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Urine Turbidity and Microhaematuria as Rapid Assessment Indicators for *Schistosoma haematobium* Infection among School Children in Endemic Areas

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Abstract: Problem statement: Urinary schistosomiasis is highly endemic in Nigeria and for effective control measure, an efficient, quick and yet cheap diagnosis should be integrated. This will ensure the proper management of infection due to *Schistosoma haematobium* in low resource communities of Nigeria. **Approach:** This cross-sectional study recruited a total of 456 (252 males, 204 females) school children aged 3-20 years between November 2010 and June 2011. Urine samples were examined macroscopically for turbidity and subsequently screened for microhaematuria using diagnostic reagent strips. The microscopic examination of urine samples for schistosome eggs was used as the standard for diagnosis. **Results:** The prevalence of *S. haematobium* and geometric mean intensity of infection were 54.8% and 13.9 ± 0.67 eggs/10 mL of urine respectively. The age and sex prevalence of urinary schistosomiasis showed no significant differences ($p>0.05$). The prevalences of urine turbidity and microhaematuria were 37.1 and 53.9% respectively and these varied significantly across age groups ($p<0.05$). The sensitivities of urine turbidity and microhaematuria used for the indirect diagnosis of urinary schistosomiasis were 54.8 and 59.3 ($p>0.05$) with their corresponding specificities 80.2 and 65.8% respectively. Intensity of infection was significantly correlated with the indirect diagnostic methods, urine turbidity ($r = 0.203$, $p<0.01$) and microhaematuria ($r = 0.487$, $p<0.01$). **Conclusion:** The possible use of urine turbidity as an indicator for rapid diagnosis of urinary schistosomiasis in low resource communities is implied.

Key words: Urinary schistosomiasis, urine turbidity, microhaematuria, prevalence, indirect diagnosis

INTRODUCTION

Schistosomiasis is an infection caused by a trematode parasite; a prevalent tropical disease, ranking second to malaria and posing a great public health and social economic threat in sub-Saharan Africa (Sangweme *et al*., 2010). In 2007, the World Health Organization estimated 235 million cases of schistosomiais worldwide, 732 million people at risk of infection in known transmission areas. Sub-saharan Africa accounts for 85% of the *Schistosoma* burden WHO, 2010. If these WHO values are adjusted for the probable 40-60% of missed diagnoses, the true number of active *Schistosoma* infections in 2007 was more likely to be between 391 and 587 million people worldwide (King, 2010). In Nigeria, urinary schistosomiasis is known to have existed from time immemorial and the low resource communities are mostly plaqued by the disease. With approximately 20% of the population of sub-Saharan Africa, Nigeria is or once was the most highly affected country in Africa for schistosomiasis (Njepuome *et al*., 2009). Young individuals are mostly infected with peak prevalence and intensity of infection in the age group 11-15 years (Biu *et al*., 2009; Sarkinfada *et al*., 2009). Reports have established that both natural (McManus *et al*., 2011) and artificial water bodies (Duwa and Oyeyi, 2009) are transmission foci of the parasite.

One of the problems faced in the control of schistosomiasis in Nigeria is the choice of rapid and yet efficient diagnostic method to cater for the ever increasing population of infected individuals. Urine microscopy for detection of *S. haematobium* ova is considered one of the most conclusive diagnostic criteria for urinary schistosomiasis. Unfortunately, this procedure is expensive, cumbersome, time consuming and too technical for lay use. This limitation therefore dictates the need for a simple, fast, cheap and reliable

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diagnostic method for the detection of infected persons. Screening using rapid using rapid, indirect tests have been proposed as a procedure to simplify mapping surveys (Brooker and Utzinger, 2007).

Indirect diagnosis of urinary schistosomiasis through the use of chemical reagent strips has been proven highly sensitive and specific in many endemic areas of the world (Robinson *et al*., 2009; Ugbomoiko *et al*., 2009a). Proteinuria and haematuria are recognized clinical features of *S. haematobium* infection and often used as indicators of infection in endemic areas. One limitation of haematuria reagent strips is that they can be affected by factors such as menstruation and genitourinary infections (Poggensee *et al*., 2000). A further shortcoming is that they are semi-quantitative and have a limited concentration detection range. Such limitations have prompted researchers to investigate alternative rapid diagnosis methods.

