



Molecular Epidemiological Investigation of Urinary Schistosomiasis Among Individual Residing Along Selected Riparian Communities in Bauchi State, Nigeria

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Abstract

Schistosomiasis, also known as snail fever or bilharziasis, is a prevalent tropical parasitic disease caused by blood flukes, Schistosoma species. It poses a great public health and socio-economic threat in sub-Saharan Africa. This study investigated the epidemiology of urinary schistosomiasis among individuals residing along selected riparian communities of Bauchi State, Nigeria. Ethical approval was obtained before sample collection. A total of 321 individuals were randomly selected from the riparian areas of Bauchi State for the study. A structured questionnaire was administered to the respondents following informed consent. Urine samples were analysed using microscopy, urinalysis, and molecular techniques. DNA was extracted from positive urine sediment, followed by Polymerase Chain Reaction amplification, sequencing, BLAST, and phylogenetic analysis. The demographic study of the respondents revealed that they were 4 - 65 years old, with a mean age of 15.1 ± 9.2 years; made up of 244 males and 77 females. The overall prevalence of urinary schistosomiasis was 22.7%, which was significantly associated with age, gender, education, the reason for going to the river, frequency of coming in contact with a river, and the household's source of water supply. The urinalysis results showed strong associations of schistosomiasis with leukocyturia, proteinuria, and haematuria. Molecular analysis revealed genetic diversity among Schistosoma haematobium populations. The present study indicated that urinary schistosomiasis is prevalent in the study area, and a need for an effective urinary schistosomiasis control programme in the riparian communities of Bauchi State, Nigeria.

Keywords: Bilharziasis, Urine, Haematuria, endemic, and Schistosoma haematobium.

INTRODUCTION

Background of the Study

Schistosomiasis, also known as bilharziasis, is a disease transmitted through freshwater and caused by parasites of the genus *Schistosoma*. These parasites are digenic trematodes that live in the blood vessels of both humans and livestock. Humans can be infected by five different species of schistosome: *Schistosoma haematobium*, *S. mansoni*, *S. japonicum*, *S. mekongi*, and *S. intercalatum*. *S. intercalatum* is known to parasitise cattle in West Africa and sporadically leads to disease in humans (Ponzo *et al.*, 2024). The intestine and liver are affected by all *Schistosoma* species, except *Schistosoma haematobium*, which invades the urinary tracts. Children's physical growth and cognitive development are also affected by chronic schistosomiasis (Kasambala *et al.*, 2022). Bilharziasis is one of the most significant concerns for public health, a prevalent tropical parasitic disease that affects over 200 million individuals. It ranks second only to malaria in terms of fatalities, claiming the lives of approximately 280,000 people annually in the African region alone (Lubanga *et al.*, 2024). Urinary schistosomiasis is caused by *S. haematobium*, which is

primarily found in Africa. The disease remains a major problem among people living in low-income countries, especially those with poor sanitation. The transmission of the disease is limited to people in contact with contaminated waters in endemic countries (Mohammed *et al.*, 2022).

Schistosomiasis is one of the Neglected Tropical Diseases (NTDs) prevalent in Africa. It has a significant impact on the economies and health of African nations (Mawson, 2023). The disease remains largely unnoticed in developed regions due to its rarity and the limited resources available in many developing African countries to understand their biological and social aspects (Abdulkadir and Ahmed, 2020). Globally, schistosomiasis holds third position, in terms of its impact on human health among tropical diseases, following intestinal helminthiasis and malaria (Mawa *et al.*, 2021). It is considered to be the most severe and widespread parasitic disease in Africa, South America, the Caribbean, the Middle East, and Asia due to the high morbidity and mortality associated with it (WHO, 2022).

Nigeria has a land area of 923,768 Km² with 1,300 Km² covered by water bodies; the remaining 2,930 Km² are lakes, either naturally occurring or of artificial water resource initiatives. Over 70% of Nigerians are classified as subsistence farmers, and their average life expectancy is 60.87 years. An updated report on schistosomiasis showed that Nigeria requires about 20 million medications per annum to treat the afflicted people, as the country is the most badly affected globally (Adedze-Kpodo et al., 2023).

Schistosomiasis transmission occurs when the water body is contaminated by the cercariae of the worms, which infect their host mainly through contact with snail-infested freshwater (Kamel et al., 2021). Numerous freshwater snails hold significance in clinical and veterinary settings by acting as intermediate hosts for various helminthic parasites in humans and animals (Wiroonpan et al., 2021). Freshwater snails from the *Planorbidae* family are primarily identified as the intermediate hosts of the highly infectious trematode larvae of the genus *Schistosoma*, which is responsible for causing schistosomiasis. The three genera *Biomphalaria*, *Bulinus*, and *Oncomelania* are the primary intermediate hosts for human schistosomiasis (Gboeloh and Ike-Ihunwo, 2022; Zhang et al., 2022). *Biomphalaria* and *Bulinus* are the intermediate hosts of *Schistosoma mansoni* and *Schistosoma haematobium* in Nigeria, respectively (Chibwana et al., 2020; Okeke et al., 2020). The aquatic snail hosts can be found in shallow water near lake shores, ponds, marshes, streams, and irrigation channels. They inhabit water plants and muddy areas abundant in decaying organic material. The presence of snail intermediate hosts in a region is a key factor in sustaining schistosomiasis transmission. The expansion of water projects has significantly contributed to the spread of schistosomiasis in certain areas. The construction of dams has boosted the likelihood of establishing irrigation channels and given residents in nearby areas access to a substantial water supply (Pennance et al., 2020). This has ultimately provided suitable habitats and breeding sites for the freshwater snail vector, leading to an eventual increase in the population density of the snail vectors.

The most important risk factor for an individual to acquire urinary schistosomiasis is coming into contact with infected water. The final host becomes infected when exposed to fresh water contaminated with cercariae, parasitic larvae secreted by infected snails (the intermediate host) (Hailegebriel et al., 2020). The process involves contaminating surface water with

infected urine and excreta, the specific freshwater snails as intermediary hosts, and human interaction with water (Adekiya et al., 2020).

Urinary schistosomiasis exhibits high global prevalence, affecting over 243 million individuals across 78 countries. The disease predominantly occurs in tropical and subtropical regions, particularly within impoverished communities lacking access to potable water and adequate sanitation facilities. It is estimated that at least 90% of those requiring treatment for Schistosomiasis live in Africa (Aula et al., 2021; WHO, 2023). Nigeria is a region where the parasite is widespread, with approximately 29 million individuals being infected. Due to the vast rivers and numerous snails present, urinary schistosomiasis poses a public health concern in the riparian areas of Bauchi State, Nigeria. This disease is prevalent in all 36 states and the Federal Capital Territory of Nigeria (Isa et al., 2023).

Schistosomiasis has distinct morbidity and pathophysiology in human hosts. Although the variations among *Schistosoma* species may account for numerous unclear facets of the parasite, including epidemiology, susceptibility to infection, and response to therapy (Stothard et al., 2020). Studies have shown genetic heterogeneity within the *S. haematobium* species, but no information is available regarding the genetic variation of the indigenous *Schistosoma haematobium* population in the study locations. Since it is challenging to distinguish the infectious cercariae that come from snails morphologically, all *Schistosoma haematobium* assemblages depend on *Bulinus* spp. for transmission. As a result, reliable molecular markers are needed to enable the distinction of different species. This is important in regions where *S. haematobium* is widespread and efforts are being made to control transmission (Osondu-Anyanwu et al., 2022). *S. haematobium* is known to be able to adapt to a wide range of environments, even though its morphology is believed to be consistent. Variations in population genetics may be the cause of these traits (Pennance, 2020).

