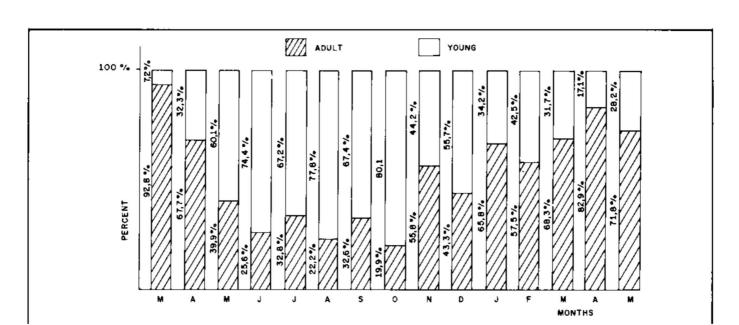


Figure 5, which shows the monthly variation in mean shell diameter of *B. tenagophila*, illustrates an uncommon situation in which there was practically no overlapping of snail diameter during the months in which mean diameter stayed above 10 mm (March 1985 and April and May 1986). This is clearly demonstrated by the broken line in the graph.

When the adjusted curve for monthly variation in mean snail diameter is superimposed on adjusted curve for population density, an inverse relationship between population density and mean diameter is clearly observed. This population growth curve is logistic and of sigmoidal configuration (Fig. 3).

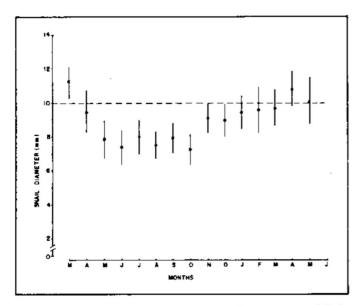


Fig. 5: month variation of *Biomphalaria tenagophila's* medium diameter colected from March 85 to May 86 in Alto da Boa Vista region, RJ.

#### DISCUSSION

Physicochemical analysis of the water — Even though the general habitat conditions inside the water cress garden under study were apparently homogeneous, differences in density were observed between patches, with a complete absence of snails in some of them. An attempt was made to determine the causes of this phenomenon and the results of comparative analyses of the physicochemical parameters of the water for A1, A2 and A3 are discussed.

pH values were statistically different between areas; however, pH values determined for B. glabrata at breeding sites for both snail species have show a variation from 5.8 to 9.1 (Pieri 1985) indicating that this difference is

Significant difference in chlorides were observed among the three areas. The highest value being detected in A2 (15.2 mg/l). However, the variations observed (from 12.0 mg/l in A1 to 14.8 mg/l in A3) do not represent critical values for the species, which is found at sites with concentrations as high as 2.562 mg/l or low as 10.0 mg/l. In contrast, the extreme values for *B. glabrata* are 1.0 to 3,500 mg/l.

Even though significant differences in DO values were detected among areas, the mean values for area 2 (5.74 mg/l) were very close to chose for area 3 (5.38 mg/l) and were within the range detected for this species at other locations. In addition, when mean DO concentrations were transformed to percent saturation with oxygen dissolved in water, the value exceed 120% saturation in all three areas, indicating that oxygen availability is not a biologically limiting factor for the snails at any of the three areas.

The mean values obtained for the other parameters (conductivity, Ca and Mg concentration, total Ca hardness in CaCO<sub>3</sub> and CaCO<sub>3</sub> hardness) did not differ statistically and were comparable to those observed in *B. tenagophila* breeding sites at other locations described by Grisolia & Freitas (1985) and Andrade et al. (1955).

The influence of each parameter on snail populations has been extensively discussed in the literature (Sioli, 1953; Chernin & Michelson, 1957; Schutt & Frank, 1964; Grisolia & Freitas, 1985; Appleton, 1978; Harry et al., 1957).

Frequency of infected snails — Examination of human feces for the determination of the rate of infection by S. mansoni may often produce false negative results. Thus, the ideal procedure is to use a serologic test for comparison though keeping in mind that the latter test may give false positive results or indicate positivity in subjects who used to have the disease and were treated. Silva et al. (1986), by examinating the feces, detected 5.5% infection by S. mansoni among individuals aged 13 to 59 years in the Alto da Boa Vista region, while the serum test gave a result of 51% infection. These authors discussed the possibility of infection by parasites of a single sex.

Paraense & Santos (1953) have commented that during times when the infection rate among planorbids is low, the mollusks tend to emit cercariae of a single sex, a fact that may explain in part the high infection rates detected by the serum test used by Silva et al. (1986), since the rate of snail infection was found to be low throughout most the year. Furthermore, cercariae of a single sex have been identified in the reproduction of the cycle in mice starting from samples collected in the present study (Correia L., personal communication).

Many investigators have demonstrated that the thermal conditions of shallow waters usually reflect the ambient temperature of the air (see Burky, 1969; Eversole, 1974; and McMahon, 1975, 1983, among others).

The correlation coefficient obtained, when mean monthly air temperature at Alto da Boa Vista was correlated with mean monthly water temperature, was r=0.91 (p=0.005). Thus, when the mean maximum and minimum air temperature recorded in June and July are 23.4  $^{\rm O}$ C and 14.04  $^{\rm O}$ C, respectively, and the daily range of variation is as much as 13  $^{\rm O}$ C, corresponding daily fluctuations in water temperature would be expected to occur.

