# SCHISTOSOMIASIS MANSONI IN THE MUNICIPALITY OF PEDRO DE TOLEDO (SÃO PAULO, BRAZIL) WHERE THE Biomphalaria tenagophila IS THE SNAIL HOST. I — PREVALENCE IN HUMAN POPULATION (1)

L. C. S. DIAS (2), U. KAWAZOE (2), C. GLASSER (3), S. HOSHINO-SHIMIZU (4), H. Y. KANAMURA (4), J. A. CORDEIRO (2), O. F. GUARITA (3) & G. J. ISHIHATA (3)

#### SUMMARY

Due to the scarce information about the epidemiological features of schistoso miasis in which the vector is Biomphalaria tenagophila, an investigation was carried in Pedro de Toledo in 1980 where such peculiarity is observed. Stool examinations (Kato Katz method) were performed in 4,741 individuals (22.8% positive to Schistosoma mansoni eggs) of this 583 had previously received chemoterapy and 4,158 remainders, untreated. The schistosomiasis prevalence in those two groups where respectively 31.7% and 21.6%. Epidemiological investigation showed that 83.6% were autochthonous cases from the studied area: the autochthonous prevalence rate, and the intensity of infection in the untreated autochthonous cases were higher in males than in females; the intensity in the latter untreated group was low, 58.5 eggs/g feces (geometric mean). Moreover, according to the age groups the intensity of infections correlated well ( $r_s = 0.745$ ) with the prevalence rates. Schistosomiasis was verified to occur mostly during the leisure time and by the use of water streams for housework in rural zone. Only 0.4% out of 1,137 snails was positive for S. mansoni cercariae, apparently unchanged from the 1978 study when the human prevalence was 12.0%. The studied area presented differences and similarities in relation to the other Brazilian areas were the main intermediate host is B. glabrata.

KEY WORDS: Schistosoma mansoni; Biomphalaria tenagophila; Epidemiology.

### INTRODUCTION

In the Valley of Ribeira (State of São Paulo — Brazil), the human schistosomiasis was first recorded in 1953<sup>21</sup>, with 43 autochthonous cases in the District of Ana Dias, Municipality of Itariri, neighboring the Municipality of Pedro de Toledo, where the present study was performed. Since that time only **Biomphalaria tenagophila** 

has been found in the molluscan breeding sites but, those snails were already positive for **Schistosoma mansoni** cercariae in about 1.7%. In the Municipality of Pedro de Toledo, autochthonous cases of schistosomiasis were also detected as reported by PIZA & RAMOS<sup>21</sup>, however, in only 6 of the studied population.

<sup>(1)</sup> Supported by SUCEN, FAPESP and CNPq.

<sup>(2)</sup> Professor of Universidade Estadual de Campinas. Inst. Biologia, Depto. Parasitologia, C. P. 6109. CEP 13081 Campinas, SP. Brazil.

<sup>(3)</sup> Epidemiologist of Superintendência de Controle de Endemias. São Paulo, SP, Brasil.

<sup>(4)</sup> Professor of Universidade de São Paulo, São Paulo, SP, Brazil.

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More recently (1970), the Superintendency of Endemic Disease Control (SUCEN) of the São Paulo State Health Department has established a Schistosomiasis Mansoni Control Programme in that area. This programme includes yearly stool examination for all the population exposed to the risk of transmission, chemoterapy of infected individuals, periodical snails examinations from the water collection, health education, and, application of molluscicide drugs in the streams whenever infected snails with cercariae of S. mansoni are found.

Despite this effort the prevalence has increased from 4.0% (1970) to 12.0% (1978); of the latter percentage 85.0% consisted of autochthonous cases. The rate of infected **B. tenagophila** however seemed unchanged since 1970, with 1.0%.

The present study was undertaken due to that increasing rate of schistosomiasis mansoni verified in Pedro de Toledo and also because of the lack of studies in relation to the natural history of schistosomiasis in regions where B. tenagophila is the intermediate host. The whole project which started in 1980 includes a detailed investigation on parasitological an epidemiological aspects of urban and rural areas during, a period of approximately 8 years. These studies will be performed as following: prevalence of the infection in the human population and in the small mammals; prevalence of snail infection with S. mansoni; evaluation of parasitological and immunological methods for diagnosis of schistosomiasis; clinical study of infected individuals; specific treatment and assessment of pa rasitologic cure of treated patients; application of molluscicide drugs in all natural snail bree ding sites, whenever infected molluscs are detec ted; evaluation of transmission by the incidence of schistosomiasis using control groups; use of sentinel-mice; infection rate and density of the snail host.

