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Diagnostic Potentials of Haematuria and Proteinuria in Urinary Schistosomiasis among School-Age Children in Aliero Local Government Area, Kebbi State, North-Western Nigeria

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Authors' contributions

This work was carried out in collaboration between all authors. Authors JH and KM designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors SAO and TA managed the analyses of the study. Authors SUN, MKG, MB and NMB managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

Aims: The study was conducted to evaluate the diagnostic value of Haematuria (HU) and Proteinuria (PU) detected using reagent strips compared with microscopy in the examination of *Schistosoma haematobium* eggs in urine in Aliero Local Government Area of Kebbi State, Nigeria.

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Methodology: This was a cross-sectional, descriptive study, conducted in Aliero Local Government Area between March, 2015 to June, 2016. A total of 400 participants were enrolled for the study.Ten (10 ml) of urine samples were collected from each participant in to universal containers. Samples were examined macroscopically for gross Haematuria and then tested for micro-haematuria and Proteinuria using Combi-9 reagent strip. Samples were preserved with 10% formal saline and then transported to laboratory for analysis. Samples were filtered using Vacuum pump filtration machine and Whatman No.1 filter paper and were then examined under the microscope to determine the presence of ova of *S. haematobium eggs in urine.*
Results: Out of 400 urine samples 128(32.0%) had infection with mean egg intensity of 63.4/10 ml of urine, with significant difference ($p<0.005$). Altogether, 158 samples were positive for Haematuria (HU) and 145 samples positive for Proteinuria (PU). The Sensitivity and Specificity of Haematuria were 74.4% and 86.8% respectively with Positive predictive value 81.0%, and Negative Predictive Value 81.8%. Proteinuria has Specificity of 69.2% and Sensitivity of 91.2% ($P < 0.05$).
Conclusion: The high sensitivity and specificity of Haematuria (HU) and Proteinuria (PU) testing, shows that it can be used as good screening test for *S. haematobium* infection. The overall result of the study (32.0%) shows the establishment of moderate *S. haematobium* infection in the study area with higher prevalence among males 35.1% than females 19.7%, fishermen 64.15% and children 10-14 years having 38.8%. Prevention and control of Schistosomiasis based on mass chemotherapeutic treatment of population at risk using praziquantel is therefore warranted to reduce morbidity.

Keywords: Diagnostic potentials; haematuria; proteinuria; urinary schistosomiasis; children.

1. INTRODUCTION

Schistosomiasis is a chronic and debilitating disease caused by digenetic trematode flat worms of the genus Schistosoma. Schistosomiasis is a major public health problem and second after malaria in terms of Public Health Importance. The disease affects people from tropical countries in Africa, East Asia, and South America [1]. Despite the high burden of Schistosomiasis especially in Africa which accounted for more than 85 percent of the estimated 238 million people infected with the disease in 2010 [2,3], Schistosomiasis is still considered a neglected tropical disease.

Recent estimates from sub-Saharan Africa indicate that 280,000 deaths per year can be attributed to schistosomiasis [4]. Schistosomiasis is associated with water resources development projects such as dams, irrigation schemes, rice and fish-farming, which seems to increase the human contact and thus increase the risk of infection [5].

Detection of Haematuria (HU) and Proteinuria (PU) has been used as an indirect diagnostic assay for *S. haematobium* [6]. Many studies have validated HU and PU tests against standard urine filtration and concluded that the detection of microhaematocrit is a valid proxy for urinary schistosomiasis and the related morbidities [7], showing that Haematuria testing for *S. haematobium* has an overall sensitivity and specificity of 75% and 87% respectively [8]. HU and PU are widely accepted as indirect markers for screening of urinary schistosomiasis [9]. However, the presence of blood in urine due to menstruation or presence of protein in urine due to urinary tract infections (UTI) and other pathologies are confounding issues with regards to reagent strip results [9].

Numerous studies conducted over the years, have compared detection of HU and PU with filtration methods and were shown to be reliable, but sensitivity and specificity values differ considerably from one endemic area to another. In view of the above, it was stressed that the diagnostic performance should be assessed in every epidemiological setting before using this approach for a large community diagnosis [9].

