Biodiversity and Density of Larvae and Adults of Anopheles Mosquitoes in El Obied City – Sudan

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To cite this article:
Adam Musa Adam Eissa, Mutman Ali Abdghder Kehail. Biodiversity and Density of Larvae and Adults of Anopheles Mosquitoes in El Obied City – Sudan. International Journal of Health Economics and Policy. Vol. 4, No. 2, 2019, pp. 58-66. doi: 10.11648/j.hep.20190402.14

Received: July 4, 2018; Accepted: August 15, 2018; Published: May 30, 2019

Abstract: The weather changes especially rainfall affects the distribution and densities of mosquitoes. There are about 380 species of Anopheles, recorded, sixty of them act as vectors of many diseases. This work was carried out to study the biodiversity and density of Anopheles mosquitoes {adults and larvae} in El Obied City. A cross-sectional survey of Anopheline mosquito larval habitats was conducted during {April 2014 - April 2016}. Larvae were collected by using the standard dipping and netting techniques weekly for the whole year from five selected stations, while adult stages were collected by spray sheet method using Permethrin 25% E. C. The climatic factors and the malaria cases among the study areas were also recorded. The DNA from the identified adults and larvae was extracted in order to make the molecular confirmation for these species. The results revealed that, all Anopheline mosquito larvae {100%} which were found and collected from three breeding sites during all seasons were classified as probably Anopheles squamosus. The study also showed that, all {100%} of adults Anopheline mosquito were classified as most probably A. squamosus and then this result was confirmed by the National Laboratory for Public Health, Medical Entomology Department, Khartoum {as a first record for this sp.}. A. squamosus were found only in the shallow pond water habitat in El Obied City. The mean Anopheline density in the study area for larvae was 0.42 per dip while the mean density of adults was 0.55 per room. The high mosquito larval density in El Obied City indicated that, it is at risk of mosquito-borne diseases including malaria. The correlation analysis between mosquito abundance and density to the malaria cases within the selected stations in El Obied City, was positive. The macro and microclimate within which the larvae and the adults A. squamosus lived were also compared to that of other Anopheles species studied before, and new limits were recorded concerning mainly: temperature {15.5 - 41.1°C}, relative humidity{16–82%} and rainfall {6.3–88.2 mm} in respect to outdoor and indoor resting mosquitoes specially for adults, and temperature {24–27°C}, pH {6.5–7.2} and salinity {1.6–1.9}of larval breeding site waters in addition to the floral types. This study is recommended to be conducted in the other Cities and Sudan.

Keywords: Anopheles, squamosus, Mosquitoes, El Obied

1. Introduction

Mosquitoes are important vectors of several tropical diseases. About one hundred species act as vectors of human diseases. Examples of such disease are malaria, Filariasis, Japanese encephalitis and yellow fever some species were reported to transmit arbovirusis. However, Anopheles spp, beside transmitting malaria, they are capable to transmit Filariais. Other biting members of Anopheles spp are nuisance of man [1]. Every 30 seconds a child dies of malaria with the vast majority of deaths occurring in Africa, South of the Sahara. Infection is primarily among pregnant women and children under five years of age, accounting for around 20% of deaths and 10% of the continent's overall disease burden [2]. There are at least 300 million acute cases of malaria, each year globally, resulting in a million deaths. Ninety percent of these deaths occur in Africa, mostly in young children. The disease...
has been estimated to cost Africa more than $12 billion every year in Gross Domestic Product, even though it could be controlled to a fraction of that amount. Malaria in Africa as a whole accounts for 40% of public health expenditures, 30-50% of inpatient admissions, and up to 50% of outpatients visits in areas with high malaria transmission. Malaria kills more people today than it did 30 years ago. In Sudan Malaria is a disease of considerable magnitude and serious impact, it ranks on the top of the list of diseases with highest rates of morbidity and mortality estimated pointed that each year 7.5 million and 35,000 deaths; this represents 50% and 70% of all EMRO cases and deaths. This is in addition to its indirect impact on health services utilization and loss of working days and school absenteeism [3] more details mentioned by [4].

Based on climate models, it is estimated that 75% of the population of Sudan {37 millions} are at risk of malaria, while 25% are at risk of epidemic malaria [5].

Malaria is one of the public health problems in El Obied City. The proportion of malaria cases {probable and confirmed} from the total Outpatient attendance are 22.5%, 16%, 11.5%, 8.5%, 8% in 2009, 2010, 2011, 2012 and 2013, respectively [6] The general objective was To identify different species and habitats of Anopheles in El Obied City. when the Specific objectives were to record the breeding sites and habitats of Anopheles mosquitoes in El Obied City, To check the distribution of Anopheles mosquitoes within El Obied City and to determine the diversity and density of Anopheles mosquitoes in El Obied City during the study period.

