MONTHLY ASSESSMENT OF THE RELATIVE ABUNDANCE AND DEVELOPMENT OF IMMATURE STAGES OF *Culex* MOSQUITOES (DIPTERA: CULICIDAE) OF THE SELECTED BREEDING HABITATS IN NIGER, NIGERIA

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ABSTRACT

The information on relative abundance and distribution of *Culex* mosquitoes are essential for determining illness risk and this is especially critical when a disease spreads quickly and that prompt assessments for disease mitigation techniques are required. This study was conducted to assess and provide an epidemiological report on the relative abundance and development of immature stages of Culex in conventional larval breeding habitats in selected LGAs of Niger State, Nigeria. Gutter, swamps, and large water bodies were chosen as larval habitats, and samples were taken weekly from May to November 2019. The findings revealed the presence of three vector mosquito species in the areas namely: *Culex quinquefasciatus* (887.25±121.7), Cx. nigripalpus (434.50±46.34) and Cx. salinarius (351.92±32.48). Habitats-wised showed large water habitats had the abundance of Culex (686.75±98.11) followed by gutters (516.67 ± 60.20) and the least was swamps (471.25 ± 42.08) mosquitoes. Based on the monthly abundance rate (MAR), the peak abundance of the immature stages was June and July and declined in November in both habitats. While the age survival rate (ASR) differed significantly (P < 0.05) from one another across the months in all the habitat types. On the age distribution within the habitat types, L4 (109.78±30.50) was the highest, in gutters, while in swamps, and large water habitats, L1 larvae were the most abundant (29.88±11.14 and 39.46±11.69), respectively. While between the habitats, gutters had a significantly higher abundance of L1, L2, L3, and L4, while swamp and larger water habitats were insignificant (P>0.05) from one another for L1 and L4 except for the L3 immature stage that was differentiated with larger water being the most abundant. The findings of this study imply that anthropogenic changes to the ecosystem are causing a severe hazard of culex-borne diseases (CBD) to public health in Niger State.

Keywords: Conventional breeding habitats, abundance, immature stages, *Culex quinquefasciatus*, *Cx. nigripalpul*, *Cx. salinarius*, *Culex*-borne disease (CBD)

ABSTRAK

Maklumat mengenai kelimpahan relatif dan pembiakan nyamuk *Culex* adalah penting bagi menentukan risiko bawaan penyakit dan adalah kritikal apabila penyakit meningkat dengan cepat. Oleh itu, penilaian segera bagi teknik mitigasi penyakit adalah diperlukan. Kajian ini dijalankan bagi menilai dan menyediakan laporan epidemiologi untuk kelimpahan relatif dan tumbesaran larva nyamuk Culex pada habitat konvensional pembiakan larva di negeri Niger, Nigeria. Habitat pada longkang, paya, dan kawasan tadahan air yang besar dipilih sebagai habitat larva dan sampel diambil pada setiap minggu dari bulan Mei sehingga November 2019. Dapatan menunjukkan kehadiran nyamuk bagi tiga spesies vektor di dalam kawasan kajian iaitu Culex quinquefasciatus (887.25±121.7), Cx. nigripalpus (434.50±46.34) and Cx. salinarius (351.92±32.48). Selain itu, kawasan tadahan air besar menunjukkan kadar kelimpahan tinggi Culex (686.75±98.11) disusuli dengan longkang (516.67±60.20) dan yang terakhir adalah nyamuk di paya (471.25±42.08). Berdasarkan kadar limpahan bulanan (MAR), limpahan tertinggi bagi larva Culex adalah pada bulan Jun dan Julai dan menurun pada November di kedua-dua habitat. Manakala kadar umur kelangsungan hidup (ASR) berbeza dengan bererti (P<0.05) antara satu sama lain di sepanjang bulan di kesemua jenis habitat. Pada pembahagian peringkat larva di antara jenis habitat, L4 (109.78±30.50) adalah yang paling tinggi di dalam longkang, sementara di paya dan di habitat tadahan air besar merekodkan L1 larva adalah yang paling melimpah (29.88±11.14 dan 39.46±11.69), masing-masing. Manakala antara habitat, longkang adalah tertinggi dan beerti pada L1, L2, L3, dan L4, sementara paya dan habitat kawasan tadahan air besar tidak mempunyai perbezaan yang bererti (P>0.05) antara satu sama lain bagi L1 dan L4 kecuali pada larva L3 yang dibezakan dengan kawasan tadahan air besar menjadi kawasan yang paling tinggi. Dapatan dari kajian ini mendapati bahawa perubahan antropogenik kepada ekosistem boleh mengakibatkan ancaman serius terhadap penyakit bawaan Culex (CBD) kepada kesihatan awam di negeri Niger.

