Factors related to transmission of and infection with *Schistosoma mansoni* in a village in the South-eastern Region of Brazil

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In this transversal study, factors related to infection with and transmission of Schistosoma mansoni *were explored. Based on stool examinations of two Kato-Katz smears of a single sample, the prevalences of schistosomiasis and geohelminths were established. In a multivariable analysis, sets of demographic, socio-economic and water contact pattern variables were tested for strength of relation with infection. Males presented a 3.39-times higher risk for infection than females. The age groups between 10-19 years and 20-30 years showed risks of infection 7.1- and 7.5-times higher, respectively, than the control age group between 0-10 years. Individuals practicing leisure activities had a 1.96-times higher risk than those without these activities. The malacological survey identified snails of the species* Biomphalaria glabrata, Biomphalaria straminea *and* Biomphalaria tenagophila. *Two exemplars of* B. glabrata *(0.53%) proved positive for* S. mansoni. *The socio-economic improvements observed in the locality suggest a protective and preventive effect towards infection with schistosomiasis, which requires further investigation with a longitudinal and more detailed study design. Considering our findings, a proposal for an integrated control program should be based on two pillars: one horizontal, which involves social empowerment and health education, and another more vertical, which delivers treatment and infrastructure improvements.*

Key words: schistosomiasis - *Schistosoma mansoni* - socio-economic factors - risk factors - area of low transmission - Brazil

There is much scientific evidence that socio-demographic variables and contact with unsafe water are associated with infection with schistosomiasis. Studies in different settings have been carried out describing vulnerable parts of the population, such as school children, types of behaviour related to a higher risk of acquiring the infection, as is the case with household, occupational and leisure activities, or involving socio-economical status and its correlation to the disease (Lima e Costa 1983, Guimarães et al. 1985, Cairncross et al. 1996, da Silva et al. 1997, Barbosa & Barbosa 1998, Moza et al. 1998, Watts et al. 1998, Bethony et al. 2001, 2004, Massara et al. 2004, Gazzinelli et al. 2006). Studies carried out in Africa investigating infections with *Schistosoma haematobium* present similar associations, demonstrating the importance of studies involving snail hosts, water contacts and micro-spatial distribution for an integrated epidemiological approach (Stothard et al. 2002).

The data presented in this paper are part of a broader project with a primary focus on diagnostic and control strategies for *Schistosoma mansoni* in an endemic area (Enk et al. 2008). This study intended to investigate some aspects of the association between infections with *S. mansoni* and general demographic, socio-economic and water contact pattern variables in the village of Chonim de Cima and to give an overall view about the transmission and maintenance of the disease in this area. Therefore, a malacological survey was carried out and an evaluation of safe water supply and sanitation issues was also part of this study. It also tried to gather knowledge on specific local features of water contact that determine schistosomiasis infection, providing support to elaborate control strategies involving local communities (Barbosa et al. 1996). Seeking such conditional characteristics allows investigators to realise local people's behaviour, enabling an association between the disease and activities that may result in an increased risk for infection, in order to propose specific preventive strategies (Laurrell & Gil 1975).

PATIENTS, MATERIALS AND METHODS

Study area and population - The village of Chonim de Cima is situated in the county of Governador Valadares in the north-east of the state of Minas Gerais (MG), Brazil. The district of Chonim de Cima covers an area of 175 km^2 in the north of the county. The area is characterised by multiple water sources, including brooks, swamps and small lakes that are used for professional and leisure activities of the local population.

The brook, before it enters the village, is used as the main water source for drinking and domestic needs of the population. For this purpose, the water is pumped to a water treatment station, which is run by the municipality-owned Water and Sewerage Company of Governador Valadares. The treated and chlorinated water is stored in water tanks and finally delivered through a pipe system to the consumers in all households of the village. As the brook

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crosses the village, it serves as the principal recipient for sewage of in numerous individual drainages of households nearby. At the brook's exit from the village, the canalisation system delivers untreated sewage into its water.

