Urinary   Schistosomiasis And Concomitant Bacteriuria In The Federal Capital Territory Abuja Nigeria

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ABSTRACT
Urinary schistosomiasis and concomitant bacteriuria was investigated in the Federal Capital Territory (FCT) Abuja. Single urine samples collected   from subjects aged 5 years and above between 1000 hours and 1400hours were examined for the presence of *S.haematobium* eggs using centrifugation technique and for bacteriuria by standard bacteriological methods. A total of 1,150 subjects comprised of 667 males and 483 females were studied from the 6 Area Councils of the FCT. Overall, 360 (31.3%) had the eggs of *S. haematobium* in their urine while 289 (80.3%) of the 360 who had eggs of *S. haematobium* in their urine, had bacterial growth. Prevalence of bacteriuria in urinary schistosomiasis ranged from 74-86% with no significant difference in the distribution of the prevalence of the co-infection in the 6 area councils surveyed (P=0.125). The distribution of bacteria colony count in relation to different ova intensity was significantly different (P<0.001) and assumed a weak positive linear relationship (r=0.2). There was no significant difference in the results of the methods used to investigate for bacteriuria (P=0.05). The bacteria isolated included: *klebsiella species, Escherichia coli, Enterococci species, Staphylococcus aureus, Staphylococcus saprophyticus, Salmonella species, Proteus species, and Pseudomonas species. Eshericha coli* occurred more frequently (70%) than the rest of the bacteria species isolated. The antimicrobial susceptibility pattern of isolates revealed varying percentage susceptibilities by all isolates. This study clearly suggests that bacteriuria is a potent complication in the management of urinary schistosomiasis. Therefore the complimentary incorporation of antibacterial therapy appear essential. [New York Science Journal. 2009;2(1):22-28]. (ISSN: 1554-0200).

Keywords: Schistosomiasis, Concomitant Bacteriuria , Prevalence, Susceptibility,

INTRODUCTION
Urinary tract disease is a specific trait of infection with *Schistosoma haematobium* which affects in a diffuse manner the entire genitourinary tract (King, 2001; Pereira *et al*., 1997). Bacteria infections are often recurrent and important complications of the inactive stage of urinary schistosomiasis which may be instrumental in precipitating renal failure (Farid, 1993). In schistosomiasis of the urinary bladder, secondary bacterial infections are common and in men can involve the seminal vesicles, spermatic cord, and to a lesser extent, the prostate. In women, infection can involve the cervix and fallopian tubes and can cause infertility. Mostafa *et al*., (1999) opined that it seems possible that agricultural workers and others who are regularly exposed to contaminated water are occasionally simultaneously infected with both the schistosome parasite and pathogenic bacteria. The risk factor of agricultural practices the major occupation of indigenous residents of the Federal Capital Territory (FCT) Abuja, Nigeria is capable of breeding urinary schistosomiasis and concomitant bacteriuria. In the light of the relative high level of schistosomal and bacteria infection, active assessment and reporting of bacteriuria in urinary schistosomiasis and the complementary incorporation of antibacterial therapy to the integrated morbidity control approach to urinary schistosomiasis deserves emphasis. This study examined the terminal urine sample of individuals with or without signs of urinary disturbance and infection for evidence of urinary schistosomiasis as well as evaluated associated bacteriological burden and susceptibility pattern of bacteria isolated.

MATERIALS AND METHODS
The study included 1,150 subjects both males and females between the ages of 5-50 years recruited directly through surveillance out-reaches to district/village schools and health related institutions. Informed consent of adult subjects was obtained, while consent to obtain specimen from 'minors'/pupils

was obtained through parents/guardian and the Education department of the Ministry of the Federation Capital Territory (MFCT) Abuja.

The urine samples were collected between 1000 hours and 1400 hours and were examined for colour, naked eye haematuria, turbidity and these observations were noted. Ten millilitres of urine were transferred aseptically into centrifuge tube and centrifuged for 5 minutes at 5000 rpm (Anosike *et al*., 2001). After discarding the supernatant the entire sediment was transferred to a slide covered with cover glass examined for red blood cells, pus cells (pyuria) and counting of eggs of *S. haematobium*. Using the 10x objective with the condenser iris closed sufficiently to give good contrast, the entire sediment preparation was examined systematically and ova count reported per 10ml of urine (Chessbrough, 1981; Richards *et al.,* 1984).

