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Influence of Environmental Factors on Breeding Habitats of Mosquito Species in Kosti City, White Nile State, Sudan

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Abstract

This study was conducted in Kosti town, White Nile State, Sudan, to determine the effect of the temperature and turbidity of water on breeding habitats of different species of mosquitoes. Data of water, temperature and turbidity were recorded. Immature stages of mosquitoes, collected by the dipping method from breeding habitats in three residential areas in Kosti town, were reared to adult stage and identified. It was found that the mean temperature of the water body from where large numbers were recovered ranged between 28-35°C. Aquatic stages of the mosquitos also, preferred water with high turbidity ranging from 451 to 650 ppm. The result concluded the early stage of most mosquito species were significantly positively correlated with temperature and turbidity of water.

Keywords: Breeding habitats; Mosquitoes; Temperature of water; Turbidity of water

Introduction

Water temperature is an important determinant of the growth and development rate of immature mosquitoes [1-3]. Mosquitoes are ectotherms, each life stage is dependent on temperature in the developmental and mortality rates [4]. The ability to withstand high temperatures differs among species and even among larval instars [2]. The immature stages of most species are extremely sensitive to temperatures above 40°C during development [5]. Water temperature is positively correlated to larval density [6]. Higher temperatures allow *Aedes* spp. to grow faster and reach the adult stage earlier [7].

Turbid water is preferred by mosquitoes over clear water [2]. However, et al. [8] found that the sites with relatively clear water produced more *An. Arabiens* is pupae and larger adults than habitats with turbid water. Larvae of *An. Gambiae* are found in both clear and turbid water [3]. *A. gambiae* are seen to breed more prolifically in temporary and turbid water bodies, such as ones formed by rain while, in contrast, *A. funestus* prefers more permanent water bodies [9]. However, a study for Teklu et al. [6] reported that the *A. gambiae* to exist in greater quantity in slightly turbid aquatic habitat than in turbid aquatic habitat. For instance, pH of 7.4 was found to be suitable for *Aedes* spp. [10,11]. Similarly, the work [12] established that water of a near neutral pH 6.8 to 7.2 was found most optimal for the weakening of the egg shells for the first instar larvae to emerge. [13] and Adebote et al. [10] suggested that pH <5.0 and slightly higher than 7.4 produced a lethal effect on mosquito species. Mosquito species were borne-diseases that cause serious public health problems in most states in Sudan. This situation causes high mortalities among the population of the aforementioned states. However, there are lack of an efficient surveillance system and published reliable data on ecology and bionomics of mosquito species. Therefore, this study is proposed to contribute to better understanding of ecology of mosquito species in areas at risk of diseases and to help health authorities to design proper vector control strategies that will reduce disease burden. The Objectives of this study to determine the effect of water temperature and turbidity on the breeding habitats of mosquitoes species in White Nile state, Sudan.

Materials and Methods

Study design

A two seasons study was conducted in selected houses and larval habitats in Kosti town, White Nile State, Sudan.

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Study area

Kosti town is located in Central Sudan. It lies between longitudes (13° 12' -13° 40'E) and between latitudes (13° 39'–13° 45' N), and at altitude 382 m above sea level. It has a long rainy season which lasts for five months (June-October). The mean annual rainfall is 600 mm; the monthly mean temperature is 22.5°C in winter and 34.5°C in summer and the mean annual relative humidity is 55% [14].

Collection of mosquitoes larvae

Larvae were collected from 5 breeding sites according to water habitats (water of broken pipes), clay pots, manholes near the bathroom, ponds and streams of water from the study area.

Collection was carried out in the morning hours from (7 am to 10 am) for three times a month (at the first week in each month) in autumn and winter in 2015, from three residential areas in Kosti town (Elnasr, Elshatee and Elrabaa). From each residential area, larvae were collected by the dipping method which was described by World Health Organization [15]. The dipper was lowered gently at an angle of about 45° to minimize disruption and either skim the top of the water or gentle lower it to cause the water and nearby larvae to flow into the dipper. Care was taken not to disturb the larvae to prevent them to swim downwards. The surface of the water was skimmed with the dipper, then, the dipper was left out of the water and poured in a white tray containing some water. Size of the dipper, number of dipping and method of scooping were kept uniform throughout the study period. Five replicates of dipping were carried out at each site. Larvae and pupae in each dip were counted and recorded according to their instars and placed in containers with water volume of about 800 mL. The containers were covered by gauze and kept at a temperature of between 28 to 30°C **(Figure 1)**.