In this paper, part of the epidemiological stu dies concerning the prevalence of urban and ru ral schistosomiasis, identification of autochtho nous cases and assessment of previously treated subjects will be reported.

#### MATERIAL AND METHODS

#### 1. Description of the studied area

Pedro de Toledo is located in the Valley of Ribeira, near the Southern coast of São Paulo State, Brazil. The studied region comprises an area of 631 km<sup>2</sup> and the town is located 43.5 m above sea level. The landscape is hilly with many small streams which are shallow with many stones on its margin and plentiful vegetation. The Itariri River is the main stream and belongs to the Ribeira de Iguape basin. The climate according to Kopenn's classification can be considered as tropical and humid. The mean temperature is about 22°C in the hottest month whereas, in the coldest month, 18°C. In the dryest month the rainfall is about 60 mm, however, in January and February the rainfall is heavier reaching 203 to 220 mm. According to the 1980 census of the Brazilian Institute of Geography and Statistics. Pedro de Toledo has 6,076 inhabitants, with 2,336 living in the urban zone and 3,704 in the rural zone. The majority of the people is white. The main local activity is agriculture, mostly in banana plantation. According to SUCEN, about 5,000 inhabitants live either in places where the transmission of schistosomiasis has ocurred or is presently occuring. These areas are called FO CI. The remaining 1,000 inhabitants live in pla ces where this parasitosis has not been reported yet.

The studied population from the FOCI was 2,370 inhabitants from the urban zone and 2,371 from the rural zone. The urban inhabitants lived in a small town with the following facilites: tap water and power supplies, around 85.0% of the dwellings had sewer system, and 50.0% of the streets were paved; the sewage was treated. The rural inhabitants, generally, lived in poor houses scattered along non-paved roads in 15 areas namely: Rio do Peixe, Rio do Peixe I, km 106, km 110, Fazenda São José, Braço do Meio, Braço do Meio I, Mariano, Mariano I, Ribeirão do Luiz, Três Barras, Água Parada, Jardim Caju, Água Fria and Vila Batista. There were no irrigation canals in the area; there were drain channels and small streams in the banana plantation which were the main breeding places of B. tenagophila; there was no sewers system available.

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## 2. Laboratory method for diagnosis

Stool examination was carried out in 1980 according to Kato-Katz quantitative method<sup>11</sup>, by collecting one fecal sample from each individual and preparing three slides for searching eggs of **S. mansoni** and of other non-related helminths.

## 3. Statistical analysis

Results of laboratory test and personal data regarding to sex, age, skin color, occupation, places of birth, living sites, epidemiological classification, as well as, previous chemotherapy were transcribed into individual computer cards.

The intensity of infection was expressed as geometric mean of number of excreted eggs per gram feces, due to the irregular distribution of eggs<sup>15</sup>.

The following methods were used for the statistical analyses: analyses of variance (ANOVA), Tukey's test for contrasts, Kolmogorov-Smirnov goodness-of-fit test and confidence intervals. The level of 5.0% (p < 0.05) was adopted as minimum significant difference for rejecting the null hypothesis. Sperman correlation coefficients were

estimated<sup>25</sup> to verify the correlation between the intensity of the infection observed in different age groups or localities and the prevalence of schistosomiasis.

#### RESULTS

A total of 4,741 subjects could be studied (Table 1) and the overall schistosomiasis mansoni prevalence in Pedro de Toledo was 22.8% (1083/4741). The rural zone presented higher infection rate (25.6%) in relation to the urban area (20.2%) but no significant at the 5.0% level. This parasitosis also prevailed in rural even after the separation into two sub-groups, one that received treatment and other, no-treatment (Table 1)

The prevalence rates verified in rural untreated population to schistosomiasis varied greately according to the locality: 5.9% (Rio do Peixe I), 14.5% (Km 110), 17.8% (Km 106), 20.9% (Rio do Peixe), 25.0% (Fazenda São José), 25.5% (Braço do Meio I), 27.5% (Mariano I), 30.2% (Braço do Meio), 31.4% (Ribeirão do Luiz), 31.6% (Mariano), 31.9% (Três Barras), 36.9% (Água Parada), 42.9% (Jardim Caju), 43.6% (Água Fria) and 61.2% (Vila Batista).

