## **Research Articles**

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# Studies on mosquitoes breeding in rock pools on inselbergs around Zaria, northern Nigeria

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#### Abstract

*Background & objectives:* Rainwater often collects in depressions on rocks to form pools that are ideal breeding sites of mosquito vectors of diseases. Knowledge on the existence of disease vectors in these remote and relatively inaccessible locations could improve epidemiologic understanding and control capabilities. This study identifies mosquito species, their relative abundance and physicochemical characteristics of breeding microhabitats in rock pools on four inselbergs in northern Nigeria.

*Methods:* Soup ladle dipper was used to obtain representative samples of larval mosquitoes breeding in 141 rock pools on four inselbergs. Physicochemical parameters (depth, electrical conductivity, pH, surface area, temperature and total dissolved solids) of the pools were determined. Larvae were preserved in 70% alcohol and identified microscopically to species using taxonomic keys. Statistical correlation analysis and ANOVA were used to test the associations between physicochemical parameters and mosquito abundance, and for differences amongst inselbergs.

*Results:* Of 2991 larvae, five species of mosquito distributed in three genera (*Anopheles, Aedes* and *Culex*) including *Ae. vittatus* (92.88%), *An. ardensis* (0.13%), *An. distinctus* (1.67%), *An. wilsoni* (0.13%) and *Cx. ingrami* (5.18%) bred in the rock pools, Up to five species occurred per pool in various conspecific and heterogeneric combinations. Except for *Ae. vittatus*, the physicochemical parameters of the pools correlate significantly with species abundance.

*Conclusion: Ae. vittatus*, a potential vector of yellow fever in Nigeria breeds profusely in rock pools on inselbergs around Zaria. For comprehensive vector implication and control, rock pools should be amongst the habitats of focus in yellow fever epidemiology.

Key words Aedes vittatus - inselbergs - mosquitoes - Nigeria - rock pools - yellow fever

### Introduction

The disease transmission and biting nuisance problems arising from the occurrence and interaction of mosquitoes with humans appear to have defied several scientific advances and health services instituted to combat them. Part of the problems militating against effective and sustained control of mosquitoes and the diseases transmitted by them is the overt advantages available to mosquitoes to breed in diverse aquatic media that are naturally occurring and or the creation of human activities. Diversity of mosquito breeding environment stems from innate preferences shown by different taxa to the locations and conditions of various aquatic habitats<sup>1,2</sup>. These myriads of pre-imaginal mosquito habitats are often neglected or are not within easy reach of control operations. There is, therefore, a complexity in the epidemiology of vector-borne diseases generally, due to variability in the ecology of hosts, parasites and vectors<sup>3</sup>. For mosquito-borne diseases, contrasting local epidemiological situations may arise that demand a broad view of the overall environment to quantify the interfaces between hosts and vectors, as a form of environmental audit<sup>4</sup>. To critically audit the environment for the sources of mosquito that may be involved in disease transmission, entails searches of aquatic habitats in rock pools amongst several other breeding sites.

The Nigerian northern Guinea savanna is naturally endowed with inselbergs which possess their characteristic flora and fauna. These crystalline igneous rocks possess several depressions in which rainwater collects to form discrete pools during the rainy season. These collections of water serve as breeding habitats for mosquitoes. There is a dearth in the knowledge of the mosquito fauna associated with these rock pools. There are reports on mosquitoes associated with rock pool habitats in northern Nigeria<sup>5,6</sup> and in coral rock holes in coastal areas of Tanzania<sup>7,8</sup>. Several other publications also report on mosquitoes observed in rock pools as part of crosssectional surveys of their distributions<sup>9,10</sup>. This paper presents the mosquito fauna breeding exclusively in rock pools, available on four inselbergs around Zaria, northern Nigeria. The physicochemical conditions affecting mosquito occurrence and abundance in these microhabitats are also presented. This information is important in widening the knowledge base of breeding habitats of potential vectors of human diseases that could have applicability in their control.