Each sample was labeled to indicate date of collection, site and description of the habitat, locality and environmental determinants. A mercury thermometer was used to measure the water surface temperature while Paline Test was used to measure the turbidity of water.

Rearing of mosquitoes

It is often necessary to breed mosquitoes to the adult stage for the purpose of identification. Larvae were kept in water, to assist successful development to the adult stage [16]. Very small amount of rice was added to the water with larvae in trays for feeding. Water in trays was daily changed by clean water and rice. The pupae were sorted out by pipette and put in paper-cups with netting; each cup containing 50 pupae were placed into cages (75 × 75 × 75 cm) with fine meshes and with cotton impregnated with sugar solution inside the cages for adult feeding. After the emergence of the adult the mosquito, they were collected by an aspirator, put into nylon bags and exposed briefly to direct sunlight at ground level or put in a deep freezer to deactivate them. They were later kept in clean dry test tubes and covered by cotton wool until identified.

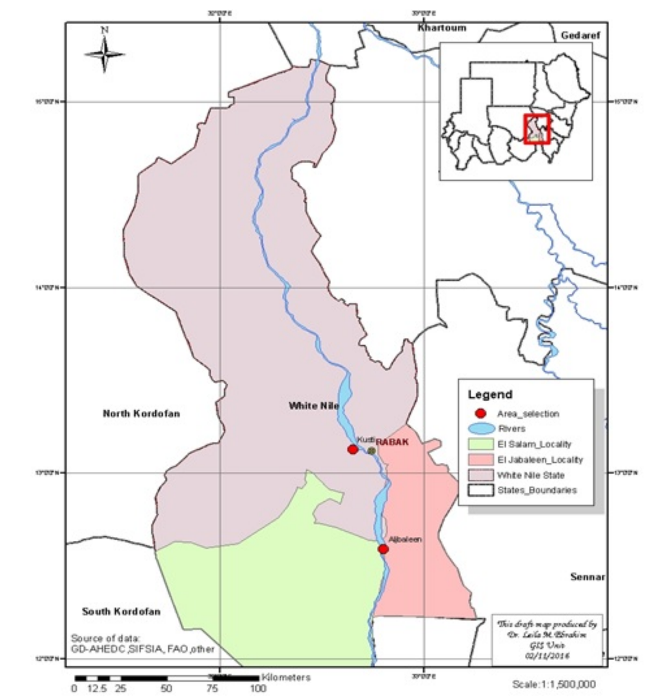

Figure 1 White Nile state showing localities (red region) from where samples were collected.

Identification of adult mosquitoes

Adult mosquitoes were identified under a dissecting microscope according to Edward [17], Russel et al. [18], Reuben et al. [19], Harbach [20], Rueda [21] and Andreadis et al. [22] and using the Anopheles mosquito keys of the Sudan Gillies et al. [23] and Gillies and Coetzee [24] methods. The identified adults were recorded according to the genera and species.

Statistical analyses

Data collected from different habitats were subjected to an appropriate general linear model (GLM) procedure of the statistical analyses using the SAS package. The SAS was used to perform mean separation using Ryan-Einot-Gabriel-Welsch Multiple F-test (REGWQ) [25]. Data when needed were subjected to transformation using (log10 ($\bar{x}$+1)).

Correlation analysis was carried out to determine the relationship between distribution and abundance of mosquitoes (immature and adults) among the different habitats, in different residential areas in autumn and winter seasons. Correlation analysis was also carried out to determine the relationship between distribution and abundance of mosquitoes among the different physical and chemical characteristics of water breeding sites [25].

Results

Effect of water temperature on the number of immature stages

Water temperature had significant effect on the number of immature stages. At the water temperature of (28-31°C) the mean number of immature stages of mosquito was found with the mean (40.13±2.50) that was significantly higher than (27.00 ± 5.67) at (24-27°C) **(Table 1)**. The first instar larvae, was recorded with the highest mean (14.64 ± 1.57) was at water temperature (28-31°C), and the lowest mean (3.13 ± 1.22) at water temperature (24-27°C) **(Table 1)**.

