Human Contamination and Schistosome Infection Intensity in Bulinid and Planorbid Snail Vectors in Kadawa Irrigation Area, Kano State, Nigeria

M. U. Ali\textsuperscript{1*}, U. A. Umar\textsuperscript{2}, A. Yahaya\textsuperscript{1}, M. Lawal\textsuperscript{1} and M. S. Danhassan\textsuperscript{2}

\textsuperscript{1}Department of Biology, Kano University of Science and Technology, Wudil, Kano State, Nigeria.
\textsuperscript{2}Department of Biochemistry, Ahmadu Bello University, Zaria, Nigeria.

Authors' contributions
This work was carried out in collaboration among all authors. Author MUA designed the study and wrote the protocol. Authors MUA and UAU performed the statistical analysis and wrote the first draft of the manuscript. Authors AY and ML managed the analyses of the study. Authors MUA, UAU and MSD managed the literature searches. All authors read and approved the final manuscript.

ABSTRACT

Objectives of the Study: An Epidemiological Research, a cross-sectional study, was conducted to determine the magnitude of human contamination of irrigation canal perimeter as it relates to the prevalence and intensity of schistosome cercarial infection in snail vectors.

Place and Duration of Study: The study was conducted along water canal located within an irrigation area, Kano River Project Phase I, Kadawa, between January and June, 2012.

Methodology: The study area was categorized into Zone of Heavy Contamination (ZHC), Zone of Light Contamination (ZLC) and Zone of Free Contamination (ZFC) based on the density of faecal lumps observed along the canal perimeter using 1 m\textsuperscript{2} quadrat sampling technique. Snail vectors of schistosomiasis were collected from these zones, identified and subjected to cercarial shedding. Brevifurcate apharyngeate cercariae were identified as schistosome cercariae.

Results: Of the 827 snails collected 28.54\% shed schistosome cercariae. The breakdown of...
infection prevalence was 31.37%, 27.69% and 26.26% for ZHC, ZLC and ZFC respectively. Three snail species recovered in the study area, Bulinus globosus, B. rohlfsi and Biomphalaria pfefferi had infection intensity of 8.6, 5.67 and 3.94 respectively, with total mean intensity of 4.67. A Chi-square analysis did not show any significant difference in infection prevalence in the three zones ($\chi^2_{\text{cal.}} = 0.025$, $\chi^2_{0.05} = 5.99$). However, infection intensity was significantly different in the three zones and among the three snail species using analysis of variance (P<0.05).

**Conclusion:** Human environmental contamination with faeces and urine around irrigation canals remains the source of infection to snail hosts and then to humans. It is presumed that contact control through avoidance of defaecation in the open and building of pit latrines near water contact points along irrigation canals will be effective means of drawing a barrier to infection with schistosomes in the study area.