The economy of the village is based on cattle breeding for meat and milk production, subsidiary agriculture, small businesses and local trade. The only school of the district is situated in the centre of the village and receives pupils from the rural and urban areas. The school enrolment rate is above 90%, representing the Brazilian average. Basic medical support is provided by a team of the Community Health Assistance Program (PACS), consisting of five health technicians, based in a little health facility. A medical doctor, together with a fully trained nurse, attends patients of the district each Monday and Wednesday. In the cases of specific medical examinations, treatment or hospitalisation, patients are taken to Governador Valadares, which is 30 km from the village and can be reached by driving 10 km on a dirt road and 20 km on an asphalt street.

This study area was selected because it is situated in an area at the low range of medium prevalence for schistosomiasis, where the community never received previous chemotherapy treatment through the National Schistosomiasis Control Program (SCP) and the village's population was adequate in size for the study purpose and characterised by a low rate of migration.

During a census carried out in cooperation with the local health authorities (PACS), 1,448 residents living in 404 households were registered. Non-permanent residents (183 individuals) and 204 residents of the rural area of the district were excluded from this study, resulting in a total population of 1,061 participants investigated.

Parasitological survey - A cross-sectional parasitological survey was carried out in all households. A single stool sample of each household member was examined according to the Kato-Katz thick smear technique using two slides for each sample (Katz et al. 1972). The slides were prepared in a field laboratory in Chonim de Cima by a well trained technician of the regional health agency. After that, two experienced laboratory technicians of the Laboratory of Schistosomiasis performed the microscopic examination in the René Rachou Research Institute (CPqRR). Approximately 10% of all examined slides were subjected to a control system for diagnostic efficacy. The results for soil-transmitted helminths (STHs) were expressed qualitatively and those for *S. mansoni* were expressed quantitatively to obtain the number of eggs per gram (epg). For each individual, the arithmetic mean epg of faeces based on the two samples was calculated. For analysis on group and population levels, the intensity of infection was calculated as the geometric mean of the egg counts of positive individuals.

Treatment - All participants positive for schistosomiasis were treated with praziquantel according to the recommendation of the Brazilian Ministry of Health (adults, 50 mg/kg in a single oral dose, children ≤ 15 years old, 60 mg/kg in a single oral dose). Those positive for STHs were treated with albendazole (adults and children, 400 mg in a single oral dose) in the local health facility.

Malacological survey - A senior technician from the regional centre of disease control (Regional Section of Decentralized Health Assistance), trained by the CPqRR and with large experience in snail collection, carried out the survey in the presence of one member of the research team. Snails were collected from the brooks crossing the village and swamps, little ponds, irrigation ponds and fish reservoirs within the urban area. Additionally, two sites used by the local population for leisure activities were examined for the presence of molluscs. The collection process was performed with perforated metal dippers fixed on a wooden stick and wooden tweezers (Thiengo 1995).

After capture, the molluscs were packed in boxes especially designed for transport and taken to the laboratory. Those alive were measured and examined in small plastic flasks with de-chlorinated water under artificial light in order to verify the presence of *S. mansoni* cercariae. Five snails from each collection site were identified based on comparative morphology of the reproductive organs and shell as described by Deslandes (1951) and Paraense (1975) and submitted to molecular analysis according to Vidigal et al. (2000). The other snails were submitted to smashing between glass plates for verification of the presence of sporocysts and cercariae.

Demographic, socio-economic, water contact variables and data collection - Different sets of variables, which have shown association with transmission and infection in other study areas, were chosen to evaluate their relation to the disease in our setting (Bethony et al. 2004, Massara et al. 2004, Gazzinelli et al. 2006). In order to investigate these different sets of variables, a questionnaire based on a model developed by Firmo (1994) during research activities in the region of Gorduras, Belo Horizonte, MG and adapted to the local conditions of this study area was used.

The following four sets of variables were investigated in this study: (i) a demographic set, including name, age, gender and family's origin, (ii) a socio-economic set, consisting of information about water supply, presence of electrical energy, household possessions (car, bicycle, television set, radio, refrigerator), regime of ownership (proper or rent), total household income (less or more than 2 minimum wages), education (none, 1st grade, 2nd grade and more than 2nd grade) and occupation (children and school children, manual and rural worker, employed in service sector, housewife, housekeeper, retired person and pensioner and unemployed), (iii) a set of information about sanitation devices, including sewage disposal (canalisation, septic pit, rudimentary pit and direct disposal into the brook) and infrastructure of toilet (presence of toilet and type of floor) and (iv) a set about water contact patterns categorised into leisure activities (swimming and fishing), horticultural activities, fetching water, occupational exposure to untreated water and crossing natural water collections, which means water contact with the bare feet when crossing brooks or swamps.