The first instar larvae were positively correlated with temperature ($r2=0.06462$, $n=168$, $P>0.05$) while the pupae was negatively correlated with temperature ($r2=-0.01481$, $n=168$, $P>0.05$). The first instar larvae were highly significantly positively correlated with water turbidity ($r2=0.26185$, $n=168$, $P\leq0.001$) but the pupae were significantly negatively correlated ($r2=-0.00946$, $n=168$, $P\leq0.001$) **(Table 2)**.

Effect of water turbidity on the number of immature stages

Water turbidity had modest effect on the number of immature stages of mosquitoes. The highest mean number of immature stages of mosquito (40.48 ± 3.64) was observed at water turbidity of 451-550 ppm and the lowest mean number (34.95 ± 5.19) was at water turbidity of 150-250 ppm **(Table 3)**. The first instar larvae were found with the highest mean (14.48 ± 2.03) at water turbidity of (451-550 ppm) and the lowest (10.03 ± 1.19) was at (351-450 ppm) **(Table 3)**.

Effect of water temperature on the adult composition reared from breeding habitats

The highest mean number (12.64 ± 1.33) of *Anopheles gambiae* was reared at water temperature of 32-35°C and the lowest mean number (6.38 ± 1.41)was reared at water temperature of 24-27°C. the highest mean number (15.25 ± 4.37) of *C. univitattus* was reared at water temperature of 24-27°C and the lowest mean number (2.84 ± 0.94) at (32-35°C) **(Table 4)**.

In this study *Culex quinquefasciatus*, was highly significantly negatively correlated with water temperature ($r2=-0.50714$, $n=168$, $P\leq0.001$) while *C. pipiens* was significantly positively correlated with water temperature ($r2=0.25072$, $n=168$, $P\leq0.001$). In this study *Culex quinquefasciatus* was highly significantly positively correlated with water turbidity ($r2=0.29832$, $n=168$, $P\leq0.001$) and *C. theileri* was significantly negatively correlated ($r2=-0.18737$, $n=168$, $P\leq0.05$) **(Table 5)**.

Effect of water turbidity on the adult composition reared from breeding habitats

In this study *Anopheles gambiae* complex was reared with the highest mean (13.86 ± 1.81) in water turbidity of (451-550 ppm) and the lowest (8.50 ± 1.06) in (351-450 ppm), *C. univitattus* was reared with the highest mean (14.40 ± 2.36) in (351-450 ppm) and the lowest (5.95 ± 1.97) in (150-151 ppm) **(Table 6)**.

Table 1 Mean (± SE) total number of immature stages of mosquitoes collected at different water surface temperature in autumn and winter in Kosti town.

Ranges of water surface temperature (°C)				
Immature stages	24-27 (8)*	28-31 (64)	32-35 (25)	36-39 (71)
Larva 1	3.13 ± 1.22^b	14.64 ± 1.57^a	13.92 ± 1.78^a	12.51 ± 1.02^a
Larva 2	4.38 ± 2.06^a	8.45 ± 0.10^a	5.92 ± 1.15^a	6.04 ± 0.74^a
Larva 3	3.63 ± 1.87^b	7.08 ± 0.68^a	6.40 ± 1.19^a	10.00 ± 1.36^a
Larva 4	7.62 ± 2.92^a	6.47 ± 0.63^a	6.08 ± 1.17^a	6.41 ± 0.77^a
Pupa	8.250 ± 2.84^a	3.48 ± 0.66^b	1.36 ± 0.63^b	1.58 ± 0.39^b
Total	27.00 ± 5.67^b	40.13 ± 2.50^a	33.68 ± 3.70^{a,b}	36.54 ± 2.70^{a,b}

[a]Mean separation was based on values transformed to log10 ($\bar{x}$+1).

[b]Means (± SE) with the same letter in each row are not significantly different at 5% level according to REGWQ range test

*Values in parenthesis=number of observations

Total=larva 1+larva 2+larva 3+larva 4+pupa

Table 2 Correlation analysis between water variables and number of immature stages of mosquitoes during autumn and winter in Kosti town.

