

Full Length Research Paper

Reassessment of endemicity level of urinary schistosomiasis in the Kotto-Barombi focus (South West Cameroon) and impact of mass drug administration (MDA) on the parasitic indices

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The level of infestation and mechanism of persistence of urinary schistosomiasis was determined in two villages (Kotto Barombi and Marumba II) by conducting a survey on school children from May 2007 to May 2008. Urine samples were collected from 418 children and examined using filtration technique. All participants were subsequently treated by Praziquantel. Drug efficacy and incidence rate were assessed 3 months and 1 year later respectively. The initial prevalence (50.8%) of *S. haematobium* was 69.5% (Kotto Barombi) and 41.3% (Marumba II). The prevalence differed significantly between the villages ($P = 0.004$). The mean parasite load of 211.3 (Kotto Barombi) and 39.62 (Marumba II) eggs/10 ml of urine differed significantly ($P = 0.0001$) and between the quarters (Mainland and Island) in Kotto Barombi ($P = 0.007$). Praziquantel was highly efficacious on schistosome worms, with a global cure rate (CR) and egg reduction rate (ERR) of 97.2 and 99.2% respectively. These values were 100% in Kotto Barombi, 88.1% (CR) and 79.8% (ERR) in Marumba II. The overall incidence rate was 13.5%: Kotto Barombi (13.5%), Marumba II (13.7%). These results suggest that the initial prevalence (50.8%) may be reached soon, if the WHO recommendations (appropriate health education and snails' control) are not effectively implemented.

Key words: Urinary schistosomiasis, *S. haematobium*, prevalence, parasite load, drug efficacy, cure rate, egg reduction rate, incidence rate, Kotto Barombi, Marumba II.

INTRODUCTION

Schistosomiasis is a parasitic disease caused by blood-flukes of the genus *Schistosoma* (Trematoda, Schistosomatidae). Transmission occurs through direct contact with infective larvae emitted by freshwater Gastropods. Three forms of human schistosomiasis (intestinal, urinary and rectal) occur in Africa, caused by the species *Schistosoma mansoni*, *S. haematobium* and *S. intercalatum*, respectively. It is estimated that 779 million people are at risk with about 207 million people infected worldwide (WHO, 1999; Chitsulo et al., 2000; Fenwick et al., 2006;

Steinmann et al., 2006; Martyn et al., 2007; Zhang et al., 2007). Schistosomiasis is responsible for 200,000 deaths per year in Sub-Saharan Africa (Savioli et al., 2004). Despite this fact, the control of this disease has not previously been given priority in some foci in many countries. Therefore, the true burden of this parasitic infection seems to be underestimated and then needs to be reassessed (Zhang et al., 2007).

In Cameroon, 3 forms of Schistosomiasis occur, but the foci are very unequally distributed. *S. mansoni* and *S. haematobium* are more prevalent in the Northern part of the country than in the Southern part, while *S. intercalatum* is found only in 2 regions of the country (Littoral and Centre). *S. mansoni* and *S. intercalatum* are

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transmitted by *Biomphalaria pfeifferi* and *Bulinus forskalii* respectively, while *S. haematobium* is transmitted by *Bulinus truncatus*, *B. senegalensis*, *B. globosus* and *B. camerunensis*, with *B. truncatus* being its primary intermediate host (Greer et al., 1990).

Urinary schistosomiasis foci in the Southwest province are concentrated in the Meme Division (Ratard et al., 1990). The main transmission sites are 2 crater lakes, Kotto Barombi and Barombi Mbo where transmission is assured by *B. truncatus* and *B. camerunensis*. In the Kotto Barombi focus, a control program carried out from 1970 to 1975 by Duke and Moore (1976) considerably reduced the prevalence of schistosomiasis. Since 1975, studies conducted have shown repeated resurgence of the disease despite the treatment administered to patients after every survey (Moyou et al., 1987; Rattard et al., 1990; Ndamukong et al., 2000, 2001).

Cameroon now follows the WHO recommendations that lay great emphasis on regular treatments of high risk groups, particularly school children and appropriate health education, as the most appropriate means of reducing morbidity (WHO, 2001). This study was designed to assess the current level of endemicity of urinary schistosomiasis amongst school aged children of the Kotto-Barombi focus and to evaluate, 1 year after treatment, the impact of a mass chemotherapy campaign on the parasitic and disease indices (prevalence, intensity, incidence rate, Egg reduction rate, cure rate). These results will provide an essential background for focussed treatment and appropriate control strategies of the parasite among children and the entire community in the Kotto Barombi focus and nationwide.