Several ongoing research efforts are aimed at developing quick indicators for assessing endemicity of urinary schistosomiasis in communities. Urinary turbidity is one of the macroscopic observations during investigation of urinary schistosomiasis in endemic regions of Africa, however, the validity and reliability of urinary turbidity as a screening criterion in hyper-endemic communities like Yewa North in Ogun State has not been fully elucidated. This study reports the assessment of specificity and sensitivity, both of which are indicators of validity in the use of urinary turbidity in screening for urinary schistosomiasis.

MATERIALS AND METHODS

Study area: The study was carried out in Yewa North Local Government Area (LGA), Ogun State, Nigeria. It lies between latitude $7° 15^1$ N and longitude $3^o 3^1$ E in a deciduous/derived savannah zone (Oyesiku and Jegede, 1992). It has a land size of about 200, 213-5 Km^2. The area is dominated largely by Yoruba speaking people. The Local Government has the following communities: Ayetoro, Ijale, Ketu, Ijoun, Isetu, Igan, Eggua, Igan Alade, Igan Okoto, Igbogila, Ijako, Ibese, Joga Orile, Imasayi and Iboro. It is bounded by Ewekoro Local Government Areas in the East, Imeko-Afon Local Government Area in the North, Ijebu-Ode and, Republic of Benin in the West and Yewa South local government area in the south. Trading, timber logging and farming are the main occupation of the inhabitants. There are 97 public primary schools, 19 public secondary schools and one Technical College. There are many flowing river bodies in the Local Government Area which serve as major sources of water supply. Human-water contact activities with rivers include bathing, swimming and laundry.

Ethical approval, informed consent and exclusion criteria: Approvals were granted by the University of Ibadan/University College Hospital Institutional Ethical Review Committee and the State Universal Basic Education Board of Ogun State, Nigeria. A pre-survey visit was made to the study area during which time, discussions were held with the Community Heads and school teachers who assisted in mobilizing the pupils for the study. Written informed consents were obtained from those willing to participate in the study. Those who do not reside in the area and those showing multiple signs of illness were excluded from the study.

Parasitological examination and indirect diagnoses: The cross-sectional survey which employed a systematic random sampling technique, using the class register was conducted between November 2010 and June 2011. Pupils were recruited from three Public Primary Schools; Community Primary School (CPS) Eggua, Ebenezer African Church Primary School (EAC) Ile-Ijoun and Yewa Central African School (YCAS) Igan-Alade. A total of 456 pupils were enrolled for the study with each given a clean, dry screw-capped universal bottle carrying the same identification number with the one on the record sheets. The freshly passed mid-day urine samples (collected between 10-14 h) were inspected macroscopically (for gross haematuria and visual opacity i.e., turbidity) and then screened for microhaematuria using commercially available urine reagent strips (UroColor^{TM}10, Manufactured by Standard Diagnostics, Inc., Korea). The strip testing was performed in accordance with the manufacturer's instructions. The specimens were then transported to the laboratory within 4 h of collection and processed for microscopic examination of schistosome eggs.

Data analysis: Data were entered into an Excel spreadsheet, checked for entry errors and transferred into Statistical Package for Social Science Windows (version 11.0, SPSS Inc, Chicago, USA) for analysis. Chi-square test was used to determine significant differences in the prevalence of infection. Pearson's correlation test was used to assess the potential statistical relationships between intensity of infection and morbidity indicators of urinary Schistosomiasis. Sensitivity (SS), Specificity (SP) Positive Predictive Value (PPV) and Negative Predictive Value (NPV) were calculated against the 'gold standard' of parasitological diagnosis of urinary schistosomiasis (microscopy).

