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ORIGINAL ARTICLE

Surveys for Schistosomiasis and Soil Transmitted Helminths in Luangwa, Kalabo and Serenje Districts of Zambia

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ABSTRACT

***Background**:* Earlier publications have indicated that human infections with schistosomes and soil transmitted helminths (STH) are widespread in Zambia. Despite being classified as a moderate endemic country for these infections, and a recent modelling study suggesting an average schistosomiasis prevalence of 26% among school aged children, there is a lack of published data on the present occurrence and distribution of these infections.

Design: Cross sectional surveys for schistosomiasis and STH were carried out among individuals aged 4-20 years at three sites in Luangwa, Kalabo and Serenje districts, as an added component to comprehensive studies on lymphatic filariasis. Urine samples were examined for hematuria and Schistosoma haematobium ova using the Nuclepore filtration technique. Stool samples were examined for S. mansoni and STH ova using the Kato-Katz technique.

Results: The prevalence of S. haematobium was generally low, with the Serenje site recording the highest of 3.0%. S. mansoni was only seen at the Kalabo site, which had a moderate prevalence of 37.5%. Hookworm infections were relatively common at all three sites, with prevalences ranging from 12.1 to 35.0%. Other more rare STH infections noted were Ascaris lumbricoides, Hymenolepis nana and Enterobius vermicularis.

Conclusions: The study confirmed that schistosomiasis and STH infections were endemic at the study sites, but also suggested that the prevalences had declined compared to earlier reports. There is need for more surveys to be carried out to assess the current distribution and prevalence of these infections to provide guidance for implementation of mass drug administration for their control.

INTRODUCTION

Studies carried out in the past have shown that schistosomiasis is endemic in most parts of Zambia, with prevalences as high as 70% [1-5]. In 2000, it was estimated that schistosomiasis affected 2.4 million individuals in Zambia and that approximately 9 million were at risk of acquiring infection [6]. Occurrence of dual infection with *S. haematobium* and *S. mansoni* is common although the overall prevalence of *S. haematobium* appears to be higher than that of *S. mansoni* [1,2,5,7]. High prevalences have been observed in both rural and urban settings [1,2,3,7]. However, it is likely that these numbers have now changed, partly because of population increase and partly because mass drug administration (MDA) has been implemented among school children in some districts. More recent modelling

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studies, using observations from available field surveys in combination with climate and other environmental data, have estimated the average prevalence for schistosomiasis among school aged children to be 26% [8] and thus has classified Zambia as a moderately endemic country [9]. Prevalence varies from place to place, from many areas with low endemicity to a few high endemicity areas especially in the northern and eastern parts of Zambia [8].

Soil transmitted helminths (STH), especially hookworms, are also well known to be endemic in Zambia. Surveys have shown the prevalence of hookworm infection varying between 11% and 77% across the country [10,11]. A few studies have moreover shown that *Ascaris lumbricoides* and *Trichuris trichiura* infections are present in some districts [3,12-14].

There is a general lack of recent published information about schistosomiasis and STH in the various districts and provinces around Zambia. Increased awareness of the harmful effects of these and other Neglected Tropical Diseases (NTDs), - especially on the growth and development of children, - and the current global mobilization for their control [9, 15, 16], has highlighted the need for more knowledge on their occurrence and prevalence in the country. The surveys on schistosomiasis and STH infections presented here were undertaken as an added component to a larger research study on the epidemiology of lymphatic filariasis (LF) in three districts of Zambia [17,18].

METHODS

Study sites

The surveys were carried out in three districts namely Luangwa, Kalabo and Serenje. The study sites were originally selected for detailed studies on the epidemiology of LF (17, 18), as earlier mapping surveys had indicated high LF prevalences at these sites (19). The surveys described here were thus carried out as an added component to the LF studies. Luangwa District is located in Lusaka Province in the eastern part of the country at the confluence of the Zambezi and Luangwa rivers and comprises of plateau areas and valleys with altitudes below 600 m. The study sites were two neighbouring villages (Janeiro and Yapite) located about 35 km North-West of Luangwa town, and samples were collected in May 2011. Kalabo District is situated in the Zambezi floodplains in Western Province and is mainly a flat plateau with an altitude of about 1000 m. The study site was Liumba Hill Mission about 25 km West of Kalabo town, and samples were collected in October 2011. Serenje District is located in Central Province and is partly peri-urban and partly rural due to its central location between the two larger cities of Mansa and Kasama. The study sites were two neighbouring areas namely Muchinka and Mwimbula sections about 60 km North-East of Serenje town, and samples were collected in February 2012.