The remainder of urine samples positive for *S. haematobium* ova were homogenized by inverting the container severally and 0.002 ml of the urine inoculated and spread on Cysteine lactose electrolyte deficient medium (CLED-BIOTEC, UK)and blood agar (Blood agar base-BIOTEC, UK). Afterwards 10ul of the homogenized uncentrifuged urine were applied unto a glass slide allowed to dry without spreading at ambient temperature and stained by Grams method. Using 100x objective the slide was examined for bacteria per oil immersion field (Celso *et al.,* 1998).

The uncentrifuged urine samples were diluted 1:20 (20 ul of urine + 380 ul of Turks solution - 2% Acetic acid tinged with gentian violet).This is to destroy the red blood cells and stain the white blood cell nuclei. The dilutions were transferred to Neubauer haemocytometer chamber. The chamber was   examined using 10x objective and 4 squares counted applying the margin rule for including and excluding cell lying on the peripherial lines to quantify pyuria (Campbell *et al.,* 2002). Reagent strip urinalysis was performed using L-Combur reagent strip (Boehringer Mannhein).

The culture plates were examined after 24 hours of incubation for bacterial growth and colony count. Bacteria growth less than 10^5 organisms per ml produced less than 30 colony forming units per ml of urine (Chessbrough, 2000).Bacteria isolates were identified and characterized using methods prescribed by Cowan and Steel,1974; Chessbrough,2000 and Graham and Galloway, 2001. Susceptibility testing of all pathogenic baceteria were performed using the standard disc diffusion method according to British Society for Antimicrobial Chemotherapy (Andrew, 2001).

Statistically Analysis

The data analysis was done using X^2 (chi-square) test to determine significant relationships between variable and coefficient of correlation for test of linearity of relationship.

RESULTS

The overall prevalence of urinary schistosomiasis was 31.1% (95% CI 26.2 – 36.4) in the Federal Capital Territory Abuja and ranged between 25 – 36.3% in the six area councils surveyed. Prevalence followed the typical age group pattern for urinary schistosomiasis attaining a peak 78.4% in subjects 10 – 14 years age, decreasing to 47.6% in subjects $\geq$ 50 years and lower in subjects within 20 – 39 year. Prevalence of urinary schistosomiasis was higher at all ages in males ranging between 0 – 42.1% and in females 0 – 36.3% (Table1). *S. haematobium* infection prevalence had a statistical significant difference between males and females at different age groups (x^2=48; P<0.001).

In all, of the 360 subjects that had ova in their urine, 275 was positive for uncentrifuged gram microscopy, 305 was positive for pyuria (WBC) count, 330 was positive for leucocytes esterase, 350 was positive for protein, 336 was positive for erythrocytes (urinary blood), 240 was positive for nitrite (Table 2). Overall, 289 samples from subjects had bacteria growth of varying count. 261 (90.3%) samples had overt significant bacteriuria ($\geq 10^5$cfu/ml) in both males and females. Between males and females, there was a statistical significant difference in bacteria colony count in urinary schistosomiasis (x^2=9.9; P=0.025).

Bacteriuria in urinary schistosomiasis in F.C.T. had a prevalence of 80.3% ranging between 74 to 86% in the six area councils of F.C.T. surveyed. Bacteriuria and urinary schistosomiasis co-infection had no statistical significant difference (x^2=9.8; P=0.125).The distribution of bacteria colony count (cfu/ml) according to different ova intensity i.e. egg/10 ml of urine (Table 2); had a weak positive linear relationship (r=0.2). Albeit, there was a significant difference between bacteria colony count and different ova intensity in urinary schistosomiasis (x^2=39.0; P<0.001). The statistical analysis of results from culture and non – culture methods (enhanced microscopic urinalysis and reagent strip tests) for investigating bacteriuria are shown in Table 4. There is no significant difference in percentage positive results of culture and a combination of the non-culture methods for investigating bacteriuria (x^2=5.9; P=0.05). Various bacteria

species were isolated with *Escherichia coli* occurring more frequent than the rest in males (Table 4). Notwithstanding, there was no significant difference in the bacterial isolates between males and females (x^2=7.5; P=0.65)

Antimicrobial susceptibility pattern of bacteria isolates are shown in Table 8. All the isolates had susceptibility in varying percentage to Ofloxacin Ciprofloxacin, Gentamicin and Cefuroxime in order of percentage effectiveness respectively. However all the isolates except 3 were susceptible to Nitofurantion, 2 species of the isolates *(Proteus species and Pseudomonas species)* were not susceptible to Co-trimoxazole while 1 species was not susceptible to Co-amoxiclav.