Keywords: Habitat pembiakan konvensional, kelimpahan, larva nyamuk, *Culex quinquefasciatus*, *Cx. nigripalpus*, *Cx. salinarius*, penyakit bawaan *Culex* (CBD)

INTRODUCTION

Culex mosquitoes are among the Culicidae family that often need blood meals from animals including humans, essentially transmit pathogens and ensure they emerge and re-emerge (Barba et al. 2019). Mosquito-borne diseases account for more than 17% of all contagious diseases worldwide and cause the death of over 700,000 people each year (Moise et al. 2021; Tabachnick 2010), and infectious disease outbreaks aided by *Culex* species are becoming more common (Gorris et al. 2021). *Culex* mosquitoes are a major public health issue, ranking with the *Anopheles* and *Aedes* genera in the spread of epidemiologically important diseases in Africa and Nigeria, respectively. The economic impact of these diseases is considerable in Nigeria, as the country spends billions of dollars on fighting these diseases, money that could have been spent on other productive sectors of the economy (Ukubuiwe et al. 2016). Filarial fever (FF), West Nile fever (WNF), St. Louis encephalitis fever (SLEF), and other mosquito diseases are

spread by Culex species like *Culex quinquefasciatus*, *Cx. nigripalpul*, and *Cx. salinarius* (Duguma et al. 2019; Mores et al. 2007; Nwana et al. 2021).

Filarial fever (FF), caused by the parasite *Wuchereria bancrofti*, is the most common disease in Nigeria, with 106 million people at risk. *Cx. nigripalpul and Cx. salinarius* whose introduction of WNF and SLEF to African counties and Nigeria, due to the global increase in human population have mild diseases impact. United Nation reported that, currently the estimation of human population is projected to be >7 billion people with a likely increase to roughly 9.6 billion people in 2050 (Braack et al. 2018).

The spread and effect of disease transmitted by mosquitoes, especially *Culex* species, will very certainly be aided by such population growth. Large increases in human population will be coupled with increased population density, which will enhance disease transmission either directly or through vectors. There will also be more worldwide movement of individuals due to migration, tourism, or business travel, increasing the possibility of infective sources spreading more frequently. Similarly, the global movement of freight and commerce items from other nations to Africa will rise, supporting the spread of these *Culex* vectors. In addition, as the human population grows, so will the number of breeding places and habitats available to *Culex* vectors. The movement of water bodies along with eggs and larvae across the world can facilitate the increase in the distribution and abundance of these mosquitoes leading to the increase in the high percentage of the mosquito-borne diseases by *Culex* species in African countries (Braack et al. 2018; Duque et al. 2019; Wilke et al. 2021). The larvae of these species have been in both semi-permanent and temporary habitats including ponds, freshwater swamps, gutters, discarded empty containers.

Culex species such as *Cx. salinarious* and *Cx. nigripalpus* are regarded as the most important vectors for Saint Louis encephalitis (SLE) and has been responsible for multiple epidemic mosquito diseases in some parts of the world despite their appearance as less nuisance mosquitoes to humans (Akaratovic et al. 2021). Although, these two species are less abundance with mild effects in Niger State, however highly propagated in some part of Nigeria, thus their distribution and the quality of their breeding habitats need to be frequently checked (Ikeh et al. 2017).

Like other mosquitoes, *Culex* occurs widely in various habitats (Duguma et al. 2019) and grows in four stages: Egg, larva, pupa and adult, while the first three stages occur in water (Hancock & Godfray 2007). Even though their larval occurrence and abundance vary by species, habitat, location, and season, they are all linked to the availability of adequate larval breeding sites. As a result, the water quality of mosquito breeding habitats is a key predictor of female mosquito oviposition and larval development success (Garba et al.2018; Ikeh et al. 2017). Habitats include gutters, swamps, pools, and several others that are known to be supportive breeding sites to various mosquitoes including *Culex* mosquitoes (Mores et al. 2007). Thus, managing water bodies that are potential larval habitats is crucial to curb any mosquito-borne disease outbreaks in urban and rural areas (Odero et al. 2018).

Culex mosquito distribution in breeding areas can be used to assess mosquito-borne disease risk and estimate future dangers from developing diseases transmitted by these *Culex* species (Gorris et al. 2021). The proliferation potential, survivorship, adaptability, and vectorial capacity of mosquitoes in a given location are all positively related to the distribution and number of these vector species (Olayemi et al. 2014). A thorough understanding of mosquito breeding ecology, including types of and preferences for larval habitats, distribution of breeding

grounds, and physical, biological, and chemical properties of the habitats, is necessary for successful larval management (Akaratovic et al. 2021; Amao et al. 2018; Ikeh et al. 2017).

Some of these characteristics have been carefully examined and understood in Nigeria (Gatton et al. 2013), therefore increasing understanding of the threat posed by these vectors is critical. This study evaluated and raised awareness about the impact of relative abundance and development of immature stages of *Culex* species in several types of breeding habitat in Niger State. This study also document changes in transmission patterns and propose appropriate control interventions using a tool that was already available.