2. Materials and Methods

2.1. Study Area

A cross-sectional study was conducted during the dry season in randomly selected five stations to reveal the distribution of Anopheles mosquitoes in El Obied City during the period [April 2014 – April 2016]. El Obied is a City located in the center of the state of North Kordofan in Sudan. It has an average height of 650 meters above sea level and lies about 588 kilometers south-west of Khartoum, the capital. It is one of the largest cities of Sudan and is renowned as a market for cash crops in Sudan, the largest Bursaha of gum Arabic in the world, and has a historic place in the revolution of Mahdia. It is also an important crossroads, and commercial and agricultural center prominently. Also passes through an oil pipeline extending from south to east to Port Sudan.

2.2. Measurement of Climate and Microclimate Factors

El Obied occupies a strategic position in central Sudan between its geographic regions and different climatic and economic: between the desert and semi-desert area, and between savannah zone area dry and wet and between the dry tropics and rainforest areas, western Sudan, south and east. The climatic factors data include temperatures, humidity and rainfall were obtained from El Obied Airport Map and geographical information [2014].

The macro and microclimate within which the larvae and the adults A. squamosus lived were also measured concerning mainly: temperature {thermometer}, humidity {Hygrometer} and respect to outdoor and indoor resting mosquitoes specially for adults, and temperature {thermometer}, pH {ph-meter} and salinity {Measuring Semaphore} of larval breeding site waters.

2.3. Collection and Mounting of Mosquito’s Samples

Mosquitoes larvae were collected from different selected breeding sites. Permanent bodies of water such as lakes or stream pools also contained larvae of mosquito species. In addition to that adult stages were collected by rearing larvae in special cages and from different rooms {for identification of the species} and by using spray sheet method to determine Anopheles density during the study period.

A sampling frame was constructed and it consisted of five stations {which were enumerated} according to their geographical locations {from north to south, east to west and middle}. The selected stations were randomly picked by using random sampling. Mosquitoes larvae were collected during 12 months from all available breeding sites in the five stations included in the study. From each station, one type of breeding site with positive larvae was photographed {one from broken pipe or Bricks factory, etc}. The positive Anopheles breeding sites in the five stations were sampled during the all seasons of the year. Larvae {all instars} were collected from shallow, bricks factory habitats, jars and broken pipes.

Larval surveys were done weekly using dipping and netting techniques as was recommended by [7] according to the type and size of the breeding place. Identification of larvae was done according to the key of of Common Anopheles larvae in Sudan [1960]. However, in practice, ten dips were usually taken arbitrarily from each positive site. Larvae were transferred to El Obied Central laboratory and kept in 70% alcohol then mounted on slides and identified probably then confirmed from national laboratory for Public Health, Medical Entomology Department {Khartoum, Sudan – P. O. Box 287} done according to the key for Identification of [8]. The material which were used for larvae mounting process, include {pipette, slides, cover glass, pins, D. P. X, filter paper, Petri dish and dissection microscope}.

Adult mosquitoes collected from Five rooms were selected randomly from each selected area by using White Sheets Spray method, techniques as was recommended by [9] according to type and size of the rooms. Identification of adult was done according to the key for Identification of Common Anopheles adult in Sudan {1960}. However, in practice, adult were usually taken at the morning {08:30, AM} from each positive room. Adults were transferred to El Obied Central laboratory and kept in silica gel then pined and identified probably then confirmed from national laboratory for public health, medical entomology department {Khartoum, Sudan – P. O. Box 287} done according to the key for Identification of [8]. The following materials were used: Hudson Pump {8 L}, Permethrin pesticide 25% E. C, and white sheets and silica gel.
2.4. Data Analysis

The obtained data were first summarized and loaded to Excel program 2007 worksheet. The Pie chart and the descriptive statistics, were run, so as to evaluate the diversity and the density of mosquitoes in El Obeid City.

3. Results

3.1. Mosquito’s Diversity and Density

Results presented in {Tables 1 & 2 and 3} during year 2014 which depended on the morphological characteristics presented in Figure {1 and 2}, showed that, the number of Anopheles larvae collected from different types of breeding habitats in El Obeid City during 12 months and indicated that, a total of only 1635 Anopheles mosquito larvae were found and collected from three locations {Shallow pond of Alrahma west {north}, Almodereua {west} and Alseka hadied {middle} } during the test period, whereas the other two locations, were not inhibited with mosquito larvae. The collected Anopheles larvae were identified according to their morphological characteristics and the identification proved that {1635} 100% larvae were identified as probably Anopheles squamosus according to the [10]. and then confirmed from National laboratory for public health, Medical Entomology Department. Other Anopheles spp larvae were not found in all sampled larvae.