#### **Material & Methods**

Study area: This investigation was done in the northern Guinea savanna on four inselbergs, around the

ancient city of Zaria, Kaduna State, Nigeria. They included Dumbi (on latitude 10° 57.7' N and longitude 7° 39.3' E at an elevation of 111.56 m above the surrounding), Hanwa (on latitude 11° 07.211' N and longitude 7° 42.659' E at an elevation of 19 m above the surrounding), Kufena (on latitude 11° 04.980' N and longitude 7° 39.416' E at an elevation of 10 m above the surrounding) and Wusasa (on latitude 11° 04.597' N and longitude 7° 40.475' E at an elevation of 32 m above the surrounding). Dumbi inselberg, and its environs are associated with thickets of trees, shrubs and grasses. It provides at its foot a permanent settlement for nomadic cattle herdsmen and the Dumbi village is located within one kilometer radius. Hanwa and Wusasa inselbergs are located in close proximity to human settlements, with the base of Hanwa inselberg virtually surrounded by structures utilised as worship centres. Kufena inselberg is located close to poorly-flooded paddy fields and humans engaged in stone quarrying are always present on it, but it is distal to human residences. The four inselbergs have several depressions that collect water during the rainy season which constitute effective rock pool breeding microhabitats for mosquitoes. These rock pools were examined for mosquito breeding between June and September 2006.

Sampling technique: Ten dips of the water in every other rock pool were obtained with a plastic soup ladle dipper (0.105 L capacity)<sup>11</sup>. The water was collected in a white plastic bowl and carefully observed for the presence of pre-imaginal mosquitoes. Culicine larvae collected were concentrated in a sieve and carefully picked with dropping pipette into labeled specimen bottles, the water drained and 70% alcohol preservative added. Anopheline larvae were collected alive in plastic bottles and reared to adults in the laboratory in small plastic bowls (11x 11x 5.5 cm) on a diet of bakers' yeast.

Determination of physicochemical parameters of rock pools: Depths of water in rock pools were obtained by lowering a metre rule to the bottom of the pools at three locations and the mean depths recorded. The surface areas of rock pools were determined from length and width measurements with a metre rule. The pH, electrical conductivity, total dissolved solids and temperature of water in each rock pool were determined by means of a HANNA HI 991300 pH/ EC/TDS/Temp meter.

*Species identification:* Mosquitoes collected were identified to species and counted under the x50 magnification of a stereo-microscope using pictorial keys for culicines<sup>12</sup>, and anophelines<sup>13</sup>. Dominant aquatic macrophytes were uprooted from rock pools and identified to species at the Herbarium Unit of the Department of Biological Sciences, Ahmadu Bello University, Zaria, Nigeria.

*Statistics:* Pearson correlation analysis was done to establish relationships amongst the physicochemical parameters of water, in rock pools, and abundance of species of mosquito breeding therein. One-way analysis of variance (ANOVA) was employed to test for significant differences in the relative abundance of mosquitoes amongst inselbergs; using least significant difference to separate means that differ significantly.

#### **Results**

A total of 141 rock pools were examined for larval stages of mosquito on four inselbergs in this study. These range from seven pools examined on Wusasa inselberg to 93 pools examined on Dumbi inselberg. The numbers examined were reflective of their relative availability on the inselbergs. By far the highest number of pools was observed on Dumbi inselberg, which also had the highest percentage positivity (78.49%). The least percentage positivity (64%) of rock pools with larval mosquitoes was observed on the Hanwa inselberg. Kufena and Wusasa inselbergs had similar percentage positivity (71.43%) of mosquito laden pools. A total of 2991 mosquito larvae were collected from 109 positive rock pools. The maximum number of larvae (1201) were collected from Dumbi while least number (416) were collected from Kufena. The relative abundance of larvae in the rock pools was highest (98) on Wusasa and least (16.45) on Dumbi inselbergs (Table 1).