MATERIALS AND METHODS

Study Area's Description

Niger is one of the states is in the Middle Belt region of Nigeria located between longitude 6[°] 33E, and latitude 9°37N covering 88km^2 (Figure. 1) and representing 9.30% of the total land area. It consists of 85% arable land with a population of about 4.8 million inhabitants. This area has a tropical climate with mean annual rainfall, temperature, and relative humidity of 1334 mm, 30.2°C and 61%, respectively. Niger State is bordered by several states: Kaduna (northeast), Federal Capital Territory Abuja (FCTA) (southeast), Zamfara (north), Kebbi (west), Kogi (south), Kwara (southwest), and the Republic of Benin, along Agwara LGA borders (northwest). Furthermore, this state experiences two distinct climates: the rainy season (May – October) and the dry season (November – April). In addition, the vegetation in the area is typically grass-dominated savannah with scattered trees (Akpan et al. 2018; Ukubuiwe et al. 2012).

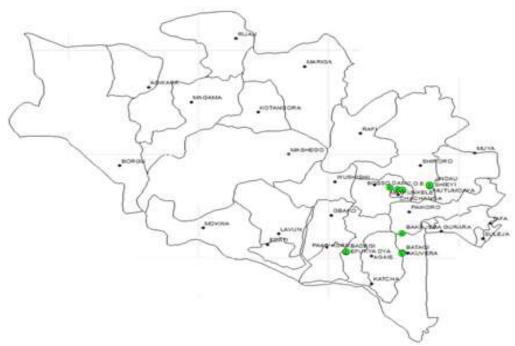


Figure 1. Map of Niger State, showing study location in the local government area

This study was conducted in three breeding habitats, which are classed as temporary or semi-permanent breeding habitats based on their nature and availability. Temporary larval habitats, for example, are a site that holds water during the rainy season and then dries up two

Table 1.

to three weeks later, but semi-permanent larval habitats may or may not dry up one month after the rainy season has ended (Mereta et al. 2013). Temporary larval habitats include gutters and swamps, and semi-permanent breeding habitats include large water bodies such as rivers, lakes, and seas. In 12 sites in Niger State's Boss, Katcha, Lapai, and Shiroro LGAs, the dipping technique was used to determine sampling spots for the presence of mosquito larvae (Table 1). The site selection was finished in April 2019, just before the rainy season began, and the breeding areas were swamped with precipitation.

Replicates						
Habitats/Sites		Site 1	Site 2	Site 3	Site 4	No. of Sites
Gutters		BGT	KGT	LGT	SGT	4
Swamps		BSW	KSW	LSW	SSW	4
8	ıter	BLW	KLW	LLW	SLW	4
bodies						
Total						12

Distribution of mosquitoes breeding habitats in different local government areas

BGT=Bosso gutter, BSW=Bosso swamp, Bosso large water, KGT=Katcha gutter, KSW=Katcha swamp, KLW=Katcha large water, LGT=Lapai gutter, LSW=Lapai swamp, LLW=Lapai large water, SGT= Shiroro gutter, SSW=Shiroro swamp, SLW=Shiroro large water.

Mosquito sampling was conducted from May to November 2019 at selected sites to identify and evaluate the breeding habitats in the four LGAs. For the sampling of immature mosquitoes, monthly abundance, and age survival rates (MAR and ASR), occurred weekly in breeding habitats, and instars (L1-L4) were sampled between 0700hrs and 0900hrs. These times correspond to the needed temperature (range 16-32°C) for most larvae and adult mosquito species, including Anopheles, Culex, and Aedes, to reproduce in the tropics (Aneesh 2014). The dipping process was carried out at random in the breeding habitat's surface areas where the concentration of larvae was detected, using a standard pint 350ml dipper at a rate of 20 per sampling location, as specified by (Azari-Hamidian 2011). On each week of the collection, the immature mosquitoes in each breeding habitat were estimated.

Larval Rearing for Adult Identification

Mosquito instars were collected and maintained to adult stage in troughs under laboratory conditions (28°C and 73%), fed with 0.32ml yeast solution. Monitored until the adult stage, when they were morphologically identified and separated into culicine mosquitoes using a voucher key, as described by Olayemi & Ande (2009) and Shapiro et al. (2005).

Mosquito Preservation and Identification

The larvae were placed in a 10% formalin preservative and then classed macroscopically depending on the position of the larvae's breathing tube in the water. If the respiratory tube was small and oriented parallel to the surface of the water, they were most likely Culex. Mosquito larvae that drifted horizontally at the water's surface were most likely Anopheles, and mosquito larvae that drifted vertically to the water's surface were most likely Aedes surface (Wang et al. 2020a). In the laboratory, the instars (L1-L4) were divided into morph groups and classified to genus level based on observable traits such as colour presence or absence of siphon, the position of hair tufts, length of the siphon, arrangement of comb scales, presence of caudal setae, and presence and number of hairs on the saddle. Also, using a dissection microscope and morphological criteria, life phases were separated by the length of the larva. In this study, only

Culex mosquitos of the subgenus Culicinae were employed, and their morphs-groups were given names based on the genus abbreviation (i.e., *Culex* – *Cx.*, and *Aedes*- *Ae.*, (Rueda 2004). Adult *Culex* mosquitoes were separated into morph groups in the lab and identified at the species level using a dissecting microscope (40x) and morphological criteria based on physical traits (e.g., femora, tibia, air tube, and wings) (Snell 2005).