A team of four trained interviewers, all from the CPqRR, was in charge to carry out the survey. Prior to the survey, they were trained to conduct the interviews by using personal digital assistants. They visited all house-

holds. After signing a consent form for each resident of the household, a representative, in most cases the head of the household, was interviewed about general conditions and possessions of the residence. Individual data, including water contact pattern, were obtained by questioning each member of the household. In the case of children with less than seven years of age, this information was gathered by asking the household representative who answered the questionnaire about the residence.

Data analysis - The Chi-square test was applied to compare proportions between groups. The odds ratio was used to determine the strength of association of results from stool examinations between groups of the population. The relationship between infection with *S. mansoni* and demographic, socio-economic and water contact variables was explored with univariate logistic regression. Considering all variables revealing a p value < 0.20 and sequentially removing the variable with the highest p value until all variables showed a p value < 0.005 was the adapted procedure of the multivariate logistic regression to develop the final model. All tests were carried out with a 95% level of confidence. The software package Minitab 14.0 was used to carry out these calculations.

Ethical considerations - All participants were informed about the purpose of this study, the possible risks and inconveniences. The study protocol was received and approved by the Ethical Review Board of the CPqRR (04/2005). The study was carried out between 2004-2006.

RESULTS

Population - The study population of 1,061 individuals consisted of 597 (56.3%) females and 464 (43.7%) males living in 351 residences. There was a significant difference between the proportion of female and male populations (Chi-square test: 16.672, p value = 0.000). The age of this population ranged from 0-96 years, revealing a mean age of 27 years (23-30 years, CI 95%) and a median age of 32 years (30.6-33.4 years, CI 95%). The extreme age groups, less than 10 years and more than 60 years, represented 19.2% and 17.2%, respectively, of the entire population. More than 40% of the population was younger than 20 years. The detailed figures of the population correlating age and gender are shown in Table I.

A total of 336 households, representing 95.7% of the population, had access to electrical energy and 299 households, representing 86.1% of the population, received treated water. Regarding the issue of sewage disposal, 188 households, representing 53.3% of the population, were connected to the public sewage system, 99 households, or 29.2% of the population, used some sort of pit and the rest of the households, or 17.5% of the population, drained their sewage directly into the brook.

Parasitological survey - The parasitological survey of all 1,061 participants revealed a total of 119 individuals infected with *S. mansoni*, indicating a prevalence of 11.2%. Males, with a proportion of 18.1% schistosomiasis positive, showed a 3.1-times higher infection rate than females, with a proportion of 5.9%. The overall geometric mean egg count was 61.1 (63.6 for females and 60.1 for males). As shown in Fig. 1, the highest infection rates occurred in the age group from 10-30 years and as shown in Table II, the highest epg were found in the age group of children up to 10 years.

The results of prevalence of STHs were equal to or greater than 2%, except for hookworm infections, which reached a level of 10.4% (Table III). A proportion of 1.8% of the population revealed co-infection with two or more STHs and 2% showed co-infection with schistosomiasis and one or more STH.

Malacological survey - Out of the 30 collection sites in the urban area, 491 snails of the genus *Biomphalaria* were found at 22 (73.3%) locations. At both sites used for recreation and leisure activities outside of the village, 27 snails were captured. Of 391 live molluscs, 235 (60.1%) were identified as *Biomphalaria straminea*, 152 (38.9%) as *Biomphalaria glabrata* and four (1%) as *Biomphalaria tenagophila*. Remarkably, each snail species seemed to prefer a specific habitat. All specimens of *B. straminea* were found in brooks, those of *B. glabrata* only in little

TABLE I

The population of Chonim de Cima, Governador Valadares, Minas Gerais, Brazil, by age and gender

	Females		Males		Total	
Age group	n	%	n	%	n	%
0-10	102	17.1	102	22	204	19.2
10-20	142	23.8	116	25	258	24.3
20-30	59	9.9	36	7.8	95	9
30-40	68	11.4	39	8.4	107	10.1
40-50	66	11.1	47	10.1	113	10.7
50-60	64	10.7	38	8.2	102	9.6
> 60	96	16.1	86	18.5	182	17.2
Total	597	56.3	464	43.7	1,061	100