Schistosomiasis commonly occurs in impoverished rural areas where inhabitants engage primarily in fishing and farming activities. Women and children are particularly susceptible to infection due to their involvement in household and leisure pursuits such as laundering clothes, collecting water, and engaging in recreational swimming (WHO 2022). Residents of these communities utilise the

river for laundry and bathing purposes. Bauchi State possesses numerous freshwater bodies, and a significant portion of the population in the state engages in irrigated rice cultivation. The river is a freshwater source and contributes to the high prevalence of schistosomiasis in the region (Tefera *et al.*, 2020).

Therefore, this study aimed to investigate the epidemiology of urinary schistosomiasis among individuals residing in riparian communities in Bauchi State. Utilising molecular techniques will enhance our understanding of the genetic variation, transmission patterns, and population characteristics of *Schistosoma haematobium* parasites in the study area. The findings will help in designing specific control measures and actions to reduce the impact of urinary schistosomiasis in Bauchi State.

Justification of the Study

Urinary schistosomiasis is reported to affect 29.7% of school-age children in Bauchi State (Mohammed *et al.*, 2022). Previous studies on the prevalence of schistosomiasis in Bauchi State involved the use of microscopy only (Usman and Babeker, 2017; Isa *et al.*, 2023). This method has limitations such as low sensitivity and its inability to classify the parasite to the subspecies level, making it inefficient and ineffective. This makes it necessary to devise alternative ways, especially specific and sensitive molecular techniques, to provide solutions for easy and effective diagnosis. Due to their high specificity, Polymerase Chain Reaction (PCR) techniques are successfully employed to identify genetic material from various parasites, such as *Schistosoma* species (Fuss *et al.*, 2021).

No reported investigation on the use of molecular techniques on urinary schistosomiasis in Bauchi State. Understanding the molecular epidemiology of urinary schistosomiasis can lead to better prevention, treatment, control, and ultimately, this will improve the progress towards achieving Sustainable Development Goal 3, which focuses on promoting good health and well-being.

Aim and Objectives

This study investigated the epidemiology of urinary schistosomiasis among individuals residing along selected riparian communities of Bauchi state, Nigeria. The specific objectives of the study were to: (i) determine the prevalence and distribution of *Schistosoma haematobium* infection among individuals residing in riparian areas of

Bauchi State, (ii) evaluate the risk factors associated with urinary schistosomiasis transmission in the study area, and (iii) identify the strains of *Schistosoma haematobium* predominant in the study area.

Scope of the Study

This research investigated individuals living in riparian communities of Bauchi State in North-East Nigeria to detect *Schistosoma haematobium* eggs in their urine samples. The research was conducted in seven months from December 2023 to June 2024. The sampling and analysis of samples were done concurrently. The study focused on determining the prevalence and identifying the risk factors associated with urinary schistosomiasis among individuals living in riparian communities of Bauchi State.

MATERIALS AND METHODS

Study Area

The investigation was conducted in four communities along the Jama'are River in Bauchi State, Nigeria. The river begins in the mountainous region near Jos in Plateau State, flows in the northeast direction, passing through Bauchi State and Yobe State before eventually meeting the Hadejia River (Figure 2.1). It is about 400 Km long and has a catchment area of about 20,000 square Km. People who reside along the river use it for fishing, irrigation, domestic use, transportation, swimming, and other purposes (Ayeni *et al.*, 2019).

Ethical Clearance

An introductory letter from the Head of the Biotechnology Centre, Afe Babalola University, Ado-Ekiti, was provided to the Bauchi State Ministry of Health and the Hospital Management Board of the Federal Medical Centre, Azare, to obtain ethical approval before starting the research. Ethical clearance with the following approval reference number was obtained: Ministry of Health, Bauchi State (MOH/GEN/S/1409/II) and Hospital Management Board of Federal Medical Centre Azare (FMCA/COM/35/VOL.I).

Study Design

A cross-sectional research approach was utilised to examine the prevalence of urinary schistosomiasis among people living in riparian areas of Bauchi State. The research was carried out from December 2023 to June 2024.

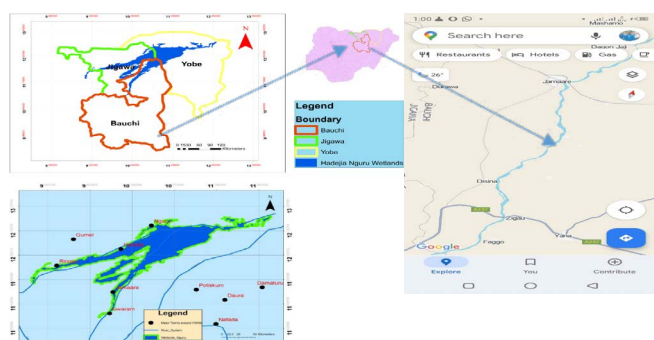


Figure 2.1: The study area (Ayeni et al., 2019)

Sample Size Determination

The study utilised the equation (Charan and Biswas 2013) below to establish a sample size of 321 individuals.

$$n = \frac{Z_{1-\alpha/2}^2 p(1-p)}{d^2}$$

Where:

n = sample size

$Z_{1-\alpha/2}$ = the standard variable ($P < 0.05$) = 1.96.

p = proportion in population as indicated by prior research = 29.7% (Mohammed et al., 2022).

d = precision = 0.05

Sample Collection

Urine Collection: A 30 ml specimen bottle was provided to the participant, with instructions to collect a midstream urine sample along with the final drops between 11:00 a.m. and 1:00 p.m. on the designated day (WHO 2011). The collected urine samples were transported to the Federal Medical Centre in Azare for analysis, utilising ice packs during transportation.

Administration of Questionnaire

Structured questionnaires were provided to evaluate socio-demographic characteristics and possible risk factors.

Sample Processing

Microscopic examination

Analysis of the urine in the laboratory was conducted using the centrifugation and sedimentation method (Cheesbrough 2006). A volume of 10 mL of urine was spun in a centrifuge at 1,500 revolutions per minute for 5 min, the supernatant was discarded, and the sediment was examined under $\times 10$ and $\times 40$ objective lenses of a microscope for the presence of terminal spine ova of *S. haematobium*.

Urinalysis

Urinalysis of the urine samples was assessed using a Combi 10 urinary strip. The quality of the urine samples was determined using the following biomarkers:

bilirubin, blood, glucose, ketone, leukocyte, nitrite, pH, protein, specific gravity, and urobilinogen.

Deoxyribonucleic Acid (DNA) Extraction of *Schistosoma haematobium* from Urine

DNA extraction from urine samples positive for *Schistosoma haematobium* eggs was performed using the Quick-DNA™ Miniprep Plus Kit (NG2024/59018), manufactured by Zymo Research for biological fluids. The manufacturer's protocol was strictly adhered to for optimal results. A 2 μ L volume of urine was transferred to an Eppendorf tube and centrifuged at 1,400 rpm for 5 minutes to concentrate the ova of *S. haematobium*. The supernatant was discarded, and 100 μ L of DNA elution buffer was added to resuspend the pellet. A volume of 100 μ L of Biofluid & Cell Buffer and 10 μ L of Proteinase K were added to the mixture, which was then vortexed for 15 seconds. The mixture was incubated in a water bath at 55 °C for 10 minutes. An equal volume of Genomic Buffer was added and mixed by vortexing for 10 seconds. The mixture was centrifuged at 1,200 rpm for 1 minute using a Zymo spin and collection tube to collect the sample. A total of 400 μ L of pre-wash buffer was added and centrifuged at 1,200 rpm for 1 minute. Subsequently, 700 μ L of gDNA wash buffer was added and centrifuged at 1,200 rpm for 1 minute. The collection tube was discarded, and 100 μ L of DNA elution buffer was added and allowed to stand for 5 minutes at room temperature inside the centrifuge machine. Centrifugation at full speed was performed for 1 minute, and the eluted DNA was collected and immediately stored at -20 °C (Umar et al., 2017).