Statistical Analysis

The statistical analysis was carried out using SPSS software analyses (version 23 for Windows, SPSS Inc., Chicago, IL). The monthly relative abundance and age survival rates were determined using one and two-way analysis of variance (ANOVA), which were then converted to plotted graphs for distribution within and between breeding habitats P=0.05 was used to determine the significance of the discrepancy, and Duncan Multiple Range was used to separate the means (DMR).

RESULTS

Three primary mosquito species from the *Culex* genus namely, *Culex quinquefasciatus*, *Cx. nigripalpus*, and *Cx. salinarius* was encountered in different habitats (gutters, swamps, and large water bodies) within four local government areas (LGAs) (Table 2). The three *Culex* species, abundance was insignificant (P>0.05), in gutters and swamps, but their population significantly differed (P<0.05) from large water bodies. The large water habitats being the highest follow the order of abundance: *Cx. quinquefasciatus* (1122.75±178.56)>*Cx. nigripalpus* (524.50±68.09) >*Cx. salinarius* (413.00±47.68). The total mean abundance of the three different species in the breeding habitat types established *Cx. quinquefasciatus*, as the most populated (887.25±121.7) followed by *Cx. nigripapus*. (434.50±46.34) and the least was *Cx. salinarius* (351.92±32.48%). Based on the results obtained, large water bodies were significantly higher as breeding habitats for mosquitoes (686.75±98.11%), followed by gutters (516.67±60.20) and swamps (471.25±42.08).

 Table 2.
 Mosquito species relative abundance at different breeding habitats

Mosquito Species	Breeding Habitats					
Mosquito Species	Gutters	Swamps	Large Waters	Total Mean		
Culex quinquefasciatus	813.25±109.52 ^a	725.75±76.64 ^a	1122.75±178.56 ^b	887.25±121.7		
Cx. nigripalpus	406.25 ± 41.84^{a}	372.75±29.09 ^a	524.50±68.09b	434.50±46.34		
Cx. salinarius	330.50±29.25 ^a	312.25 ± 20.52^{a}	413.00±47.68 ^b	351.92±32.48		
Aggregate	516.67±60.20	471.25 ± 42.08	686.75±98.11	557.69±66.83		

Values are mean \pm SE. Value followed the same superscript along the row are not significantly different (*P*>0.05)

The month-wise abundance and development of immature stages of *Culex* mosquitoes in gutters are presented in (Table 3). Generally, the total mean of the monthly abundance rate (MAR) was very high in the 3rd sampling (July) (170.68±22) and varied significantly (P<0.05) from the remaining sampling months with the 7th sampling (November) as the least (13.18±0.19). For the specific immature stages across the months in the same habitats, L1 and L4 had the highest abundance in the 3rd sampling (July) with values of 174.75±35.34 and 210.00±71.82, respectively. The highest abundance for L2 and L3 were obtained in the 4th sampling (August) with values of 174.25±46.40 and 164.50±51.93. These higher abundances of the immature stages varied significantly (P<0.05) from the abundance of their respective stages across the sampling months except for L3 that was insignificant (P>0.05) from the abundance of 3rd sampling (July) (162.50 ± 44.36). The least specific abundance between the months was obtained in the 7th sampling (November) for all immature (i.e. L1, L2, L3, and L4).

Table 3.	<i>Culex</i> genus larval abundance and age structure of gutter breeding habitats						
	Larval Stages						
Sampling (Months)	L1	L2	L3	L4	MAR		
1 st (May)	64.50±23.69° _c	$31.50 \pm 9.31^{b}_{a}$	31.25±6.98 ^b _a	$41.75 \pm 10.52^{b}_{b}$	42.25±7.25b		
2 nd (June)	92.75±31.8 ^c _b	$70.00 \pm 16.59^{d}_{a}$	$117.25 \pm 29.00^{d}_{c}$	$141.75 \pm 61.30^{d}_{c}$	105.43 ± 18.50^{d}		
3 rd (July)	$174.75 \pm 35.34^{e}_{b}$	135.50±25.47 ^e a	$162.50 \pm 44.36^{e}_{b}$	210.00±71.82 ^e c	$170.68 \pm 22.34^{\rm f}$		
4 th (August)	$126.00 \pm 37.01^{d}_{b}$	$174.25 \pm 46.40^{f}_{c}$	164.50±51.93°c	112.00±16.75° _a	144.18±19.22 ^e		
5 th (September)	123.75±37.31 ^d _c	$70.25 \pm 19.89^{d}_{a}$	106.75±23.99 ^d _b	106.00±17.29 ^c _b	101.68 ± 12.58^{d}		
6 th (October)	47.00±12.19 ^b _a	$51.00 \pm 26.85^{\circ}_{a}$	$70.25 \pm 18.00^{\circ}{}_{b}$	$131.25 \pm 37.99^{d}_{c}$	74.87±14.41°		
7 th (November)	11.50±4.97 ^a b	5.00±2.38 ^a a	$10.50 \pm 3.66^{a}_{b}$	25.75±15.15 ^a c	13.18±0.19 ^a		
ASR	91.46±26.04 ^b	76.78 ± 20.98^{a}	94.71±25.42 ^b	109.78±30.50°	93.18±14.36		