Five species of mosquitoes, distributed in three genera, including *Ae. vittatus* (Bigot), *Anopheles ardensis* (Theobald), *An. distinctus* (Newstead and Carter), *An. wilsoni* Evans and *Culex ingrami* 

Inselbergs (Coordinates)	Altitude above sea level (m)	Height above surrounding area (m)	No. of pools examined	No. of pools positive (%)	No. of larvae collected (%)	Relative abundance of larvae	Larval No. per pool
Dumbi (10° 57.7' N, 7° 39.3' E)	756.82	111.56	93	73 (78.5)	1201 (40.2)	16.45	12.91
Hanwa (11° 07.211' N, 7° 42.659' E)	692	19	20	16 (64)	884 (29.6)	55.25	44.2
Kufena (11° 04.980' N, 7° 39.416' E)	715	10	21	15 (71.4)	416 (13.9)	27.73	19.8
Wusasa (11° 04.597' N, 7° 40.475' E)	715	32	7	5 (71.4)	490 (16.4)	98	70
Total			141	109 (77.30)	2991	27.44	21.21

Table 1. Occurrence of larval mosquitoes in rock pools on four inselbergs around Zaria, Nigeria

Edwards, were observed breeding in rock pools on the inselbergs. The dominant species (92.88%) being Ae. vittatus, which was encountered in high proportion of all the inselbergs studied. Cx. ingrami was encountered on three inselbergs, with the exception of Wusasa. All the five species of mosquito occurred in rock pools on the Dumbi inselberg. The three anophelines generally occurred in low density on Dumbi, but An. distinctus also bred on the Kufena inselberg. The relative abundance of Ae. vittatus larvae differ significantly (p < 0.05) amongst the four inselbergs with the trend Wusasa >Hanwa >Kufena> Dumbi. The relative abundance of An. distinctus and Cx. ingrami each did not differ significantly (p > 0.05)between the two and amongst the three inselbergs, where they occurred, respectively (Table 2).

Four breeding patterns, ranging from sole mosquito species breeding per pool to five species breeding per pool were observed on the inselbergs (Table 3). *Ae. vittatus* as sole breeding species per pool predominated in 94 (86.24%) rock pools, occurring on all the four inselbergs. Three different combinations of heterogeneric pairs of mosquitoes per pool were encountered on three inselbergs. Three and five species of congeneric and heterogeneric mosquitoes breeding per pool occurred on the Dumbi inselberg, only in one pool each.

The five species of mosquitoes bred in rock pools ranging between 0.5 and 25 cm in depth. *Ae. vittatus* and *An. distinctus* bred in the shallowest (0.5 cm)

pools, while *Cx. ingrami* bred in the deepest (25 cm) pools. *Aedes vittatus* was also observed in pools with the least surface area while *Cx. ingrami* and the anophelines (with the exception of *An. wilsoni*), were associated with pools having large surface areas (21.08–945 m<sup>2</sup>). Temperature of water in the rock pools at the time of sampling ranged from 14 to 40°C; the high extremes were associated with *Ae. vittatus* and *Cx. ingrami*.

The pH of the breeding media in the rock pools varied from slightly acidic (pH 5.86) to mild alkalinity (pH 9.85). The three anopheline species were associated with pools of acidic nature (pH 5.86-6.55). The culicines occurred in partly acidic and partly alkaline pools (pH 5.86-9.85). Ae. vittatus occurred in pools with the widest range of electrical conductivity and total dissolved solids (Table 4). On the Dumbi inselberg, the six physicochemical parameters monitored did not correlate significantly with the abundance of Ae. vittatus larvae in rock pools (p >0.05). However, the abundance of An. ardensis larvae correlated significantly with the electrical conductivity, depth and surface area of the pools (p < p0.05). Highly significant positive correlation existed amongst the abundance of An. distinctus larvae, total dissolved solids, electrical conductivity, depth and surface area of the pools (p < 0.01). Abundance of An. wilsoni larvae strongly correlate with electrical conductivity (p <0.01), and significantly correlate with total dissolved solids and depth of water in the pools (p <0.05). Abundance of Cx. ingrami larvae signifi-