**Keywords:** Human contamination; schistosome cercaria; infection intensity; snail vectors; irrigation canal.

### 1. INTRODUCTION

Human schistosomiasis is a water-based disease and one of the neglected tropical diseases that is more prevalent where there is high frequency of human contact with infested water. Water resource schemes for power generation and irrigation have resulted in the increase in the transmission and outbreaks of schistosomiasis in several African countries [1]. In sub-Saharan Africa schistosomiasis is widespread with foci of high prevalence and high morbidity found adjacent to rivers, lakes and irrigation schemes [2]. The disease epidemiology is attributable to water contact pattern, biology and distribution of the potential snail vectors and the local geographical, geological and climatic conditions [3,4]. Contamination of surface waters or their surrounding with faeces and urine containing schistosome eggs is essential for transmission of the parasite [4]. Humans become infected with schistosome following contact with contaminated water through various water contact activities [5]. A combination of environmental and anthropogenic parameters controls the distribution of schistosomes within a surface water network [6]. Heavy rains aid contamination by carrying the schistosome eggs to water bodies where they can successfully hatch into viable miracidia [7]. A wet climate is an important contributor to water contamination as seen in the decreased viability of S. mansoni eggs exposed to the sun within a few days after fecal deposition. The level of contamination is thus dependent on both direct factors such as defaecation patterns and indirect factors such as rain events, overflowing latrines, and level of community sanitation [7]. Faecal contamination of surface water with schistosome eggs occurs in rural endemic regions with low sanitation infrastructure [7,8]. The continuum of infection is linked to the continuous water contact activities and anthropogenic faecal and urine contamination, coupled with prevailing snail vector population [9]. The bulinid and planorbid snail vectors, implicated in the transmission of human schistosomiasis, have been reported in Kano State and many parts of Nigeria [10,11,12,13,14]. Human activity of gross contamination of the water body perimeter is a major factor for infection of the snails with human schistosome species. Even though snail infection rates may be low, the presence of infected snails portends potential transmission of schistosomiasis. However, marked seasonal fluctuation in snail infections may occur [14]. In the North-western parts of Nigeria, comprising Sokoto, Katsina and Kebbi States, there are about 16 large and many small-scale formal irrigation and many private ones. Here, the general prevalence of urinary schistosomiasis was shown to be 22.3% [15]. In Kano State, Nigeria, a considerable amount of water development projects has been carried out and more are being proposed which will enhance transmission [13]. In a prevalence study in Katsina State, Idris et al. [16] reported infection rates of 12% and 3.3% for S. haematobium and S. mansoni among primary school pupils. Tukur and Galadima [17] reported a prevalence and intensity of S. haematobium infection of 50.9% and 151.0 eggs/10 ml urine respectively, in Bakolori irrigation project area of Zamfara State. They also found that persons aged 10-19 years had the highest prevalence rate of 70.3% and mean intensity of 324.33 eggs/10 ml urine, while those aged 40 years and above had the least prevalence of 20.8%. Adamu et al. [18] reported 41% prevalence for urinary schistosomiasis in Wurno district of Sokoto State, with intensity of 310 eggs/10 ml urine. However, low prevalence and intensity of 5% and 10 eggs/gm stools were recorded for intestinal schistosomiasis. In Kano
and Bauchi States, where a number of irrigation schemes and other water projects have been executed and still more are expected, there was high rate of schistosomiasis recorded from these water projects [19]. Schistosoma haematobium infection prevalence rates in some parts of Kano State and its neighbours have been monitored. Umar [20] recorded as high as 28.4% prevalence rate for S. haematobium among pupils of 8-10 years in Kura Local Government Area of Kano State which is an area that is extensively irrigated as well as being very rich in ponds and rivers. Betterton et al. [10] showed the presence of S. haematobium among the 813 school children and adults from Tomas and Rimin Gado dam areas of Kano State, with prevalence of 26.6% and 36.8% respectively. They observed that the prevalence and intensity of S. haematobium were low and similar in both study areas and no cases of S. mansoni infection were found. The study area is sandwiched between two village communities, Dakasoye and Dorawar Sallau, with a reported overall prevalence of 32.8% and 16.8% for S. haematobium and S. mansoni infections respectively [14]. Ali and Ndams [14] further reported an association between infection prevalence and water contact activities in both communities. This research work reports an investigation on the magnitude of human environmental contamination and its epidemiological implication in relation to the prevalence and intensity of schistosome infections in snail vector population in the study area, with a view to highlighting the role of defaecation in the open in maintaining schistosome infection in susceptible snail vectors.

2. MATERIALS AND METHODS

2.1 Study Area

The study area is an irrigated area lying about 35 km southwest of Kano City (Lat. 11°59'N, Long. 8°30'E) on both sides of Kano-Zaria trunk road. The irrigation water is conveyed from Tiga Dam to the project site through an 18 km-long main canal, which splits into East and West branches of canals and earthen field channels from where water is finally abstracted for crops irrigation using plastic siphon tubes. Canals are designed such that the west branch canal and the lateral canal are lined with the side slopes kept at 1: 1½ and maximum velocity of 1.8 m/s while the earthen distributary canals have side slopes kept at 1:2, with a velocity below 0.3m/s to prevent erosion [21]. The study area is bordered by two villages, Dakasoye (Lat. 11°44'N, Long. 8°25'E) and Dorawar Sallau (Lat. 11°39'N, Long. 8°23'E) within the Kano River Project Phase I (KRP I), which is one of the largest and successful irrigation projects in Nigeria. The study area comprised established communities with the irrigation agriculture-based economy and whose lives are directly or indirectly linked to the water that is constantly present in the irrigation canals.

