

Intestinal Schistosomiasis and the Associated Transmission Factors in Pre-School Aged Children in Villages Surrounding Lake Rweru in Bugesera District, Rwanda

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Abstract

Background: Schistosomiasis is a waterborne snail-transmitted parasitic disease and is a major public health problem in sub-Saharan Africa, affecting populations living where water supply and sanitation are poor and inadequate. Pre-school aged children (PSAC) have often been ignored in disease control and prevention programs, as a result very little information is available on the prevalence or intensity of schistosomiasis in this age group.

Objective: This study was conducted to determine the prevalence and intensity of intestinal schistosomiasis among PSAC, and determine factors associated with infection transmission in PSAC in villages surrounding Lake Rweru in Bugesera District, Rwanda.

Methods: This was a cross-sectional study involving 256 PSAC aged 12-59 months. Double Kato-Katz (KK) stool smears and the point of care circulating cathodic antigen (CCA) test were used to diagnose *S. mansoni* in stool and urine samples, respectively. A questionnaire in the local language (Kinyarwanda) was administered to parents/guardians of the PSAC to determine factors associated with schistosomiasis transmission in the area.

Results: Based on the KK stool smears, no *S. mansoni* infection was found in any of the PSAC identified. However, using the point of care CCA test, 16.9% of the PSAC tested positive, when trace was considered as positive. The information collected on questionnaire showed a significant association between infection with *S. mansoni* (based on the CCA test) and visits to the lake especially, when the children accompanied their parents/guardians, or older children ($P < 0.000$).

Conclusion: The prevalence of *S. mansoni* in the study population was low on CCA test and the infection was associated with going to the lake by the children when they accompanied their parents/guardians or older siblings. The children who go to the lake should prevent direct contact with lake's water. These results call for the need to consider including a point of care screening tool for *S. mansoni* infections among preschool children as well as a prevention program targeting this age group in the study area.

Keywords: Intestinal schistosomiasis prevalence, Pre-school children, Rwanda

Background

Schistosomiasis is a waterborne snail-transmitted parasitic disease and a major public health problem in sub-Saharan Africa, mostly, affecting populations living in low-resource countries where, water supply and sanitation are poor and inadequate (Chitsulo *et al.*, 2000). It is estimated that more than 200 million people worldwide, mostly in tropical and subtropical countries are infected (Gryseels *et al.*, 2006; King *et al.*, 2005; Sady *et al.*, 2013). It is difficult to accurately estimate the disease burden due to schistosomiasis, but recent work suggests that a loss of 3-70 million disability-adjusted life years (DALYs) is attributed to the disease (King *et al.*, 2005). Significant reduction of infection and morbidity has been achieved in some countries of Africa through global efforts to eliminate schistosomiasis and other neglected tropical diseases (NTDs) by deworming (Kabatereine *et al.*, 2007).

In Rwanda several studies have been done on intestinal schistosomiasis especially in school children, and the prevalence in the country ranges from 0 to 69.5%, but no cases of urinary schistosomiasis have been reported so far (Rwanda Ministry of Health, 2008; Mupfasoni *et al.*,

2009; Ruberanziza *et al.*, 2015).

For many years, it was generally believed that schistosomiasis prevalence and intensity curves show peaks in school aged children (SAC) aged 6-15 years, and this age group was often used in providing an index for assessing community infection prevalence. Accordingly, epidemiological studies and control interventions on schistosomiasis have frequently directed towards school-aged children by regular administration of Praziquantel (Kabatereine *et al.*, 2007).

Because of lower frequency of active water contact of pre-school-aged children (PSAC) [< 6 years] compared to school aged children (SAC) and the development of an acquired protective immunity against schistosomiasis in adolescents and adults (≥ 16 years), PSAC have often been ignored in disease control and prevention programs (Verani *et al.*, 2011). As a result very little information is available on the prevalence or intensity of schistosomiasis in infants and PSAC in much of the endemic areas of the world including Rwanda. There is emerging evidence that, however, infants and PSAC are also infected in moderate to high schistosomiasis transmission areas. (Odogwu *et al.*, 2006; Opara *et al.*, 2007).