Constitution of Cox1 gene primer for polymerase chain reaction

A total volume of 707.02 μ L and 648.66 μ L of sterile water was added to Cox1 forward and Cox reverse gene primers, respectively, following the manufacturer's instructions/guidelines. The primer was thoroughly mixed by a vortex machine and concentrated by a centrifuging machine.

Polymerase Chain Reaction (PCR) mix

A working solution of 20 samples was made using 12.5 μ L of Master Mix (one Taq), 1 μ L of forward and reverse gene primer, 3 μ L of the extracted DNA, and 7.5 μ L of sterile water.

Amplification by Polymerase Chain Reaction (PCR)

The extracted DNA was amplified by Cytochrome oxidase sub-unit 1 primer (Cox1 mitochondrial gene of *Schistosoma haematobium*) forward 5'-CCTATGGGTGGTGTATCC-3' and Reverse 5'-ACACGAGACCCACAGCTTTT-3' (Umar et al., 2017). The PCR mix used includes 6 μ L of the DNA template, 15 μ L of nuclease-free water, 25 μ L

of master mix, and 2 μ L of each primer. This reaction was carried out on a thermal cycler (Applied Biosystems, Japan). The PCR protocol included an initial denaturation step at 95 °C for 3 minutes, followed by 35 cycles of denaturation at 94 °C for 30 seconds, annealing at 45 °C for 30 seconds, and extension at 72 °C for 1 minute, with a final extension step at 72 °C for 5 minutes. The amplified DNA fragments were observed on a 1.5% agarose gel stained with a Safe View.

Gel electrophoresis

Electrophoresis of PCR amplicons was conducted for size separation using a 1.5% agarose gel. To prepare the agarose gel, 1.5 g of agarose was dissolved and boiled in 100 mL of sterile water along with 2 mL of TAE buffer solution. The gels were cooled to approximately 45 °C before adding and mixing 10 μ L of Safe View, then poured into the electrophoresis chamber equipped with combs. After the gel had become firm, the PCR product was placed into the wells that had been made. Electrophoresis was conducted at a voltage of 100 V for 45 minutes. The DNA was observed and captured in a photograph using an ultraviolet light machine, and the resolution was at 338 bp.

DNA Sequencing and Phylogenetic Analysis

The PCR products were sequenced using Sanger sequencing at Inqaba Biotech Laboratory, Ibadan, Nigeria. Sequence alignment and BLAST analysis were conducted on the National Centre for Biotechnology Information (NCBI) website, followed by phylogenetic analysis using MEGA 11 software.

Statistical Analysis

Descriptive statistics were applied in IBM SPSS Version 29.0.2.0 (20) to estimate the average prevalence of *Schistosoma haematobium* in the study area. The association between factors and prevalence was determined using a goodness-of-fit test and log-linear model selection. If the P-value was less than or equal to 0.05, it was deemed to be statistically significant.

RESULT AND DISCUSSION

Results

Places and population of the study

An investigation of urinary schistosomiasis and potential risk factors was carried out on individuals living in the riparian areas of Bauchi State. The places covered in Shira LGA were Zigau and Fadamar Andiwa, while those in Jama'are LGA were Kofar Gabas and Nassarawa (Figures 1 and 2). A total of 321 individuals from the two LGAs were enlisted in the study, made up of 220 and 101 from Shira and Jama'are, respectively.

The socio-demographic features of the participants in the study

The 321 respondents in riparian areas of Bauchi State were 4-65 years old, with a mean of 15.1 ± 9.2 years and both median and modal age ranges of 11-20 years. There were 244 (76.0%) males and 77 (24.0%) females, with half of them (50.8%) having no formal education. Two hundred and one (62.6%) of them depend on the borehole as the household's water source (Table 1).

Urine quality

Half of the respondents (55.1%) reported drops of blood toward the end of urination; 74.1% reported pain during urination, and 35.5% had been dewormed within the last 1 year (Table 2).

Urinalysis of the urine samples indicated leukocyturia (25.2%), nitrituria (3.1%), proteinuria (14.6%), 11.5% of the urine were alkaline, haematuria (28.7%), ketonuria (3.7%), bilirubinuria (0.6%) and glucosuria (4.4%) (Table 3).

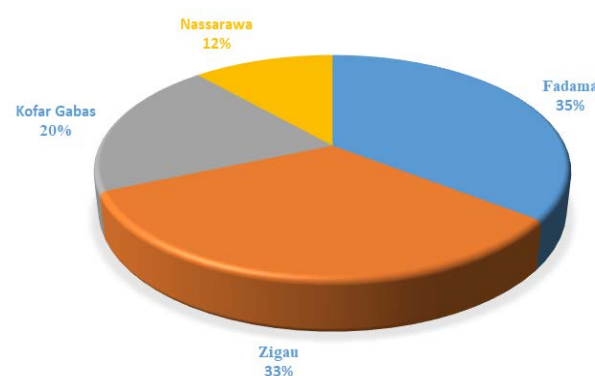


Figure 1: Distribution of the respondents and location investigated for urinary schistosomiasis in Bauchi State, Nigeria

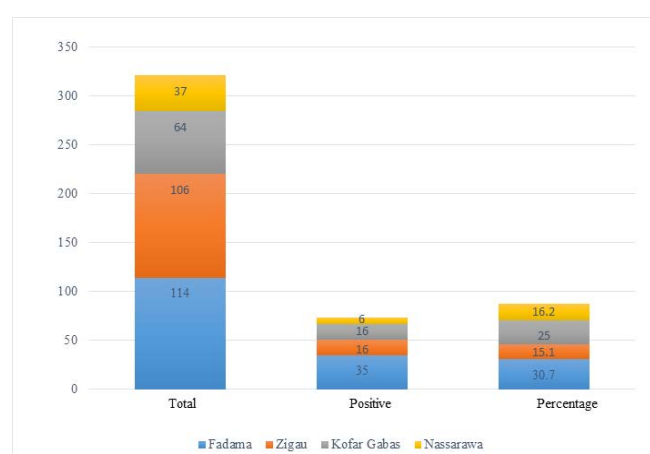


Figure 2: Frequency of *Schistosoma* eggs detection in urine samples of the respondents in the riparian areas of Bauchi State, Nigeria

Table 1: Socio-demographic characteristics of the respondents

Characteristics		Frequency	Percentage
Age	≤10	106	33.0
	11-20	170	53.0
	21-30	22	6.9
	31-40	12	3.7
	>40	11	3.4
Gender	Male	244	76.0
	Female	77	24.0
Marital Status	Single	288	89.7
	Married	27	8.4
	Divorced	6	1.9
Education	No formal education	163	50.8
	Primary	79	24.6
	Secondary	51	15.9
	Tertiary	28	8.7
Occupation	Skilled/employed	26	8.1
	Semi-skilled	37	11.5
	Unskilled	84	26.2
	Farming	24	7.5
	Fishing	60	18.7
	Others (e.g students)	90	28.0
L. G. A of residence	Jama'are	101	31.46
	Shira	220	68.54
Duration of residency near river	0-1	54	16.9
	2-3	125	38.9
	≥4	142	44.2
Household's source of water	Public water supply	91	28.3
	Borehole	201	62.6
	Well	25	7.8
	Stream/river	4	1.3
Reason for going to the river	Washing clothe	76	23.7
	Fishing	65	20.3
	Bathing/swimming	125	38.9
	Irrigation farming	30	9.3
	Others (e.g collecting water for livestock)	25	7.8
Frequency of coming in contact with the river	Occasionally	109	34.0
	Monthly	28	8.7
	Weekly	105	32.7
Season more frequent to river	Daily	79	24.6
	Dry	297	92.5
	Rainy	24	7.5