Values are mean \pm SE. Values followed by the same superscript along the column are not significantly different, values followed by the same subscript along the row are not significantly different. (*P*>0.05) L1=Larval stage 1, L2=Larval stage 2, L3=Larval stage 3, L4=Larval stage 4. MAR=Monthly abundance rate, ASR=Age survival rate.

In terms of age survival rate (ASR) of the same genus, L1 was favoured by 1st sampling (May) (64.50 ± 23.69) and 5th sampling (September) (123.75 ± 37.31). These values significantly differed (P<0.05) from the abundance of immature (L2, L3, and L4) for the sampling months. While L2 was favoured by 4th sampling (August) (174.75 ± 46.40 L/S/S), L3 was unfavoured by any month. The favourable sampling months for L4 larvae were June, July, October, and November with peak survival rate in 3rd sampling (210.00 ± 31.82). In general, the total mean of age survival rate was obtained for L4 larvae (109.78 ± 30.50) and the lowest was recorded for L2 (76.78 ± 20.98).

In swamp habitats (Table 4), specific immature stages of L1, L2, L3, and L4 across the months revealed their peak abundance in 3rd sampling with the values being significantly higher (60.50 ± 14.71 , 44.00 ± 17.59 , 45.25 ± 22.43 , and 44.25 ± 22.99 , respectively). However, only in the 3rd sampling these larvae (L1, L2, and L4) were substantially different (P<0.05) from stages of the remaining months. As it occurred in gutters, in swamps, the least abundance was in the 7th sampling for all immature. The total mean of the monthly abundance rate (MAR) was significantly high (P<0.05) in the 3rd sampling (48.50±9.00) as compared to the remaining sampling months with 7th sampling as the least (12.18±3.41).

Table 4.	<i>Culex</i> genus larval abundance and age structure of swamp breeding habitats						
	Larval Stages						
Sampling (Months)	L1	L2	L3	L4	MAR		
1 st (May)	29.75±12.68 ^c _b	$15.00 \pm 0.00^{a}_{a}$	15.00±0.00 ^a a	15.00±0.00 ^a a	18.68 ± 3.28^{b}		
2 nd (June)	29.00±11.95° _a	$38.25 \pm 9.04^{b}_{b}$	$42.00 \pm 8.05^{d}_{b}$	$31.75 \pm 5.96^{b}{}_{a}$	35.25 ± 4.24^{d}		
3 rd (July)	60.50±14.71° _b	44.00±17.59° _a	$45.25 \pm 22.43^{d}_{a}$	44.25±22.99° _a	48.50±9.00 ^e		
4 th (August)	$50.25 \pm 11.15^{d}_{b}$	$35.75 \pm 10.28^{b}_{ab}$	36.25±7.27 ^c _{ab}	$29.00 \pm 12.00^{b}_{a}$	37.81 ± 5.03^{d}		
5 th (September)	31.00±13.52 ^c _c	$23.75 \pm 11.64^{a}_{b}$	$32.00 \pm 7.62^{\circ}_{c}$	$18.75 \pm 4.71^{a}_{a}$	$26.37 \pm 4.68^{\circ}$		
6 th (October)	21.00±9.22 ^b c	$17.25 \pm 7.06^{ab}{}_{b}$	$21.25 \pm 7.79^{b}_{c}$	12.75±2.59 ^a a	18.06 ± 3.30^{b}		
7 th (November)	$8.50 \pm 4.73^{a}_{b}$	$13.00 \pm 7.76^{a}_{a}$	13.80±6.64 ^a a	$14.25{\pm}10.00^{a}_{a}$	12.18 ± 3.41^{a}		

ASR $29.88\pm11.14^{\circ}$ $26.71\pm9.05^{\circ}$ $29.25\pm8.54^{\circ}$ 23.68 ± 8.32^{a} 28.12 ± 4.28 Values are mean±SE.Values followed by the same superscript along the column are not significantly different, values followed by the same subscript along the row are not significantly different (P>0.05). L1=Larval stage 1, L2=Larval stage 2, L3=Larval stage 3, L4=Larval stage 4.MAR=Monthly abundance rate, ASR=Age survival rate.