Table 1: Distribution of the prevalence of S. haematobium infection in FCT according to age and sex; statistical test of significance between make and female

AGE GROUP (YEARS)	FEMALE			MALE		
	TOTAL NUMBER EXAMINED	NUMBER INFECTED	% EXAMINED INFECTED	NUMBER EXAMINED	NUMBER INFECTED	% INFECTED
5 – 9	213	120	21	93	30	32.3
10 – 14	557	179	65	378	159	42.1
			36.3			
15 – 19	200	110	27	90	30	33.3
			24.5			
20 – 24	50	12	2	38	5	13.2
			16.7			
25 – 29	29	10	1	19	3	15.8
			10			
30 – 34	34	21	3	13	2	15.8
			14.3			
35 – 39	22	10	2	12	1	8.3
			20			
40 – 44	20	12	1	8	3	37.5
			8.3			
45 – 49	15	6	1	9	2	22.2
			16.7			
>50	10	3	1	7	1	14.3
			33.3			
TOTAL	**1150**	**483**	**124**	**667**	**236**	**35.4**
			25.7			
X^2_{cal}	**42**		X^2_{tab}	**18.25**		

Table 2: Analysis and statistical test of significance of percentage positively for culture and non culture tests for urinary
schistosomiasis and bacteriuria.

NON-CULTURE TESTS						URINE CULTURE
	%	NUMBER + VE OF 360 EXAMINED/METHOD PER METHOD	NUMBER . -VE PER METHOD	%	%	NUMBER + VE PER METHOD
UNCENTRIFUGED URINE GRAM MICROSOCOPY > 1 ORGANISM/OIL IMMERSION FIELD	87.3	275	35	12.7	76.4	240
PYURIA (WBC) COUNT $\geq 1.0 \times 10^{9}$	93.4	305	20	6.6	85	285
LEUCOCYTES ESTERASE > 25 LEU/UL	86	330	46	14	91.6	284
PROTEIN > 30 MG/DL	77.1	350	80	22.9	97.2	270
ERYTHROCYTES URINARY BLOOD > 10ERY/UL	86.5	336	49	14.5	93.3	287
NITRITE POSITIVE	84.2	240	38	15.8	66.7	202
TOTAL	514.50%				510.20%	
$X^2{}_{cal}$		5.5		$X^2{}_{tab}$		11.03
Pvalue	0.05					

Table 3: Distribution of bacteria colony count (cfu/ml) according to different ova   intensity (egg/10ml urine); Statistical test of significance and coefficient of correlation in urinary schistosomiasis.

BACRERIA COUNT cfu/ml	NUMBER OF SUBJECTS				
	0-20 (egg/10ml urine)	21-40 (egg/10ml urine)	41-50 (egg/10ml urine)	>50 (egg/10ml urine)	TOTAL
$\geq 10^5$	13	20	10	218	261
10^4	0	3	4	5	12
10^3	3	2	2	3	10
10^2	0	0	2	4	6
TOTAL	**16**	**25**	**18**	**230**	**289**

X^2_{cal} **39**
X^2_{tab} **16.92**
Pvalue *<0.001*
r *0.2*

Table 4: Bacteria pathogens associated with *S.haematobium* infection and their antimicrobial susceptibility pattern.

ANTIMICROBIAL AGENT	PERCENTAGE SUSCEPTIBILITY							
	Eschericha Species r =101	*Klebsiella coli* n=30	*Enterococci Species* n=27	*Staphylococcus aureus* n=78	*Staphylococcus saprophyticus* n=19	*Salmonella Species* n=11	*Proteus Species* n=9	*Pseudomonas Species* n= 14
Ciprofloxacin	98	89.6	67.5	74	78	68.7	95	61.7
Cephalexin	25	38.5	39.2	25	30	16.9	25.4	0
Cefuroxime	65	70	67	63	50	46.3	72.5	45
Oxfloxacin	80.4	90	85	82.1	80	78	95	8.6
Gentamycin	76	82	47	45.9	49.6	85.2	58	70.5
Co-trimoxazole	26.4	56	61	68	54	25	0	0
	38.4	85	65.1	70	64.3	0	0	0
Nitrofurantion	32.8	25	41	33.5	40.2	20.4	63.2	0
Co-amoxiclav								

DISCUSSION AND CONCLUSION

The findings in this study demonstrates that the overall estimated prevalence of urinary schistosomiasis as determined by ova in the urine was high (31.3%; 95% CI 26.2-36.4%). Recent researchers estimate prevalence of 29.4% in the Eastern Nigeria (Anosike *et al.,* 2001) and 57.4% in the West (Adeyaba and Ojeaga, 2002). The result of this study is agreeable with these reports.