Table 2: Urine characteristics of the respondents

Characteristics		Frequency	Percentage
Blood drops toward the end of urination	No	144	44.9
	Yes	177	55.1
Pain during urination	No	83	25.9
	Yes	238	74.1
Deworming within the last 1 year	No	207	64.5
	Yes	114	35.5
Genital washing after urination	No	6	1.9
	Yes	315	98.1

Table 3: Biomarkers of urinary disorder in the urine of respondents

Analyte		Frequency	Percentage
Leukocytes (cell/μL)	Negative	240	74.8
	Trace	45	14.0
	Moderate 70-125	36	11.2
Nitrite	Negative	311	96.9
	Positive	10	3.1
Urobilinogen (μmol/L)	Normal 3.2	289	90.0
	Normal 16	32	10.0
Protein (g/L)	Negative	274	85.4
	Trace ±	20	6.2
	0.3-1.0	27	8.4
pH	<7	235	73.2
	7.0	49	15.3
	>7.5	37	11.5
Blood (cell/μL)	Negative	229	71.3
	Positive	30	9.4
	Large 80-200	62	19.3
Specific gravity	1.000-1.010	263	82.0
	1.015-1.030	58	18.0
Ketone (mmol/L)	Negative	309	96.3
	Trace 0.5	12	3.7
Bilirubin	Negative	319	99.4
	Small +	02	0.6
Glucose (mmol/L)	Negative	307	95.6
	Trace 5	11	3.4
	15+	03	0.9

Prevalence of urinary schistosomiasis and socio-demographic characteristics of the respondents

The prevalence of urinary schistosomiasis was documented in all the towns/villages investigated in the research area. *Schistosoma* eggs (terminal spine) (Plate 1) were microscopically observed in 73 (22.7%) out of the 321 urine samples. A higher prevalence was recorded in females (31.2%) than in males (20.1%) (Table 4). The overall prevalence of urinary schistosomiasis in riparian areas of Bauchi State was 22.7%.

Relationship between prevalence of urinary schistosomiasis and socio-demographic indices of the respondent

The prevalence of urinary schistosomiasis in riparian areas of Bauchi state showed a significant association with age ($P=0.021$), with a higher prevalence among children within the age group 11-20 years. The occurrence of urinary schistosomiasis was significantly associated with gender (female greater than male, $P=0.048$), education ($P=0.001$), Household's source of water ($P=0.001$), Reason for going to the river ($P=0.001$), and frequency of coming in contact with the river ($P=0.001$) (Table 4).

Relationship between the prevalence of urinary schistosomiasis and urine characteristics of the respondents

Prevalence of urinary schistosomiasis in the study area was significantly associated with blood drops towards the end of urination ($\chi^2=45.224$, $P<0.001$), pain during urination ($\chi^2=18.265$, $P<0.001$) and deworming within last 1 year ($\chi^2=22.327$, $P<0.001$), (Table 5).

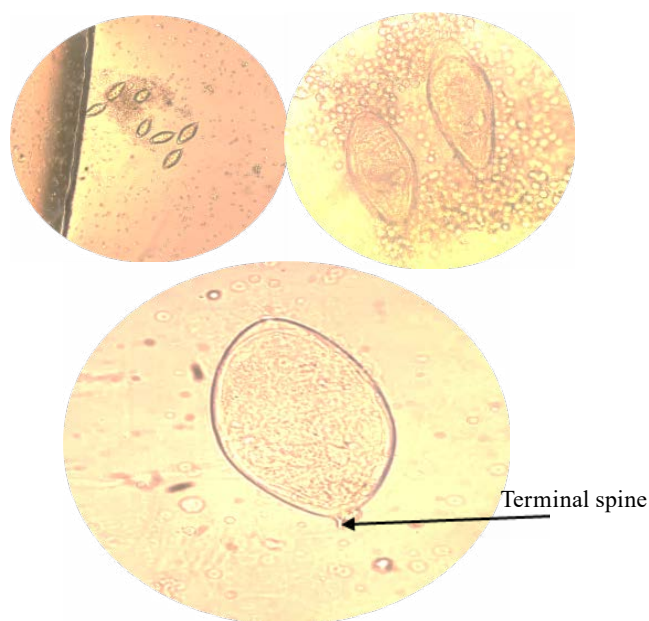


Plate 1: Microscopic image of *Schistosoma haematobium* ova at x400 magnification from this study

Table 4: Relationship between prevalence of urinary schistosomiasis and socio-demographic indices of the respondents

Characteristics		Total	<i>Schistosoma haematobium</i> Eggs in urine (%)		Loglinear Model Selection	Pearson Chi-square
			Negative	Positive		
Age (years)	≤10	106	92 (86.8)	14 (13.2)	$\chi^2=11.53$	$\chi^2=10.967$
	11-20	170	120 (70.6)	50 (29.4)	*P=0.021	*P=0.027
	21-30	22	19 (86.4)	3 (13.6)		
	31-40	12	9 (75.0)	3 (25.0)		
	>40	11	8 (72.8)	3 (27.2)		
Gender	Male	244	195 (79.9)	49 (20.1)	$\chi^2=3.90$	$\chi^2=4.095$
	Female	77	53 (68.8)	24 (31.2)	*P=0.048	*P=0.043
Marital Status	Single	288	226 (78.5)	62 (21.5)	$\chi^2=7.556$	$\chi^2=6.804$
	Married	27	16 (59.3)	11 (40.7)	*P=0.023	*P=0.033
	Divorced	6	6 (100.0)	0 (0)		
Education	No formal education	163	128 (78.5)	35 (21.5)	$\chi^2=17.810$	$\chi^2=16.086$
	Primary	79	66 (83.5)	13 (16.5)	*P=0.001	*P=0.003
	Secondary	51	30 (58.8)	21 (41.2)		
	Tertiary	28	24 (85.7)	4 (14.3)		
Occupation	Skilled/employed	26	23 (88.5)	3 (11.5)	$\chi^2=10.370$	$\chi^2=10.070$
	Semi-skilled	37	28 (75.7)	9 (24.3)	P=0.065	P=0.073
	Unskilled	84	70 (83.3)	14 (16.7)		
	Farming	24	21 (87.5)	3 (12.5)		
	Fishing	60	45 (75.0)	15 (25.0)		
	Others (e.g students)	90	61 (67.8)	29 (32.2)		
L. G. A of residence	Jama'are	101	79 (78.2)	22 (21.8)	$\chi^2=0.053$	$\chi^2=0.053$
	Shira	220	169 (76.8)	51 (23.2)	P=0.818	P=0.818
Duration of residency near the river	0-1	54	44 (81.5)	10 (18.5)	$\chi^2=3.286$	$\chi^2=3.303$
	2-3	125	101 (80.8)	24 (19.2)	P=0.193	P=0.192
	≥4	142	103 (72.5)	39 (27.5)		
The household's source of water	Public water supply	91	63 (69.2)	28 (30.8)	$\chi^2=15.513$	$\chi^2=16.429$
	Borehole	201	169 (84.1)	32 (15.9)	*P=0.001	**P=<0.001
	Well	25	14 (56)	11 (44.0)		
	Stream/river	4	2 (50)	2 (50.0)		
Reason for going to the river	Washing clothes	76	58 (76.3)	18 (23.7)	$\chi^2=13.666$	$\chi^2=8.326$
	Fishing	65	48 (73.8)	17 (26.2)	*P=0.008	P=0.139
	Bathing/swimming	125	97 (77.6)	28 (22.4)		
	Irrigation farming	30	24 (80.0)	6 (20.0)		
	Others	25	21 (84)	4 (16)		
Frequency of coming in contact with the river	Occasionally	109	97 (89.0)	12 (11.0)	$\chi^2=17.763$	$\chi^2=17.042$
	Monthly	28	23 (82.1)	5 (17.9)	*P=0.001	P=0.002*
	Weekly	105	77 (73.3)	28 (26.7)		
	Daily	79	51 (64.6)	28 (35.4)		
Season more frequent to river	Dry	297	228 (76.8)	69 (23.2)	$\chi^2=0.717$	$\chi^2=1.665$
	Rainy	24	20 (83.3)	4 (16.7)	P=0.397	P=0.435