Urinary Schistosomiasis caused by *S. haematobium* which results to passing of eggs through the bladder wall causes damage leading to the passage of small amounts of blood and protein in to the urine [10,11]. Reagent strips can detect such small amounts of blood and protein present in urine and can thus be used as indicators of infection with *S. haematobium* especially in field surveys [10].

The symptoms of Schistosomiasis include; dry cough with changes on chest x-ray, fever, fatigue and muscle aches. Others include malaise,

abdominal pains and enlargement of spleen and liver [12].

The aim of his study is to evaluate the diagnostic values of HU and PU in urinary schistosomiasis among School-Age Children in Aliero Local Government Area of Kebbi State, Nigeria.

2. MATERIALS AND METHODS

2.1 Study Area

The study was conducted in Aliero Local Government Area. Aliero local government area is approximately located at latitudes 4°23'S by 12°26' N and longitudes 3°6'W by 4°27' E. The local government was created in 1996, with a total land mass of 412.25 km^2, [13]. Aliero local government area has a total population of 67,078, and is one of the 21 local governments which make up the present Kebbi State [14].

Aliero local government Area shares common boarders with Gwandu Local government area by the east, Jega Local government area to the West, and Birnin Kebbi Local government area to the north. The Local Government has three (3) districts which comprise Aliero, Sabiyal, and Danwarai districts. The area has an annual rainfall ranging from 500 mm to 1,300 mm. Rainfall begins early May and ends in October each year. Major occupation of the inhabitants include, farming, irrigation works, rice farming, fishing and trading with a reasonable proportion of the population working in private and public sectors [15].

Large-scale production of millet during the raining season is the major practice and vegetables especially *onion* and pepper are grown through irrigation farming. The major tribes in Aliero local government area are Hausa, Fulani, Arawa and some minority tribes that include Yoruba's and Igbo's. Aliero residents are known for traditional bone setting across West and Central Africa. However, some areas of Aliero local government are blessed with several water bodies which enable the inhabitants to engage in water contact activities such as fishing, swimming, rice farming and other irrigation works as their source of daily income. The availability of water and the tropical type of weather may provide suitable breeding grounds for the snail intermediate host, leading to the possible transmission of Urinary Schistosomiasis in the area.

2.2 Study Population

The study population consists of 400 school aged children, in Aliero Local Government Area, North-West, Nigeria.

2.3 Study Design

This was a cross-sectional, descriptive study. The research was conducted during the raining season. The procedure was explained to all participants and were each given the consent forms to sign. Questionnaires were distributed to generate information on their bio-data and other Socio-demographic information of the respondents.

2.4 Sampling Method

Simple random sampling technique was used to recruit four hundred (400) School Children into the study.

2.4.1 Inclusion criteria

The study included all consented, apparently healthy, Children within the age range of (5 – 19 years) that were in Kashinzama and Sabiyal village who consented to participate in the study, and those that have not been on any sort of schistosomiasis treatment

2.4.2 Exclusion criteria

All Children that did not meet the inclusion criteria were excluded from the study and children on anti-schistosomal therapy, and those that did not consented as well as those that were less than (5 years or greater than 19 years) of age and those that have any form of internal bleeding or bladder injury are excluded.

2.5 Sample Size Determination

Sample size determination for this research was based on the findings of *S. haematobium* screening study which reported a prevalence rate of 38% [6]. Number of sample size was determined using the formula;

$n = Z^2 P Q/d^2$

where,

n = Minimum sample size
Z (standard deviation of normal) = 1.96

P (prevalence rate) = 38% (0.38) [6].
Q (1-P) = (1 - 0.34) = 0.62
d = confidence interval = 5% (0.05)
$n = (1.96)^2 \times 0.38 \times 1\text{-}0.38/ (0.05)^2$
n = 362

Due to attrition, 10% of 362 were added to the sample size

362 + 38 = 400

Therefore the minimum sample required was approx. 400

2.6 Ethical Consideration

Ethical clearance was obtained from the Ethical Committee of the Ministry of Health, Kebbi State in accordance with the code of Ethics for Biomedical Research involving Human subjects. The relevance and benefit of the study was explained to all of the subjects to ensure their voluntary participation and a written informed consent was taken from each subject.