MATERIALS AND METHODS

Study area

The study was conducted in 2 villages of a highly endemic area, the Kotto Barombi focus, located in the Mbonge subdivision (South West Cameroon) (Moyou et al., 1987; Ndamukog et al., 2001). The main transmission site of this focus is the crater lake of Kotto Barombi (altitude of 400 m), which harbours *B. truncatus* and *B. Camerunensis* as the main intermediate hosts of *S. haematobium* (Duke and Moore., 1976; Moyou et al., 1987). 2 small streams meet with the lake, one emptying into it (the inlet) and the other flowing out of it (outlet) which meets with other water bodies that lead to other villages. This focus belongs to the equatorial forest zone, Cameroon-type climate, with one long rainy season (9 months) and one short dry season (3 months). The total annual rain fall varies between 2000 – 4000 mm. (Greer et al., 1990; Rattard et al., 1990).

2 villages were prospected for the study.

i) Kotto Barombi village (4° 28' 4"N; 9° 15', 2"E) is made up of 2 quarters: the Island situated in the middle of the lake where part of the inhabitants live and the mainland (around the lake and along side the road leading to other villages) where the majority of the population lives. There is a total absence of water sources (wells, taps, forages) what makes most of human activities necessitating water (laundry, bathing and fishing) to be done only in the lake. Also, the population living in the island carries out their activities (Farming, marketing, schooling etc) in other villages after crossing

the lake.

ii) Marumba II (4° 34' 2.7"N; 9° 20' 27"E), situated at about 9 km from Kotto Barombi, is characterised by the presence of wells, so that the freshwater collections are visited only for particular purposes (swimming, laundry).

Sampling and analysis of urine

The study was done from May 2007 to May 2008 on pupils from Kotto Barombi and Marumba II villages. The traditional leaders, chief of district health centre, school directors and teachers and school children were informed 2 weeks before the survey. In each school, prior to the sample collection, the participants were briefed on the disease (causes, manifestations, consequences, diagnosis and prevention). Of the 523 pupils registered, 418 were successfully sampled. After registration urine sample was collected from each volunteer in a 10 ml plastic screw-cap vials, between 10 am and 2 pm. The samples were transported to the General Biology laboratory (Faculty of Science) of the University of Yaounde I. The urine was examined using the filtration technique (Plouvier et al., 1975) for the identification of *S. haematobium* eggs following their morphology (Sturrock, 1993). Each tube was then agitated to ensure adequate dispersal of *S. haematobium* eggs followed by filtration, using a polyamide Nytrell filter fixed to a 10 ml syringe. The filter was subsequently removed, put on a slide and stained with 1% Lugol solution. The slide was examined under a light microscope magnification X100. The eggs found were counted and their number in 10 ml of urine was estimated and recorded. Treatment was administered to all the participants 2 weeks after the collection of baseline data with a single dose of praziquantel (40 mg/kg of body weight). The National schistosomiasis control program supervised the drug distribution. A control was done 3 months after treatment, urine samples being collected only from children who were found positive before treatment.

The incidence rate was evaluated 1 year after treatment on children who were negative before treatment and during the control. The population sampled was between the age of 1 - 5, 6 - 10, 11 - 15 and above 15 years.

Data analysis

The following parameters were assessed:

1. The prevalence (P), defined as the percentage of infected individuals (NP) among the total number of individuals examined (N) ($P = (NP/N) \times 100$) (Hamit et al., 2008).
2. The cure rate (CR) which is the ratio of the difference between the number of children who were positive before treatment and those positive 3 months post-treatment on the number of positive cases before treatment, expressed as a percentage.
3. The parasite load which is the mean number of *S. haematobium* eggs per 10 ml of urine of each patient.
4. The egg reduction rate (ERR) which is the ratio of the difference between the average parasite load before and three months after treatment on the pre-treatment parasite load, expressed as a percentage (Cure rate, parasite load and egg reduction rate are used following Saathoff et al. (2004).
5. The incidence rate (IN) which is defined as the ratio of the number of new positive cases detected 1 year after treatment on the number of negative cases obtained before treatment and during the control phases expressed as a percentage (Chandiwana et al., 1987).

The Chi-square test was used to compare the prevalence in relation to sex, age groups, quarters and villages while 1 - way ANOVA or Kruskal-Wallis tests were used to compare the parasite load in

Table 1. Prevalence and parasitic load of urinary schistosomiasis among age groups.