Zambia has 3 main seasons; the cold season from May to August (temperature range 6-30ºC), the hot season from September to October (17-35ºC) and the rainy season from November to April (8-35ºC).

Study design

The study comprised of cross sectional surveys for schistosomiasis and STH. Communities were sensitized to the study with the help of the teachers and community health workers. After examination of all community members according to the LF study protocol [17, 18], individuals aged 4-20 years were requested to provide a urine and stool sample for the present study. Efforts were made to screen 100 young individuals for schistosomiasis and STHs at each site. However, collection of urine and stool samples was challenging as the children and youngsters were generally reluctant to submit the specimens because they felt uncomfortable. This also resulted in a lower proportion of children being examined in Kalabo compared to the other sites.

Each individual was provided with two containers, for urine and stool respectively. The stool containers had spatulas attached to the lids, which were used by the individuals to scoop the faeces. The individuals were requested to bring back the stool and urine samples immediately. Samples collected were stored in a cool box and transported to a nearby district hospital or health centre for laboratory examination.

Laboratory examinations

Urine samples were first examined for visible haematuria and then tested for micro-haematuria by using reagent strips (Hemastix, ACON Laboratories Inc., San Diego, USA). The Nuclepore filtration technique was used for examination for *S. haematobium* eggs [15]. The samples were examined on the day of collection to avoid crystals forming. Briefly, 10 ml of urine was drawn into a syringe after careful mixing and pressed down vertically through a filter holder with a Nuclepore filter (12.0 µm pore size). The syringe was then detached from the filter holder, air was drawn into the syringe and the expulsion process was repeated. The filter was placed on a microscope slide top side up. A drop of iodine was added for easy visibility of the eggs, and after 15 seconds the filter was examined under a microscope for *S. haematobium* eggs. The number of eggs were counted and expressed as eggs per 10 ml of urine.

The Kato-Katz technique [15] was used for examination of stool samples. Briefly, coverslips made from cellophane strips were soaked in 50% glycerine-malachite solution for at least 24 hours before use. A smooth faecal sample sieved through nylon mesh was transferred to the well of a template (41.7g) on a microscope slide. After removing the template, the faecal sample was covered with the pre-soaked cellophane coverslip, spread out evenly under the coverslip and examined under a microscope for *S. mansoni* and other helminths eggs. Slides were prepared in duplicate and examined immediately to prevent hookworm eggs from clearing. The mean egg count of the two slides was calculated and the eggs per gram of faeces (epg) thereafter calculated by multiplying the mean egg count with 24 [15].

Data analysis

Data collected from field and laboratory (study location, and name, age, gender, and infection status and intensity for study individuals) were entered in Excel for cleaning and storage and exported to STATA (version 12) for analysis. Chi-square (chi^2) test was used to compare categorical variables and Student's t-test was used for continuous variables. *P*-values less than 0.05 were considered statistically significant.

Ethical Considerations

Ethical approval for the study was provided by the University of Zambia Research and Ethics committee. Permissions were also provided by the Ministry of Health, the Provincial and District Health offices, and the local area chiefs and village headmen. Oral consent was obtained from parents/guardians of children willing to participate in the survey. Children unwilling to take part were not included. Children found positive for STHs or schistosomiasis were referred to the local health centre for treatment with albendazole or praziquantel as appropriate.