*Significant (i.e P<0.05)

**Highly Significant (P≤0.001)

Table 5: Relationship between prevalence of urinary schistosomiasis and urine characteristics of the respondents

Characteristics		Total	<i>Schistosoma haematobium</i> Eggs in urine (%)		Loglinear Model Selection	Pearson Chi-square
			Negative	Positive		
Blood drops toward the end of urination	No	144	135 (93.7)	9 (6.3)	$\chi^2=45.224$ **P<0.001	$\chi^2=40.425$ **P<0.001
	Yes	177	113 (63.8)	64 (36.2)		
Pain during urination	No	83	77 (92.8)	6 (7.2)	$\chi^2=18.265$ **P<0.001	$\chi^2=15.404$ **P<0.001
	Yes	238	171 (71.8)	67 (28.2)		
Deworming within the last 1 year	No	207	144 (69.6)	63 (30.4)	$\chi^2=22.327$ **P<0.001	$\chi^2=19.916$ **P<0.001
	Yes	114	(104) 91.2	10 (8.8)		
Genital washing after urination	No	06	04 (66.7)	2 (33.3)	$\chi^2=0.336$ P=0.562	$\chi^2=0.366$ P=0.545
	Yes	315	244 (77.5)	71 (22.5)		

**Highly significant (P≤0.001)

Relationship between prevalence of urinary schistosomiasis and biomarkers in the urine of the respondents

Table 6 shows the results of urinalysis as biomarkers for urinary disorders among the patients. Prevalence of schistosomiasis was found to be significantly associated with leukocytes ($\chi^2=104.85$, P<0.001), protein ($\chi^2=15.980$, P=0.007), and blood ($\chi^2=213.818$, P<0.001). Consequently, there was no significant association of urinary schistosomiasis with the presence of nitrite, uroilinogen, specific gravity, bilirubin, ketone, and glucose in urine (Table 6).

Molecular characterization of *Schistosoma haematobium* in urine

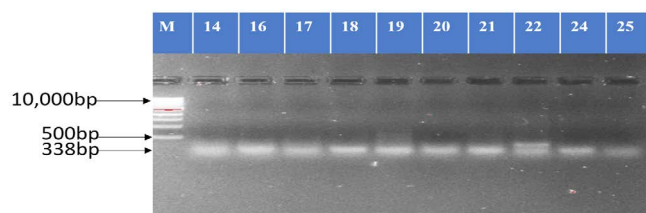
Out of the 25 extracted DNA samples from urine samples, because of limited resources, ten (10) of the DNA samples were found to be of good quality. The

PCR amplification of the 10 DNA using the COX1 gene primer is presented in Plate 3.2. Ten of the DNA samples with single bands were sequenced, and the BLAST result is presented in Table 7. The BLAST produced ten (10) *Schistosoma haematobium* nearest relatives. Moreover, *S. haematobium* isolates were found to belong to a variety of nearest relatives, an indication of genetic diversity (Table 7).

Phylogenetic comparison of the *Schistosoma haematobium* isolates grouped them into two clades, and the isolates from Bauchi State were evenly distributed into two clades (Figure 3). The *Schistosoma haematobium* isolates from the LGAs were compared constructively with *Schistosoma haematobium* isolates from Genbank (downloaded from NCBI), Figure 4.

Table 6: Relationship between prevalence of urinary schistosomiasis and biomarkers in the urine of the respondents

Analyte		Total	<i>Schistosoma haematobium</i> Eggs in urine (%)		Loglinear Model Selection	Pearson Chi-square
			Negative	Positive		
Leukocytes (cell/ μ L)	Negative	240	220 (91.7)	20 (8.3)	$\chi^2 = 104.85$	$\chi^2 = 1.159 \times 10^2$
	Trace 15	45	19 (42.2)	26 (57.8)	**P<0.001	**P<0.001
	Moderate ≥ 70	36	9 (25.0)	27 (75.0)		
Nitrite	Negative	311	240 (77.2)	71 (22.8)	$\chi^2 = 0.045$	$\chi^2 = 0.449$
	Positive	10	8 (80.0)	2 (20.0)	P= 0.831	P= 0.834
Urobilinogen (μ mol/L)	Normal 3.2	289	224 (77.5)	65 (22.5)	$\chi^2 = 0.101$	$\chi^2 = 0.103$
	Normal 16	32	24 (75.0)	8 (25.0)	P= 0.999	P= 0.748
Protein (g/L)	Negative	274	222 (81.0)	52 (19.0)	$\chi^2 = 15.980$	$\chi^2 = 10.312$
	Trace $\pm$	20	13 (65.0)	7 (35.0)	*P= 0.007	**P= <0.001
	0.3-1.0	27	13 (48.1)	14 (51.9)		
Ph	<7	235	183 (77.9)	52 (22.1)	$\chi^2 = 2.783$	$\chi^2 = 2.516$
	7.0	49	35 (71.4)	14 (28.6)	P= 0.836	P= 0.774
	>7.5	37	30 (81.0)	7 (19.0)		
Blood (cell/ μ L)	Negative	229	224 (97.8)	5 (2.2)	$\chi^2 = 213.818$	$\chi^2 = 2.191 \times 10^2$
	Small	30	16 (53.3)	14 (46.7)	**P<0.001	**P<0.001
	Large 80-200	62	8 (12.9)	54 (87.1)		
Specific gravity	1.000-1.010	263	204 (77.6)	59 (22.4)	$\chi^2 = 3.658$	$\chi^2 = 2.770$
	1.015-1.030	58	44 (75.9)	14 (24.1)	P= 0.723	P= 0.837
Ketone (mmol/L)	Negative	309	240 (77.7)	69 (22.3)	$\chi^2 = 0.730$	$\chi^2 = 0.796$
	Trace 0.5	12	8 (66.7)	4 (33.3)	P= 0.981	P= 0.372
Bilirubin	Negative	319	247 (77.4)	72 (22.6)	$\chi^2 = 0.711$	$\chi^2 = 0.851$
	Small +	02	1 (50)	1 (50)	P= 0.871	P= 0.356
Glucose (mmol/L)	Negative	307	237 (77.2)	70 (22.8)	$\chi^2 = 0.313$	$\chi^2 = 0.322$
	Trace 5	11	9 (81.8)	2 (18.2)	P= 0.997	P= 0.851
	15+	03	2 (66.7)	1 (33.3)		

*Significant (i.e P $\leq$ 0.05)**Highly Significant (P $\leq$ 0.001)**Plate 2:** Product of PCR amplification of DNA extract of sediments of *Schistosoma* egg-positive urine samples using (COX 1) gene**Key:**