2.7 Sample Collection

Dark and labeled plastic containers were given to each participant for collection of urine sample which was done between the hours of 10:00 am to 14:00 because it is the period of egg production by the female *S. haematobium*. A total of 400 urine samples were received and were preceded for analysis.

2.8 Laboratory Test

2.8.1 Test for haematuria and proteinuria

After collection, samples of urine were examined macroscopically for gross haematuria and then tested for micro-haematuria and proteinuria using Combi-9 reagent strips. The reagent strips were dipped in to the freshly collected urine and the result were read by comparing with the colour chart on the container.

2.8.2 Preservation and transportation of samples

Urine samples were then preserved using 1 drop of 10% Formal Saline and then transported to Laboratory for analysis.

2.9 Filtration

Urine samples were filtered using vacuum-pump filtration machine (Millipore Cooperation Bedford, Massachusset 01730, USA). The filtration techniques were used for the detection of urinary schistosomiasis [15,16,17]. The sample was mixed and 10 ml of each urine sample was collected from the container using 20 ml syringe. The filter paper was then removed from the vacuum, fixed with Ninhydrine solution and stained with Lugol's iodine solution and allowed to stain overnight [16].

2.10 Microscopy and Egg Counts

The stained filter papers containing urine deposits were examined under the microscope using x10 objectives to determine the presence of eggs of *S. haematobium.* Terminal spine eggs, characteristic of *S. haematobium*, were counted from several fields of each positive sample and number of eggs was recorded [17].

2.11 Research Tools

2.11.1 Questionnaire

Data collection was carried out using questionnaire in order to obtain socio-demographic characteristic of the respondent. During data collection, research investigator ensures that the data were collected accurately and correctly.

2.11.2 Validation of questionnaire

After the questionnaire was designed, it was sent to 3 experts in order to seek for their opinion as part of expert review panel to evaluate questionnaire test validity.

2.11.3 Questionnaire administration

The questionnaire survey domain consists of items socio-demographic characteristics of the participants as well as the risk factors associated with *S.haematobium* infection. The questionnaire has 3 domain which includes socio-demographic domains in section A consisting of age, gender, ethnicity and religion. Section B socio- economic data consisting of occupation, type of family, etc. section C Laboratory investigation results.

2.12 Statistical Analysis

Data obtained was analyzed using SPSS statistic version 20 (2013 Chicago, Illinois).The prevalence of infection was calculated in percentages. *P-value* less than 0.05 were considered significant. The sensitivity and

specificity of HU and PU were calculated using two by two contingency table to determine their diagnostic value in urinary Schistosomiasis.

3. RESULTS

A total of four hundred (400) participants were enrolled for this study and are within the age range of (5-19 years) old. All the participants are from Kashinzama and Sabiyal Communities in Aliero Local Government Area of Kebbi State. Out of the number examined, 158 (39.5%) were positive for haematuria and 128 (32%) were positive for *S. haematobium* infection using microscopy while 145 (36.2%) were positive for Proteinuria and 128 (32%) were positive for *S. haematobium* infection using microscopy for Kashinzama and Sabiyal Communities respectively (Table 1).

Table 1. Detection of urinary schistosomiasis using haematuria versus microscopy (n= 400)

Screening test	Microscopy		Haematuria strip	
	n	%	n	%
Positive	128	32.0	158	39.5
Negative	272	68.0	242	60.5
Total	400		400	

Out of the (400) participants used in this study' 319(79.5%) were males and 81(20.2%) were females. The distribution of the disease based on gender shows that males have higher prevalence 112(35.1%) than females with 16(19.7%) (Table 3).

Among the participants 270(67.5%) are between 10-14 years old and 75(18.7%) are between 15-19 years while 55(13.7) among them are 5-9 years old. The highest prevalence of infection was among children 10-14 years of age 105(38.8%) followed by 5-9 years age-groups with 13(23.6%) and 15-19 years age-group with 10(13.3%) (Table 1).