Age survival rate (ASR) of *Culex* mosquitoes in swamps, revealed significant variation among the immature stages with different stages being favoured by different months. The 1st, 3rd, and 4th samplings were particularly favourable for the abundance of L1 with high values of 29.75 ± 12.68 , 60.50 ± 14.71 , and 50.25 ± 11.15 , and substantially different (*P*<0.05) from the other months' larval stages (L2, L3, and L4). L3 was favoured by 2nd, 5th and 7th samplings with a range survival rate value of 42.00 ± 8.05 to 13.10 ± 6.64 . However, L2 was not favoured by any of the months while L4 was favoured by the 7th sampling. In swamps, the mean age survival rate was obtained for L1 (29.88±11.14) and the lowest was recorded for L4 with the value of 23.68 ± 8.32 .

While, in the large water bodies, abundance and development of the instars (Table 5), revealed the highest L1 abundance in 3rd sampling (78.25±26.12). This highest value of L1 abundance was insignificant (P>0.05) from the abundance of 4th sampling, but significantly differed (P<0.05) from the remaining months. L2 larvae were high in the 4th sampling (36.50±9.13), while the highest L3 and L4 were favoured by 2nd sampling with the abundance values of 74.00±33.67 and 33.50±9.34 respectively. These values of L3 and L4 were significantly different (P<0.05) from the abundance for all other months. The lowest larval stage abundance for LI, L2 and L3 were in 7th sampling with the values of 14.50±3.66, 15.75±7.21, and 16.00±5.49 respectively, while the lowest L4 stage abundance was recorded in 1st sampling (9.50±2.72). The total mean abundance (MAR) was very high in 2nd sampling (45.00±9.40), while the lowest was recorded in the 7th sampling (16.81±2.92).

Sampling (Months)	L1	L2	L3	L4	MAR
1 st (May)	34.50±10.78 ^b _c	$18.25 \pm 6.54^{a}_{b}$	19.75±5.57 ^b _b	9.50±2.72 ^a a	20.50±3.90b
2 nd (June)	$36.25 \pm 9.25^{b}_{a}$	34.25±9.25° _a	$74.00 \pm 33.67^{d}_{b}$	35.50±9.34 ^c _a	45.00±9.40e
3 rd (July)	78.25±26.12 ^c c	32.00±6.09 ^c b	$29.50 \pm 8.79^{b}_{b}$	$23.00 \pm 9.85^{b}{}_{a}$	40.68±8.76e
4 th (August)	52.00±13.00 ^c c	36.50±9.13°b	$34.50 \pm 6.30^{b}_{b}$	19.75±9.74 ^b a	35.68±5.29d
5 th (September)	36.00±11.21 ^b _b	$26.75 \pm 9.56^{b}_{a}$	23.00±4.18 ^b _a	$25.00 \pm 12.56^{b}_{b}$	27.68±4.61c
6 th (October)	$24.75 \pm 7.78^{b}_{a}$	$23.75 \pm 5.63^{b}_{a}$	$47.75 \pm 24.75^{c}_{b}$	$18.00 \pm 9.32^{b}_{b}$	28.56±6.94c
7 th (November)	14.50±3.66 ^a a	15.75±7.21 ^a a	16.00±5.49 ^b _a	$21.00 \pm 8.18^{b}{}_{a}$	16.81±2.92a
ASR	39.46±11.69°	26.75 ± 7.36^{a}	34.93±12.68 ^b	21.68±8.82 ^a	30.70±5.97

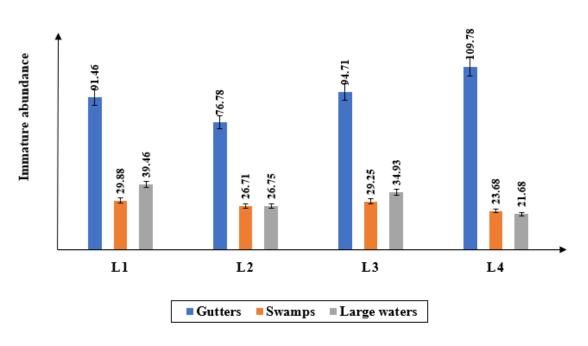
 Table 5.
 Culex genus larval abundance and age structure of large water breeding habitats

Values are mean \pm SE. Values followed by the same superscript along the column are not significantly different, values followed by the same subscript along the row are not significantly different (*P*>0.05). L1=Larval stage 1, L2=Larval stage 2, L3=Larval stage 3, L4=Larval stage 4. MAR=Monthly abundance rate, ASR=Age survival rate.

In terms of aging, the survival rate (ASR), L1 larvae were favoured by 1st, 3rd, 4th, and 5th (34.50 ± 10.78 , 78.25 ± 26.12 , 52.00 ± 13.00 , and 36.00 ± 11.21 respectively). These values of L1 were significantly different (P<0.05) from the abundance of other age structures (L2, L3, and L4) for the remaining months. L2 was not favoured by any of the months while the highest L3 stage was favoured by 2nd sampling (74.00 ± 33.67) and 6th (47.75 ± 24.75). These L3 values

were significantly different (P<0.05) from the values recorded in the remaining stages (L1, L2, and L4) of the same months, and L4 was favoured by the 7th (21.00±8.18). The highest mean abundance of all the larval stages was recorded for L1 (39.46±11.69L/S) and the lowest was recorded for L4 with the value of 21.68±8.82.