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The detection of *S. mansoni* eggs in stool samples using Kato-Katz (KK) smears by microscopy is the most commonly used method, and its benefits include high specificity, low cost, and simplicity. However, the sensitivity of this method is low particularly in areas of low endemicity, and low-infection intensities (e.g. in young children), and may be affected by day to day variability in the rate of egg excretion (Barreto *et al.*, 1978; Shane *et al.*, 2011; Teesdale *et al.*, 1985).

Immunodiagnosis, on the other hand, is generally more sensitive than examination of stool samples, particularly in low transmission areas where infection intensities are light (Shane *et al.*, 2011). Studies done in Uganda and Kenya to assess the Circulating Cathodic Antigen (CCA) urine dipstick test and the Point of Care-Circulating Cathodic Antigen (POC-CCA) cassette test in PSAC, recommended these rapid tests as useful tools for the detection of *S. mansoni* in that age group (Sousa-Figueiredo *et al.*, 2013; Verani *et al.*, 2011). In one study conducted in Ivory Coast where *S. mansoni* is common among school-aged children (SAC), a single POC-CCA cassette test was shown to have similar sensitivity as triplicate KK thick smears (Coulibaly *et al.*, 2011).

However, many studies have shown that day-to-day fluctuations occur in *S. mansoni* CCA test scores and in egg counts in KK smears, and therefore, it is recommended that a collection of several samples from individuals is necessary on consecutive days to increase the sensitivity of these methods (Coulibaly *et al.*, 2013; Degarege *et al.*, 2014; Sousa-Figueiredo *et al.*, 2013).

But this may have some limitations like cost and obtaining stool specimens from individuals on different days which would be challenging (Knopp *et al.*, 2008), so it is easier to use double KK smears for single stool sample and even a single urine-CCA cassette test would be enough for mapping and screening of *S. mansoni* infection at a reasonable cost (Degarege *et al.*, 2014).

As the degree of exposure and intensity of infection increases, it is necessary to document the prevalence and intensity of infection in PSAC in schistosomiasis endemic areas so that they can benefit from large scale disease control programs.

The aim of this study was to determine the prevalence, intensity of *S. mansoni* infection and the factors associated with intestinal schistosomiasis transmission in pre-school-aged children in areas surrounding Lake Rweru in Bugesera district using both KK as specific test and POC-CCA cassette test as sensitive test.

Method and materials

Study area

The Lake Rweru is located in the South-East of Rwanda (Rweru sector in Bugesera district), at an altitude of 1350 m. The total surface area of Lake Rweru covers some 100 km², of which 20% is in Rwanda and the remaining lie in Burundi. The mean depth is around 2.1 m with a maximum of 3.9 m

(Ruberanziza *et al.*, 2010). Compared to other regions of the country, Bugesera's climate is dry with a temperature varying between 20°C and 30°C with an average ranging between 26 and 29°C. The district has two dry periods and two rainy periods; a short dry season (January to mid-March), long rainy season (mid-March to June), long dry season (mid-June to September), a short rainy season (mid-October to December). This climate influences agriculture in Bugesera. About water and sanitation in this district, a big proportion still use unprotected water sources like rivers and lakes (27%) and 7.9% households don't have toilet facilities (Republic of Rwanda, 2012).

Study design and population

This was a cross sectional study of 256 randomly selected PSAC aged 12-59 months from 10 villages surrounding Lake Rweru in Bugesera district, Rwanda. These PSAC had been living in the study area at least 3 months before the beginning of the study.

Data collection

Questionnaire

A structured questionnaire was administered to the enrolled children's parents/guardians, to collect the socio-demographic data of the parents/guardians of study participant, hygiene, sanitation, and lake's water contact of study participant

Laboratory procedure

Single urine and stool samples were collected from enrolled children. CCA tests (Rapid Medical Diagnostics; Pretoria, South Africa) were conducted according to the manufacturer's instructions on single urine samples. The CCA results were read after 20 minutes after adding the buffer solution. CCA results were graded as negative, trace, 1+, 2+, and 3+. For stool samples double KK thick smears were prepared for each sample by using template which can hold 41.7g of stool, the smears were left 24 hours for clearing before examination for *S. mansoni* eggs. KK and slides were examined at Nzangwa Health center for Schistosoma and STH eggs by light microscopy. A and B smears were read by different technicians. After the survey, a 10% of slides were randomly selected and sent to National reference laboratory division/ Rwanda Biomedical Center for control.

