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Abstract

Objective

The influence of age and the presence of secondary sporocysts in the miraxonal attraction exercised by *Biomphalaria glabrata* on miracidia of *Schistosoma mansoni* of the BH strain were studied.

Material and Method

A glass apparatus containing two compartments joined by a tube and previously tested in other experiments, was used. Specimens of *B. glabrata* or its snail conditioned water (SCW) selected before the first oviposition (sexually immature), after the first oviposition (adult), with or without secondary sporocysts, were used to attract the miracidia.

Results

It was noted that snails or their SCW containing secondary sporocysts lost the ability to attract miracidia. The sexual maturity of the snail did not influence miraxonal attraction.

***Biomphalaria*, parasitology. *Schistosoma mansoni*, physiology. Host-parasite relations.**

Resumo

Objetivo

Estudou-se a influência da idade e da presença de esporocistos secundários na atração miraxonal exercida por *Biomphalaria glabrata* sobre miracídeos de *Schistosoma mansoni* da linhagem BH.

Material e Método

Utilizou-se um aparato de vidro composto de duas câmaras unidas por um canal, previamente testado em outros experimentos. Como atraentes utilizaram-se exemplares de *B. glabrata* ou a água de condicionamento destes moluscos (SCW) com idade inferior à primeira oviposição (infantil) e com idade superior à primeira oviposição (adulta), contendo ou não esporocistos secundários.

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Resultados

Verificou-se que os moluscos (ou sua SCW) que continham esporocistos secundários perderam o poder de atrair miracídeos. A condição dos moluscos serem infantis ou adultos não interferiu na atração miraxonal.

Biomphalaria, parasitologia. Schistosoma mansoni, fisiologia. Relações hospedeiro-parasita.

INTRODUCTION

Miraxonal attraction caused by vectors snails of *Schistosoma mansoni* was demonstrated by Kloetzel^{8, 9} (1958, 1960), Etges et al.⁶ (1985).

The miracidia attraction to intermediate hosts results from substances emanated by the snail in water. The water containing these substances is named SCW (snail conditioned water) (Chernin⁴, 1970).

The hemolymph and the SCW of the vector mollusks were analysed by Brasio et al.^{2,3} (1985) where they appointed some cations and aminoacids as probable substances responsible for miraxonal attraction.

Magalhães¹¹ (1987) deemed the miraxonal attraction as one of the biological events essential in the study of epidemiology of *mansonic schistosomiasis*.

Niemann et al.¹⁴ (1990) observed that the susceptibility to the infection of *B. glabrata* caused by *S. mansoni* was inversely related to the size of the snail and not necessarily to its age.

Zanotti-Magalhães et al.¹⁷ (1991) verified that the intensity of miraxonal attraction is directly related to the degree of susceptibility of the snail to the schistosome infection. To get to this conclusion the authors utilized populations genetically selected by self-fecundation. The authors also concluded that the degree of miraxonal attraction depends exclusively on snail population, not interfering the fact that the miracidium had been originated of trematode population maintained by several generations in snails partially resistant or highly susceptible.

In the present work it is considered the possible relationships of snail sexual maturity (before and after first oviposition) and infection (present, absent) with miraxonal attraction.

MATERIAL AND METHOD

Attractants were *B. glabrata* selected before the first oviposition (sexually immature) or after the first oviposition (adults), having or not secondary sporocysts (infected or not). SCW (Chernin⁵, 1972) obtained from the snail pools, maintaining the same biomass constant, classified according to the above characteristics were also used as attractants.

The factors to be studied were thus:

- sexual maturity (immature, mature)
- infection (no: no secondary sporocysts; yes: presence of secondary sporocysts)
- source (mollusk, SCW)

Every combination of the levels of the three factors, comprising eight experimental settings, were tested in a completely randomized experiment. Ten replications were performed. The resulting eighty uses of the artifact described below were performed in random order. An additional setting, with only dechlorinated water, was tested for the sole purpose of checking the experimental device.

A previously tested glass artifact was used for the study of miraxonal attraction. The artifact is composed of two circular chambers, with 30 mm diameter and 20 mm depth, joined by a channel of 40 mm length, 11 mm width and 10 mm depth (Brasio et al.², 1985).

Each run of the experiment was carried out by allocating a mollusk or SCW in a randomly chosen chamber of the artifact and depositing ten BH strain miracidia in the channel. The result of the run was the frequencies of miracidia in each compartment of the artifact - chamber with mollusk or SCW, empty chamber or channel, after fifteen minutes. A stereoscopic microscope with $\times 0.5$ reductor frontal glass was used for the observation and counting of miracidia.

Standard categorical data statistical techniques were employed to analyze the results: Grizzle et al.⁷ (1969) model and derivations thereof for the evaluation of statistical significance of main effects and interactions; and correspondence analysis (Benzécri¹, 1973), for the visualization of these effects.

RESULTS

A summary of the experimental results are in Table 1.

The statistical analysis (Table 2) evidenced that: a) infection had a definite effect, in those snails with secondary sporocysts showed no attraction to miracidia; b) sexual maturity was immaterial to attraction.