2.2 Study Design

The study area was categorized into three (3) zones: Zone of Heavy Contamination (ZHC), Zone of Light Contamination (ZLC) and Zone of Free Contamination (ZFC). The categorization was on the basis of the observed level of human faecal and urine contamination during pre-sampling visits to the study area, and the established presence of snail intermediate hosts in the water canals reported in previous studies [9,14]. The degree of contamination was determined by the density of faecal lumps in each zone, using 1 m² quadrat sampling technique. The quadrat was thrown three times at random, on each landing the area covered by it was observed. The number of visible faecal lumps within the quadrat was recorded and the average number of faecal lumps calculated as lumps/m². Selection of contamination zones in the study area was made on the basis of faecal density as: ZHC (>3 lumps/m²), ZLC (1-3 lumps/m²) and ZFC (<1 lump/m²). The distance between ZHC and ZFC was about 2.6 km and that between ZFC and ZLC was roughly 1.2 km. The distance between points of faecal contamination and the edge of the water canal was also determined using meter rule. The study area covers a distance of about 4 km along the water canal and Kano-Zaria Trunk Road, with the direction of the water course from ZHC to ZFC to ZLC. The source of water in the irrigation canal was Tiga Dam. The topography of the three zones, in particular, the vegetation covers and the nature of gradient around the perimeter of the water canal, were also noted. All the three zones were measured approximately 8 m by 150 m to obtain an approximate canal perimeter area of 1200 m² along the water canal. ZHC was located proximal to Kwanar Gafan seasonal Vegetable Market. The people attending the market come from various parts of Nigeria transacting in green vegetables which were harvested from the surrounding irrigation area; although majority were from the neighboring communities. ZLC is located near the town of Dakasoye, where it forms a partial open latrine to some members of
the village community and visiting irrigation farmers, who do not have access to standard latrines during water exposure for occupational or recreational purposes. ZFC interspersed ZHC and ZLC. Throughout the research period, rain boots, protective and disposable hand gloves, and nose cover, were worn during each sampling.
2.3 Snail Collection and Identification

Collection of snails was done between January and June, 2012 directly from 3 or 4 points adjacent to the respective zones of contamination. The peak period of snail abundance and anthropogenic environmental contamination in the hot dry seasons as well as water contact activities reported earlier by [12] and [14], informed the decision of confining the study to six months. The snails were searched from aquatic substrata such as macrophytes, plant twigs, rock surfaces and floating objects and collected by hand picking with the aid of a tea strainer from the three zones (ZHC, ZL and ZFC), during the study period, taking into cognizance of the substrates to which the snails were attached. Protective hand gloves were worn during each sampling. The snails were then transferred to labeled plastic beakers containing the canal water and transported to the laboratory for identification and cercarial shedding. Identification of the snail was based on gross morphology of snail shells as in Brown [22].

2.4 Snail Cercarial Shedding and Counting

Snails were examined for schistosome infection by immersing each snail in 5ml of dechlorinated water in a Petri dish after exposure to light from a lamp-bulb for 2-3 hours according to [11]. The cercariae observed in the water contained in each Petri dish were counted by adopting the method of [23] as follows. Water sample in each Petri dish was passed through 7cm Whatman No.1 filter paper in a Buchner funnel under partial vacuum. Dechlorinated water was used to rinse the Petri dishes to ensure washing out of all shed cercariae. Cercariae trapped on the filter paper were stained and immobilized with Lugol's Iodine (jective) of dissecting microscope. Only the heads of cercariae were counted since tails may become detached during sample preparation [23].

3. RESULTS

3.1 Snail Vector Abundance and Temporal Distribution

The results for the abundance and temporal distribution of snail vector species in the three zones of contamination have been presented in Figs. 1 and 3. There was a monthly variation in snail abundance in the three zones of contamination. Snail count was generally low in the months of January and February, high between the months of March and May and highest in May in the zones of heavy and light contamination. However, snail count dropped in all the three zones in June, although the highest snail count was recorded in April in ZHC, during the research period. Only three species of snail intermediate hosts of human schistosomiasis were recovered in the study area; viz.: Biomphalaria pfeifferi, Bulinus globosus and B. rohlfisi, the former species being predominant in all the three zones.

