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Original Article

Influence of Land-use on the Fitness of *Anopheles gambiae*, the Principal Vector of Malaria in Nigeria

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Abstract:
Background: Urbanization often results in profound environmental alterations that may promote the transmission of malaria. Though, land-use practices in urban areas have been linked with proliferations of suitable larval breeding habitats of malaria vectors, no attempt has been made to systematically investigate the influence of land-use practices on malaria transmission in Nigeria. **Objectives:** To elucidate the influence of land-use practices on larval development and adult body size of *Anopheles gambiae* (Diptera: Culicidae) mosquitoes in Minna, Nigeria. **Materials and Methods:** Newly-hatched larvae of *An. gmbiae* mosquitoes were reared in semi-natural habitats stationed in five different sites, each representing the major land-use types in the area. The larvae were monitored daily for Duration of Immature Development (DID) and Immature Survival Rate (ISR); while Wing Length (WL) was used as an index of adult body size. **Results:** DID, ISR and WL varied significantly ($P < 0.05$) among the land-use categories; with lager numbers of bigger mosquitoes produced at a faster rate in the artificial than natural land-use sites. Water temperature for larval development was best in the Refuse Dump (RD) site (mean = 28.11 ± 2.50°C) and consequently the shortest DID (mean = 9.70 ± 0.74 days), as well as, the largest mosquitoes (mean WL = 3.10 ± 0.90 mm), were recorded in this land-use category. However, while ISR was highest (mean = 96.30 ± 2.78%) in Farm Land (FL), the mosquitoes that emerged from this site were the smallest (mean WL = 1.96 ± 0.51mm). The Natural Vegetation (NV) land-use category was the least productive, as the larvae took the longest time (13.29 ± 1.69 days) to develop, and survived least (42.94 ± 7.50%) in this site. **Conclusion:** The land-use practices in Minna enhanced the fitness of *An. gambiae*, and may increase the vectorial capacity of the species for malaria transmission in the area. Targeted larviciding interventions will greatly contribute to malaria control efforts in Minna, Nigeria.

Key Words: Malaria, Mosquitoes, Survival Rates, Temperature, Wing Length.

Introduction:

Malaria remains the most important disease in the Tropics, where it is responsible for an estimated 300 to 500 million clinical attacks and over 1 million deaths each year.[1] The disease accounts for an estimated loss of about 44.7 million disability adjusted life years (DALYs), more than 80% of which are concentrated in sub-Saharan Africa.[2] In Nigeria, malaria is the number one public health problem and the country has been classified as one of the main high burden malaria-endemic countries. Nigeria bears Africa's greatest malaria burden, with at least 50% of the country's 140 million human population suffering from at least one episode of malaria every year.[3]

An. gambiae is the principal vector of malaria in Nigeria and other West African countries.[4-5] Over 90% of malaria transmission in Ilorin, Nigeria, is due to this important vector.[6] The vectorial capacity of *An. gambiae* is greatly enhanced by its breeding ecology; being a typical r-strategist, the species colonize temporary sunlit habitats in which selection favours rapid population increase.[7-8] Such habitats are rare in natural settings due to continuous vegetation cover, as common in rural areas.[9] On the other hand, proliferations of ideal habitats for *An. gambiae* in urban areas have been reported; primarily the results of changes in land cover.[10]

During the past decades, Nigeria has experienced very rapid population growth in its urban centers, due mainly to rural-urban human migration; a phenomenon that is changing the context for human population and natural systems interactions. Rapid urbanization results in profound demographic, ecological and socio-economic changes that may promote the transmission of parasitic diseases.[11]

Minna, being a state capital, is one of the fastest growing cities in Nigeria. In recent years, Minna, the once predominantly agricultural community,[12] has been gradually replaced by urban development. This development has led to unprecedented land-use changes in the city; such environmental changes often result in the creation of productive mosquito breeding sites and might promote the transmission of malaria in the area.[13]

In view of the endemicity of malaria in Minna,[14] and hence the potent risk factor to the rapidly growing human population, there is an urgent need to improve our understanding of the epidemiology of malaria, especially, those factors that promote its transmission. This study, the first of its kind in Nigeria, was therefore carried out to elucidate the influence of land-use practices on larval development and adult body size of *An. gambiae* mosquitoes in Minna, Nigeria, with a view to understanding which of the present land-use practices in the city promote the fitness of the species and hence, malaria transmission.

Materials and Methods:

Study Area

Minna, the capital of Niger state, Nigeria is located within longitude 6° 33¢E and latitude 9° 37¢N, covering a land area of 88 km^2 with an estimated human population of 1.2 million.[15] Minna has a tropical climate with mean annual temperature, relative humidity and rainfall of 30.20°C, 61.00% and 1334.00mm, respectively. The climate presents two distinct seasons: a rainy season between April and October, and a dry season (November - March) completely devoid of rains. The vegetation in the area is typically grass dominated savannah with scattered trees.