The immature abundance of *Culex* within and between the breeding habitats revealed significant variations in the mosquitoes (Figure 2). In gutters, a significantly higher abundance was for L4 (109.78±30.50), and the lowest was L2 (76.78±20.98). These values were significantly differed (P<0.05) from one another and the abundance of L1 and L3, however, these two stages were insignificant from one another. In swamps, L1 larvae were the most abundant (29.88±11.14), however, insignificant (P>0.05) from L3 but significant from the remaining immature stages. L4 stage was the least abundant (23.68±8.32) and significantly different from L2 with latter being the abundant mosquitoes (26.71±9.05). In the same manner, L1 had the highest abundance in the large waters (39.46±11.69) followed by L3, L2, and L4 immature mosquitoes. For the abundance of L1, L2, L3, and L4 with immature r recorded 91.46±26.04, 76.78±20.98, 94.71±25.42, and 109.78±30.50 respectively, while, swamp and larger water habitats were insignificant (P>0.05) from one another for L1 and L4 except for L3 immature stage that was differentiated with larger water being the most abundant.



Breeding habitats

Figure 2. Mean age structure of mosquito genera abundance of different breeding habitats

DISCUSSION

Immature stages indicate a step in the metamorphosis of mosquitoes to terrestrial adult form, and their assessment for relative abundance and development are the best proxy quantifying the adult production from the breeding habitats (Hancock & Godfray 2007; Mutuku et al. 2006; Mwangangi et al. 2007). This research aimed to assess the *Culex* vectors abundance and their

development in different breeding habitats of four local government areas of Niger State, Nigeria. These findings will aid future planning and development of mosquito control measures against *Culex*- borne diseases (CBD), particularly filarial fever (FF), in specific LGAs where the disease occurs. Based on the current findings, three species of *Culex* mosquitoes were encountered namely *Culex quinquefasciatus, Cx. nigripalpus,* and *Cx. salinarius* with their abundance based on habitat preference and adaptability in the three breeding habitats investigated (Table 2). Thus, regardless of whether the breeding sites were clean, dirty, or contaminated, these findings indicated mosquitoes' varied habitat usage.

The presence and composition of these *Culex* mosquitoes in both habitats suggest their distribution and abundance in the study areas. Therefore, they can cause the transmission of different *Culex*-borne diseases such as Filarial fever (FF), Saint Louis encephalitis (SLE), West Nile fever .(WNF) and several others leading to *culex*-borne diseases (CBD) outbreaks (Duguma et al. 2019), in the areas were their prevalence is high. Although in Niger State, there are mild cases of SLE and WNF, however, cases presented by FF are at a peak due to the high abundance of *Cx. quinquefasciatus*. The current findings corroborated previous research on mosquito larvae distribution and abundance of the different genus in which *Culex* mosquitoes are included in different parts of Nigeria, (Akpan & Nwabueze 2015; Egwu et al. 2018). Similar mosquito species such as *Culex pipiens, Cx. restuans* (Olayemi et al. 2014), was previously reported in Minna, Niger State in addition to *Cx. quinquefasciatus, Cx. nigripalpus*, and *Cx. salinarius* in this current study. These findings further clarified the nature of mosquito breeding habitats, demonstrating that swamps and large water bodies can be equally conducive as gutters for *Culex* propagation.

The significantly high density of the three *Culex* species in large water bodies and gutters could be attributed to the high quantity of organic matter present from different sources due to the alteration of the aquatic habitat ecosystem. Consequently, these habitats promote mosquito propagation (Olayemi & Ande 2008). The density of adult mosquitoes is determined by the density of immature stages that are influence by positive physico-chemical conditions of the breeding habitats and not negatively affected by predators and competitors. According to Mutuku et al. (2006), Mwangangi et al. (2007), the size of an adult is influenced by the conditions in which it develops as a larva, longevity, fecundity, and blood meal volume are all affected by body size, and all these parameters may influence the vector's fitness for mosquito pathogen spread.

Similarly, each habitat has been identified as having unique ecological features that are critical to the success of anti-larval biological control measures aimed at reducing juvenile mosquito populations (Mala & Irungu 2011; Rydzanicz et al. 2016). Since resources are limited and cannot cover all mosquito breeding sites in the state, larval control (i.e. application of insecticides, environmental manipulation & management) should be focused on large water bodies and gutters.