Village	PE	Age groups			P value	
		1 – 5	6 – 10	11 – 15	> 15	
Kotto Barombi	P	75.9	64.7	72.4	74.4	0.9
	PL	263.7	213.0	197.0	187.4	0.84
	sd	597	367	327	160	
Marumba II	P	20.0	42.9	55.0	-	0.2
	PL	36.87	22.2	75.1	-	0.06
	sd	37	31	105		

P: prevalence in percentage, PL: parasitic load (number of eggs per 10 ml of urine) given with the standard deviation in italic, PE: parameter estimated, sd: standard deviation.

Table 2. Evolution of the prevalence of urinary schistosomiasis with the survey phases.

Villages	Survey phases	Number of children examined	Number infested (P)
Kotto Barombi	Before treatment	275	193 (69.5)
	3 months after treatment	132	0 (0.0)
	One year after treatment	193	26 (13.5)
Marumba II	Before treatment	143	59 (41.3)
	3 months after treatment	38	7 (18.4)
	One year after treatment	95	13(13.7)
Total	Before treatment	418	250 (50.8)
	3 months after treatment	170	7 (4.1)
	One year after treatment	288	39 (13.5)

P: prevalence in percentage.

relation to sex, age groups, quarters and villages. The Kruskal–Wallis test was used when the conditions of parametric ANOVA were not fulfilled (Sokal and Rohlf, 1981). The level of statistical significance was at 5% ($P \leq 0.5$).

RESULTS

Prevalence before treatment

A total of 418 school children were sampled (275 out of 380 expected in Kotto Barombi and 143 out of 171 expected in Marumba II), giving an overall participation rate of 75.9%. The sample included 201 (48%) boys and 217 (52%) girls. 250 children harboured *S. haematobium* eggs, giving an overall prevalence rate of 50.8% (Table 2). The proportion of infected children differed significantly ($P = 0.004$) between the 2 villages.

Out of the 127 boys and 148 girls examined in Kotto Barombi, 91 (71.7%) and 100 (67.6%) were positive respectively, while in Marumba II, 30 (40.5%) boys and 29 (42.0%) girls out of the 74 and 69 examined, harboured *S. haematobium* eggs respectively. These prevalence values were not significantly different between the sexes in Kotto Barombi ($P = 0.80$) and in Marumba II ($P = 0.90$).

Children of all age groups were infected, the difference observed was not significant in neither of the 2 villages; Kotto Barombi ($P = 0.9$) and Marumba II ($P = 0.2$) (Table 1).

In Kotto Barombi, 106 (60.9%) children out of 174 from the mainland and 86 (85.1%) out of 101 from the Island harboured *S. haematobium* eggs. These prevalence values were not significantly different between the 2 quarters ($P = 0.08$).

Parasitic load before treatment

The mean parasitic load was 211.30 and 39.62 eggs/10 ml of urine in Kotto Barombi and Marumba II respectively (Table 3). This parameter varied considerably among children in the two villages. The parasite load was significantly higher in Kotto Barombi than in Marumba II ($K = 21.62$; $P = 0.0001$).

In Kotto Barombi, the mean parasitic load was 213.9 eggs/10 ml of urine in males and 208.9 in females, while in Marumba II it was 40.39 and 38.83 eggs/10 ml of urine respectively in males and females. The difference observed among sex was not significant within each village ($K = 1.07$; $P = 0.30$ in Kotto Barombi and $F = 0.01$;

Table 3. Cure rate and Egg reduction Rate of *S. haematobium* infection three months after treatment. The number of positive cases is given with the mean parasitic load in the brackets.

Village	Number of positive Cases before treatment	Number of positive Cases 3 months after treatment	Cure rate (%)	ERR (%)
Kotto Barombi	191 (211.3)	0 (0.0)	100	100
Marumba II	59 (39.62)	7 (8.0)	88.1	79.8
Total	250 (172.18)	7 (8.0)	97.2	99.2

ERR: egg reduction rate.

P = 0.92 in Marumba II).

There was no significant difference of parasitic load observed among age groups in Kotto Barombi (K= 0.83; P = 0.9) as well as in Marumba II (K= 5.56; P = 0.84) (Table 1). In Kotto Barombi, the mean parasitic load in the mainland (132.2 eggs/10 ml of urine) was significantly (K= 11.58, P = 0.0007) lower than that of the Island (310.4 eggs/10 ml).

Infection rates 3 months after treatment

Among the 250 children who took part in the control 3 months after treatment, 170 came from Kotto Barombi and 38 from Marumba II. No child was found positive (0.0%) in Kotto Barombi giving a reduction rate of 100%, while 7 (18.4%) were positive in Marumba II with a reduction rate of 55.4% (Table 2). The global parasitic Cure Rate was 97.2%. The CR was 100% in Kotto Barombi, while in Marumba II, it was 88.1%. The global ERR was 99.2%. The ERR was 100% in Kotto Barombi and 79.8% in Marumba II (Table 3).

