

PRODUCTION OF *SCHISTOSOMA MANSONI* CERCARIAE BY *BIOMPHALARIA GLABRATA* FROM A FOCUS IN BELO HORIZONTE, MINAS GERAIS

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SUMMARY

The snail density, levels of infection and the monthly production of *Schistosoma mansoni* cercariae by *Biomphalaria glabrata* were determined in a focus of Barreiro de Baixo (Belo Horizonte, MG, Brazil). During a period of 38 months (1984 to 1987) 5,366 snails were collected of which 324 (6.03%) were infected with *S. mansoni*. The total number of cercariae shed was 5,667,312. Each snail shed an average of 17,422 cercariae during the time that it was under study in the laboratory. The greatest longevity of infected snails was 218 days. Natural cure was observed in 42 (12.9%) of the infected specimens about 130 days after collection. The average snail density in the focus during the period of study was 16.3 snails per scoop.

The shedding of cercariae by snails collected from the field was compared with laboratory bred specimens infected in mass with the LE strain of *S. mansoni* from Belo Horizonte. The laboratory infected snails shed an average of 6,061 cercariae each, a value 2.8 times less than the field specimens due to a shorter life span.

The prevalence of schistosomiasis in the focus was 14.3%.

KEYWORDS: *Schistosoma mansoni*; *Biomphalaria glabrata*; Focus; Cercarial production.

INTRODUCTION

Following the study of the first cases of schistosomiasis in Belo Horizonte by TEIXEIRA²³, when a prevalence of 0.5% was found, several workers have investigated transmission foci of this disease in the region^{2, 3, 7, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20}. With the exception of the "Julien Rien" Park⁷ all the foci are situated on the periphery of the city or in neighboring municipalities. The mean levels of infection of the snails in these foci in the last ten years are not normally greater than 10%^{2, 20, 22}. The highest densities of *Biomphalaria glabrata* are found in horticultural irrigation systems as noted by some years ago by PAULINI et al¹⁸ and which is still true today. The repopulation of irrigation ditches after molluscicide treatment is common depending principally on the type of substrate in the habitat²².

In the present work a focus in Belo Horizonte was studied which is situated in a horticultural small holding in Barreiro de Baixo. The capture and examination of snails was undertaken during 38 months. The objectives were to establish the density of snails, the levels of infection and the production of cercariae per snail. The study was undertaken in the laboratory due to the difficulties involved in following the shedding of cercariae in the field.

MATERIALS AND METHODS

Specimens of *Biomphalaria glabrata* were captured in a system of irrigation ditches and pools measuring about 425 m² in a horticultural small holding situated in Barreiro de Baixo, an area of about 10,000 m² which we have used for a number of studies and

experiments with snails. The existence of this breeding site was registered more than 20 years ago by PAULINI & DIAS¹⁹. Since then the focus has been treated with molluscicide on several occasions but repopulation has always occurred. The source of the water in the ditches is a nearby spring into which sewers from neighboring buildings empty directly. The small holder and members of his family live in the area beside the small holding which they have cultivated for some years.

Snail capture was undertaken on a monthly basis using the scoop method and the density was calculated on the basis of the average number of mollusks recovered in 10 scoop¹⁸. Following collection, the snails were taken to the laboratory where they were counted, measured and examined individually using a stereomicroscope after being exposed to light for 30 minutes. The specimens shedding cercariae were separated and maintained in plastic aquaria with four liters of circulated water and fed on fresh lettuce and rat food pellets supplemented with 10% calcium carbonate. The snails were exposed to light twice a week for four hours in an incubator at $30 \pm 1^\circ\text{C}$ in order to induce cercarial shedding. The cercariae were counted in two aliquots of one ml after being fixed in Lugol. The cercariae eliminated in the aquaria were also counted three times a week in four aliquots of one ml taken from the aquaria before changing the water. Dead specimens were counted and removed from the aquaria on the days when the water was changed. In parallel, 68,818 snails were exposed in mass in the laboratory to 50 LE strain miracidia each during the time period of the survey²¹. The specimens shedding cercariae were separated and exposed to light

twice a week in order to obtain larvae as described for those snails collected in the field.