The occupational distribution of *S.haematobium* infection shows that 78(19.5%) are children of fishermen with the highest prevalence of 50(64.1%), followed by children whose parents are farmers 178(44.5%) with the prevalence of 65(36.5%), and then children whose parents are Civil servants 55(13.7%) with prevalence of 6(10.9%) and children whose parent are traders 89(22.2%) with the least prevalence of 7(7.8%).

The sensitivity and specificity of Haematuria obtained was significant, the sensitivity was 74.4% and specificity of 86.8% when compared with the gold standard (microscopy). Positive predictive value obtained 81.0%, while the Negative predictive value (NPV) obtained was 81.8% (Table 2).

Table 2. Detection of urinary schistosomiasis using proteinuria versus microscopy (n= 400)

Screening test	Microscopy		Proteinuria strip	
	n	%	n	%
Positive	128	32.0	145	36.2
Negative	272	68.0	255	63.7
Total	400		400	

4. DISCUSSION

Detection of Haematuria and Proteinuria is widely used as indirect marker in the diagnosis of *S. haematobium* infection. This is due to the fact that spine eggs of *S. haematobium* could rupture bladder wall causes damage leading to the passage of small amounts of blood and protein into the urine [10,11].

HU and PU can be used as indicators of infection with *S. haematobium* especially in field surveys [10], so they can provide a semi quantitative result. Numerous studies conducted over the years, have compared HU and PU testing with filtration methods and were shown to be reliable, but sensitivity and specificity values differ considerably from one endemic area to another.

The result of this study shows high sensitivity of Haematuria 74.4% and specificity of 86.8%, and Proteinuria has Specificity of 69.2% and Sensitivity of 91.2% which is very much similar to results obtained by [18]; with a high sensitivity and specificity of 75.0% and 87.0% respectively.

Other studies conducted in many African countries reported sensitivities of HU and PU ranging from 67-93%, with specificities of up to 66-99% [19], including a sensitivity of 87% in White Nile province in Sudan [20]. HU and PU testing can thus be proposed as a simple indirect method for identifying children with *S. haematobium* infection, and hence may be a useful tool for the rapid mapping of the prevalence of schistosomiasis to identify high risk areas which requires mass treatment with praziquantel [21].

Table 3. Prevalence and distribution of *S. haematobium* infection with respect to gender, age-group and occupation in the study area

Variables	Infection n %	No infection n %	Total n %	p-value
Gender				0.005[a]
Male	112(35.1)	207(64.8)	319(79.7)	
Female	16(19.7)	65(80.2)	81(20.0)	
Age-group (yrs)				0.001[a]
5-9	13(23.6)	42(76.3)	55(13.7)	
10-14	105(38.8)	165(61.1)	270(67.5)	
15-19	10(13.3)	65(86.6)	75(18.7)	
Occupation				0.001[a]
Fishing	50(64.1)	28(35.8)	78(19.5)	
Farming	65(36.5)	113(63.4)	178(44.5)	
Civil service	6(10.9)	49(89.0)	55(13.7)	
Trading	7(7.8)	82(92.1)	89(22.2)	

Table 4. Sensitivity and specificity of haematuria in the diagnosis urinary schistosomiasis

Screening test	Infected	Not infected	Total
Positive	True positive = a	False positive = b	a + b
158	128	30	158
Negative	False negative= c	True negative= d	Total
272	44	198	272
Total	a + c	b + d	a + b + c + d
400	172	228	400

Sensitivity= a / a + c x 100 /1 =128/ 128 + 44x100/1= 128/ 172 x 100/1=74.4%
Specificity= d/d+b x100/1 =198/198+30x100/1= 198/228x100/1=86.8%

Table 5. The sensitivity and specificity of proteinuria using reagent strips as indirect indicator of urinary schistosomiasis

Screening test	Infected	Not infected	Total
Positive	True positive = a	False positive = b	a + b
145	128	17	145
Negative	False negative= c	True negative= d	Total
255	57	198	255
Total	a + c	b + d	a + b + c + d
400	185	215	400

Sensitivity= a / a + c x 100 /1 =128/ 128 + 57x100/1= 128/ 185 x 100/1=69.2%
Specificity= d/d+b x100/1 =198/198+17x100/1= 198/215x100/1=92.1%

The findings in this study showed the establishment of moderate *S. haematobium* infection in the study area which is below the WHO range which consider 40% to be endemic or high. The result agrees with the result obtained by [22] in a study conducted in Argungu Local government Area of Kebbi State with a moderate prevalence of 34.0%.