3.2 Snail Infection Prevalence and Intensity

The prevalence of infection in the snail intermediate hosts was presented in Table 1 and Figs. 2 and 4. The prevalence of schistosome cercarial infection in the snail vectors in the three zones was in the following order: ZHC, 31.37%; ZLC, 27.69% and ZFC, 26.26%, with overall infection prevalence of 28.54% in the study area (Table 1). Fig. 2 showed that the rate of infection with schistosome cercariae followed a spatio-temporal pattern. Infection was highest in the month of May, followed by April and January. The ZHC has the highest infection prevalence in 5 out of 6 months of the study. This is followed by ZFC and ZLC. All the three snail species were infected with schistosome cercariae (Fig. 4). Infection prevalence was highest in Biomphalaria pfeifferi and lowest in Bulinus globosus. Infection in B. pfeifferi was highest in ZHC and lowest in ZLC. Conversely, in B. globosus, infection was highest in ZFC and lowest in ZLC. The order of increasing infection prevalence in B. rohlfisi was: ZFC, ZLC and ZHC. However, there was no statistically significant difference in infection prevalence in the three zones (χ² = 0.025). Tables 2 and 3 revealed the results of the mean intensity of schistosome cercarial infection in the snail species. The mean infection intensities for Bulinus globosus, B. rohlfisi and Biomphalaria pfeifferi were 8.6, 5.67 and 3.94, respectively; with total mean intensity of 4.67. Moreover, infection intensity was significantly different in the three zones and among the three snail species using analysis of variance at P<0.05 (Table 3).
Fig. 1. Temporal distribution of snail vectors in the three zones

Fig. 2. Relative number of infected snails in the three zones

Fig. 3. Relative abundance of snail species in the three zones
Fig. 4. Relative abundance of snail vector species shedding schistosome cercariae

Table 1. Schistosome cercarial infection prevalence in snail vectors

<table>
<thead>
<tr>
<th>Zone of contamination</th>
<th>No. of snail vectors</th>
<th>No. of infected snails</th>
<th>% Infection prevalence</th>
</tr>
</thead>
<tbody>
<tr>
<td>ZHC</td>
<td>306</td>
<td>96</td>
<td>31.37</td>
</tr>
<tr>
<td>ZLC</td>
<td>242</td>
<td>67</td>
<td>27.69</td>
</tr>
<tr>
<td>ZFC</td>
<td>278</td>
<td>73</td>
<td>26.26</td>
</tr>
<tr>
<td>Total</td>
<td>827</td>
<td>236</td>
<td>28.54</td>
</tr>
</tbody>
</table>

$\chi^2 = 0.025$

Table 2. Mean intensity of schistosome cercariae infection

<table>
<thead>
<tr>
<th>Zone of contamination</th>
<th>Snail vector species</th>
<th>Bulinus globosus</th>
<th>Bulinus rohlfsi</th>
<th>Biomphalaria pfeifferi</th>
<th>Mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>ZHC</td>
<td>12.2</td>
<td>8.24</td>
<td>5.19</td>
<td>6.09</td>
<td></td>
</tr>
<tr>
<td>ZLC</td>
<td>6.75</td>
<td>3.94</td>
<td>3.96</td>
<td>4.12</td>
<td></td>
</tr>
<tr>
<td>ZFC</td>
<td>6.83</td>
<td>4.33</td>
<td>2.67</td>
<td>3.29</td>
<td></td>
</tr>
<tr>
<td>Mean</td>
<td>8.6</td>
<td>5.67</td>
<td>3.94</td>
<td>4.67</td>
<td></td>
</tr>
</tbody>
</table>

Table 3. Analysis of variance for snail cercarial infection intensity in the three contamination zones

<table>
<thead>
<tr>
<th>Source of variation</th>
<th>Degree of freedom</th>
<th>Sum of squares</th>
<th>Mean squares</th>
<th>$F_{calculated}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Snail Species</td>
<td>2</td>
<td>33.65</td>
<td>16.83</td>
<td>13.304 $^*$</td>
</tr>
<tr>
<td>Zones of Contamination</td>
<td>2</td>
<td>28.94</td>
<td>14.47</td>
<td>11.439 $^*$</td>
</tr>
<tr>
<td>Error</td>
<td>4</td>
<td>5.06</td>
<td>1.265</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>8</td>
<td>67.65</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