Inselberg	Specieswise mosquito populations and percentages (in parentheses) of occurrence							
	Aedes vittatus	Anopheles ardensis	Anopheles distinctus	Anopheles wilsoni	Culex ingrami	-		
Dumbi	1025 (85.35)	4 (0.33)	36 (3)	4 (0.33)	132 (10.99)	1201		
Hanwa	883 (99.89)	0 (0)	0 (0)	0 (0)	1 (0.11)	884		
Kufena	380 (91.35)	0 (0)	14 (3.37)	0 (0)	22 (5.29)	416		
Wusasa	490 (100)	0 (0)	0 (0)	0 (0)	0 (0)	490		
Total	2778 (92.88)	4 (0.13)	50 (1.67)	4 (0.13)	155 (5.18)	2991		

Table 2. Species of mosquito breeding in rock pools on four inselbergs around Zaria, Nigeria

Species composition per rock pool	No. (%) of positive rock pools (n = 109)		Inselberg against (No.) of rock pools	
(a) Sole species				
Aedes vittatus	94	(86.24)	Dumbi (61), Hanwa (15), Kufena (13), Wusasa (5)	
Culex ingrami	2	(1.83)	Dumbi (2)	
Total	96	(88.07)		
(b) Double species				
Aedes vittatus + An. distinctus	1	(0.92)	Dumbi (1)	
Aedes vittatus + Culex ingrami	5	(4.59)	Dumbi (4), Hanwa (1)	
Anopheles distinctus + Culex ingrami	5	(4.59)	Dumbi (3), Kufena (2)	
Total	11	(10.09)		
(c) Triple species				
Anopheles ardensis + An. distinctus + Culex ingrami	1	(0.92)	Dumbi (1)	
Total	1	(0.92)		
(d) Pentad species				
Aedes vittatus + An. ardensis + An. distinctus + An. wilsoni Culex ingrami	+ 1	(0.92)	Dumbi (1)	
Total	1	(0.92)		

cantly correlate with depth, surface area and electrical conductivity of water in the rock pools (p < 0.01); and with total dissolved solids in the water (p < 0.05). On the Kufena inselberg, abundance of *An. distinctus* 

larvae in rock pools had strong correlation with the surface area of the pools (p < 0.01). Significant correlation also existed between abundance of *Cx*. *ingrami* larvae and surface area of the rock pools in

 Table 4. Range (mean ± SE) of physicochemical parameters of rock pools supportive of mosquito species breeding on inselbergs around Zaria, Nigeria

Species		Physical paramete	Chemical parameters			
	Depth (cm)	Surface area (m <sup>2</sup> )	Temperature (°C)	рН	Total dissolved solids (ppm)	Electrical conductivity (µScm <sup>-1</sup> )
Aedes vittatus	0.5–17 (5.18±0.35)	0.5–150 (19.25±3.46)	25.3–40	5.97–9.85	0–188	0–376
Anopheles ardensis	14 –17 (15.50±1.06)	21.08–254.4 (137.7±82.74)	14–17	6.30–6.55	15-80	28–165
Anopheles distinctus	4–17 (9.5±2.01)	4.93–945 (241.42±134.62)	27.2–28.2	5.86-6.55	10–120	22–234
Anopheles wilsoni	17 (17±0.0)	21.08 (21.08±0.0)	27.4	6.30	80	165
Culex ingrami	3.5–25 (10.64±1.47)	4.59–945 (170.16±64.71)	27.2–40	5.86–9.17	10–120	22–234

which they bred (p <0.05). On the Hanwa and Wusasa inselbergs, the physicochemical variables monitored did not correlate significantly (p >0.05) with larval abundance of the mosquito species breeding in rock pools.

#### Discussion

Discrete microhabitats for breeding mosquitoes exist in rock pools on inselbergs and each pool constitutes an independent replicate for determining the ecology of mosquito species. A distinct and predictable disturbance to the fauna that colonise these rock pools is seasonal drying. This was evident at the preliminary stage of this study at the onset of the rainy season in May, when the depressions were devoid of water and could not then serve as effective mosquito breeding habitats. Following prolonged rainfall, the depression later turned to important mosquito breeding habitats. The volume of water in the pools was usually small and in October the pools dried within few days following the cessation of rainfall. Ae. vittatus is the dominant species of mosquito breeding in rock pools around Zaria, northern Nigeria. Results show that the species is catholic in its choice of breeding microhabitat in rock pools and was the least affected by the physicochemical conditions of the rock pools. Ae. vittatus is widely distributed in Africa and has been associated with breeding principally in rock pool habitat<sup>5,6</sup>. Together with Ae. aegypti, Ae. vittatus is a potential yellow fever vector in northwestern Nigeria, adjacent to the study area, and in several foci in Africa<sup>6</sup>.