It is obvious that, the mean density of Anopheles larvae in shallow habitats {pond} were found {2.725} per dip {1635 out 50; 5 location and 10 dips, 12 months} and the mean density of Anopheles larvae in El Obeid City were 0.908 {1635 out 150; 5 location, 3 habitat and 10 dips}.

As can be seen from {Table 4 & Table 5 & Table 6} during year 2015 the descriptive statistics analysis revealed that, the mean Anopheles larvae was 0.520 per dip with standard error SE of 0.171, the standard deviation SD was 4.204, the range {the difference between maximum and minimum} was 41, the minimum {min} number collected was 0, the maximum {max} number collected was 41, while the total {sum} was 250 larvae.

It is obvious that, the mean density of Anopheles larvae in shallow habitats {pond} were found {0.416} per dip {250 out 50; 5 location and 10 dips, 12 months} and the mean density of Anopheles larvae in El Obeid City were 0.138 {250 out 150; 5 location, 3 habitat and 10 dips}.

Table 1. Percentage of larvae Mosquito’s diversity which were identified in El Obeid City during 2014.

| Area         | Number of Collected Larvae | % of An. Squamosus | % of An. nili | % of An. Arabiensis |
|--------------|----------------------------|--------------------|---------------|---------------------|
| Alrahma west | 825                        | 100                | 0             | 0                   |
| Almodereua   | 450                        | 100                | 0             | 0                   |
| Almatar      | 0                          | 0                  | 0             | 0                   |
| Alseka hadied| 360                        | 100                | 0             | 0                   |
| Arafat       | 0                          | 0                  | 0             | 0                   |

Table 2. Number of Anopheles squamosus larvae and Adults collected from El Obeid City during 2014.

| Number of Area | Number of larvae | Number of Adults |
|----------------|------------------|-----------------|
| Alrahma west   | 825              | 680             |
| Almodereua     | 450              | 222             |
| Almatar        | 360              | 600             |
| Alseka hadied  | 360              | 0               |
| Arafat         | 0                | 0               |
| Descriptive statistics | | |
| Mean          | 27               | 25              |
| SE            | 1.618            | 1.528           |
| SD            | 12.537           | 11.834          |
| Range         | 0-825            | 0-680           |
| Min           | 0                | 0               |
| Max           | 825              | 680             |
| Sum           | 1635             | 1502            |

Table 3. Number and Density of Anopheles squamosus larvae collected from El Obeid City during 2014.

| Month | Area         | Alrahma west | Almodereua | Almatar | Alseka hadied | Arafat | Total |
|-------|--------------|--------------|------------|---------|---------------|--------|-------|
| Jan.  | 0            | 0            | 0          | 0       | 0             | 0      | 0     |
| Feb.  | 0            | 0            | 0          | 0       | 0             | 0      | 0     |
| Mar.  | 0            | 0            | 0          | 0       | 0             | 0      | 0     |
| Apr.  | 0            | 0            | 0          | 0       | 0             | 0      | 0     |
Results explained in that {Table 7} & {Table 8}, during year 2014, the number of Anopheles adults collected from different types of rooms in El Obeid City and showed that, a total of only 1502 Anopheles mosquito adults were found and collected from three locations {5 rooms per area randomly}: Alrahma west, Almodereua and Almatar during the test period, whereas none was found in the other two locations. The collected Anopheles {1502} 100% Adults were identified as probably Anopheles squamosus according to their distinguished morphological characteristic features described according to the [10]. And then confirmed by National laboratory for Public Health, Medical Entomology Department. Other Anopheles spp larvae were not found in all sampled Adults.