Categorization and selection of land-use sites

Land-use in Minna was initially classified into two broad categories namely, natural (essentially consisting of areas with typical Natural Vegetation (NV) cover, with little or no human interference) and artificial (areas with profound environmental alterations due to human interference). The artificial land-use types were further subdivided into the following categories: Residential Area (RA), Farm Land (FL), Refuse Dump (RD) and Commercial Area (CA). Five sites were selected for the study, each representing a land-use category. Initially, obvious larval habitats in each site were visually inspected to ascertain the presence of anopheline larvae.

Rearing of Mosquito Larvae at the Sites

In June 2008, ten semi-natural larval habitats (two replicates per site) were created using earthen pots (about 35cm diameter and 25cm deep). Two kilograms of dry soil, collected from the study site, and eight liters of rain water were placed in each pot. Two holes (about 3cm in diameter) were carefully bored into opposite ends of each pot near the top edge, to maintain a constant water level during periods of rainfall. Both the holes and open top of the earthen pots were screened with nylon netting (mesh size = 1mm) to prevent larvae from being washed away and colonization by wild mosquitoes.

Fifty, approximately three hours old, first instar larvae of *An. gambiae* from a colony maintained in the laboratory of the Federal University of Technology, Minna, Nigeria, were transferred to each earthen pot and transferred to the study sites. At the sites, the pots were partially buried in the ground at the same spot where the particular dry soil was originally excavated. The earthen pots were strategically placed among obvious anopheline larval habitats at each site. All experiments ran simultaneously at the study sites.

Initially, the experiments were monitored once per day from 0900 to 1200 hours, during which the temperature of the earthen pots were measured using mercury thermometer; but when the larvae began to pupate, the earthen pots were also examined from 4:00 P.M. to 7:00 P.M. Emerged adult mosquitoes were carefully collected from the water surface or under the netting, using an oral aspirator.[9] The adult mosquitoes were sexed and preserved in 4% formaldehyde solution prior to further analysis in the laboratory. The experiments were again repeated in July and August 2008 thus, resulting in the observation of 300 larvae per land-use site.

Determination of Adult body size

The adult mosquitoes were analyzed within 48 hours of collection. Wing length was used as an index of adult body size because it is closely correlated with dry weight.[16-17] The left wing of each specimen was removed, spread in a drop of distilled water on a microscope slide and covered with another slide to keep the wing immobile and flat. Wing length was measured from the alular notch to the apical margin, excluding the fringe of scales, using an ocular micrometer fitted to a dissecting microscope.[18] Only the left wing was measured, unless where damaged, because preliminary measurements showed no significant difference between the left and right wings.

Data Analysis

Duration of Immature Development (DID) was estimated as the time between egg-hatching and adult eclosion. The Immature Survival Rate (ISR) was determined as the proportion of introduced larvae (i.e., 100) that emerged as adults per site. Differences in a variable among sites were determined using Chi-square at P = 0.05 level of significance.

Results:

Table 1 shows mean water temperature and entomological variables of *Anopheles gambiae* in the five land-use categories investigated. Water temperature in the land-use categories during the study period was lowest in Farm Land (FL) (25.13 ± 0.60°C) and highest in the Refuse Dump (RD) (28.12 ± 1.63°C). Significantly ($P < 0.05$) higher water temperature was recorded in RD than the other land-use categories.

Table 1: Mean water temperature and entomological variables of *Anopheles gambiae* in Minna, Nigeria

Land-use	Temperature (°C)	DID (days)	ISR (%)	Sex Ratio (M : F)	WL (mm) Male (M)	WL (mm) Female (F)	WL (mm) Mean (M&F)
FL	25.13±0.60^{a*}	11.11±1.25^{b}	96.30±2.78^{e}	1.00 : 1.12^{a}	1.95±0.49^{a}	1.97±0.53^{a}	1.96±0.51^{a}
RA	27.28±1.70^{a}	10.30±1.80^{a}	68.15±2.45^{c}	1.00 : 0.98^{a}	2.85±0.50^{b}	2.92±0.75^{b}	2.89±0.63^{b}
NV	26.80±0.98^{a}	13.29±1.69^{c}	42.94±7.50^{a}	1.00 : 1.20^{a}	2.85±0.32^{b}	2.80±0.26^{b}	2.83±0.58^{b}
RD	28.11±2.50^{b}	9.70±0.74^{a}	81.22±3.37^{d}	1.00 : 1.29^{a}	3.05±0.70^{b}	3.15±1.09^{b}	3.10±0.90^{b}
CA	27.12±1.63^{a}	10.52±1.30^{a}	54.39±4.81^{b}	1.00 : 1.04^{a}	2.85±0.80^{b}	3.20±0.87^{b}	3.03±0.84^{b}
Aggregate	26.89±1.48^{a}	11.18±1.36^{b}	68.60±4.18^{c}	1.00 : 1.13^{a}	2.71±0.56^{b}	2.82±0.70^{b}	2.77±0.63^{b}