$F_{0.05} (2, 4) = 6.94$; $^*$ = Significant at 5% level of Statistical Significance

4. DISCUSSION

This research reveals a variation in the distribution and abundance of snail intermediate hosts in the three snail sampling zones. There was no marked variation in species distribution in the three zones of contamination. This was similarly reported by [14] that spatio-temporal variation in snail vector distribution and abundance was due to fluctuating seasonal
temperature. Moreover, the snail vectors were observed to be mainly spatially distributed in the littoral zone of the water canal where water flow velocity was lowest, and usually attached to submerged and floating objects as is typical of periphytonic communities. The research recovered three species of snails that serve as intermediate hosts of human schistosomiasis. The snails are *Biomphalaria pfeifferi*, *Bulinus globosus* and *B. rohlfsi*. However, *Biomphalaria pfeifferi* was found to be the most abundant snail intermediate hosts in the study area. The predominance of *Biomphalaria pfeifferi* in the study area was earlier reported by [14].

The observed anthropogenic faecal and urine contamination of the irrigation water canal perimeter has been documented by several researchers [13,14,24]. Lack of standard pit latrines in the study area, especially around the canal perimeter, was the major cause of human contamination activities along the water canal when they go there for one form of water contact activity or the other. Liu et al. [25] suggested provision of sanitary toilets in such settings. The role of environmental contamination in the spread of schistosomiasis is of immense importance since human urine and faecal matter are the sources of infection to the snail vectors. This observation was in agreement with that of Akullian [7] who reported the essentiality of contamination of waterways with human waste, and subsequent exposure to contaminated water for the parasite's continued asexual reproduction in the snail host and sexual reproduction within the mammalian host. Amadou et al. [26] and WHO [1] have linked schistosomiasis to very low standard of hygiene and inadequate potable water supply which may lead to unprotected water contact activity for occupational or recreational purposes. Ali et al. [27] attributed the endemicity of schistosomiasis to lack of basic amenities, indiscriminate disposal of human sewage and high water contact activities with snail-infested water, among other factors. The overall prevalence of schistosome infection in the snail vectors was low (28.54%). This figure was slightly higher than that of [14] who recorded an overall infection prevalence of 20.9% in the snail vectors. This may be attributed to small sample size in this research, and the varying environmental conditions. In addition, Li et al. [8] attributed schistosomiasis prevalence to levels of local surface water contamination contributed by sanitation levels and faecal contamination patterns in humans and domestic animals. They further observed that faecal contamination of surface water with schistosome eggs occurs in rural endemic regions with low sanitation infrastructure. In a cross-sectional study conducted by Abubakar et al. [28], similar observation was reported in which the high prevalence of *S. haematobium* infection (61.9%) was attributed to human contact with contaminated water. These findings were in agreement with [7] who reported that geographic distribution of schistosomes along waterways might be related to human behavior and the geographic extent of human travel than environmental factors alone.

The significant difference in infection prevalence in the three zones observed in this study proves the essential role of anthropogenic activity of faecal and urine contamination of the water canals in the epidemiology of schistosomiasis as well as maintenance of infection in snail hosts which is reliant upon infected urine and faecal matter that come in direct contact with the water peripherally present in the water canal. Similar findings have been reported by [14] who observed that human activity of grossly contaminating the water canal periphery contributed to the increased infection of snail vectors with human schistosome species. This promiscuous contamination is the sole source of human-to-snail transmission which has an attendant effect of maintaining schistosome infection in humans in the study area as a result of water contact with cercariae-infested water. This is contributory to the endemicity of schistosomiasis in the study area, as reported by several researchers [9,13,14,20,28]. Moreover, Akullian [7] further observed that in many endemic areas humans contribute heavily to both the parasite's survival and the resulting burden of disease within the human population through continued faecal and urinary contamination of heavily used waterways. This study revealed a significant difference in infection intensity in the three zones and among the three snail species, namely *Biomphalaria pfeifferi*, *Bulinus globosus* and *B. rohlfsi*. The presence of infected snail vectors in the ZFC might be attributed to the influence of water currents transporting the snail infective larval forms, miracidia, thereby seeding the near and distant snail colonies along the water course, as well as the greater chance of the surrounding contaminated soil to be blown into the water canal by the whirling wind during the hot dry season, when the water contact and contamination activities of the surrounding communities were highest, and when the water canal accommodates a higher population of...
susceptible snail vectors. This finding thus, strengthens the epidemiologic importance of anthropogenic activity of environmental contamination with human excreta in snail-to-human and human-to-snail transmission cycle. The infectivity and vector competence of the three snail intermediate hosts recovered, enabled them to be effective in maintaining transmission of schistosomiasis all year round. In this regard, bulinid species seemed to be more vector competent, thus connoting a higher prevalence of urinary schistosomiasis in the study area [9,13,20].