The findings in this study contradict the results obtained by [23] in their research on Schistosomiasis in Dutsinma, Katsina State where a higher prevalence of 72.0% was obtained. The higher prevalence of schistosomiasis in the area may be due to low level of awareness about the associated risk factors in addition to high level of poverty among the inhabitants. It may also be due to the high level of water contact activities such as fishing and irrigation farming as the major sources of income in the area and high dependence on surface water [23].

Out of 400 school children examined in the study area, the prevalence and distribution of *S. haematobium* infection with respect to gender, age-group and occupation of parent showed that males have higher prevalence of infection

112(35.1%), while their female counter parts have 16(19.7%).

The high prevalence of infection recorded among males, may be due to the fact that males usually engage more in water contact activities than females leading to their higher exposure to infection with *S. haematobium*. This agreed with the results obtained by several researchers in Nigeria such as [24]. It may also be because the number of females participating in studies is usually smaller compared to the number of participating males.

The findings from the results obtained in this study contradicts the findings of [25], in a study conducted on urogenital Schistosomiasis in children in endemic rural community in Nigeria, where prevalence of *S. haematobium* infection was higher among females (60.3%) than males (54.1%) although the difference is not statistically significant. However, the higher prevalence among females may be due to their exposure to water contact activities related to domestic works such as fetching water from the ponds, washing clothes and eating utensils. It may also be due to the fact that in some communities, women engage in agricultural and irrigation works than their male counter parts which may likely expose them to infections.

The prevalence based on age-groups showed that, children 10-14 years old have the highest prevalence of infected individuals' 105(38.8%) while 5-9 years old have 13(23.6%) infected. Children 15-19 years have 13(3%) infected. The higher prevalence recorded among 10-14 years age-groups which with significant statistical difference (x^2=19.666[a] and (p- value= 0.001), may be due to the fact that children at that age perform more water contact activities than other age-groups and may not have developed their immunity fully enough to give them the needed protection against schistosomal infection. Children at the age of 5-9 usually have less water contact activities that may expose them to infection since parents may not allow them by virtue of their age which is similar to the findings of [26].

The high prevalence of infection among children of fishermen and farmers, and the least prevalence of infection recorded among children of traders and civil servants shows a significant correlation between urinary schistosomiasis and occupation. It also shows that Urinary Schistosomiasis has a significant relationship with the economic status, level of awareness and the educational background of an individual. Traders and Civil servants may be more economically and socially fit than farmers and fishermen and so can take good care of their children and their environment which may however reduce the chances of their children acquiring the infection. Fishermen and farmers may also have higher level of water contact leading to exposure to contaminated water bodies more than the traders and civil servants with less water contact activities and thus little chances of acquiring the infection.

5. CONCLUSION

This study showed high sensitivity and specificity of HU and PU in the detection of urinary schistosomiasis. HU and PU testing can thus be proposed as a simple indirect method for identifying children with *S. haematobium* infection, and hence may be a useful tool for the rapid mapping of the prevalence of Schistosomiasis to identify high risk areas.

6. RECOMMENDATIONS

Haematuria and Proteinuria testing for diagnosis of Urinary Schistosomiasis, should further be carried out in other settings and validated for use as rapid screening test for *Schistosoma haematobium* infection. Prevention and control measures should be adopted such as provision of adequate portable drinking water, sanitation and health education by the concerned authorities to address the problem. Presence of haematuria (HU) should be reported as early as possible so as to take immediate diagnostic and chemotherapeutic measures.

CONSENT

As per international standard or university standard, patient's written consent has been collected and preserved by the authors.

ETHICAL APPROVAL

As per international standard or university standard, written approval of Ethics committee has been collected and preserved by the authors.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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