In 1993 we undertook stool examination of 14 individuals who work in the small holding and treated those infected with *S. mansoni*.

RESULTS

During the 38 months of the study 5,366 specimens of *B. glabrata* were collected measuring 2-29 mm in diameter amongst which 324 (6.03%) measuring between 7 at 29 mm were infected with *S. mansoni*. The snail density in the focus, the natural levels of infection and the mean number of cercariae shed per snail collected from the field or infected in the laboratory are shown in Figures 1 and 2. The total number of snails captured per year, the mean annual densities, the number of snails infected, the levels of infection, the mean and standard deviations and the total numbers of cercariae shed are shown in the Table 1. Each snail from the field eliminated an average of 17,422 cercariae during the period that it was maintained in the laboratory. The annual means of cercariae per snail from the field varied from 558 to 1,889 (see Table). The lowest annual mean (558 cercariae) was recorded in 1986 (Fig. 2). The maximum life span of the naturally infected snails was 218 days. Amongst the 324 naturally infected specimens 42 (12.9%) stopped shedding cercariae approximately four months after capture. The monthly levels of natural infection varied between 0 and 48.7% and were greatest in the first six months of the year (Fig. 1).

In the laboratory, 54,020 (88.6%) of the snails exposed were infected with *S. mansoni*. The total

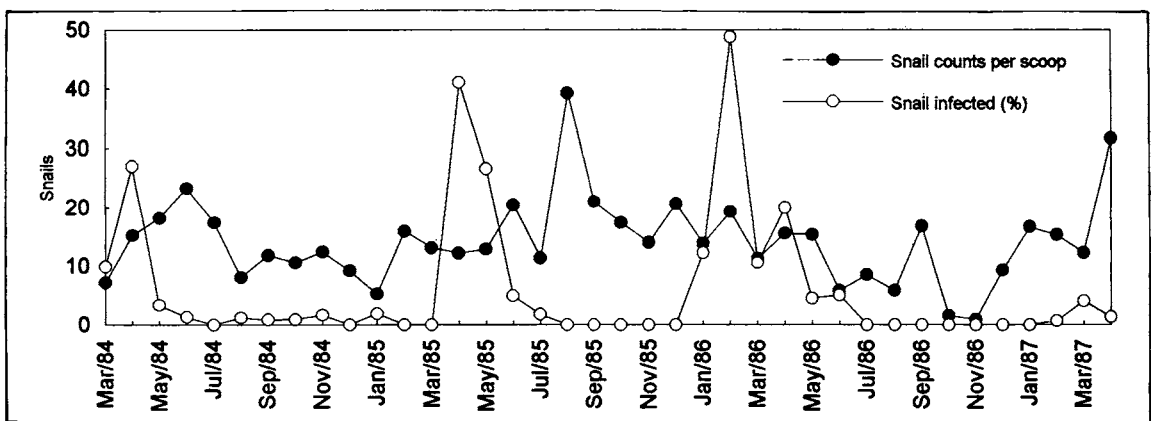


Fig. 1 - Monthly density of *Biomphalaria glabrata* and levels of infection with *Schistosoma mansoni* in a focus in Belo Horizonte, MG.