Infection rates one year after treatment

Of the 288 children tested one year post-treatment, 39 new positive cases were found, giving an incidence rate of 13.5%. The number of new positive cases was 26 children from Kotto Barombi and 13 children from Marumba II, giving an incidence rate of 13.5 and 13.7% for the 2 villages respectively (Table 2).

DISCUSSION

The parasitological studies revealed high prevalence and high density of urinary schistosomiasis in Kotto Barombi and Marumba II villages. Although the global prevalence (50.8%) was slightly lower than 76.0 and 75.9% obtained respectively by Moyou et al. (1987) and Ndamukong et al. (2001) in the same focus, it confirms the high endemicity of schistosomiasis in this focus. The differences among prevalence may be mainly due to the difference in the number of years spent since the last mass treatment among the 3 surveys. The highest preva-

lence and parasite load in Kotto Barombi compared to those of Marumba II and in the Island compared to those of the Mainland in Kotto Barombi, can be justified by frequent contacts with the infected sites. This finding agrees with those of Moyou et al. (1987) and Ndamukong et al. (2000; 2001), who found that people closer to the lake were the most infested. Similar results have also been obtained in KwaZulu-Natal/South Africa by Saathoff et al. (2004).

The factor sex seems to influence the level of infestation of children by schistosomiasis since the boys generally carry more activities in water collections than girls (Njiokou et al., 2004; Tchuem Tchuenté et al., 2003). However, in this particular biotope without other water sources, all children almost have the same frequency of contact with the lake. This makes the difference between males and females not statistically significant as previously observed by Moyou et al. (1987) and Ndamukong et al. (2001) in Kotto Barombi village and by Saathof et al. (2004) in KwaZulu-Natal/South Africa, and Tohon et al. (2008) in Nigeria.

The progressive increase of infestation rate and parasite load with age between 1 and 15 years in Marumba II may reflect the gradual augmentation of the activities in the infected water of that particular age group, as the presence of wells provide other water sources. This result is in accordance with the declarations of PNLSHI (2005) which placed the most vulnerable age group between 6 - 15 years.

The cure rate (97.2%) obtained 3 months after treatment was very close to 98% found by Tchuem Tchuenté et al. (2004) in Loum after 6 weeks and more than 92.6% obtained by Kihara et al. (2007) in Kenya. These values are high, confirming the high efficacy of praziquantel against schistosome worms in these foci. The relatively low values of Cure Rate and Egg Reduction Rate observed in Marumba II could reflect the drug distribution rather than a drug resistance. In Kotto Barombi, the drug was administered to each child by the representatives of the National control program of schistosomiasis who probably ensure that each child took the drug properly, since they are more experienced, while in Marumba II, drug distribution was done by a school director who may not have ensured that each child really took the drug. It could also be due to 'old' and mostly non infective eggs laid before treatment, that were

trapped somewhere in the tissue and were slowly finding their way through the lumen of the bladder (Saathoff et al., 2004). Unfortunately, our laboratory examination did not differentiate between viable and non-viable eggs. However, the results obtained 1 year after are consistent with the above explanation because very little change in prevalence is observed between 3 months and 1 year (18.4% versus 19.6%). This is similar to the results of Saathoff et al. (2004) who observed little change between 3 and 41 weeks with regards to infestation rate and parasitic load.

The incidence rate (13.5%) reached only 1 year after treatment was relatively high suggesting that the prevalence rate may evolve to the initial value just few years after the mass treatment. This mechanism may explain the outbreaks of urinary schistosomiasis (Moyou et al., 1987; Rattard et al., 1990; Ndamukong et al., 2000, 2001) in this focus the last decades. The new patients could have been infected by cercariae produced by the snails remaining in the water collections as the malacological control was not performed. A second source of parasite could be the patients that were absent during the treatment and who came back to the focus after this period, or the patients from the neighbouring villages that were not concerned by the medical survey.

Our study shows that prevalence and parasitic load of *S. haematobium* infection in the Kotto Barombi focus are still high, praziquantel is highly efficacious in reducing the parasite load, and a high pace of re-infection. These results suggest that WHO (2002) recommendations need to be implemented effectively (regular survey and treatment of patients, water sources supply, health education and snail control). Also, a more expanded study in the entire population within an extended period is necessary, which will permit the evaluation of morbidity and mortality rate of this parasitic infection.

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