In addition, *Cx. quinquefasciatus* were predominant compared to other *Culex* species in the breeding habitats and the LGAs. The distribution and abundance of these mosquitoes result from superior ecological adaptability to the eco-climatic conditions in these areas, promoting the rapid breeding and development of these mosquito populations (Akpan et al. 2018). The highest distribution of mosquitoes in given breeding habitat and quality of cues (physico-chemical) could be accountable for the equally high endemicity of mosquito diseases in the area (Evans & Adenomon 2014). In support of these findings, it has previously been suggested that mosquitoes' survival in each ecosystem is dependent on adaptations and specific physico-

chemical features that have a significant impact on the larval density of individual mosquito species. The existence of organic nutrients and other cues in the breeding habitat of the immature stage appears to influence mosquito abundance and development. The abundance of organic nutrients due to anthropogenic wastes such as home (refuse and sewage) and agricultural (fertilizer) waste could explain the gutter habitats' high larval abundance and immature survival when compared to other habitats (Madzlan et al. 2018; Kweka et al. 2019). Similar assertions by Austin et al. (2017), Kenawy et al. (2013), Khan and Khan (2018), Mavian et al. (2019), McClure et al. (2018) and Salihu et al. (2017), opined that ecological changes caused by human activities such as mining, marketing, farming, fishing, and household activities such as washing clothes and cooking dishes, along with a lack of basic sanitation and knowledge, allow vectors to spread and settle in inhabited regions. Also, when the population grows, the environment changes quickly, which might result in the unintentional creation of constructed environments that favour mosquito. Furthermore, agricultural activities such as rice growing have made a significant contribution to the creation of man-made mosquito breeding habitats. As a result, human actions and behaviours have continued to increase and renew the diversity of mosquito species' presence and growth (Adeleke et al. 2013).

The prevalence of several mosquito immature stages of *Culex* mosquitoes in this study indicates that *culex*-borne diseases (CBD) such as filariasis, and encephalitis may be a severe health risk. Therefore, for example, endemicity is attributable to *Culex* mosquitoes found in breeding habitats in the study area. This finding, suggests that with the known information on the ecology, breeding, and general biology of this mosquito species, it would be highly rewarding to target filarial fever vector control strategy against *Cx. quinquefasciatus* for optimum results in *culex*-borne vector (CBV) control in these study areas. Consequently, the high distribution of these mosquito species could be responsible for the rise in the endemicity of mosquito diseases in these LGAs, especially filarial fever and the entire state. Previously, studies on the prevalence of mosquito-borne diseases emphasized that filarial fever has spread in Niger State (Evans & Adenomon 2014; Olayemi et al. 2014; Omalu et al. 2015).

In addition, this study revealed monthly relative abundance peaked in July (gutters and swamps) and June (large water bodies) and declined in November in both habitats, reflecting the seasonal fluctuations on the population dynamics of different mosquitoes in habitats (Table 3, 4 and 5). This variation also affected the pathogen transmission pattern by these vectors in the state. Also, the study revealed that the mosquitoes are most abundant in June, July, and August in both habitats. This could be due to the environmental conditions which may influence the distribution and abundance of these *Culex* mosquitoes. Thus, the environmental conditions in the favourable months promoted the rapid growth and reproduction of mosquitoes. Other months, such as May, signal the start of the rainy season, whereas November is typically a dry month, preventing mosquito eggs from hatching and surviving in breeding habitats (Wang et al. 2020b).

The study found that all the chosen breeding habitats were suitable for the development of immature *Culex* with active abundance and survival at varying rates. The immature abundance of *Culex* within the breeding habitats revealed significant variations in the mosquito productions (Figure 2) with gutters, significantly higher abundance of L4, and while in swamps, and large water breeding habitats L1 were the most abundant of immature mosquitoes. Likewise, between the habitats, gutters had a significantly higher abundant of all the immature stages follow by swamp and larger water habitats.

The emergence of distinct immature stages in the breeding habitats might be explained by the fact that the breeding habitats quality nutrient contents and the immature stages are less accessible to predators. This current finding support the research work of Dida et al. (2015), that discovered a lower number of mosquito immature stages and higher abundant of predators that led to the invasion of mosquitoes breeding habitats.

CONCLUSION

Culicines mainly *Culex*, dominate the vector mosquito species in Niger State, especially in the examined LGAs. As a result, ongoing anti-anopheline control initiatives in the city should be expanded to include Culicines breeding areas, as these mosquitoes transmit diseases with loads comparable to malaria. Large water breeding habitats had the highest abundance of the three *Culex* species i.e., *Culex quinquefasciatus Cx. nigripalpus* and *Cx. Salinarious* followed by gutters and swamps. The conditions in larval habitats as well as season (rainy season) have a substantial impact on the mosquito density. Furthermore, anthropogenic activities are the primary drivers of mosquito growth in these ecosystems, and as a result, these activities should be closely monitored. The mosquito population diversity indices in Niger State show that the established species are well suited to the area's ecological characteristics and that effective control will require persistent vigorous larvicidal operations. The findings of this study will aid in a better knowledge of the epidemiology of mosquito-borne diseases in Minna, Niger State, which is necessary for long-term disease control.

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CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

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