

Efficacy of Chemical Reagent Strip in the Diagnosis of Urinary Schistosomiasis in Ikota, Ifedore Local Government Area, Ondo State, Nigeria

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ABSTRACT

Urinary schistosomiasis is a parasitic disease caused by *Schistosoma haematobium* which affects the bladder and as such is a major source of morbidity and mortality in Africa and Sub-Saharan Africa. This study was conducted to determine the efficiency and efficacy of chemical reagent strip in the diagnosis of urinary schistosomiasis among pupils in Ikota, Ifedore Local Government Area of Ondo state. The pupils' data were obtained using pretested, well-structured questionnaire while the samples were analyzed in the laboratory using sedimentation method and chemical reagent dipstick (Combi-9) was used to examine the microhaematuria and proteinuria. Of the one hundred and fifty (150) individuals that were sampled, 76 (50.66%) pupils were males and 74 (49.33%) were females. The prevalence of infection of *S. haematobium* in the area was 36 (24.0%). There was no significant difference ($p > 0.05$) in prevalence relating to sex and age. Analysis on microhaematuria and proteinuria shows that 30 (20.0%) were positive for microhaematuria and 76 (50.67%) for proteinuria. The sensitivities of the urinary symptoms were 50.0% for proteinuria and 83.33% for microhaematuria and specificity was 62.3% for proteinuria and 100.00% for microhaematuria respectively. This chemical reagent strip combine with gold standard method of diagnosis will increase the accuracy and give good predictive value.

Keywords: Urinary schistosomiasis; *S. haematobium*; Microhaematuria; Proteinuria; Chemical reagent strip

INTRODUCTION

Urinary schistosomiasis is very prevalent in many parts of Nigeria, especially rural areas that lack access to portable water and good sewage system [1-3]. It is one of the Neglected Tropical diseases (NTDs) that are predominant in Africa, especially in population with low income [4]. It is also known as bilharziasis, a chronic parasitic disease that circulate in blood vessel and caused by one of the *Schistosoma* species namely *Schistosoma haematobium* which affects the bladder.

Diagnosis is very important in the management and control programs of diseases, that is why any method chosen for diagnosis must be sensitive enough to detect infected person. Over the years, a number of methods have been developed for rapid diagnostic assessment like visual examination for blood, interview, chemical reagent strip, urine microscopy and immunological method [5,6]. Although the more reliable way to diagnose urinary schistosomiasis is the detection of eggs in urine

by sedimentation or filtration and microscopy method but urinalysis dipstick is good for rapid diagnosis, epidemiological surveys where the endemicity of urinary schistosomiasis has already been established or to measure the impact of control program in endemic areas.

Many researchers have validated the use of chemical reagent strip for microhaematuria against egg detection by urine microscopy [7-9]. Short coming associated with chemical reagent strip is that it is not appropriate for light prevalence population and in a population containing pregnant or menstruating females [10,11]. Hence, the specific objective of this study is to assess the efficacy of chemical reagent strip in the diagnosis of urinary schistosomiasis in Ikota, Ifedore local Government Area, Ondo State, Nigeria.

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MATERIALS AND METHODS

Study area

This investigation was carried out in Ikota in Ifedore Local Government Area of Ondo State. Ikota is located between latitude 7° 21' 0" North and longitude 5° 9' 0" East with an average temperature of 27°C. Members of this community are farmers and traders of the Yoruba ethnic group. Passing through the community is a flowing stream which is a source of water in the community as the water bore-hole system in the community is not functioning very well.

Study subjects and collection of samples

The study subjects include school age pupils in Ikota. Permission was obtained from Zonal Education Department of the Local Government through the school authority and also from Ondo State Ministry of Health. Verbal consent was also sought from parents of the participating pupils through the Parent Teacher Association of each school. The consent of the primary health care was also sought. Giving of samples (urine) was not made mandatory or compulsory for the students. Urine samples were collected between 10:00 am and 1:00 pm. Volunteered children were registered using a well-structured questionnaire which documented their age, sex and their awareness of the disease in the community. Each pupil was given a clean, sterilized; dry screw-capped urine tube with identification number earlier assigned to them and was instructed on the mode of sample collection. Two (2) drops of 10% formalin solution was added immediately to the urine samples as they are being collected from the pupils in order to preserve it before being examined.

Examination of urine for microhaematuria and proteinuria

A chemical reagent strip (Combi-9® urinalysis Medi-test strip manufactured by Analyticon Biotechnologies, Germany) was

carefully dipped into the sterile sampling bottle containing the urine for 5 seconds. The resulting change in colour of the strip was compared with manufacturer's colour chart to estimate the amount of blood and protein in the urine.