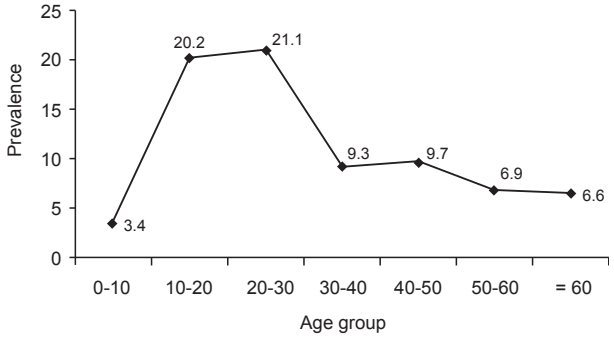

Fig. 1: relation between age group and proportion of schistosomiasis positives among the population of Chonim de Cima, Governador Valadares, Minas Gerais, Brazil (n = 1,061).

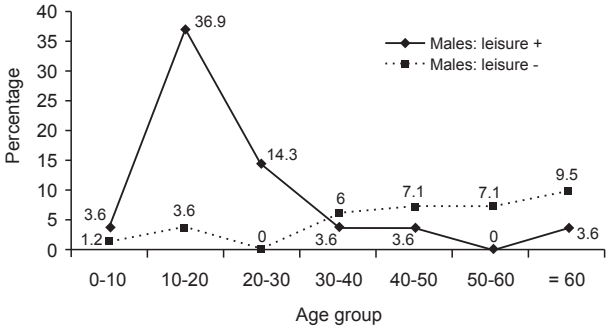

Fig. 2: relation between age groups, schistosomiasis positive males practicing leisure activities (males: leisure +) and schistosomiasis positive males not practicing leisure activities (males: leisure -) among the population of Chonim de Cima, Governador Valadares, Minas Gerais, Brazil.

TABLE II

Number of schistosomiasis positives and negatives, prevalence and egg count per gram (epg) in relation to gender and age group among the population of Chonim de Cima, Governador Valadares, Minas Gerais, Brazil (n = 1,061)

	Results				
Variable	Negatives n	Positives n	Total	%	epg
Gender					
Male	380	84	464	18.1	60.1
Female	562	35	597	5.9	63.6
Age group					
0-10	197	7	204	3.4	160.1
10-20	206	52	258	20.2	65.1
20-30	75	20	95	21.1	59.5
30-40	97	10	107	9.3	72.2
40-50	102	11	113	9.7	80.3
50-60	95	7	102	6.9	20
≥ 60	170	12	182	6.6	35.7
Total	942	119	1,061	11.2	61.1

ponds and water reservoirs and all *B. tenagophila* snails were captured in a single pond, which was used for fishing. A total of 376 (96.2%) live molluscs were examined for infection with *S. mansoni* and two snails, or 0.53%, proved positive, both for the species *B. glabrata*. Considering only *B. glabrata* snails in the evaluation, the infection rate increased to 1.6% among this species.

Demographic, socio-economic and water contact variables - A univariate logistic regression was conducted in order to identify a significant relationship between infection with *S. mansoni* and potential risk factors represented by demographic, socio-economic and water contact variables.

Among the demographic variables considered, a significant relation was found between infection and gender, as well as age group. Males showed a 3.39-times (95% CI = 2.2-5.3) higher risk for infection than females. The most notable results were found in the age groups between 10-19 years and 20-30 years, in which the risk of infection was 7.1 (95% CI = 3.2-16.0) and 7.5-times higher (95% CI = 3.0-18.5) than in the control, with an age between 0-10 years. The family's origin was also associated with infection, revealing that families with origin outside of the study area were at lower risk.

The socio-economic variables related to infection with schistosomiasis are shown in Table IV. The rest of this set of variables, including water supply, presence of electrical energy, household possessions, regime of ownership and household income, revealed no statistically significant relationship with infection.