Immature Stages	Water variables: Temperature	Turbidity
Larva 1	0.06462	0.26185***
	0.3682	0.0002
Larva 2	0.00175	0.11324
	0.982	0.1439
Larva 3	-0.12073	0.18184*
	0.119	0.0186
Larva 4	0.14079	-0.03095
	0.0687	0.6904
Pupa	-0.01481	-0.00946

	0.8489	0.9031

*P ≤0.05, **P≤0.01, ***P≤0.001, number of samples=364

Table 3 Mean (±SE) total number of immature stages of mosquitoes collected at different turbidity levels of water in autumn and winter in Kosti town.

Ranges of turbidity of water (ppm)					
Immature stages	150-250 (19)*	251-350 (65)	351-450 (30)	451-550 (29)	551-650 (25)
Larva 1	12.63 ± 2.21a	13.63 ± 1.19a	10.03 ± 1.59a	14.48 ± 2.03a	14.04 ± 2.75a
Larva 2	7.42 ± 1.72a	6.20 ± 0.80a	6.73 ± 1.47a	7.93 ± 1.31a	7.08 ± 1.27a
Larva 3	7.53 ± 1.57a	8.66 ± 1.38a	8.23 ± 1.45a	7.56 ± 1.01a	7.24 ± 1.38a
Larva 4	6.32 ± 1.08a	6.80 ± 0.91a	6.7 ± 0.95a	6.34 ± 0.96a	5.32 ± 0.88a
Pupa	1.05 ± 0.59b	1.02 ± 0.34b	4.40 ± 1.14a	4.21 ± 0.98a	3.80 ± 1.08a
Total	34.95 ± 5.19a	36.31 ± 2.99a	36.17 ± 2.97a	40.48 ± 3.64a	37.48 ± 3.65a
aMean separation was based on values transformed to log10 ($\bar{x}$+1).					
bMeans (± SE) with the same letter in each row are not significantly different at 5% level according to REGWQ range test					
*Values in parenthesis=number of observations					
Total=larva 1+larva 2+larva 3+larva 4+pupa					

Table 4 Mean (± SE) total number of adult mosquito species (and) reared from developmental stages collected from breeding site of different surface temperature in autumn and winter in Kosti town.

Ranges of water surface temperature (°C)				
Mosquito species	24-27 (8)*	28-31 (64)	32-35 (25)	36-39 (71)
A. gambiae complex	6.38 ± 1.41a	9.77 ± 1.04a	12.64 ± 1.33a	10.25 ± 0.93a
A. pharoensis	12:00 AM	0.06 ± 0.05a	0.12 ± 0.09a	0.30 ± 0.12a
C. quinquefasciatus	4.50 ± 1.61a	12.67 ± 1.60a	7.36 ± 1.97a	9.10 ± 1.27a
C. univittatus	15.25 ± 4.37a	14.75 ± 1.57a	2.84 ± 0.94b	4.01 ± 0.61b
C. theileri	12:00 AM	12:00 AM	1.76 ± 0.61a	1.62 ± 0.42a
C. pipiens	0.25 ± 0.25a	0.48 ± 0.19a	1.52 ± 0.49a	1.66 ± 0.35a
aMean separation was based on values transformed to log10 ($\bar{x}$+1)				
bMeans (±SE) with the same letter in each row are not significantly different at 5% level according to REGWQ range test				
*Values in parenthesis=number of observations.				

Table 5 Correlation analysis between water variables and number of adult mosquito species (and) collected in autumn and winter in Kosti.

	Water variables	
Mosquito species	**Temperature (°C)**	**Turbidity (ppm)**
	0.09657	0.00943
A. gambiae **complex**	0.213	0.9034
A. pharoensis	0.14623	-0.05282
	0.0586	0.4965
	-0.50714***	0.29832***
C. quinquefasciatus	0.0001	0.0001
	-0.50714***	0.29832***
C. univittatus	0.0001	0.0001
	0.30262***	-0.18737*
C. theileri	0.0001	0.015
C. pipiens	0.25072***	0.11212

	0.001	0.1479	*P≤0.05, **P≤0.01, ***P ≤ 0.001, number of samples=364

Table 6 Mean (± SE) total number of adult mosquito species (and) reared from developmental stages collected from breeding sites of different water turbidity levels in Autumn and wintering Kosti town.