The results of the correspondence analysis also show clearly the importance of infection on the miracidia attraction. The graphic (Figure) should be read by evaluating the angles between the vectors corresponding to the compartments (labeled A, B and C, respectively) and the vectors corresponding to the factors levels combination (labeled zero to 7, in the order of Table 1) (Table 3, Figure).

Table 1 - Total frequencies of miracidia in compartments, after a 15 minutes period: results of ten replicates per experimental setting.

Source	Sexual Maturity	Infection	Chamber A (with SCW or Mollusk)	Empty Chamber (B)	Channel (C)
Mollusk	Immature	No	81	19	0
		Yes	53	47	0
SCW	Mature	No	75	25	0
		Yes	44	55	1
	Immature	No	66	34	0
		Yes	59	39	2
Mature	No	86	13	1	
	Yes	54	39	7	
Total			518	271	11

SCW - Snail conditioned water.

Table 2 - Generalized Least Squares (GLS) analysis of variance table.

Source	DF	Chi-Square	Probability
Intercept	2	171.20	0.0000
Mollusk	2	3.23	0.1987
Oviposition	2	1.23	0.5402
Mollusk Oviposition	2	8.19	0.0167
Infection	2	49.18	0.0000
Mollusk Infection	2	2.27	0.3219
Oviposition Infection	2	4.23	0.1205
Mollusk Oviposition Infection	2	4.18	0.1236
Likelihood ratio	0	-	-

DF - degree freedom

Table 3 - Miracidia distribution.

Compartments	Dimension 1	Dimension 2
Chamber A	0.994827	0.005173
Chamber B	0.947816	0.052184
Channel C	0.171922	0.828078

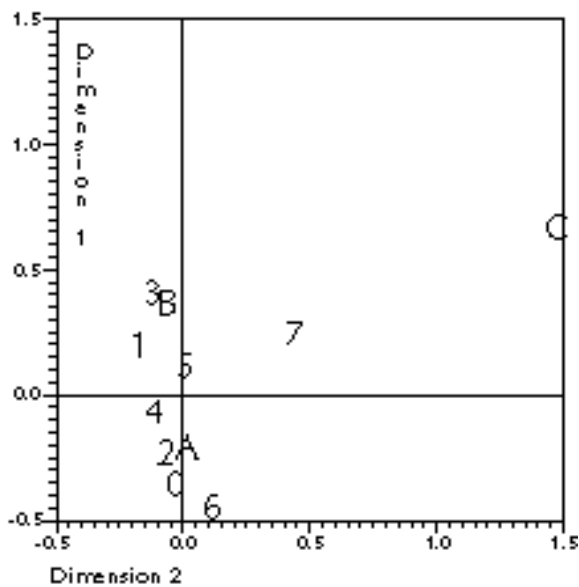


Figure - Correspondence Analysis of Miracidia Distribution.

The vectors labeled A and B are close to axis 1 and contrary to each other. This axis represents 73% of the total variation in the correspondence analysis. One sees, then, that axis 1 is associated with the chambers. Now the vectors corresponding to the factors settings show that the even numbered ones are in the same direction as A; those numbered with odd numbers, associated with B. This shows clearly that chamber A receives more miracidia in the presence of non-infected mollusk or SCW of non-infected mollusks.

There is one vector⁷, which is aligned with C. This corresponds to the situation: SCW, mature, infected. It is apparent that this situation is the one situation that leaves some of the miracidia left in the channel.

DISCUSSION AND CONCLUSIONS

It is well know that mollusks infected by digenetic larvae suffer deep physiological transformation altering, for example, their reproductive ability and the susceptibility to infection (Minchella et al.¹³, 1983; Machado et al.¹⁰, 1988).

Seta et al.¹⁶ (1993) verified that *B. glabrata* (and also the SCW) newly infected (having primary sporocysts) repulsed miracidia of *S. man-*

soni. For the *B. tenagophila* these authors obtained contrary results.

It was noted by us in the present work that *B. glabrata* having secondary sporocysts, sexually immature, as well as adults, lost attraction to miracidia. It seems, however, that the power to repel observed by Seta et al.¹⁶ (1993) is attenuated during the infection of the snail.

The repellence to miracidia of *S. mansoni* had already been described by Magalhães et al.¹² (1991) in a sample of *Biomphalaria straminea*. These authors concluded that snails can have the ability to emanate substances repulsive to the miracidia.

The fact that infected snails do not attract miracidia or even repulse these larvae, results in a better

profit of the larvae in water, because that they are only attracted by non-infected snails.

Richards et al.¹⁵ (1972) verified that the sexual maturity of snails affects the susceptibility of the infection caused by *S. mansoni*.

The present results show that the mollusks maturity does not influence the miraxonal attraction.

During the miracidia observation, it was verified in all experiments that the larvae were more active in the chambers where the attractants (snails or SCW) were. This fact demonstrates that miraxonal substances act not only attracting miracidia but also increasing their vitality. The increased of vitality was evidenced by a more active movement of the larvae and also, by their increased length.

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