Data analysis

Double data entry were performed in Microsoft Excel 2010 spreadsheet, and checked for entry errors. The STATA version 11 (Stata Corp.; College Station, Texas) was used in analysis. The mean and standard deviation of age variable were described and categorized in age groups: 12-24, 25-36, 37-48, and 49-59 months. Binary variables were compared using the Chi-square test (χ^2) or the Fisher exact test, where appropriate. Socio-demographic, hygiene, sanitation, and lake's water contact of study participant to the Schistosomiasis were assessed using logistic regression.

Ethical consideration

The study was approved by Rwanda National Ethic Committee (RNEC) No.331/RNEC/2015 and Malaria & OPDD-RBC. Before starting the study, meetings were

held at village's level with parents whom have pre-school aged children to describe the purpose of the study, the procedures to be followed, and the risks and benefits to the participation. Written informed consent was obtained from the parents/guardians of children before their enrolment into the study. The study has minimal risks to the participants, because the samples (stool and urine) that were used are obtained in normal life processes. Praziquantel and albendazole at the recommended doses were administered to the infected children under the supervision of a qualified clinician. Information obtained on the questionnaire was treated in confidential way and the records were stored in a locked cabinet and data were entered in a password protected file in a computer.

Results

Demographic characteristics of the study children

A total of 251 PSAC living in 10 villages on the shoreline of lake Rweru aged 12 to 59 months with a mean of age 35.3± 13 months (95% CI=12, 13.8) participated in this study, the compliance was 98% and 96.8% for stool and urine submission respectively. Among these children 51.8% were males and 48.2% were females. Participants were distributed in villages as following: 19.5%, 17.1%, 12.7%, 11.2%, 9.2%, 7.6%, 7.6%, 6.4%, 5.2% and 3.6% for Ruzo, Kigina, Mugina, Sharita, Rusenyi, Nyiragiseke, Karizinge, Mujwiri, Gasasa, and Rukira respectively. Table 1 and table 2 show the general characteristics of participants

Table 1. Demographic characteristics of the 251 study pre-school children from villages surrounding Lake Rweru, Bugesera district in Rwanda

Age groups (months)	Children Gender				Mean	S.D
	Female		Male			
	Number	Percentage	Number	Percentage		
12-24	32	12.7%	32	12.7%		
25-36	34	13.5%	33	13.1%		
37-48	30	12.0%	39	15.5%		
49-59	25	10.0%	26	10.4%		
Total	121	48.2%	130	51.8%	35.37	13

Table 2. Distribution of the pre-school children enrolled into the study and categorized according to sex and village

Villages	Sex		Total	%
	Female	Male		
	No	No	No	%
Kigina	24 (55.8%)	19 (44.2)	43	17.1
Mugina	17 (53.1%)	15 (46.9%)	32	12.7
Mujwiri	11 (68.8%)	5 (31.2%)	16	6.4
Nyiragiseke	6 (31.6%)	13 (68.4%)	19	7.6
Ruzo	19 (38.8%)	30 (61.2%)	49	19.5
Sharita	14 (50%)	14 (50%)	28	11.2
Karizinge	7 (36.8%)	12 (63.2%)	19	7.6
Rukira	3 (33.3%)	6 (66.7%)	9	3.6
Gasasa	8 (61.5%)	5 (38.5%)	13	5.2
Rusenyi	12 (52.2%)	11 (47.8%)	23	9.2
Total	121(48.2%)	130(51.8%)	251	100.0

Intestinal schistosomiasis caused by *S. mansoni*

In a total of 251 pre-school aged children examined based on KK technique, none was found to be positive for *S. mansoni*.