DID = Duration of Immature Development; ISR = Immature Survival Rate; WL = Wing Length; FL = Farm Land; RA = Residential Area; NV = Natural Vegetation; RD = Refuse Dump; CA = Commercial Area.
*Values followed by same superscript alphabet in a column are not significantly different at P = 0.05

Mean DID of the aggregate mosquito population was 11.18 ± 1.36 days. For the land-use categories, DID ranged from 9.70 ± 0.74 days in RD to 13.29 ± 1.69 days in NV. DID was in the following increasing order: RD < RA < CA < FL < NV. DID in RA, RD and CA were not significantly different (P > 0.05); but were significantly higher in FL (11.11 ± 1.25 days) and NV (13.29 ± 1.69 days), that were in turn significantly different from each other (P < 0.05).

Mean ISR of the aggregate mosquito population was 68.60 ± 4.18%. The distribution of ISR according to land-use was distinctly different from DID. While ISR was highest in FL (96.30 ± 2.78%), the species survived least in NV (42.94 ± 7.50%). ISR were significantly different (P < 0.05) among the land-use categories.

The sex ratio of the aggregate mosquito population was 1.00 : 1.13 (Male : Female). Sex ratios recorded in the different land-use categories were not significantly different (P > 0.05) from the values recorded for the aggregate mosquito population.

Mean Wing Length (WL) ranged from 1.96 ± 0.51mm in NV to 3.10 ± 0.90mm in RD. While WL was significantly lower (P < 0.05) in NV than in all other land-use types, such values were not significantly different (P > 0.05) among these other land-use types. Mean WL of the aggregate male (2.71 ± 0.56mm) and female (2.82 ± 0.70mm) mosquito populations were not significantly different (P > 0.05). However, for both sexes, wing length was significantly (P < 0.05) shorter in NV than the other land-use categories.

Discussion:

Despite the reported contributions of human activities to malaria prevalence,[19-20] this study is the first to assess how human-induced environmental alterations enhance the fitness of the principal vector of the disease in Nigeria. The results of this study showed that land-use type significantly influenced larval development of *An. gambiae* in Minna. Relatively larger numbers of bigger mosquitoes were produced at a faster rate in the artificial land-use sites than the Natural Vegetation site. practices in the city. Duration of Immature Development was significantly shorter in RD, RA and CA than FL and NV. This observation may be due to the relatively higher, water temperature recorded in the former than latter sets of land-use categories; as the rate of anopheline larval development depends greatly on water temperature of the breeding habitats.[21]

Immature Survival Rates varied significantly among the land-use categories; and was significantly lower in NV than the other land-use categories. This may be due to the quality of soil substrate of breeding habitats on larval development in the two broad categories of land-use namely, natural and artificial. Pre-study observations indicated that the soil substrate in NV had relatively lower amounts of organic debris than the artificial land-use categories. The larvae of *An. gambiae* depend largely on suspended organic particles in their habitats for nourishment,[22] and food availability is particularly important in determining larval developmental rates.[23]

A very high Immature Survival Rate (96.30 ± 2.78%) was recorded in FL. This finding, perhaps, confirms the influence of habitat quality on larval developmental success. The study site in FL was surrounded by patches of planted agricultural crops, especially, maize corn (*Zea mays*). Thus, the larval habitats at this site, perhaps, received additional larval food in the form of pollen grains shed by the surrounding crops. Maize pollen, in particular, constituted an important source of nutriment for anopheline mosquitoes in Ethiopia.[18]

Significantly bigger adult mosquitoes were produced in the artificial than natural land-use categories, suggesting that the present land-use practices in Minna may be enhancing the transmission of malaria in the area. Large adult mosquitoes live longer and lay more eggs than those that are smaller,[24] two major factors that significantly influence the intensity of malaria transmission.[9]

Conclusion:

The land-use practices in Minna enhanced the development and body size of *An. gambiae* thus, resulting in the production of better-fit adults. These favourable attributes of *An. gambiae* mosquitoes that emerged from sites of profound environmental alterations, may increase the vectorial capacity of the species for malaria transmission in Minna. Inherently, it is of value to preserve natural vegetation cover and where this is not possible, larval breeding habitats in areas that have been subjected to gross land-use alterations must be targeted for effective larviciding interventions, as the removal of such habitats will greatly contribute to malaria control effort.

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