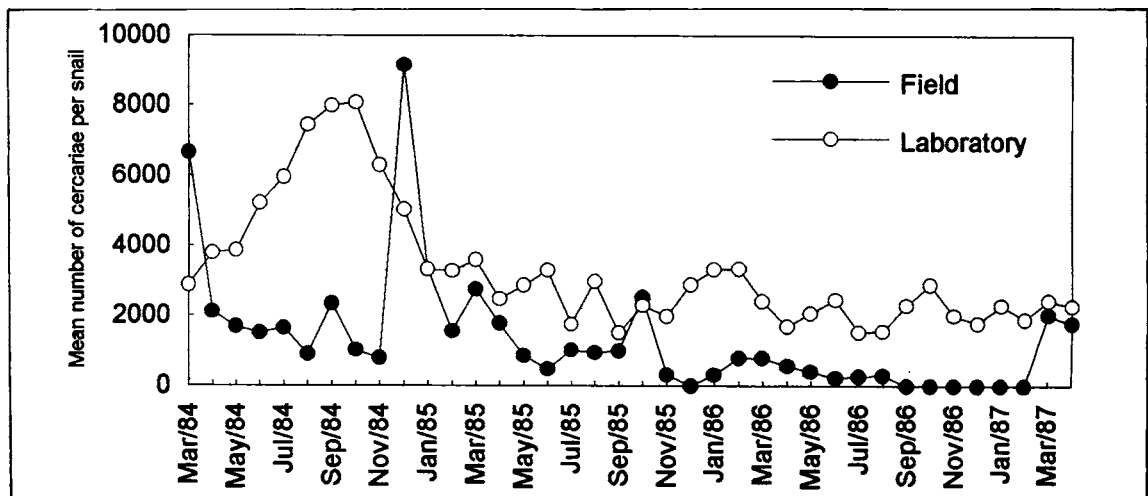


Fig. 2 - Monthly elimination of *Schistosoma mansoni* cercariae by *Biomphalaria glabrata* from field and from laboratory.

number of cercariae shed by the snails infected in the laboratory was 327,454,090 with an average of 6,061 larvae per snail. The maximum life span of the laboratory infected snails was 91 days. The annual means of the cercariae per snail in the laboratory varied between 2,224 and 4,222. Parasitological cure was not observed in these snails.

Amongst the 14 individuals working in the small holding two schoolchildren (14.3%) were infected with *S. mansoni* and were treated with oxamniquine.

DISCUSSION

The dynamics of *S. mansoni* cercarial shedding by naturally infected snails has been little studied due to the operational difficulties involved. Nevertheless the importance of such data in terms of the study of the transmission of the disease and their relevance to

prevalence levels are indisputable. In Minas Gerais, we have observed on number of occasions that the levels of infection of snails in endemic regions are low when the prevalence is high.

MILWARD-DE-ANDRADE^{15, 16}, in a three years study of a transmission focus in Horto, Belo Horizonte, recorded levels of infection varying between 1.8 and 83.5%. Of the 2,695 snails collected by the author, 6% were infected with *S. mansoni*. The highest level of infection was recorded in the month of October. The infected snails measured between 7 and 28mm in diameter.

CHU & DAWOOD⁴ correlated the number of cercariae shed by *Biomphalaria alexandrina* in Egypt with snail diameter and the variation of the seasons.

The longevity of the infected snails is another

TABLE 1

Production of *Schistosoma mansoni* cercariae by *Biomphalaria glabrata* collected during 38 months from a focus in Belo Horizonte.

Year of capture	Snails					Cercariae shed		
	Number collected	Mean diameter mm	Density in breeding site	Number infected	Percentage of infected	Number exposed*	Mean per snail	Total per year
1984	1,333	14.5	13.33	58	4.3	1,211	1,844	2,173,350
1985	2,033	12.4	20.33	92	4.5	1,874	955	1,752,390
1986	1,241	16.0	12.41	164	13.2	3,145	558	1,696,222
1987	759	13.0	18.97	10	1.3	24	1,889	45,350
Total	5,366	13.9	-	324	6.03	6,254	1,311.5	5,667,312
Mean ± S.D.	1,073 ± 1,606	13.9 ± 2.3	16.3 ± 3.9	64.8 ± 112.2	6.03	1,563 ± 2,164	1,311 ± 1,608	1,416,828

* Since positive snails were exposed to light twice a week in order to obtain cercariae this value is higher than the number of snails. S.D. = Standard deviation.

important factor in the transmission of the disease. For *B. alexandrina* from Egypt the recorded lifetime was 130 days⁴ and for *Bulinus truncatus* from the same country 329 days¹⁰. For *B. truncatus* from Malaysia longevity was 210 days⁵. THÉRON²⁴ recorded a longevity of 240 days for *B. glabrata* from Brazil similar to that found in the present study (218 days).