In respect to 4,158 individuals without any previous treatment to **S. mansoni**, the prevalen-

TABLE 1

Prevalence of schistosomiasis mansoni in urban and rural areas of the municipality of Pedro de Toledo (São Paulo State — Brazil — 1980) by parasitological Kato-Katz method, according to the treatment to the disease.

Stool Examination ——	Treatment		— Total —	Untreated		— Total
	Urban Area %	Rural Area %		Urban Area %	Rural Area %	
Positive	26.4* ( ± 4.4)	41.6 ( ± 6.7)	185	18.9 ( ± 2.1)	24.1 ( ± 1.8)	898
Negative	$73.2$ ( $\pm 4.4$ )	$58.4$ ( $\pm 6.7$ )	398	$81.1$ ( $\pm 2.1$ )	75.9 $(\pm 1.8)$	3,260
Total	385	198	583	1,985	2,173	4,158

<sup>\*</sup> Confidence interval 95%

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ce rates of helminthiasis were: Trichuris trichiura, 42.3%, Ascaris lumbricoides, 40.4% and S. mansoni, 21.6%. There also were one case of infection with Enterobius vermicularis and one with Taenia sp.. Multiple parasitism with two or more species was observed in 36.4% of the cases.

Epidemiological investigation was possible to be done in 884 (81.6%) out of 1083 carriers of **S. mansoni**, of these 83.6% (739) were autochthonous cases in Pedro de Toledo, 3.8% from other municipalities of São Paulo State, 1.3% from other Brazilian States and 11.2% undetermined. Thus, the number of individuals acquired schistosomiasis only in São Paulo State was 772 (87.3%).

All the statistical analyses were performed in untreated population or in autochthonous cases.

The analysis of variance could not be done in the total examined inhabitants (4,741) because the full information (sex, age, domicile and epidemiological classification) were not obtained; from 3,407 residents with complete informations for this analysis was performed.

The ANOVA test indicate differences between the prevalence of autochthonous schistosomiasis in 1,765 males (28.1%) and in 1,642 females (14.7%), as well as, among age groups comprising from 0 to over 60 years (Figure 1 and Table 2).

TABLE 2

Results obtained by the statistical analysis of variance (scale:  $arcsen \sqrt{-p}$ ) from data of Figure 1, corresponding to the prevalence of S. mansoni infection determined by parasitological method (Kato Katz) according to the age groups, sex and zone, in the population of Pedro de Toledo, São Paulo State, Brazil (1980)

VARIANCE	AGE GROUP						
VARIANCE	0 to over 6	0 years	5 to 29 years				
	Degree of freedom	F	Degree of freedom	F			
Zone	1	2.92	1	1.53			
Sex	1	41.09*	1	6.73			
Age group	12	7.71	4	9.49			
Zone x Sex	1	0.13	1	5.89			
Zone x Age group	12	1.42	4	1.08			
Sex x Age group	12	0.76	4	2.85			

<sup>\*</sup> Significance at the 5.0% level

In Figures 1 and 2, higher prevalence rates and intensity of infection can respectively be seen within the age groups from 5 to 29 years.

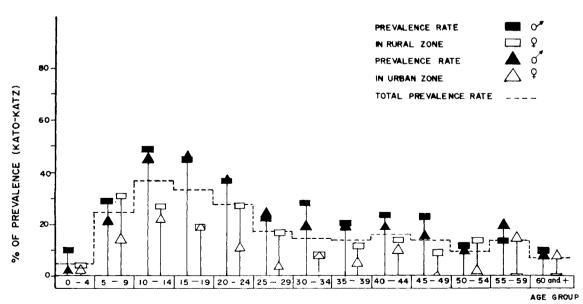


Fig. 1 — Prevalence rates of autochthonous schistosomiasis mansoni by the Kato-Katz method, according to age group, sex, rural and urban zones, in Pedro de Toledo, São Paulo, Brazil (1980).