	Turbidity of water ranges (ppm)				
Mosquito Species	150-250 (19)*	251-350 (65)	351-450 (30)	451-550 (29)	551-650 (25)
A. gambiae complex	10.16 ± 1.87^{a}	10.95 ± 1.00^{a}	8.50 ± 1.06^{a}	13.86 ± 1.81^{a}	9.36 ± 1.79^{a}
A. pharoensis	0.26 ± 0.18^{a}	0.20 ± 0.11^{a}	0.10 ± 0.10^{a}	0.03 ± 0.03^{a}	0.24 ± 0.17^{a}
C. quinquefasciatus	12.75 ± 4.25^{a,b}	8.40 ± 1.17^{ab}	5.83 ± 1.13^{b}	13.90 ± 2.22^{a}	12.44 ± 2.36^{a,b}
C. univittatus	5.95 ± 1.97^{b,c}	4.71 ± 0.77c	14.40 ± 2.36^{a}	10.72 ± 2.29 ^{a,b}	10.40 ± 2.32^{a,c}
C. theileri	1.95 ± 0.73^{a}	1.14 ± 0.43^{a}	1.17 ± 0.46^{a}	0.24 ± 0.13^{a}	0.24 ± 0.18^{a}
C. pipiens	1.26 ± 0.52^{a,b}	0.85 ± 0.25^{b}	0.70 ± 0.29^{b}	0.12 ± 0.55^{a,b}	2.44 ± 0.77^{a}
[a]Mean separation was based on values transformed to log10 ($\bar{x}$+1)					
[b,c]Means (±SE) with the same letter in each row are not significantly different at 5% level according to REGWQ range test					
*Values in parenthesis=number of observations					

Discussion

Water temperature is one of the most important factors that account for development of larvae [26]. It is an important determinant of development rate of immature mosquitoes [2,3]. A study demonstrated that the immature stages of most species are extremely sensitive to temperatures above 40°C during development [27]. The authors of that study reported that average optimum temperature for the development of most mosquito species is around 25-27°C and the development could be arrested at 10°C. In this study, the first instar larvae were more susceptible to water temperature. It is possible that the development of immature stages were suppressed by high and the low water temperature. Low water temperature prolonged the development and high water temperature caused death of the immature stages. This assumption is in line with that reported by Craig et al. [9] who found the thermal death for mosquitoes was around 40-42°C. Slightly higher water temperature results in a faster development. According to Paaijmans [2], an ability to withstand high temperatures differs among species and even among larval instars. Older larvae seem to be more susceptible to heat than younger ones. In this study, water temperature was significantly negatively correlated with number of first and second instar and it was highly significantly negatively on number of pupae. This explains that the newly emerged larvae were affected by low water temperature, which probably elongate the developmental period for the first and second instar and pupa. The third and fourth instar was significantly positively correlated with water temperature. According to Ibrahim [5] who found the immature stages of most species are extremely sensitive to temperatures above 40°C during the development. Tolerance to temperature depends on the species [28]. In the present study, the mean water temperature for development was found ranging between 24°-39°C. *A. gambiae* complex collected was significantly positively correlated with water temperature in Kosti town. This explains that *A. gambiae* complex prefers development and breeding at high degree of water temperature. A study of Teklu et al. [6] they found that water temperature was positively correlated with mean larval density of the dominant species. The development rate of *A. gambiae* complex increased up to water temperatures of approximately 37°C, after which it sharply decreased [2]. In this study, *An. gambiae* complex was also observed with tolerant to relatively low degree of water temperature (24-27°C). Water temperature was positively correlated with mean larval density of the dominant species, *A. pharoensis* which is tolerant to relatively high water temperatures [6]. However, Lindsay et al. [4] reported that water temperatures in the pools that are preferred by *An. gambiae* mosquitoes are warmer than air temperatures.

Turbid water is preferred to clear water [2]. In the current study, most stages preferred water with high turbidity ranging from (551-650 ppm). This explains that high turbidity of water was important for development of other instars and the high water turbidity possibly suppressed development of fourth instar larvae. At the same time, high water turbidity was preferred by the pupal stage.

Anopheles gambiae complex were positively correlated with water turbidity. It is possible that factors other than turbidity affected the latter genus. A study of Paaijmans [2] reported that turbid water is preferred for mosquitoes over clear water. However, another study reported that *An. gambiae* complex was greater in slightly turbid aquatic habitat than in turbid aquatic habitat *Culex* spp. preferred turbid water in the same season [6].

Conclusion

Most species were significantly positively correlated with temperature and turbidity of water.

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