RESULTS

Overall prevalence and intensity of infection of urinary schistosomiasis were 54.8% and 13.9±0.67 eggs/10 mL of urine respectively. The highest (44.2%) and the least prevalence (29.7%) of infection were observed in Community Primary School, Eggua and EAC Primary School, Ile-Ijoun respectively. Prevalence in male was higher than in female and also generally increased with age, however, the age group 9-14 years had the highest prevalence (62.9%) (Table1). Variations in prevalences of urinary schistosomiasis at the community level, sex and age of participants, showed no significant differences (p>0.05). The highest (15.9 eggs/10 mL urine) and lowest (13.3 eggs/10 mL urine) intensity of infection was recorded in Community Primary School, Eggua and EAC Primary School, Ile-Ijoun respectively. Variations in intensity of infection at the community level showed significant difference (p<0.05). Also intensity varied significantly in male (14.4 eggs/10 mL of urine) and female (14.1eggs/10 mL of urine) individuals (p<0.05) with the males showing relatively higher intensity. Intensity of infection increased with age, however, the age group 15-20 years had the highest mean egg loads (22.6 eggs/10 mL of urine).

The overall occurrences of Microhaematuria (MH) and Urine Turbidity (UT) were 53.9 and 37.1% respectively. School children in YCAS had the highest prevalence of MH (57.8%) and UT (41.4%) (Table 2). The occurrence of MH in school pupils varied significantly across the three communities (p<0.05) while variation in UT was not significant (p>0.05). The prevalence of the two morbidity indicators of urinary schistosomiasis used in the present study showed no significant difference in the male and female subjects (p>0.05). However, the prevalence of MH and UT were slightly higher in male subjects. The occurrence of MH and UT varied significantly across the age groups (p<0.05), with the age group 9-14 years having the highest prevalences 62.5 and 45.9% for MH and UT respectively (Table 2). Intensity of infection was significantly correlated with the indirect diagnostic methods, urine turbidity (r = 0.203, p<0.01) and microhaematuria (r = 0.487, p<0.01).

The sensitivities of MH (59.3%) and UT (54.8%) in diagnosing infection by *S. haematobium* were relatively low. Specificities of MH and UT were 65.8 and 80.2% respectively. The accuracy of MH (72.4%) as morbidity indicators of infection by *Schistosoma haematobium* was slightly higher than that of UT (69.1%) (Table 3).

Table 1: Prevalence of urinary schistosomiasis in schoolchildren in Yewa North LGA, Ogun State, Nigeria

	Number			
	Examined	Infected	Prevalence (%)	GMI (eggs/10 mL urine)
Schools				
YCAS	128	55	39	14.9
EAC	83	20	29.7	13.3
CPS	41	17	44.2	15.9
p value			p>0.05	p<0.05
Sex				
Male	252	139	55.2	14.4
Female	204	111	54.4	14.1
p value			p>0.05	p<0.05
Age groups				
8-Mar	154	67	43.5	10.3
14-Sep	224	141	62.9	13.8
15-20	78	42	53.8	22.6
p value			p>0.05	p>0.05

Table 2: Prevalence of microhaematuria and urine turbidity in schoolchildren in Yewa North LGA, Ogun State, Nigeria

	Microhaematuria		Urine turbidity	
	Number Positive (examined)	Prevalence	Number Positive (examined)	Prevalence
Schools				
YCAS	74(128)	57.8	53(128)	41.4
EAC	41(83)	49.4	26(83)	31.3
CPS	18(41)	43.9	16(41)	39.0
p value		p<0.05		p>0.05
Sex				
Male	136(252)	54	95(252)	37.7
Female	110(204)	53.9	74(204)	36.3
p value		p>0.05		p>0.05
Age groups				
3-8	63(154)	40.9	36(154)	23.4
9-14	140(224)	62.5	103(224)	45.9
15-20	43(78)	55.1	30(78)	38.4
p value		p<0.05		p<0.05

Table 3: Reliability and validity of diagnostic indicators for detection of urinary schistosomiasis

Diagnoses	Microhaematuria	Urine Turbidity	Microscopy
Sensitivity	59.3	54.8	100
Specificity	65.8	80.2	100
Positive predictive value	63.5	58.0	100
Negative predictive value	82.6	76.9	100
Accuracy	72.4	69.1	100