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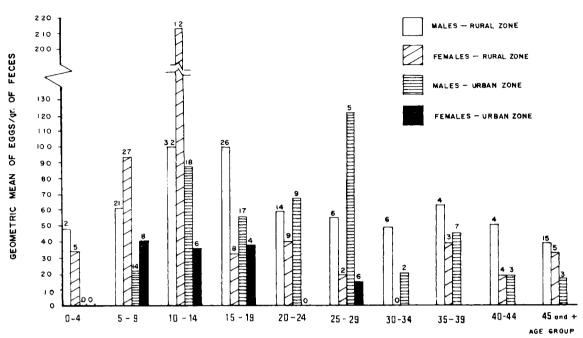


Fig. 2 — Geometric mean of the number of. S. mansoni eggs per gram of feces by the Kato-Katz method, in 307 autochthonous non treated individuals from Pedro de Toledo, São Paulo, Brasil (1980).

Obs. The 25 to 29 female age group in the urban zone also includes individuals from subsequent groups. Numbers placed above the columns indicate the number of subjects examined.

ANOVA was also applied for these age groups and the results obtained (Table 2) were similar to the afore mentioned study for all age groups, besides the significant difference verified in the interaction between zone and sex. More detailed studies done using Tukey's test showed that, in general, the age groups 10-14, 15-19 and 20-24 had significantly higher prevalence rates than the remainder age groups. However, these three groups did not differ among them. The age group 5-9 had significantly higher prevalence than the 0-4 and those more than 45 years of age and lower than the age group 10-14 (Tukey's test difference 5% = 7.308; k = 11 and v = 7.1).

The intensity of infection in 307 untreated autochthonous cases was 58.5 eggs/g feces (geometric mean). The confidence intervals test showed statistically significant diference (p value less than 0.01) for the following conditions: in the rural zone the intensity of infection (66.7 eggs/g) was higher than in the urban area (44.6 eggs/g); the males presented higher infection (61.8 eggs/g) than females (53.3 eggs/g), while the males from the rural area had the egg excretion

rate (69.9 eggs/g) higher than those from urban area (49.8 eggs/g); the males from the rural zone also eliminated more eggs (61.4 eggs/g) than those from urban zone (32.1 eggs/g).

The data which provided Figure 2 were analysed by Kolmogorov-Smirnov test and it was observed that the distribution of the intensity of infection, expressed in terms of geometric mean of excreted eggs per gram feces, between sexes in the rural zone was significantly different [ $D_{max}$  (0.2498)  $D_{5\%}$  (0.1972);  $n_1=130$  and  $n_2=75$ ] and this phenomenom did not occur in urban area.

A good correlation was obtained ( $r_s=0.745$ ) between the intensity of infection and prevalence rate according to the age groups (Figure 3) Nevertheless, this correlation was poor ( $r_s=0.196$ ) between the same parameters in relation to the town and 15 studied areas in Pedro de Toledo.

### DISCUSSION

The overall prevalence rate (22.8%) found in Pedro de Toledo in 1980 was considerably higher

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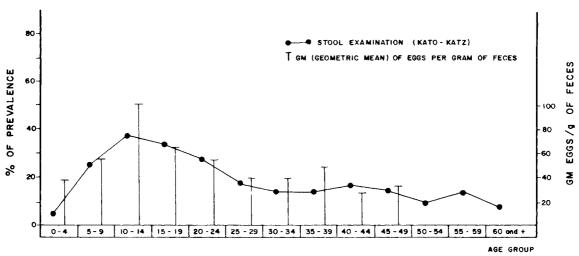


Fig. 3 — Prevalence rates of autochthonous cases of schistosomiasis mansoni obtained by Kato Katz method and intensity of infection according to age groups, in Pedro de Toledo, São Paulo, Brazil (1980).

than the rate of 12.0% observed in 1978 when SUCEN performed stool examinations by Kato's qualitative methods in 1.000 inhabitants.

This prevalence is considered high, specially by the fact that, **B. tenagophila** is known to be an intermediate host of low suceptibility for **S. mansoni**<sup>19, 23</sup>. In that area, from March to May of 1980, only 5 specimens out of 1,137 **B. tenagophila** collected were realising cercariae of **S. mansoni**. Those positive snails were collected from two breeding sites, one located in rural and other in urban areas. Since high prevalence rate of schistosomiasis in non-treated cases was detected in Vila Batista, the causes of high transmission in that population are now under investigation.