TABLE III

Prevalence of soil transmitted helminths among the population of Chonim de Cima, Governador Valadares, Minas Gerais, Brazil (n = 1,061)

Type of soil transmitted helminth	Positives n	Negatives n	%
Ascaris lumbricoides	17	1,044	1.6
Hookworm	110	951	10.4
Enterobius vermicularis	21	1,040	2
Trichuris trichiura	0	1,061	0
Taenia sp.	19	1,042	1.8
Hymenolepis nana	7	1,054	0.7

TABLE IV

Risk and 95% confidence intervals (CI) for schistosomiasis infection of individuals (n) according to socio-economic variables among the population of Chonim de Cima, Governador Valadares, Minas Gerais, Brazil (n = 1,061)

Variable	n	p value	Risk	95% CI	
Education		**0.002**			
Non	270	-	1.0	-	-
1st grade	644	**0.003**	2.4	1.3	4.2
2nd grade	131	**0.000**	3.8	1.9	7.6
More than 2nd grade	16	0.907	1.1	0.1	9.2
Occupation		**0.000**			
Children and schoolchildren	376	-	1.0	-	-
Manual and rural worker	140	**0.000**	3.9	2.3	6.5
Service sector	94	0.576	1.2	0.6	2.6
Housewife, housekeeper, retired and handicapped	371	0.139	0.7	0.4	1.1
Unemployed	79	**0.004**	2.6	1.4	5.1

The variables connected with the household's sanitation revealed no relationship with infection in the case of sewage disposal (p value = 0.25) and presence of toilet (p value = 0.42). The infrastructure of the toilet, evaluated by the type of floor, presented a significant association with the disease. The risk decreased according to the increase of the infrastructure's quality, showing values of 0.4 (95% CI = 0.2-0.8) for ceramic floors and 0.5 (95% CI = 0.3-1.0) for concrete floors compared to floors without improvement.

The evaluation of the water contact pattern of the population in relation to infection is shown in Table V. The strongest association was found for the variable of leisure activities, showing a 3.5-fold increased risk (95% CI = 2.4-5.2) for those individuals practicing these activities.

Table VI shows the final multivariable model for schistosomiasis infection. According to the results of this model, males were at higher risk than females and the age groups between 10-30 years showed the highest values of association with the infection. Members of the community who practiced leisure activities involving water contact with natural water sources were also at higher risk than those without these activities. Further analysis of the schistosomiasis-positive male population in relation to age group and leisure activities showed that 51.2% of this population was between 10-30 years old and practiced leisure activities. Fig. 2 provides more detailed information about these distributions.

DISCUSSION

Bio-ecological data obtained in the present study reveal that the district of Chonim de Cima provides adequate environmental conditions for the vector reproduction and parasite survival expressed by schistosomiasis-positive individuals, as well as by infected snails. Considering the fact that the population never received treatment through the SCP raises the question why the infection rate among the residents of this village is not higher. Based on data from the socio-economic survey, the most probable answer is that measures related to social improvement, especially a safe water supply (86.1% of the population), indicate a protective and preventive

TABLE V

Risk and 95% confidence intervals (CI) for schistosomiasis infection according to water contact variables among the population of Chonim de Cima, Governador Valadares, Minas Gerais, Brazil (n = 1,061)

Variable	n	p value	Risk	95% CI	
Leisure activities		**0.000**			
No	718	-	1.0	-	-
Yes	343	-	3.5	2.4	5.2
Watering garden		0.999			
No	1,051	-	1.0	-	-
Yes	10	-	0	0	0
Fetching water		0.695			
No	1,028	-	1.0	-	-
Yes	33	-	0.8	0.2	2.6
Occupational exposure to untreated water		0.158			
No	1,023	-	1.0	-	-
Yes	38	-	1.8	0.8	4.3
Crossing natural water sources		**0.016**			
No	937	-	1.0	-	-
Yes	124	-	1.9	1.1	3.1

effect on infection with schistosomiasis. This assumption is confirmed by two studies recently carried out in other endemic areas in MG (Castro 2009, Vasconcelos et al. 2009). In our study, a difference in the distribution of infection according to gender was found. Females were at lower risk because their pattern of water contact involves, due to their more domestic activities, mainly safe water sources. No relation of infection with schistosomiasis and the occupation category, which includes this section of the population (housewife, housekeeper, handicapped and retired persons), could be found.