M: Molecular Standard Marker
 14-25 well: positive sample by microscopy

Table 7: BLAST report of *Schistosoma haematobium*

Sample Code	Nearest Relatives	Homology	Accession Number
Sample 1	<i>Schistosoma haematobium</i> isolate RT16.1_NdgR.5 cytochrome c oxidase subunit I (COX1) gene, partial cds; mitochondrial	100.00%	MT579447.1
Sample 2	<i>Schistosoma haematobium</i> isolate Sh1_Group1 cytochrome c oxidase subunit I (COX1) gene, partial cds; mitochondrial	100.00%	MT380523.1
Sample 3	<i>Schistosoma haematobium</i> isolate Sh4 cytochrome c oxidase gene, partial cds; mitochondrial	100.00%	MK757165.1
Sample 4	<i>Schistosoma haematobium</i> isolate Sh7 cytochrome c oxidase gene, partial cds; mitochondrial	100.00%	MK757168.1
Sample 5	<i>Schistosoma haematobium</i> haplotype H1 cytochrome oxidase subunit I (cox1) gene, partial cds; mitochondrial	100.00%	MK358853.1
Sample 6	<i>Schistosoma haematobium</i> isolate SE2b cytochrome c oxidase subunit I (cox1) gene, partial cds; mitochondrial	100.00%	JQ397332.1
Sample 7	<i>Schistosoma haematobium</i> isolate SE1 cytochrome c oxidase subunit I (cox1) gene, partial cds; mitochondrial	100.00%	JQ397330.1
Sample 8	<i>Schistosoma haematobium</i> isolate Sh3_Group1 cytochrome c oxidase subunit I (COX1) gene, partial cds; mitochondrial	100.00%	MT380524.1
Sample 9	<i>Schistosoma haematobium</i> isolate sample A207 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial	100.00%	MK293624.1
Sample 10	<i>Schistosoma haematobium</i> isolate EG1 cytochrome c oxidase subunit I (cox1) gene, partial cds; mitochondrial	100.00%	JQ397368.1

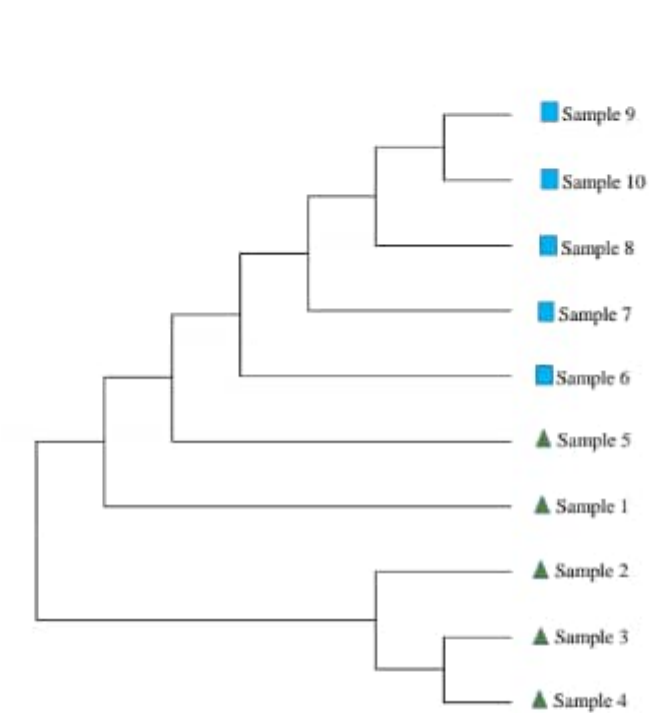


Figure 3: Phylogenetic relationship of *Schistosoma haematobium* from urine of infected individuals from Shira and Jama'are LGA, Bauchi State, Nigeria

Key:

■ Jama'are, Bauchi

▲ Shira, Bauchi

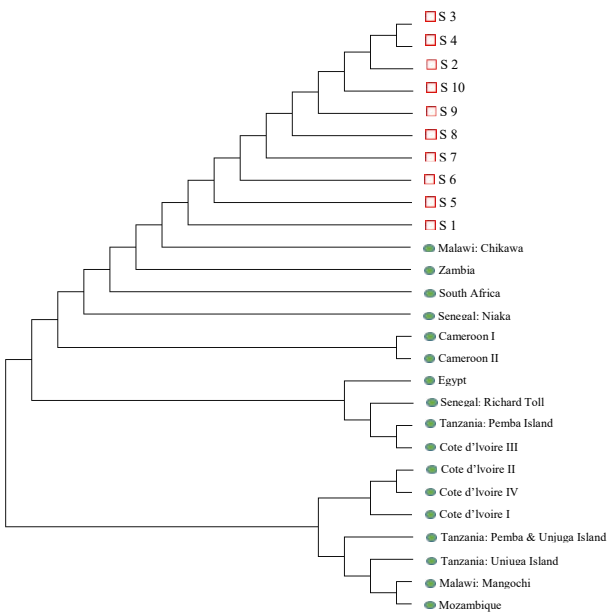


Figure 4: Phylogenetic comparison of *Schistosoma haematobium* from the urine of infected individuals from Bauchi State, Nigeria, with those from Genbank

Key:

■ Bauchi, Nigeria

● Genbank

DISCUSSION

The present study conducted in riparian community dwellers of Bauchi State has established the existence of *Schistosoma haematobium* infection in the State. The infection was found in 22.7% of people living in the riparian area of Bauchi State, indicating a moderate level of risk within the community (WHO 2011) (more than 10% but less than 50%). The recorded prevalence of urinary schistosomiasis was similar to the research carried out in different regions of Nigeria, such as Bauchi State (29.7%) (Mohammed *et al.*, 2022), Jigawa State (20.3%) (Alhaji *et al.*, 2021), and Ondo State, Nigeria (23.77%) (Kone *et al.*, 2023).

The prevalence in the present study is higher than the findings of previous researchers in Nigeria; 14.34% was reported in Ekiti State (Oyedemi *et al.*, 2022), 13.7% in Cross River State (Opara *et al.*, 2021) and 15.20% in Osun State (Alade *et al.*, 2023). The variations between the present finding and those of earlier researchers can be attributed to the variations in the level of exposure to the infection, knowledge about the infection and the different environmental conditions needed for the snails that act as hosts for the parasites to reproduce.

However, the current prevalence of urinary schistosomiasis is lower compared to other studies conducted in various regions of Nigeria, such as Ebonyi State 68.00% (Okpeta and Ani, 2024), Oyo State 45.60% (Odaibo *et al.*, 2024) and Kano State, 53.30% (Ahmad and Bagwai, 2024).