This study evaluated bacteriuria in urinary schistosomiasis revealing that of the 360 subjects (31.3%) who had Ova of *S. haematobium* from 1,150 examined; 289 (80.3%) had bacteriuria by culture characterization. The percentage positive results of culture and a combination of non-culture had insignificant difference (P> 0.05). Though King (2001) noted that urinary tract disease is a specific trait of infection with *S. haematobium*; The 80.3% prevalence of bacteriuria in urinary schistosomiasis need further categorization since by the definition of Gallagher and Hemphil (2004) it may simply be taken as referring to the presence of bacteria in the urine of individuals infected with *S. haematobium* and not necessarily implying infection. This is cogent as bacteriuria and urinary schistosomiasis co-infection assessed in the study had no significant difference (P>0.05). Gallagher and Hemphil (2004) and Franz and Horl (1999) had equally noted that in general terms urinary tract infection (UTI) is infection by Pathogen along the urinary tract causing inflammation depicted by pyuria indicating significant inflammatory response to bacteriuria such as occur with infection even in asymptomatic setting. These views mentioned above explicitly suggests that bacteriuria may be significant or non- significant depending on the quantity of bacteria in the urine which imply infection and is traditionally urine culture containing $\geq 10^5$ cfu/ml. The result of our assessment of bacteriuria in urinary schistosomiasis agreeably categorized 261 (90.3%) subjects by urine culture as having significant bacteriuria ($\geq 10^5$cfu/ml) with their sex distribution being significantly different (P<0.05) . This finding in consonance to that by Rushton (1997) which suggest that it may be possible to eliminate the urine culture when enhanced microscopic urinalysis and reagent strip urinalysis are negative and clinical suspicion is low. Nonetheless, isolation of significant number of single organism on culture remains the definitive diagnosis.

The finding of lower threshold of bacteria counts (10^2-10^4 cfu/ml) and the distribution of bacteria colony count according to different ova intensity which had a weak positive linear relationship (r = 0.2) deserves critical scrutiny because bacteria colony count and different ova intensity in urinary schistosomiasis was significantly different (P<0.05).This is pertinent to obtaining the best combination of sensitivity and specificity in the diagnosis of urinary tract infection. Franz and Horl (1999) reported that the utility and consistency of the criterion ≥105 cfu/ml of clean-catch urine for the diagnosis of UTI has been validated repeatedly. Thus, Stamm and Hooton (1993) noted that in dysuric patients, an appropriate threshold value for defining significant bacteriuria is 10^2cfu/ml of a known pathogen. Considering the foregoing and that dysuria is common in both early and late urinary schistosomiasis where ova count correlate with morbidity, it might be prudent to consider these thresholds significant for the diagnosis of UTI. More so, community –based epidemiological survey of bacterial count in Egypt (Mostafa *et al*., 1999) of subjects with *S. haematobium* infection had similar low bacteria counts (10^3 cfu/ml). However, interpretation of low threshold counts as significant for diagnosis must be in the absence of mixed bacteria growth with a predominant organism typical of contamination.

Infection of the 289 (80.3%) subjects with one bacteria or the other was the trend in our study and had no significant difference (P>0.05) in bacteria isolates between males and females. The isolation of *Esherichia coli* more frequently than the rest conforms to reports of many researchers (Farid, 1993; Mostafa *et al*., 1999) about it's association with schistosome infection.

The antimicrobial susceptibility pattern of the bacteria isolates to routinely tested first line antimicrobial agents were quite diminished. There were notable pockets of resistance to all first line agents tested except for Ciprofloxacin and Cefuroxime (Table 4). This antimicrobial susceptibility results would be an invaluable premise for empirical therapy where suspicion exists but cultures are impracticable whereas enhanced microscopic urinalysis and reagent strip are positive since comparative analysis of these methods had no significant difference (P>0.05). The main goal of most initiatives to control schistosomiasis is morbidity control. The reported complications of bacteria infections in urinary schistosomiasis are odious. Clinical and pathological conditions arising there from had been enunciated (Farid, 1993; Ganem *et al.,* 1998). Hence, this research further documents and authenticate the importance of a database for continued valuation and evolution of control programmes. The complementary incorporation of antibacterial therapy to the integrated morbidity control approach of diagnosis, drugs treatment, snail control, provision of safe, adequate water supply, sanitation and health education is

advocated. More over, our results infer that urinary schistosomiasis is endemic in FCT Abuja Nigeria and deserves urgent intervention.

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