Prevalence of *S. mansoni* based on CCA cassette test

The prevalence of *S. mansoni* based on CCA cassette test considering trace as positive (CCA t+), 42/248, 16.9% (95% CI: 12.6-21.8) of participants were infected with *S. mansoni*. According to the sex, the prevalence in male children was 19.4% and in the female children the prevalence was 14.3%, this difference was not statistically significance ($X^2=1.14, P=0.28$), according to age group the prevalence of *S. mansoni* was 19.4%, 19.7%, 13%, and 15.7% for 12-24, 25-36, 37-48, and 49-59 months respectively, the difference was not statistically significant (Fisher's exact test=1.45, P= 0.72). When we consider the trace as negative (CCA t-), the prevalence of *S. mansoni* was 7/248, 2.8% (95% CI: 0.8-4.8). The prevalence in male children was 3.1% while in female children was 2.5%, the difference was not statistically significance ($X^2=0.76, P=1$). According to age group of PSAC, the prevalence was 0.0%, 3.0%, 4.3%, and 3.9% for 12-24, 25-36, 37-48, and 49-59 months respectively, the difference was not statistically significant (Fisher's exact test=2.89, P= 0.5). Figure 1 shows the prevalence of *S. mansoni* infection based on CCA cassette test in general whereas, Table 3 shows infection prevalence according to gender and age groups.

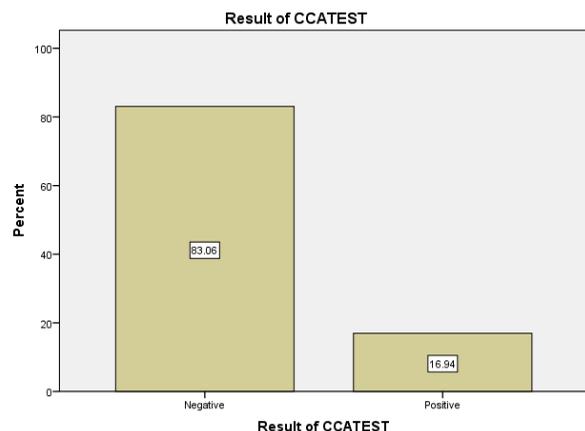


Figure 1. prevalence of *S. mansoni* based on CCA cassette test in PSAC living in villages surrounding Lake Rweru (trace considered as Positive)

Table 3. Prevalence of *S. mansoni* based on CCA cassette test according to sex and age group of PSAC in villages surrounding Lake Rweru

	CCA test results		Total	X^2	P-value
	Negative	Positive			
Sex					
Male	104 (80.4%)	25 (19.4%)	129	1.14	0.28
Female	102 (85.7%)	17 (14.3%)	119		
Age group					
12-24	50 (80.6%)	12 (19.4%)	62	1.45	0.72
25-36	53 (80.3%)	13 (19.7%)	66		
37-48	60 (87%)	9 (13%)	69		
49-59	43 (84.3%)	8 (15.7%)	51		

Sanitation and lake's water contact habits

94.8% of surveyed children were living in household with the presence of toilet. 62.5% of children were defecating in the toilet while 35.1% were defecating around the house. 88.4% of the children had parents/guardian who use water from the lake as first choice of water use at home while only 4.4% use tap water, 81.3% of the study children use water from lake as first choice of water bathing. Approximately, 97% of the parents/guardians of PSAC reported that their children got frequently into contact with lake water. 79% of children were in contact with water from lake when bathing while 13.5% had contact when accompanying adults. Sanitation and lake's water contact habit is presented in table 4.

Factors associated with *S. mansoni* infection in PSAC

The association between *S. mansoni* infection and sanitation and lake's water contact habit by the children is presented in Table 5.

Although not significant ($p > 0.05$), Children living in household without toilet had roughly 2 times the odd to have *S. mansoni* infection compared to children who had toilet (OR 2.16(0.46-10.12)). Even though not significant ($p > 0.05$), children defecating around the house had 1.5 odds (OR 1.59 (0.69-3.61)) of having *S. mansoni* infection compared to those defecating in the toilet ($p > 0.05$). Although not significant ($p > 0.05$), children who had contact with lake had nearly 5 times the odds (OR 4.8 (0.87-27.16)) to have *S. mansoni* infection compared to those who did not have contact with lake ($p > 0.05$).

Accompanying parents and older children by the PSAC to the lake was significantly associated with *S. mansoni* infection with 10 times the odd than other children ($p < 0.000$) as presented in Table 5.