DISCUSSION

The prevalence of this disease like many other endemic diseases is affected by the socio-cultural characteristics of the area. The overall prevalence (54.8%) of schistosomiasis among school pupils in the three communities was similar to 50.8% reported in four schools of Ilobu and Erin-Osun communities in Osun State, Nigeria (Ugbomoiko *et al*., 2009a). The infection pattern in this study showed typical peak prevalence in the early adolescence with males having

higher prevalence of infection. The distribution of schistosomiasis among school pupils based on age and sex is similar to other reports on prevalence studies (Biu *et al*., 2009; Ansong *et al*., 2011). The differences observed in age and sex related prevalence and intensity of infection in present study could be due to changes in the extent of exposure to infective fresh water. The insignificant difference observed in prevalence amongst sexes agrees with the past studies conducted in Abeokuta North Local Government Area of Ogun State, Nigeria (Ekpo *et al*., 2010). The significant positive correlation observed between infection intensity and urine turbidity ($r = 0.203$, $p<0.01$) and microhaematuria ($r = 0.487$, $p<0.01$) confirmed they are good morbidity indicators of urinary schistosomiasis.

The rapid assessment indicators of morbidity used in this study, which are the visual observation for turbid urine and the diagnostic chemical reagent strip for microhaematuria, gave different results. Microhaematuria has been widely used as indicator for urinary schistosomiasis since 1980 and has been considered more sensitive and specific than other diagnostic indicators (Ugbomoiko *et al*., 2009b). However urinary turbidity is not often associated with urinary schistosomiasis but often used as diagnostic indicator of bacterial related or epithelial cells infection. The sensitivity of microhaematuria in the present study was lower than values reported in other studies both in Nigeria and other regions of the world (Ugbomoiko *et al*., 2009b). The discrepancy observed in the sensitivity value of microhaematuria in our study with other works could be due to regional differences and varying quality of reagent strips from different producers. The lower sensitivity of diagnostic indicators of urinary schistosomiasis could also be due to reduced intensity of infection. In population with high prevalence as observed in this study and corresponding high egg burden, a more reliable diagnostic performance is expected.

The higher sensitivity and positive predictive value of microhaematuria than that of urine turbidity showed its (microhaematuria) greater ability to produce a true positive result when used on an infected population. However, the probability that urine turbidity will produce a true negative result when used on a non infected population was higher because of its higher specificity. The combination of microhaematuria with urine turbidity can therefore be considered a cost-effective measure for increasing the quality of data on schistosomiasis infection in school children located in highly endemic communities.

The positive predictive value for microhaematuria (63.5%) and urine turbidity (58.0%) were rather low indicating that about 36 and 42% individuals not tested positive were indeed infected. However, since the predictive values depend on the prevalence of disease and intensity of infection, in higher endemic settings the positive predictive value may be different. The negative predictive values of 82.6 and 76.9% for microhaematuria and urine turbidity respectively indicate that the probability of school children having the disease, in the absence of the two indicators of morbidity, are about 17 and 23%.

This investigation is limited in scope because the survey only included school children and as such, the data cannot be inferred to the general population however, in most epidemiologic investigations, school settings are always targeted. It is known that the diagnostic performance of haematuria decreases with increasing age, but is usually stable in children and teenagers (Robinson *et al*., 2009). Nevertheless, combining proteinuria and microhaematuria obtained from reagent strips with the urine turbidity will probably increase the diagnostic accuracy when applied to the general population. Furthermore, the cross-sectional nature of this study did not allow for assessment of day-to-day variation of intensity of infection through egg excretion, haematuria and urine turbidity which may have influenced diagnostic performance. When the assessment of these is done repeatedly during a period of a few days, the accuracy of this indicator is expected to increase.

CONCLUSION

It is therefore concluded that in addition to the already established diagnostic indicators of urinary schistosomiasis, urine turbidity is also a reliable diagnostic indicator and can be combined with proteinuria and haematuria to further improve the diagnostic efficiency of schistosomiasis in low-resource endemic communities.

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