Age is commonly acknowledged as a key determinant of the intensity of an infection. This study found that young adults (11-20 years) and older age (greater 40 years) were more susceptible to *Schistosoma* infections and the disease was highest amongst children within (11-20 years) this is analogous to the findings of Alhaji *et al.* (2021); Folahan *et al.* (2021); Umoh *et al.* (2020) who have separately stated that young adults are more at risk of infection. This is consistent with the overall pattern seen in Nigeria, where the prevalence and severity of infection rise as individuals get older and reach a peak in school-aged children, who spend a lot of time playing in and fetching water (Bishop *et al.*, 2023). Consequently, these age categories include highly energetic youths who frequently participate in swimming, athletics, and river fishing. This implies that 53.3% of schistosomiasis cases in the communities were children of school age. The educational qualification of the participants showed that 24.6% had primary education, 15.1% had secondary education, and only 8.9% had tertiary education. This lack of formal education was considered a potential risk factor, as individuals without proper knowledge may

face various challenges. In other terms, individuals with only primary (24.6%) and secondary (15.9%) education accounted for 40.5% of schistosomiasis cases. Based on the higher infection rate reported among school-aged children, the WHO recommendation is to use the prevalence of school-aged children (SAC) to determine the frequency of chemotherapeutic treatment with Praziquantel (WHO, 2022). In this, the age group had a significant association with the prevalence of infection in the riparian community of Bauchi State. The higher rate of urinary schistosomiasis recorded among the (greater than 40 years) age group, with a prevalence of 27.2%, can be linked to their occupation, which may have necessitated their frequent contact with water. Increased farming and fishing activities in a cercariae-infected/contaminated environment are capable of increasing the prevalence of schistosomiasis (Iduh and Bwari, 2021). Needful to note is the fact that the occurrence rate of the infection was found to be highest among those whose reason for going to the river/stream was for fishing (26.2%) and farming (20%) activities.

It is known that schistosomiasis can infect people when they interact with snail-infested water. Urinary schistosomiasis risk factors include direct contact with open freshwater while bathing/swimming (Mohammed *et al.*, 2022). The majority (62.6%) of the respondents used boreholes as the household's source of water. This validated the initiative of the SDG programme of providing clean water to reduce NTDs. In this study, the highest occurrence of the infection was documented among those who used the river as a household's source of water compared to those who used boreholes and public water supply. There is a statistically significant association between the prevalence of urinary schistosomiasis and household sources of water. Although the occurrence of urinary schistosomiasis was not dependent on the season, 92.5% of the respondents visited the river during the dry season, which is characterized by hot weather. Due to water scarcity and the necessity to escape the intense heat, individuals might visit water bodies more frequently.

Frequent exposure to contaminated water posed a significant risk for acquiring urinary schistosomiasis, as the highest prevalence of (35.4%) was recorded in respondents who visited the river daily. The current findings align with the research that identified regular exposure to untreated water as a contributing factor to the spread of schistosomiasis in Zimbabwe (Nyati-Jokomo & Chimbari, 2017). Additionally, it supported the conclusions by documenting that activities connecting individuals to water were closely linked to the spread of the infection (Hajissa *et al.*, 2018). Furthermore, the

findings from research support the idea that engaging in activities such as swimming and fishing, which involve prolonged exposure to water, increases the risk of transmitting schistosomiasis (Angora *et al.*, 2019; Amuta *et al.*, 2020). The present study area is known for its sizable river, which is utilized for everyday tasks like domestic chores, swimming/bathing, agriculture, and fishing.

Gender was identified as a contributing factor to the risk of contracting urinary schistosomiasis, as the majority of cases of urinary schistosomiasis occurred among a specific gender was in female (31.2%) than male (21.5%) respondents, this report supports the findings of the study carried out in certain regions of Nigeria and South Africa showed that the females were more infected than the males (Lawiye *et al.*, 2020; Maseko *et al.*, 2023), when females in such endemic communities engaged in more water-related activities than males. The study also found that, regardless of their cultural or religious beliefs, women were free to engage in activities that could expose them to infection. Females have a higher occurrence of the disease, possibly due to inadequate healthcare, limited knowledge about how the disease spreads, economic challenges, and the heavy burden of domestic responsibilities like fetching water for animals, washing clothes, and working in water (Maseko *et al.*, 2023). The significant number of cases in females may also be linked to the fact that many elementary school students regularly cross the river for their daily education, explore peri-urban regions, and engage in swimming activities. This goes against the general pattern in this region of the nation, where men are more likely than women to participate in activities that put them at risk of contracting schistosomiasis. This may be because women in this society were tasked to participate in water-related tasks like fetching water and washing dishes and clothes, particularly near the river, more often than men. Females were likely at a higher risk of contracting urinary schistosomiasis compared to males due to this. Male and female infections differed significantly from one another. The connection between gender and susceptibility to infection is uncertain and depends on the cultural context of the individuals (Odaibo *et al.*, 2024; Omondi *et al.*, 2021; Corstjens *et al.*, 2020). The extent of the relationship can differ between different regions of the country and during different times of the year, even within the same communities that are regularly affected (WHO, 2022). The reasons behind certain situations can often be attributed to variations in the social and professional responsibilities of each individual, which can impact their interaction with water. Factors such as how often, how long, and how deeply one comes into contact with water may be influenced by age and cultural practices

(Odaibo *et al.*, 2024). Hence, any comprehensive plan to prevent and manage schistosomiasis should be based on the specific data gathered from each community or region where the disease is prevalent.

However, some other studies in different regions of Nigeria (Ojo *et al.*, 2021; Onyekwere *et al.*, 2022; Abbas *et al.*, 2023; Okpeta and Ani, 2024) and other endemic countries in Africa, such as Malawi (Nyangulu *et al.*, 2022), Sierra Leone (Kargbo-Labour *et al.*, 2024) and Cameroon (Sumbele *et al.*, 2021), research has demonstrated that male individuals consistently exhibited a greater prevalence and average intensity of infection compared to female individuals, especially during periods when males engaged in activities that involved higher levels of exposure to water.

The current study revealed that the respondents who experience blood toward the end of urination (36.2%) and pain during urination (28.2%) have a higher prevalence of urinary schistosomiasis than those who do not. This corroborates previous research in Nigeria (Alexander *et al.*, 2021) and Ethiopia (Deribew *et al.*, 2022). The presence of red-coloured urine indicated haematuria and blood accompanied by pain was the symptom or indicator connected to the disease (WHO, 2022). This study shows a statistically significant association between blood at the end of urination, pain during urination (dysuria), and prevalence of urinary schistosomiasis. This agrees with the findings of a study conducted by Dahal *et al.* (2023) in North-central Nigeria. Urinary schistosomiasis may cause symptoms similar to those of a urinary tract infection, such as abdominal discomfort, diarrhoea, and blood in the urine. In more serious cases, liver enlargement is common and often leads to the accumulation of fluid in the abdomen and high blood pressure in the abdominal blood vessels. In such circumstances, the spleen may also expand (WHO, 2022). In this research work, urinalysis using Combi 10 revealed a statistically significant association of urinary schistosomiasis with leukocytes, protein, and blood, implicating these biomarkers to confirm the disease and can be used for diagnosis as well as early detection of urinary schistosomiasis. This agrees with previous studies that reported these biomarkers can be used in the identification of urinary schistosomiasis (Archer *et al.*, 2020; Vaillant *et al.*, 2024; Chikwendu *et al.*, 2022). Consequently, if not diagnosed on time, it can have a devastating impact on the urinary tract, which is often unacknowledged and unevaluated. This omission could have led to end-stage renal failure and death (Duarte *et al.*, 2020).

Findings from the current study correspond to the fact that urinary schistosomiasis poses a major risk to life in tropical and sub-tropical countries, with the rising number of infections. Nigeria is regarded as the country with the highest schistosomiasis endemicity, with approximately 101 million individuals susceptible to contracting the infection, and an estimated 29 million people already affected (Mohammed *et al.*, 2022). Schistosomiasis continues to be a significant issue for public health in many countries, leading to both illness and death, particularly in sub-Saharan Africa (Abbas *et al.*, 2023). School-age children are particularly vulnerable to *S. haematobium* infections, being among the high-risk groups. Urinary schistosomiasis is more widespread among riparian rural dwellers than the imagined number (Oyedeji *et al.*, 2022).