Furthermore, the occupation category of manual and rural worker, mainly males, showed a strong relationship to infection, due to contact with unsafe water. On the other hand, the effect of the protective factor - safe water - is hampered by the lack of another important social achievement: the insufficient sanitation system. According to our data, 70.8% of all households deliver untreated sewage into the main brook of the village, significantly facilitating the maintenance of the transmission of schistosomiasis in the area. The natural water resources, which are used for leisure purposes, become contaminated and an important source of infection in this way. As described in the final multivariable model, leisure activities represent an important risk factor to acquire infection with schistosomiasis. Putting this finding into the broader context of an integrated schistosomiasis control program, the importance of social improvements, which include the majority of the population, becomes evident. The current strategy, which focuses mainly on the treatment of the positives, has a paramount role on the individual level without any doubt. However, on the community level, the option of treatment without concurrent social improvements, especially in the water and sanitation sector, seems to be less effective, efficient and in the long term, less sustainable, because re-infection occurs after treatment as long as the environment remains contaminated with the parasite (Coura-Filho et al. 1996).

This study also intended to identify subsets of the population that are more vulnerable to infection with schistosomiasis. According to the final model, males in the age group between 10-30 years practicing leisure activities related to contact with unsafe water show the highest accumulative risk of infection. Although vulnerability for infection depends on environmental, demographic and socio-economic factors specific for each endemic area, our data follow, in some aspects a pattern that was found in other study areas. In particular, the age group of 10-30 years represents such a subset of the population, which seems to be common (Lima e Costa 1983, Gazzinelli et al. 2006).

This information may be useful for the planning of more differentiated control programs that consider an approach involving target groups in areas of low or medium prevalence. Considering our findings, a proposal for an integrated control program may be based on two pillars: one, horizontal, which involves social empowerment and health education and another one more vertical, which delivers treatment and infrastructural improvements.

In conclusion, this study shows that the factors related to the transmission of and infection with schistosomiasis cannot be understood only as a consequence of a variety of demographical, socio-economical and biological variables, which differ among study areas, but must also be seen in a framework in which different and complex biological and social systems interact and influence each other in a permanent search for homeostasis.

TABLE VI

Multivariable model of the association between demographic, socio-economic and water contact variables and infection with schistosomiasis among the population of Chonim de Cima, Governador Valadares, Minas Gerais, Brazil (n = 1,061)

Variable	n	p value	Risk	95% CI	
Gender		**0.000**			
Female	597	-	1.00	-	-
Male	464	-	**3.39**	2.17	5.28
Age group		**0.000**			
0-10	236	-	1.00	-	-
10-20	235	0.000	6.50	3.05	13.86
20-30	96	0.000	7.54	3.20	17.76
30-40	110	0.020	3.08	1.20	7.96
40-50	110	0.013	3.26	1.29	8.24
50-60	104	0.210	1.99	0.68	5.87
≥ 60	170	0.073	2.29	0.93	5.67
Leisure activity (swimming and fishing)		**0.003**			
No	718	-	1.00	-	-
Yes	343	-	1.96	-	3.06
Total	1,061	-	-	-	-

Finally, it is worth mentioning here that data of another study, which analysed in depth the proportion of schistosomiasis positives among a representative sample of the same population, revealed a significant variation (Enk et al. 2008). According to the methodology applied in this study, the prevalence showed levels of 11.4% for two Kato-Katz slides of a single stool sample. In another study, the prevalence found was 35.4% for 10 Kato-Katz slides of three stool samples combined with stool examinations carried out according to the formol ether centrifugation technique (Blagg et al. 1955).

This evidence indicates that prevalence is roughly underestimated, because approximately two-thirds of the positives are not detected if the routine sampling effort is applied. This finding strongly suggests that the association of the above evaluated variables with schistosomiasis has to be seen with caution whenever one or two slides of a single stool sample are used to define prevalence. That also means that in our case, a comparison of the demographic and socio-economic data with the higher and lower prevalence values might bring a deeper insight into this issue. Nevertheless, this evidence shows that the real situation of infection with schistosomiasis in areas considered of low and medium prevalence requires more attention and a more sophisticated approach in order to obtain reliable information.

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