Examination of *Schistosoma haematobium* ova

Ten (10) ml of the urine sample was centrifuged at 4000 rpm for 5 minutes. The supernatant was discarded to leave sediment which was transferred to the center of a clean grease-free glass slide to which was added a cover slip. This was mounted on a light microscope and examined at 10x objective to identify *Schistosoma haematobium* ova which is characterized with a terminal spine.

Statistical analysis

Data were subjected to descriptive analysis and Chi-square test using SPSS version 21.0. Sensitivity and specificity were carried out using Graph Pad Prism 6.

RESULTS

Urine centrifugation for microscopic detection of *S. haematobium* egg was used as a gold standard method to evaluate the performance of the chemical reagent strip (Combi-9® urinalysis test strip). However, 150 urine samples was collected from the school pupils with 76 males and 74 females, no female students was menstruating as they were all asked before collecting the samples from them. More than half 83 (55.3%) of the pupils were less than or 10 years old while majority 53 (35.3%) of the pupils source water from stream (Table 1). Pit latrine (48.6%) and bush (40.7%) were identified as a major sewage disposal used by the pupils, and also 44.0% bath and swim in the stream, 31.3% (fetching water) and 18.0% had no activities with stream.

Table 1: Sociodemograph factor of the studied pupils in Ikota.

Characteristics		Frequency (%)
Sex	Male	76 (50.7)
	Female	74 (49.3)
Age	≤ 10 Years	83 (55.3)
	>10 years	67 (44.7)
Water source	Tap water	42 (28.0)
	Well	48 (32.0)
	River	7 (4.7)
	Stream	53 (35.3)

Toilet	Water Closet	16 (10.7)
	Bush	61 (40.7)
	Pit Latrine	73 (48.6)
Activities with stream	None	27 (18.0)
	Washing Cloth	10 (6.7)
	Fetching Water	47 (31.3)
	Bathing or swimming	66 (44.0)

Out of a total of 150 urine specimens collected from schoolchildren of Ikota, ifedore local government, Ondo state, 36 (24.0%) were positive for *S. haematobium* eggs (Table 2). All positive samples had light infections as described as <50 egg/10

ml of urine samples. Using chi square test, no significant difference ($\chi^2=0.73$, $p=0.39$) between sex in relation to prevalence and likewise in age.

Table 2: Prevalence of Urinary Schistosomiasis among students in Ikota, Ondo state.

Variables		Status		df	χ^2	p-value
		Positive	Negative			
Sex	Male	16.0 (21.05%)	60.0 (78.94%)	1	0.73	0.39
	Female	20.0 (27.03%)	54.0 (72.97%)			
Age	≤ 10	31.0 (37.35%)	52.0 (62.65%)	1	0.39	0.53
	>10	5.0 (29.41%)	12.0 (70.58%)			

df: Degree of freedom, χ^2 :chi-square value, p-value: Significant value

As a secondary reference, urinalysis dipstick (Combi-9® Medi-Test) was also performed on all the 150 urine samples to evaluate the chemical reagent strip performance in testing for microhaematuria and proteinuria. For microhaematuria, there was no significant difference ($\chi^2=0.69$, $p=0.4$) between the

efficacy of the two methods, 30 (20.0%) pupils were tested positive using strip while 36 (24.0%) were positive using gold standard method. However, the two methods were significantly different ($\chi^2=22.8$, $p=0.01$) in proteinuria outcome, 76 (50.7%) tests were positive using strip (Table 3).

Table 3: Comparison between chemical reagent strip and microscopy test.

Variables	Status		df	χ^2	p-value
	Positive	Negative			
Microscopy	36.0 (24.0%)	114.0 (76.0%)	1	0.69	0.4
Microhaematuria	30.0 (20.0%)	120.0 (80.0%)			
Microscopy	36.0 (24.0%)	114.0 (76.0%)	1	22.8	0.01
Proteinuria	76.0 (50.7%)	74.0 (49.3%)			

df: Degree of freedom, χ^2 :chi-square value, p-value: Significant value

As shown in Table 4, 30 samples were truly positive for microhaematuria using strip while 6 were falsely negative. All

114 samples that were negative through microscopy also tested negative to microhaematuria. Sensitivity of the strip to

microhaematuria was 83.3% and specificity was 100%, these were statistically significant (Table 5).

Table 4: 2 × 2 Table on microhaematuria for chemical reagent strip testing.

Variables		Microscopic examination of urine		Total
		Positive	Negative	
Microhaematuria	Positive	30 (83.33%)	0 (0.00%)	30 (20.00%)
	Negative	6 (16.67%)	114 (100.00%)	120 (80.00%)
Total		36	114	150

Table 5: Sensitivity and specificity of chemical reagent strip in the test for microhaematuria.