Thus, the thermal limits recorded for this region are close to the limit at which schistosomes develop in the snails. As reported by Pflüger (1981), this limit is about 10 °C. Also, myracidia have no motility at temperatures below 10 °C (De Witt, 1965). Furthermore, Upathan (1973) clearly demonstrated that the maturation time of *S. mansoni* increases and its sporocyst numbers decrease in *B. glabrata* maintained at suboptimal temperatures. This evidence may explain the occurrence of periods of time in which the collected sample did not show elimination of *S. mansoni* cercariae.

In addition, it should be pointed out that S. mansoni cercariae were detected during the times when the collected snail samples contained large numbers of adults, such as March 85 (92.8% adults) and April 86 (82.9% adults).

Population structure — There are five variables that affect the fitness of an organism with a Mendelian inheritance system and they refer to the juvenile and adult phases in the population; age at first reproduction (the lower

that can be produced over the same time interval), survival up to this time (which represents progeny viability, i. e., only individuals that reach maturity will be able to transmit their genome), fertility (number of fertile eggs generated per progeny), interval between reproductive seasons (the shorter the interval, the larger the number of generations), survival during this period (increased number of progenies per individual). Thus, we assume that only two classes of individuals exist in the population (juveniles and adults) since the variables that affect fitness differ between through not within classes.

Analysis of the frequency table (Table III) shows that the population behaves according to same basic principles of population ecology, such as: 1) an expanding population contains a larger proportion of young individuals, a situation indicated by the gradual change in the proportion of young individuals with an increase in density from April 1985 and from May 1986; 2) a stationary population maintains the proportion of distribution of young and adult individuals as well as the proportion of density, a principle that was demonstrated between April and September 1985; 3) the predominance of adults with a decline in density in November agrees with the prediction that in a declining population there is a greater proportion of adults.

Despite the massive addition of young individuals to the population from June to October, which maintained a high concentration of individuals measuring up to 8 mm (80%), there was no proportional increase in the frequency of animals of larger size, but this frequency remained constant and at much lower diameters than the value given above.

Three hypotheses could be raised to explain this phenomenon, which may have been caused by factors acting together or separately. Two of them are supported by indirect evidence: (i) a division of microhabitat may be occurring between young and adults, whereby the young, upon reaching maturity, would change their ecophysiological requirements and migrate towards deeper strata in the sediment. McMahon (1983) stated that planorbids are the only basommatophores which burrow routinely. With respect to *Biomphalaria*, Paraense et al. (1955) observed that *B. glabrata* is able to bur-

nutrition and protection. Thomas & Assefa (1979) and Thomas & Sterry (1985) demonstrated the existence of marked qualitative and quantitative differences in food niche between young and adults of B. glabrata, as well as on their behavior, such as the response to different amino acids. This hypothesis could not be tested here, since the specimens were collected on the surface; (ii) differential growth rates related to maturation and temperature. Rey (1959) reported for B. glabrata that after the first few weeks following hatching, the growth rhythm is slow and is followed by a rapid increase up to the onset of sexual maturity, decreasing again thereafter and becoming slow and regular. In addition to this biological characteristic, growth rate has been shown to be directly related to temperature (Sturrock, 1966; Sturrock & Sturrock, 1972). For example, Sturrock & Sturrock (1972), by experimentally manipulating B. glabrata at different temperatures, demonstrated a decrease in growth rate with decreasing temperatures. Consequently, in the Alto da Boa Vista region, where the mean monthly temperature is about 19 °C during this period, individuals measuring 7 and 8 mm will tend to concentrate, since up to this size the growth curve is marked, declining thereafter. In support of this hypothesis, we noted a slight increase in adult frequency and a reduction in young frequency in September, suggesting that this elevation was due to an increase in mean temperature during the previous month (August), since temperature increase from 19 °C in May, June and July to 21 °C in August. This produced an accelerated growth rate from one month to the other, culminating in the elevation of adult frequency and decrease in young frequency observed in September. This is in agreement with the prediction of Cole (1954) about the populational consequences of the phenomenon of life cycle history, whereby each age class must grow at exactly the same rate as all other classes. The third hypothesis (iii) is an empirical one suggesting a constant mortality rate for adults, with a proportional entry of young to the adult phase.

#### RESUMO

Estudo de uma população de *Biomphalaria* tenagophila (Orbigny, 1835) e da transmissão da esquistossomose no Alto da Boa Vista - RJ — O presente trabalho foi realizado com dados

tenagophila em uma horta de agrião na região do Alto da Boa Vista, foco isolado de esquistossomose na cidade do Rio de Janeiro.

Foram feitas observações sobre mudanças sazonais na densidade e estrutura etária da população de *B. tenagophila* e taxas de infecção desses caramujos pelo *Schistosoma mansoni* durante um período de 15 meses.

A população de caramujo mostrou variação sazonal com diminuição do número de animais no início da estação chuvosa, sendo constituída no final desta estação principalmente de adultos (92,8% maio 85 — 82,9% abril 86). A população apresentou uma curva de crescimento logística de configuração sigmoide.

A eliminação de cercárias de S. mansoni ocorreu durante curto tempo (março, abril e maio de 85 e abril de 86).

A liberação de cercárias de *S. mansoni* e de ave parece depender da temperatura ambiental que chegou a uma amplitude diária de até 13 °C.

Palavras-chave: Biomphalaria tenagophila — estrutura etária — ecologia — horta de agrião

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DA I D. COUDA I C. VALACUE A &

D. F. Baptista et al.

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