The descriptive statistics analysis revealed that, the mean of Anopheles adults was 25 with standard error SE of 1.528, the standard deviation SD was 11.834, the range {the difference between maximum and minimum} was 680, the minimum {min} number collected was 0, the maximum {max} number collected was 680, while the total {sum} was 1502 Adults. It is obvious that, the mean density of Anopheles squamosus adults

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Table 4. Percentage of larvae Mosquito's diversity which were identified in El Obeid City during 2015.

| Area          | Number of Collected Larvae | % of An. Squamosus (%) | % of An. nili (%) | % of An. Arabiensis (%) |
|---------------|-----------------------------|------------------------|------------------|-------------------------|
| Alrahma west  | 68                          | 100                    | 0                | 0                       |
| Almodereua    | 122                         | 100                    | 0                | 0                       |
| Almatar       | 0                           | 0                      | 0                | 0                       |
| Alseka hadied | 60                          | 100                    | 0                | 0                       |
| Arafat        | 0                           | 0                      | 0                | 0                       |

Table 5. Number of Anopheles squamosus larvae and Adults collected from El Obeid City during 2015.

| The Area          | Number of larvae | Number of Adults |
|-------------------|------------------|------------------|
| Alrahma west      | 68               | 74               |
| Almodereua        | 122              | 46               |
| Almatar           | 0                | 45               |
| Alseka hadied     | 60               | 0                |
| Arafat            | 0                | 0                |

Table 6. Number and Density of Anopheles squamosus larvae collected from El Obeid City during 2015.

| Month | Area          | Number of larvae | Number of Adults |
|-------|---------------|------------------|------------------|
| Jan.  | 0             | 0                | 0                |
| Feb.  | 0             | 0                | 0                |
| Mar.  | 0             | 0                | 0                |
| Apr.  | 0             | 0                | 0                |
| May.  | 0             | 0                | 0                |
| Jun.  | 0             | 0                | 0                |
| Jul.  | 18            | 32               | 13               |
| Aug.  | 35            | 41               | 29               |
| Sep.  | 15            | 23               | 11               |
| Oct.  | 0             | 19               | 7                |
| Nov.  | 0             | 0                | 0                |
| Des.  | 0             | 0                | 0                |
| Total | 68            | 122              | 60               |
in model room was \{2.50\} per room {1502out 50; 5 locations, 12 months} and the mean density of Anopheles squamosus adults in El Obeid City were 0.834 {1502 out 150; 5 locations, 12 month and 3 model rooms}. Results in {Table 9} & {Table 10}, showed the descriptive statistics analysis which revealed that, the mean Anopheles squamosus adults was 0.343 per room with standard error SE of 0.215, the standard deviation SD was 3.729, the range (the difference between maximum and minimum) was 38, the minimum {min} number collected was 0, the maximum {max} number collected was 38, while the total {sum} was 165 adults. And when the collected Anopheles {165} 100% Adults were identified as probably Anopheles squamosus according to their distinguished morphological characteristic features described according to the [10]. And then confirmed in national laboratory for public Health, Medical Entomology Department. Other Anopheles spp larvae were not found in all sampled Adults.

It is obvious that, the mean density of Anopheles squamosus adults in model room was found {0.068} per room {165out 50; 5 locations and 12 months} and the mean density of Anopheles squamosus adults in El Obeid City was 0.022 {165 out 150; 5 locations, 12 months and 3 model rooms}.

| Month | Area          | Alrahma west | Almodereua | Almatar | Alseka Hadied | Arafat | Total |
|-------|---------------|--------------|------------|---------|---------------|--------|-------|
| Jan.  | 0             | 0            | 0          | 0       | 0             | 0      | 0     |
| Feb.  | 0             | 0            | 0          | 0       | 0             | 0      | 0     |
| Mar.  | 0             | 0            | 0          | 0       | 0             | 0      | 0     |
| Apr.  | 0             | 0            | 0          | 0       | 0             | 0      | 0     |
| May.  | 0             | 0            | 0          | 0       | 0             | 0      | 0     |
| Jun.  | 0             | 0            | 0          | 0       | 0             | 0      | 0     |
| Jul.  | 242           | 100          | 296        | 0       | 0             | 638    | 1502  |
| Aug.  | 438           | 122          | 304        | 0       | 0             | 864    |
| Sep.  | 0             | 0            | 0          | 0       | 0             | 0      | 0     |
| Oct.  | 0             | 0            | 0          | 0       | 0             | 0      | 0     |
| Nov.  | 0             | 0            | 0          | 0       | 0             | 0      | 0     |
| Des.  | 0             | 0            | 0          | 0       | 0             | 0      | 0     |
| Total | 680           | 222          | 600        | 0       | 0             | 1502   |

| Area          | Number of Collected Adults | % of An. Squamosus | % of An. nili | % of An. arabiensis |
|---------------|----------------------------|--------------------|--------------|----------------------|
| Alrahma west  | 680                        | 100                | 0            | 0                    |
| Almodereua    | 222                        | 100                | 0            | 0                    |
| Almatar       | 0                          |                    | 0            | 0                    |
| Alseka hadied | 600                        | 100                | 0            | 0                    |
| Arafat        | 0                          |                    | 0            | 0                    |