Table 4. Sanitation and lake's water contact habit of studied PSAC

	Frequency	95% CI
Presence of toilet		
Yes	238 (94.8%)	91.6-97.5
No	13 (5.2%)	2.5-8.4
Areas where the child defecates		
Toilet	157 (62.5%)	56.7-68.8
Around house	88 (35.1%)	29.2-41.0
In bushes	6 (2.4%)	0.4-4.8
First choice of the source of water used at home		
Tap water	11 (4.4%)	2.1-7.2
Stream	10 (4%)	2.0-6.8
Pond	8 (3.2%)	1.2-5.6
Lake	222 (88.4)	84.2-92.0
First choice of the source of water used to bath child		
Tap water	15 (6%)	3.2-9.0
Stream	21 (8.4%)	4.8-12.0
Pond	11 (4.4%)	2.0-7.0
Lake	204 (81.3%)	76.9-85.7
Contact with lake water		
Yes	244(97.2%)	95.2-98.8
No	7 (2.8%)	1.2-4.8
How		
At home (bathing with lake water)	195 (79.9%)	74.6-84.7
Accompany parent/older children to the lake	33 (13.5%)	9.4-18.6
Goes him/herself to lake	16 (6.6%)	3.7-10.1

Table 5. The factors associated with the intestinal schistosomiasis in PSAC in villages surrounding Lake Rweru, Bugesera district

Characteristic	CCA test result		Univariate		P-value
	Negative	positive	Total	OR(95%CI)	
Toilet ownership?					
Yes	199(84.7)	36(15.3)	235		<0.328
No	7(53.8)	6(46.2)	13	2.16(0.46-10.12)	
Child defecates					
In toilet	134(85.9)	22(14.1)	156		<0.467
Around the house	67(77.9)	19(22.1)	86	1.59(0.69-3.61)	
In the bushes	5(83.3)	1(16.7)	6	0.66(0.03-11.5)	
Home using water					
Tap water	9(81.8)	2(18.2)	11		<0.616
Stream	7(70)	3(30)	10	6.06(0.29-122.4)	
Pond	5(62.5)	3(37.5)	8	7.24(0.35-148.5)	
Lake	185(84.5)	34(15.5)	219	2.46(0.17-33.9)	
Water used to bath child					
Tap water	10(66.7)	5(33.3)	15		<0.604
Stream	20(95.2)	1(4.8)	21	0.13(0.009-1.81)	
Pond	9(81.8)	2(18.2)	11	0.89(0.09-8.18)	
Lake	167(83.1)	34(16.9)	201	0.67(0.14-3.15)	
Child contact with lake's water					
Yes	202(84.6)	39(15.4)	241		<0.065
No	4(57.1)	3(42.9)	7	4.86(0.87-27.16)	
How					
Accompany parent/					
older children to the lake	15(45.5)	18(54.5)	33		<0.000
Use lake's water					
to bath children	172(89.6)	20(10.4)	192	0.11(0.04-0.27)	
Child goes to the lake					
her/himself	15(93.8)	1(6.2)	16	0.06(0.007-0.56)	
Total	207(83.1)	42(16.9)	248(100)		

Discussion

The aim of this study was to determine the prevalence and intensity of *S. mansoni* infection in pre-school aged children (PSAC) in an area located on the shores of Lake Rweru in Rwanda Eastern province, and considered endemic for intestinal schistosomiasis, and where infection intensity is categorized as moderate, and determine the factors associated with infection transmission among the PSAC. Intestinal schistosomiasis caused by *S. mansoni* is endemic in Rwanda, especially around the lakes Ruhondo, Burera, Rweru, Kivu and on Nkombo Island (Hanotier and Gigase 1981; Mupfasoni *et al.*, 2009; Ruberanziza *et al.*, 2010; Ruberanziza *et al.*, 2015), with data being available for school age children. Until now, virtually no information was available on the prevalence of *S. mansoni* among the PSAC. The present study conducted around Lake Rweru located in Rwanda Eastern province was a first attempt to obtain data on *S. mansoni* prevalence for this age group in Rwanda.