Considering the high yellow fever epidemic risk posture of the Zaria environment, due to elevated *Ae*. *aegypti* larval indices<sup>14</sup>, the current findings of high populations of larval *Ae*. *vittatus* in rock pools within the same area has further exacerbated the risk factor. Dumbi, Hanwa and Wusasa inselbergs are surrounded by human habitations stationed within flight range of these pool-breeding mosquitoes. *Ae*. *vittatus* could thus be amongst the species that constantly create biting nuisances on humans in the area; since it has been caught at human bait in some villages northwest of Nigeria<sup>6</sup>.

The three anophelines (An. ardensis, An. distinctus and An. wilsoni) found breeding in rock pools in this study have not been implicated in malaria or disease transmission generally. As such, rock pools did not constitute a breeding habitat for malaria vectors in Zaria, Nigeria. It is plausible that the species are zoophilic and obtain their blood meals readily from the large number of cattle and small domestic ruminants owned by nomadic Fulani herdsmen camping at the foot of Dumbi inselberg. Cx. ingrami is also a medically unimportant zoophilic species. On the Kufena and Dumbi inselbergs An. distinctus and Cx. ingrami were observed breeding exclusively in five rock pools with comparatively larger surface areas and high cluster of aquatic macrophytes, overtly providing shaded canopies. The dominant plants coinhabiting these mosquito microhabitats on the Kufena inselberg are Cynotis lanata, Drosera indica, Echinochloa species and Mariscus longibracteatus.

The dominant aquatic macrophytes coinhabiting mosquito microhabitat on the Dumbi included Cyperus denudatus, Heteranthera callifolia, Ludwigia decurrens, Murdannia simplex and Scirpus uninodis. The conditions on these preferred rock pools closely simulate those of the adjoining partially flooded rice-fields around the Kufena inselberg. An. distinctus could also be breeding in these rice-fields which are known to support profuse breeding of anopheline mosquitoes<sup>15</sup>. Larvae of An. distinctus have been reported to occur mainly in open permanent swamps, occasionally in well-shaded seepage pools<sup>16</sup>. The species was reported to breed in shallow grass swamp with reeds<sup>13</sup>. The surroundings of Dumbi inselberg constitute a forest outlier being continuously degraded by human activities. Both An. ardensis and An. wilsoni occurred exclusively on rock pools in this environment, thus confirming their preferences for forest condition. Both species have been associated with montane and intermediate forests<sup>13</sup>.

In this study, it was revealed that the more diversified the availability of rock pools on inselbergs, the less are the relative abundance and mean number of larval mosquitoes per pool. This is an indication that density related pressure could regulate population of larvae, especially on inselbergs with few rock pools. Depending on the availability of rock pools, oviposition by adult mosquitoes is communal when few pools are available and dispersed in the presence of several pools. Potential predators usually encountered in most of the rock pools devoid of mosquito larvae in this study included unidentified dragonfly nymphs (odonates) and tadpoles (anurans). Predation is known to significantly affect the population dynamics and communities of prey species in aquatic systems<sup>17</sup>. These predators might have affected the availability of aquatic stages of mosquito in the rock pools. Odonate predators depressed the abundance of mosquitoes in water-filled tree holes in tropical forests of Panama<sup>18</sup>. In Kenya, tree holes with fewer mosquitoes are associated with the presence of odonates in them<sup>19</sup>.

In conclusion, this study identified five species of mosquito breeding in rock pools around Zaria, northern Nigeria. Amongst these *Ae. vittatus* a potential yellow fever vector in the area was predominant. Therefore, for effective prevention of epidemic yellow fever, attention should be focused on the control of mosquitoes breeding in rock pools. In the face of epidemic yellow fever outbreak, rock pools should be inspected to implicate vectors and launch suitable control measures.

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