RESULTS

S. haematobium

Of the 103 individuals examined in Luangwa District, none had haematuria and no *S. haematobium* eggs were seen in the urine (table 1). In Kalabo District, S. *haematobium* infection was identified in one boy aged 15 years (3 eggs/10ml of urine) out of the 69 examined, thus giving an overall prevalence of 1.4% (table 1). In addition, one boy aged 13 years had micro-haematuria, but no *S. haematobium* eggs were seen in his urine.

Table 1: Examination of urine samples for *S. haematobium* eggs.

District	No. examined	Mean age in years (range)	Female : male ratio	No. positive (%)
Luangwa	103	9.0 (4-16)	0.77	0 (0.0)
Kalabo	69	12.0 (4-18)	1.34	1 (1.4)
Serenje	100	10.9 (7-16)	1.13	3 (3.0)

In Serenje, *S. haematobium* infection was identified in 3 individuals (all from Muchinka section) out of the 100 examined, thus giving an overall prevalence of 3.0% (table 1). Among those with the infection, 2 were girls aged 11 years (5 eggs/10ml urine) and 13 years (3eggs/10ml urine) and one was a boy aged 13 years (5eggs/10ml urine). One girl aged 9 years had micro-haematuria but no eggs in the urine.

S. mansoni

Of the 255 individuals examined for intestinal helminths from the three districts (91 from Luangwa, 64 from Kalabo, 100 from Serenje), only those from Kalabo had S. *mansoni* infection (table 2). 24 individuals from this site were found with *S. mansoni* eggs, thus giving a prevalence of 37.5%. The prevalence was higher in boys than in girls but the difference was not statistically significant (46.7 % vs. 29.4%; chi^2 test $p > 0.05$). Eleven of these individuals found with *S. mansoni* infection also had hookworm infection. Among the eleven, one boy also had *S. haematobium* infection while another had micro-haematuria. The geometric mean intensity (GMI) of *S. mansoni* infection was 4.74 epg when calculated on the basis of all examined individuals and 104.8 epg when calculated on the basis of the positives only. Among the 10 *S. mansoni* positive girls and 14 *S. mansoni* positive boys, the GMI was higher in the girls than in the boys but this difference was not statistically significant (140.1 vs. 85.1 epg; t-test, $p > 0.05$).

Table 2: Examination of stool samples for helminth eggs.

District	No. examined	Mean age in years (range)	Female : male ratio	*S. mansoni*		Hookworms	
				No. positive (%)	GMI** among positives	No. positive (%)	GMI** among positives
Luangwa	91	9.0 (4-16)	0.77	0 (0.0)	-	11 (12.1)	199.2
Kalabo	64	12.0 (4-18)	1.34	24 (37.5)	104.8	22 (34.4)	110.2
Serenje	100*	10.9 (7-16)	1.13	0 (0.0)	-	35 (35.0)	224.4

*) 1 child had *Ascaris lumbricoides* eggs, 1 had *Hymenolepis nana* eggs, 1 had *Enterobius vermicularis* eggs.
**) Geometric mean intensity, in eggs/gram faeces

STH

Hookworm infection was present at all three study sites (table 2). Eleven individuals were found with hookworm in Luangwa (prevalence of 12.1%). Six of these were females and five were males, but the prevalence was not statistically significantly different between genders (14.3% vs. 10.2%; chi^{2}test $p > 0.05$). Statistical analysis also indicated no significant differences in the GMIs between the males and females (t-test, $p > 0.05$).

In Kalabo, 22 individuals had hookworms (prevalence of 34.4%). The prevalence was slightly higher in girls than boys but the difference was not statistically significant (38.2 % vs. 30.0%; chi^{2}test $p > 0.05$). However among the 13 girls and 9 boys with hookworms, the GMI was significantly higher in boys than in the girls (181.1 vs. 78.0 epg; t-test, $p = 0.04$).

In Serenje, 35 individuals were found with hookworms (prevalence of 35.0%). The prevalence was higher in boys than in girls but the difference was not statistically significant (40.4 % vs. 30.2%; chi^{2}test, $p > 0.05$). Among the 16 girls and 19 boys with hookworms, the GMI was slightly but not significantly higher in girls than in boys, (225.5 vs. 223.4 epg; t-test, $p > 0.05$). In addition to the hookworm infections, 3 girls from Serenje all aged 12 years were found with *Ascaris lumbricoides, Hymenolepis nana* and *Enterobius vermicularis* infection, respectively. The 2 girls with *H. nana* and *E. vermicularis* infection also had hookworm infection.