5. CONCLUSION

The human ‘contaminatory’ behavior of the endemic communities around the study area and the lack of measures to improve sanitary conditions continue to predispose the community to infection and re-infection with schistosome parasites. The permanence of water in the study area provides an opportunity for irrigation agriculture and other water contact activities for domestic and recreational purposes. As long as the faecal and urine contamination of the canal perimeter continues, schistosome eggs from infected subjects will seed the surface water that harbors susceptible snail population which maintains schistosomiasis transmission in the study area.

6. RECOMMENDATIONS

For effective and lasting control of schistosomiasis, contact control strategies should be employed as a preventative tool for drawing a barrier between human definitive host and schistosome parasite. Therefore, based on the findings in this research we recommend the following:

i. Mass drug administration (MDA) of anti-schistosomal regimen, Praziquantel, following mass screening should be implemented once a year, targeting school-age children in all schistosomiasis-endemic areas in order to provide mass prevention. This exercise should be community-directed, sustainable and under the supervision of health department of state and local government authorities. However, the WHO [1] criterion for MDA is a primary school prevalence of ≥50% of infection. Moreover, the WHO Control Strategy for urinary schistosomiasis states that the major control plans of urinary schistosomiasis are provision of Praziquantel to primary school children, provision of safe tap water to the whole community and health education.

ii. Government should enact sanitation laws targeting schistosomiasis-endemic communities to include components as follows: banning any form of anthropogenic contamination of the environment around canal perimeters; building a reasonable number of public convenience in the irrigation area near water contact points along the canals by the local authorities; establishing community sanitation clubs (CSC) to curb any form of faecal and urine environmental contamination through vigilance and awareness campaign; inclusion of public health education in the curricula of primary and secondary schools which will lay emphasis on the health-risk associated with unprotected exposure to water that is laden with susceptible snail vectors.

iii. Periodic community awareness campaign on the health-risk of unprotected water contact activities through community health and agricultural extension workers in order to halt the transmission of the disease in the study area and areas with similar settings.

CONSENT

It is not applicable.

ETHICAL APPROVAL

It is not applicable.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

3. Ugbomoiko US. The prevalence, incidence and distribution of human urinary


16. Preferences of potential snail intermediate dam sites and the distribution and habitat Idris HS, Ajanusi JO, Umoh JU, Galadima patter of schistosomiasis M, Ogbugu VC. Prevalence of Nigeria. The Nigerian Journal of schistosomiasis among pupils in some parasitology. 2001;22:75-80. Local Government Areas of Katsina State, Tukur A, Galadima M. Epidemiological Nigeria. The Nigerian Journal of study of schistosomiasis in the Bakalori Parasitology. 1998;19:73-75. Irrigation Project Area of Zamfara State, Nigeria: Prevalence and intensity of Schistosoma haematobium infection. Schistosoma haematobium is a hemoparasitic infection that causes Schistosomiasis. It is transmitted by snails that are infected with the parasite. The parasites develop in the small blood vessels of the intestines and liver, causing inflammation and damage. Schistosomiasis is a common disease in many parts of the world, including Nigeria. The symptoms of Schistosomiasis can include abdominal pain, diarrhea, and fatigue. It can also cause liver damage and, in severe cases, can be fatal. The disease can be treated with medication, but prevention is the best way to avoid infection. Prevention includes avoiding contact with freshwater in areas where the parasite is found, and using swim diapers to protect the skin. Ingesting freshwater snails is another way the parasites can enter the body. Prevention also includes proper sewage treatment and infected water sources. Medical professionals can also test for Schistosomiasis and provide treatment as needed.
Department of Biological Sciences, Ahmadu Bello University, Zaria, Nigeria. 2011;91.