In areas of Brazil where the schistosomiasis is endemic, and where **B. glabrata** and **B. straminea**, are considered the intermediate hosts, the human prevalence is usually higher than the one observed in this study<sup>16.18</sup>. The autochthony of endemic cases in Pedro de Toledo is contrary to those found in other regions of São Paulo State, where 88.4% of the infections are imported and only 10.0% are autochthonous<sup>26</sup>. It should be emphasized that the transmission of this parasitic disease in São Paulo State is found in isolated foci. For instance, the Fazenda Colmeia Focus, in the county of Pindamonhangaba, showed a human prevalence of 17.5%, whereas the

rate for the entire Municipality was only 3.3%. Therefore, it is difficult to compare prevalence rates of the endemic diseases in the different regions of the State.

Other helminthiasis reported for Pedro de Toledo showed similar rates to those found in other areas of Brazil<sup>8</sup>. Although the more specific method for the diagnosis for each parasites not used.

The finding of 73.2% (Table 1) previously treated individuals presented negative egg counts in feces, as well as, the relatively lower intensity of infection associated to a lower prevalence rate in urban area in relation to the rural area (58.4%) may be explained by the improvements of the levels of basic sanitation and education received and by other epidemiological factors existing in the region. On the other hand, in rural area which have not receive sanitation improvements the prevalence of schistosomiasis became higher.

The persistence of egg excretions among previously treated subjects appears to be reinfections and probally development of some drug tolerance or resistence, since the population had been successively submitted to Niridazole, Hycanthone and Oxamniquine treatments. This last aspect is also now under investigation. The prevalence rate of schistosomiasis in Pedro de

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Toledo, according to the sex was higher in males than in females (Table 2). This may be explained by the fact that in the studied area, men usually enter water streams more frequently than women. In the Municipality of Roseira (São Paulo State), where the snail host is B. tenagophila, SANTOS<sup>24</sup> observed that among schoolchildren of the urban area, 90.0% of infected people were boys. However, in schoolchildren of the lowland area there was no difference related to the sex. In another Municipality vicinity of Roseira, there was a higher prevalence rate in males 10. However, in Brazilian States where the disease is more endemic, settled for a longer period of time, most investigators agree that there is no significant difference in regard to sex1, 2, 4, 18, 22, 24. The frequency distribution of prevalence according to the age groups determined by stool examination, showed an early prime infection (0-4 age group) in 4.8% (Figure 1). The age group between 5-24 years presented the highest prevalence rate (Figures 1 and 2). This is comparable to the results found by others authors in several Brazilian States<sup>1, 5, 13, 14, 17, 20</sup>. In São Paulo State, FRÓES et al<sup>10</sup> found a higher prevalence rate in 10-20 age group and suggested that the disease in the Municipality of São José dos Campos, where the B. tenagophila is the intermediate host, was usually acquired during recreational activities. In Pedro de Toledo there was a decrease in the prevalence rates among individuals of 25 years and older (Figures 1 and 2). These data do not agree with the results presented by SANTOS<sup>24</sup> who reported the highest percentages among older individuals working in rice fields and cleaning ditches in the lowland. According to this author, the disease has a professional character. But apparently, this is not the case for Pedro de Toledo, where the schistosomiasis is acquired mainly during recreational activities such as swimming, fishing, etc. In the rural zone, transmission is also consequence of the use of water for housework.

There is no record of differences between prevalence rates of rural and urban zones in autochthonous cases (Figure 1 and Table 2). In the present area of study there are no clearcut behavioral boundaries for the inhabitants of both areas although the physical boundaries between the rural and urban zones are well defined. Agri-

culture is the main activity of the population; banana is the predominant type of agriculture, followed by vegetables. The mean intensity of infection of 58.5 eggs/g feces was low to the usually found in other Brazilian endemic areas. COURA & CONCEIÇÃO<sup>6</sup> recorded the mean values of 207 and 138 eggs/g feces respectively in of 1973 and 1974 in a rural community, in the State of Minas Gerais, where the **B. glabrata** is the major vector. In Pedro de Toledo, the highest intensity rate was observed more in the rural than in the urban area. Male individuals presented a higher number of eggs in the feces than women.