DISCUSSION

Although high prevalences for *S. haematobium* have been reported in some parts of Zambia [2, 3, 20], no cases of urinary schistosomiasis were detected among the examined children in Luangwa District. This finding is similar to that of a study carried out prior to the current study in the same community, where only one case of *S. haematobium* was recorded among the 104 children that were examined [21]. Low prevalence of *S. haematobium* was also observed in Kalabo (1.4%) and Serenje (3.0%). The observed low prevalences of *S. haematobium* in Luangwa, Kalabo and Serenje districts suggests that these infections are rare, perhaps due to ongoing deworming activities and accompanying health education and/or due to unfavourable environmental conditions for the parasites. Most schools in rural Zambia are part of the School Health and Nutrition program (SHN) which promotes school feeding activities and deworming of the children with support from the Ministry of Health. Despite the low prevalences recorded during the surveys, it is still possible that there could be areas with moderate or high endemicity within the three districts because of the focal distribution of schistosomiasis.

No cases of *S. mansoni* infection were recorded in Luangwa and Serenje. However the Kalabo site had a moderate prevalence of *S. mansoni* infection according to WHO grading [9]. The prevalence of *S. mansoni* in Kalabo was 37.5%, and was therefore higher than the national average prevalence estimated at 15-25% [8, 22]. Recent studies carried out in neighbouring districts within Western Province have also shown high prevalences of *S. mansoni* [23-25]. This suggests that *S. mansoni* generally is a major public health problem in the province and measures should be put in place to address the situation.

The prevalence of hookworm infection was moderate among the examined individuals at the three study sites, but the mean infection intensities were low, being less than 2000 epg [9]. This is probably a reflection of the ongoing deworming programmes, and thus indicates that these have a positive effect particularly on infection burdens. Continued yearly treatment of school aged children should hopefully further reduce the number of eggs being expelled and subsequently the prevalence. In a recent study in Kafue District, hookworm infections were positively associated with *A. lumbricoides* infections in children below the age of 8 years [13]. However, only one case of *A. lumbricoides* infection was found during this study (in Serenje District). Perhaps the absence of other STHs except for hookworm in the study populations could be a result of the community engaging in good hygiene practises. However, because of economic hardships, some individuals are unable to afford to wear shoes constantly and are therefore exposed to hookworm infection. Previous studies indicated that hookworm infection was common (11% -77% prevalence range) in Central Province [10, 11] but documentation on the current prevalence of hookworm in the country is limited. Though hookworm was the most common STH found in the three study populations, few cases of *A. lumbricoides, H. nana* and *E. vermicularis* infections were detected in Serenje District.

The low prevalence of schistosomiasis and STHs observed in Luangwa District could be due to increased knowledge on how to prevent these infections as a result of health talks given by the community health care workers. For over five years, deworming of schoolchildren with albendazole or mebendazole (when albendazole is not available) has been carried out twice a year by the local health centre and the school teachers. It is important that these local deworming activities are considered and properly integrated when planning and implementing further MDA activities.

Helminth infections have been shown to affect the growth and development of children. It has also been observed that infection with helminths affects school attendance, as children with infections are more likely to be absent from school [9]. Control of helminth infections can be achieved through health education, improved sanitary conditions and deworming. Deworming is safe, effective and cheap and has been shown to improve growth and development of children [26, 27]. However, although some communities are proactive in carrying out deworming activities as observed in Luangwa District, other communities are reluctant as was noted in Kalabo District. Despite the school in Liumba Hill being part of the SHN programme and located barely 500 m from the health centre (well stocked with albendazole and praziquantel), deworming of the school-children was not carried out. The lack of compliance to treatment programs even when drugs and health workers are available emphasizes the importance of community participation, so that even in the absence of a focal person, the communities can encourage and advocate the continuity of these activities.

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