Table Analyzed	Sensitivity and specificity
p value	<0.0001
Statistically significant? (alpha <0.05)	Yes
Sensitivity (%)	0.8333 (83.33%)
95% confidence interval	0.6719 to 0.9363
Specificity (%)	1.000 (100.00%)
95% confidence interval	0.9682 to 1.000
Positive Predictive Value	1.000
95% confidence interval	0.8843 to 1.000
Negative Predictive Value	0.9500
95% confidence interval	0.8943 to 0.9814

In Table 6, 2 × 2 table on proteinuria for chemical reagent strip testing was shown. Urinalysis dipstick (Combi-9® Medi-Test) diagnosed 3 pupils to be falsely negative and 43 pupils to be falsely positive to proteinuria. Seventy one (71) pupils were truly negative for both diagnostic methods. The sensitivity and specificity of the strip to proteinuria was significant (Table 7), it

sensitivity was 50%, specificity (62.3%) and the positive predictive value was 43.4% analyzed with Graph pad prism 6 at 95% confident interval.

Table 6: 2 × 2 table on proteinuria for chemical reagent strip testing.

Variables		Microscopic examination of urine		Total
		Positive	Negative	
Proteinuria	Positive	33 (91.67%)	43 (37.80%)	76 (50.67%)
	Negative	3 (8.33%)	71 (62.28%)	74 (49.33%)
Total		36	114	150

Table 7: Sensitivity and specificity of chemical reagent strip in the test for proteinuria.

Table Analyzed	Sensitivity and specificity
p value	<0.0001
Statistically significant? (alpha <0.05)	Yes
Sensitivity (%)	0.5 (50.0%)
95% confidence interval	0.3208 to 0.5529
Specificity (%)	0.6228 (62.3%)
95% confidence interval	0.5272 to 0.7119
Positive Predictive Value	0.434 (43.4%)
95% confidence interval	0.37753 to 0.4825
Negative Predictive Value	0.9595 (95.95%)
95% confidence interval	0.8861 to 0.9916

DISCUSSION

The prevalence of urinary schistosomiasis is considerably moderate in the screened population, as 24.0% pupils tested suffered the infection. Though the prevalence recorded in this study area is lower to prevalence detected in other part of Nigeria [12,13], it is greater than prevalence of 11.2%, 9.8% and 13.8% reported among pupils in Abeokuta [14], Afikpo North Local Government Area of Ebonyi State [15] and Ifedore local government area, Ondo State [16] respectively. Prevalence was not significant in term of sex and age differences, this concurred with the findings of Nworie et al. [15] and Akinneye et al. [16] on prevalence among age and sex differences. In the research conducted by Ugbomoiko et al. [17], male was significantly having higher prevalence than female. Microhaematuria and proteinuria diagnosis is age dependent and its outcome decreases as the age goes up, so it stables with school children [18]. The prevalence observed in the studied population may be due to their daily activities with the infected stream.

Result shows that the two methods considered in this study were not significantly different in diagnosis for urinary schistosomiasis but different when detection base on proteinuria. Further analysis revealed that chemical reagent strip for microhaematuria diagnosis was 95% efficacy to detect positive result and 100% efficacy to detect negative. There were 6 positive samples detected by microscopy but appeared negative to chemical reagent strip. Therefore, this made prevalence by microscopy (24%) higher than that of the strip (20%) and this was in contrary to the findings of Fatiregun et al. [6] and Stefanie et al. [19] where strip had higher prevalence outcome than microscopy. Furthermore, result gotten for proteinuria by the strip was not reliable as it has 43.3% efficacy to detect negative result.

Review had been made by Cochrane systematic, in which overall performance of chemical reagent strip testing for *S. haematobium* was stated to be 75% for sensitivity and specificity of 87% [9]. Although, the predictive value depends on prevalence of disease and its intensity [17] and even the manufacturer quality of chemical reagent strip, thus it may vary from study to study. As such sensitivity of the chemical reagent strips to detect proteinuria was low (50%) compare to microhaematuria (83.3%). Since 1980s, haematuria and proteinuria have been a major disease biomarker for urinary schistosomiasis but Taylor et al. [20] and Eltoum et al. [21] considered microhaematuria to be more sensitive and specific than proteinuria.

CONCLUSION

Though, this research was limited in term of age group examined, nevertheless, it is obvious that urinary schistosomiasis is still endemic in this community. Thus, there is urgent need to deploy intervention programme. Also, chemical reagent strip cannot be taken as a gold standard method for diagnosis; it should be combine with microscopy method to increase its predictive value. Standardize and accurate chemical reagent strip must be developed for rapid diagnosis of urinary schistosomiasis in endemic communities.

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