The health implications for the infected human population are enormous and vary from haematuria and dysuria to pain in the supra-pubic region (Lawiye *et al.*, 2020). In addition to this long-term effects of urinary schistosomiasis include bladder wall pathology, which has been associated with cancer of the bladder, hydronephrosis, risk of anaemia because of the terminal release of blood at micturition, the anaemia has various complications leading to inadequate cognitive growth, lack of essential nutrients, and delayed puberty in children, retarded or stunted growth in children, poor disposition to work and play, school performance and gross reduction in work capacity and productivity (Isegbe *et al.*, 2017). Contributing factors to this sustained endemicity are: favourable climatic conditions for the proliferation of the snail host and a gross lack of potable water. The present findings' moderate prevalence of urinary schistosomiasis indicates significant residential proximity to polluted bodies of water.

Genetic diversity of *S. haematobium* isolated from riparian areas of Bauchi State

The molecular technique is considered the most reliable method for diagnosing a wide range of infections, including urinary schistosomiasis. This research compared the results of conventional PCR (qPCR) on urine samples with the analysis of urine samples using microscopy and PCR in a study involving 10 participants residing in riparian communities in Bauchi State. The ten urine samples that tested positive using microscopy were also positive based on PCR analysis. This indicates that the population living near rivers in Bauchi State is commonly infected with urinary schistosomiasis, which is consistent with earlier research that reported riverine areas are endemic to urinary schistosomiasis in France (Gillardie *et al.*, 2021) and Ghana (Anyan *et al.*, 2020; Armoo *et al.*, 2020).

Polymerase Chain Reaction (PCR) methods have been advocated to improve the direct detection of *Schistosoma*. Microscopy is more cost-effective and easily accessible, but it comes with its own set of difficulties, such as differences in observer interpretation, fluctuations in egg excretion from day to day, and lower accuracy in detecting low levels of infection. Similar to previous studies found more schistosomiasis cases were found by detection of *Schistosoma* DNA in urine via PCR than by microscopy (Pillay *et al.*, 2020). Although PCR offers improved specificity and sensitivity and has the potential to analyse alternative clinical samples like vaginal swabs or sanitary pads, it is limited by its high cost and lack of practical use in the field (Pillay *et al.*, 2020). Nevertheless, recent advancements in the realm of DNA detection suitable for field use appear to be full of potential. Additionally, granulomas caused by *Schistosoma* in the bladder may not always allow for the excretion of eggs, leading to potential undetected infections. This limitation is a downside of using urine PCR testing (Osakunor *et al.*, 2022).

The first use of qPCR for the diagnosis of *Schistosoma* was by Pontes in 2002 (Siqueira *et al.*, 2021). The sensitivity and precision of this technique were shown by the non-appearance of the DNA of other helminth parasites that often cause serious parasitic diseases like ascariasis, ancylostomiasis, taeniasis, and trichuriasis in the tropics (Siqueira *et al.*, 2021). Also, PCR is a valuable technique for identifying *Schistosoma* infection in individuals excreting few eggs and can be used to confirm the results of seropositive (Siqueira *et al.*, 2021). Nevertheless, this method is expensive in this region, necessitates skilled staff, and involves multiple post-DNA amplification steps like gel electrophoresis, which hinders the ease of analysing a large number of samples (Fuss *et al.*, 2021).

The evolutionary history of an organism involves the historical development of its family ancestry down to the species. The family line of development is therefore monitored from their common origin, descendants comprising the order of branching and divergence (Padial *et al.*, 2021). At the molecular level, the connection between organisms or genes is analysed by comparing similar DNA or protein sequences. Variations in the sequences suggest genetic divergence due to molecular evolution occurring over time. Nucleotide sequences of either DNA, RNA, or sequences of amino acids, when extracted through new techniques, are usually the tools for evolutionary studies at the molecular level. This allows for comparative study or analysis of homologous molecules from different organisms, as such it becomes possible to establish the level of similarity

or relatedness, or reveal a hierarchy of relationships in the form of a phylogenetic tree (Padial *et al.*, 2021). The task of establishing evolutionary relatedness within different organisms is very challenging since the living world shows a high level of diversity concerning its species content. This diversity is not only revealed in the morphological traits but also ultra-structural, biochemical, and molecular features. Morphologically related organisms may have pronounced differences in their biochemical and molecular features (Dzhalilova and Makarova, 2020).

All living organisms in its features are either direct or indirect products of developmental changes that it has gone through. It is, therefore, relevant to study and understand the evolutionary history of organisms; this will be expressed in biological terms. To fully understand their developmental history, three types of information are necessary and include: external appearance or characteristics, the genetic composition, and finally, when the similarities between DNA and protein are linked, the derived data or information will be used in tracing the organic development about such organisms and their ancestry or evolutionary lineage, this is expressed graphically in form of the phylogenetic tree (Padial *et al.*, 2021).

On the sequenced amplicons and 10 BLAST isolates from riparian areas of Bauchi States, all showed 100% homology with sequences from the National Centre for Biotechnology Information (NCBI) (Table 3.7). More supportive evidence is the alignment of the sequences seen in the comparison of the sequences from riparian areas of Bauchi State with the isolates from other parts of the globe. All the sequences shared the same ancestry with *S. haematobium*, they appeared in four different clusters. Most of the isolated sequences appeared at the top of the phylogenetic tree suggestive of closer affinity with reported sequences from Malawi (Chikawa), Zambia, South Africa, and Senegal (Niaka). Dissimilarities among sequences indicate genetic divergence as a result of molecular evolution over time (Padial *et al.*, 2021). Other plausible reasons for the highly divergent strains and species in Bauchi State could be a result of the geographical location of the state. The far-reaching implications of this lies in the fact that the only available drug of choice praziquantel will be effective when administered in the State. Schistosomiasis as a disease is assuming great public health importance to both man and animals, more importantly, the absence of a vaccine for preventive purposes is still a factor for consideration (WHO 2022).

The 10 sequenced isolates presented a diverse of 6 strains of *Schistosoma*, including *S. haematobium* Sh (4), *S. haematobium* SE (2), *S. haematobium* RT16.1_NdgR.5 (1), *S. haematobium* haplotype H1 (1), *S. haematobium* A207 (1), and *S. haematobium* EG1 (1). Genetic diversity among *Schistosoma* species has been reported in many endemic regions in Africa (El-Kady *et al.*, 2021). The scarcity of information on genetic diversity in Nigeria might not necessarily be because they are absent but rather the fact that they have not been studied enough, corroborating the report of other workers that genetic diversity among *S. haematobium* has not been thoroughly investigated like *S. mansoni* (Santos *et al.*, 2021).

The phylogenetic tree is an evolutionary relationship among a set or group of organisms (Kapli *et al.*, 2020). In this study, mitochondrial gene (COX1) gene was used as primer for *S. haematobium*. Molecular Evolutionary Genetics Analysis (MEGA 11) software was used for sequence alignment. Reference sequences of different *Schistosoma* species were selected from the NCBI database (<http://www.ncbi.nih.gov>).

CONCLUSION

The present study established the existence of urinary schistosomiasis in riparian areas of Bauchi State (22.7%), Nigeria. The prevalence of urinary schistosomiasis was significantly associated with age, gender, education, the reason for going to the river, frequency of coming in contact with a river, and the household's source of water supply. Biomarkers such as leukocytes, protein, and blood in urine can be used for the early detection of urinary schistosomiasis in riparian areas. Household sources of water and frequency of coming to the river were found to be the risk factors. The study reported the first molecular characterisation of urinary schistosomiasis in Bauchi State. This study provides baseline data that could serve as justifications for the enlistment of Bauchi State into the Schistosomiasis Control Initiative of Nigeria.

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