The fact that in the present study no *S. mansoni* infection was detected among pre-school aged children in villages surrounding Lake Rweru in Bugesera district, Rwanda using the Kato-Katz procedure, and the fact that only about 17% of the PSAC tested positive with the urine CCA test when trace results were considered as positive, suggest that the prevalence of *S. mansoni* among this age group,

in this particular area, may be too low to be of public health significance. It should be understood, however, that in this particular geographic area, *S. mansoni* among the school age children is within the moderate intensity range. It is recommended that data for this age group from other intestinal schistosomiasis endemic areas within Rwanda will be necessary, if the epidemiology of schistosomiasis and the pre-schoolage group is to be fully understood in this country.

Between the microscopy-based Kato-Katz procedure and the immune-diagnostic-based urine circulating cathodic antigen (POC- CCA) assay, it appears the latter is a more sensitive test, in terms of being able to detect even low level *S. mansoni* infection. With respect to sensitivity of the tests, the results from the present study are similar to those obtained from other studies done elsewhere, in which the two tests were also, compared (Adriko *et al.*, 2014; Colley *et al.*, 2013; Degarege *et al.*, 2014; Mwinzi *et al.*, 2015).

They do suggest the value of the urine CCA test in epidemiological studies of *S. mansoni*, particularly where infection prevalence is relatively low. In addition, the CCA-positive results where Kato-Katz results were *S. mansoni* egg-negative among the PSAC, could also, suggest the presence of pre-patent immature infections (where the parasites are not at the stage of laying eggs). Alternatively, the eggs laid by the schistosomes in the infected children were below the detection levels of the Kato-Katz procedure (Adriko *et al.*, 2014; Polman *et al.*, 2000). It is also, possible that we were dealing with residual infections after chemotherapeutic interventions, which normally reduce the prevalence and increase in the number of infections of low intensity (Coulibaly *et al.*, 2012; Knopp *et al.*, 2011). Schistosomiasis control programs have been undertaken in Rweru area since 2010 (Rwanda Ministry of Health, 2010). The low prevalence of *S. mansoni* infection observed in the Rweru area, Rwanda among the PSAC in the current study differs from the results of similar studies conducted elsewhere, within the sub-Saharan region, and where prevalence with the Kato-Katz procedure was in the range 7-44% and with the CCA test, was 40-80% (Coulibaly *et al.*, 2013; Odogwu *et al.*, 2006; Ruganuzza *et al.*, 2015).

It is noted that majority of the study children in Rweru did not have direct contact with lake water, but contacted lake water at home during bathing. Schistosome cercariae present water are known to lose their infectivity with time, (Adenowo *et al.*, 2015). Interestingly, there was a significant association between infection with *S. mansoni* and going to the lake by the PSAC especially when the children accompany their parents/guardians or older children to the lake. The similar findings have been reported in western Kenya and in Tanzania (Handzel *et al.*, 2003; Ruganuzza *et al.*, 2015).

The present findings have a geographic limitation, cannot be generalized to the all endemic area in the country. This

study didn't provide the malacology information in the area because of study design.

In conclusion the findings of this study have demonstrated that the prevalence of intestinal schistosomiasis in pre-school aged children in village surrounding lake Rweru on Kato- Katz thick smears was 0%, but 16.9% on the Point Of Care- Circulating Cathodic Antigen (POC-CCA) test, which indicates some level of the risk of *S. mansoni* infection among this age group. The control program should think to consider including them in Praziquantel mass drug administration campaigns. The results showed a significant association between infection with *S. mansoni* and going to the lake by the PSAC which indicates that the water may be contaminated with cercariae. Young children often accompany their parents/guardians or older children to the lakeshore.

We recommend assessing burden of schistosomiasis among this age group in other settings, to target this PSAC in national prevention and control program and the authority to provide safe and clean water to the population in this area. As there is insufficient information on *S. mansoni* infection among pre-school aged children in endemic area around Rwanda, further studies are needed to determine the public health significance of schistosomiasis in these areas.

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Conflict of interests

The authors declare that they have no competing interests

Authors' contributions

EN, GMM, AKM, KNN were involved in the study design. EN carried out data collection, analysis and manuscript development. GMM, AKM, KNN, IU, CK critically reviewed the manuscript and results interpretation. All authors read and approved the final manuscript.

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