In a study of variation in the rhythm of cercarial shedding of nine lines of *S. mansoni* from Guadeloupe, THÉRON²⁵ observed that intraspecific variations in the chronobiology were adaptive process that favored transmission in various hosts.

In the present study, the level of *B. glabrata* infection in Barreiro of 6.03% was similar to that recorded by MILWARD-DE-ANDRADE¹⁵ for snails from Horto, was less than that found in Belo Horizonte by PAULINI & DIAS¹⁹, 19.1%, and higher than that found in the same area by SOUZA et al.²⁰, 3.1%.

The diameter of naturally infected snails (7-29mm) was similar to that registered for the snails from Horto¹⁵. The highest levels of infection found in the snails from Barreiro occurred during the first six months of the year (Fig. 1) in contrast to that found in Horto¹⁵.

The mean snail density of 16.3 per scoop was greater than that of 1,003 previously found in Belo Horizonte²⁰. The lowest monthly densities (1.5 and 0.9) were recorded in the driest period, October and November, 1986 (Fig. 1).

There was no correlation between the peaks of cercarial shedding of the laboratory and field infected snails despite maintenance under identical conditions (Fig. 2). In 16 of the 38 months of observation infected snails were not captured (Fig. 1) although in 31 months cercariae were produced by the specimens collected in previous months (Fig. 2) which guaranteed parasite transmission despite the fact that 12.9% had stopped eliminating larvae. Self cure was not observed amongst the laboratory snails since death occurred earlier due to the higher number of miracidia.

The observations of a number of authors^{4, 5, 6, 10, 15, 18, 20, 24, 25} show that in addition to the natural level of infection, compatibility, longevity of the infected snails, chronobiology of cercarial shedding and site of foci are important factors in the maintenance of the levels of prevalence in endemic regions.

In Belo Horizonte the prevalence of schistosomiasis in schoolchildren during the last decade is between 7 and 10%^{1, 8, 9}. In the present study, amongst the 14 individuals working in the small holding two schoolchildren (14.3%) were infected with *S. mansoni* and were treated. This breeding site continues to be active although infected snails were not collected in 1992 and 1993. Molluscicide treatment was undertaken in the ditches in 1988 and at the end of 1992. In order to eliminate the focus it would be necessary to fill in the ditches or canalize the water, thus changing the system of irrigation. The small holder, however, does not have any great interest in adopting these measures.

The results obtained show that a mean level of snail infection of approximately 6% can maintain a prevalence at a level of 14.3% in a focus even when the individuals here reportedly are treated (according to the report of small holder), the snails subjected to occasional control by chemical treatment and the ditches cleaned as was the case in this study.

RESUMO

Produção de cercárias de *Schistosoma mansoni* por *Biomphalaria glabrata* de foco em Belo Horizonte, Minas Gerais.

A densidade planorbídica, as taxas de infecção e a produção mensal de cercárias de *Schistosoma mansoni* por *Biomphalaria glabrata*, foram determinadas em foco do Barreiro de Baixo (Belo Horizonte MG, Brasil). Durante 38 meses, de 1984 a 1987, foram capturados 5.366 moluscos dentre os quais 324 (6,03%) estavam infectados com o *S. mansoni*. O total de cercárias eliminadas foi de 5.667.312. Cada molusco eliminou em média 17.422 cercárias durante sua permanência no laboratório. A maior longevidade dos moluscos infectados foi de 218 dias. Foi observada a cura natural de 42 (12,9%) dos exemplares infectados, após cerca de 130 dias de captura. A densidade planorbídica média, no foco durante esse período foi de 16,3 moluscos por conchada.

A eliminação de cercárias pelos moluscos do campo foi comparada com a de exemplares criados no laboratório, infectados em massa com a linhagem LE de *S. mansoni* de Belo Horizonte. Os moluscos do laboratório eliminaram em média 6.061 cercárias por exemplar, número 2,8 vezes menor do que os moluscos do campo, devido a longevidade menor.

A taxa de prevalência da esquistossomose no foco foi de 14,3%.

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