This indicatives that the men of the rural zone have greater contact with the foci of infection. In regard to age groups (Figure 2) the highest intensity of eggs was found in the groups ranging from 5 to 24 years old. In the subsequent age groups, no further attempt was made to carry out any comparison because of the relatively small size of the sample. An interesting fact is that in the 10-14 age group, women of the rural zone showed a geometric mean of 212 eggs/g of feces (Figure 2). This value is much higher than the geometric mean of 58.5. It was expected that females would generally show a lower intensity rate of infection than men. However, intensity of infection in men was 61.8 and women 52.3 eggs/ g of feces. The high geometric mean found in the 10-14 women group is probably related to the fact that these girls frequently enter water streams to wash clothes or cookware, taking the place of their mothers who usually go to the work in the fields. In spite of an irregular excretion of eggs in the feces, their countings in populational studies should be of some value, considering that the mass evaluation offsets the causes for error<sup>1, 3, 14, 15, 27</sup>. The intensity of infection in previously treated patients was not, described here because is well know that intensity decreased in those patients.

The data here obtained regarding to the existence of positive correlation between the geometric mean of egg counts and the prevalence rate will be of value in determining the conversion factor or of the forecast the intensity and prevalence of the infection through the Kato-Katz method.

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However, in highly endemic area of schistosomiasis, having **B. glabrata** as the vector, such correlation was not verified<sup>15</sup>. The results here obtained will offer a base line for similar epidemiological works carried out under the same conditions in other areas.

The sensitivity of Kato-Katz method is known to be high intensity of infections<sup>7</sup> but as verified in previous studies<sup>12</sup>, the parasitological test is not relible, mainly in areas of low and medium prevalence rates. This fact was observed by DIAS et al.<sup>9</sup>, who had to repeat the parasitological test 4 to 5 times to confirm some positive results provided by serological tests. The exact sensitivity of Kato-Katz method is now beeing estimated in relation to skin test and immunofluorescence test. The preliminar results indicate that its sensitivity is lower than 50.0%.

In spite of the problem presented by the Kato-Katz method, as to the degree of the sensitivity, it demonstrated that, in Pedro de Toledo, infection starts before 4 years of age. It was verified that in the areas which transmission of the parasite occurs via B. tenagophila, epidemiological characteristics are not so much different from those of other Brazilian areas in which B. glabrata is the intermediate host. The main difference is related to the intensity of infection, which now is still low in Pedro de Toledo.

# It is concluded that:

- a) the prevalence rate of schistosomiasis mansoni in Pedro de Toledo, as determined by stool examination, was 22.8%. The autochthonous carriers were 80.8%;
- b) according to the tests employed, younger people (5 to 24 years old) were more affected by the infection and the prevalence was higher in males than females;
- c) the prevalence rate in the urban zone was the same as in the rural zone when determined by stool examination in autochthonous cases:
- d) more than 70.0% previously treated cases presented parasitologically negative;
- e) the intensity of infection was low, with a geometric mean of 58.5 eggs/g of feces;

- f) transmission of schistosomiasis usually occurred during leisure time and also during the use of water streams for housework in the rural zone:
- g) a positive correlation between intensity of infection and prevalence rates according to the age groups was observed.

#### RESUMO

Esquistossomose mansônica no município de Pedro de Toledo (São Paulo, Brasil) onde Biomphalaria tenagophila é hospedeiro intermediário. I — Prevalência na população humana.

Devido à escassez de dados epidemiológicos sobre esquistossomose mansônica onde Biomphalaria tenagophila é vetor foi desenvolvido em 1980 o presente trabalho, no município paulista de Pedro de Toledo. Foram examinadas fezes de 4741 pessoas (Método de Kato-Katz) com prevalência de 22,8%; entre essas, 583 foram tratadas para a endemia anteriormente e 4158 não medicadas; as prevalências nos dois grupos foram, respectivamente, 31,7% e 21,6%. Por investigação epidemiológica constatou-se que 83,6% dos casos foram autóctones da área estudada. Prevalência dos autóctones e intensidade de infecção nos portadores autóctones não tratados foram maiores no homem do que na mulher; a intensidade no último grupo foi baixa: 58,5 ovos/ g de fezes (média geométrica). De acordo com grupos etários, se correlacionaram bem (r. = 0,745) as intensidades de infecção e as prevalências. A infecção ocorreu, na zona rural, principalmente, durante lazer e trabalho doméstico. Somente 0,4% de 1137 moluscos foram positivos para Schistosoma mansoni. Esse índice foi, aparentemente, o mesmo em estudo de 1978 quando a prevalência humana era de 12,0%. A área estudada apresentou diferenças e semelhanças epidemiológicas em relação às áreas onde B. glabrata é o principal hospedeiro intermediário.

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