| Area          | Number of Collected Adults | % of An. Squamosus | % of An. nili | % of An. arabiensis |
|---------------|----------------------------|--------------------|--------------|----------------------|
| Alrahma west  | 74                         |                    | 0            | 0                    |
| Almodereua    | 46                         |                    | 0            | 0                    |
| Almatar       | 45                         |                    | 0            | 0                    |
| Alseka hadied | 0                          |                    | 0            | 0                    |
| Arafat        | 0                          |                    | 0            | 0                    |

| Month | Area          | Alrahma west | Almodereua | Almatar | Alseka hadied | Arafat | Total |
|-------|---------------|--------------|------------|---------|---------------|--------|-------|
| Jan.  | 0             | 0            | 0          | 0       | 0             | 0      | 0     |
| Feb.  | 0             | 0            | 0          | 0       | 0             | 0      | 0     |
| Mar.  | 0             | 0            | 0          | 0       | 0             | 0      | 0     |
| Apr.  | 0             | 0            | 0          | 0       | 0             | 0      | 0     |
| May.  | 0             | 0            | 0          | 0       | 0             | 0      | 0     |
| Jun.  | 0             | 0            | 0          | 0       | 0             | 0      | 0     |
| Jul.  | 36            | 24           | 21         | 0       | 0             | 81     |
| Aug.  | 38            | 22           | 24         | 0       | 0             | 84     |
| Sep.  | 0             | 0            | 0          | 0       | 0             | 0      | 0     |
| Oct.  | 0             | 0            | 0          | 0       | 0             | 0      | 0     |
| Nov.  | 0             | 0            | 0          | 0       | 0             | 0      | 0     |
| Des.  | 0             | 0            | 0          | 0       | 0             | 0      | 0     |
| Total | 74            | 46           | 45         | 0       | 0             | 165    |
3.2. The Ecology of the Study Area

3.2.1. The Climate Factors

Results presented in Table 11 showed that the temperature during 2015 ranged between 15.5 °C in December and 41.1 °C in March and May, while the relative humidity RH {Table 12} ranged between 16% in April and 82% in August, whereas the rainfall amount {Tables 13 and 14} ranged between 0 from November to April and –229.9 mm in August during which the rainy days were 11, and these values showed the ranges of the climatic factors that Anopheles squamosus can endure.

3.2.2. The Microclimate Factors

The temperature °C of indoor resting habitat in El Obeid City during 2015 ranged between 23.5 °C in Almodereua area and 28°C in Almatar area with a difference of 4.5°C, while that of outdoor ranged between 36 °C in Almodereua area and 39°C in Almatar area with a difference of 3°C {Table 15}. The relative humidity [%] of indoor resting habitat in El Obeid City during 2015 ranged between 37% in Almatar area and 82% in Almodereua area with a difference of 45%, while that of outdoor ranged between 16% in Almatar area and 40% in Almodereua area with a difference of 24% {Table 16}. The temperature °C in positive larval breeding site waters in El Obeid City during 2015 ranged between 27 (in Almatar area) and 30°C (in Almodereua area) with a difference of 3°C, while that of normal waters ranged between 24 (in Almatar area) and 28°C (in Almatar area) with a difference of 4°C {Table 17}.

The pH in positive larval breeding site waters in El Obeid City during 2015 ranged between 6.5 in Alrahma west area and 7.22 in Arafat area with a difference of 0.72, while that of normal waters ranged between 6.55 in Almatar area and 7.29 in Almodereua area with a difference of 0.74 {Table 18}. The salinity in positive larval breeding site waters in El Obeid City during 2015 ranged between 1.6 in Alrahma west area and 1.9 in Almatar area with a difference of 0.3, while that of normal waters ranged between 1.73 in Alrahma west area and 1.99 in Alseka hadied area with a difference of 0.26 {Table 19}. The An. Sp. adults and larvae correlation with Malaria cases was 0.64855 while the larvae correlation with Malaria cases was 0.92043 in El Obeid City during 2015 {Table 20}. The observed values showed the ranges of the microclimatic in which Anopheles squamosus live with in El Obeid City.

Table 11. Temperature Record (°C) in El Obeid City during the year 2015.

| Month    | Maximum | Minimum | Mean |
|----------|---------|---------|------|
| January  | 33.6    | 16.5    | 25.2 |
| February | 36.3    | 18.6    | 27.5 |
| March    | 41.1    | 26.8    | 34.0 |
| April    | 40.8    | 22.7    | 31.7 |
| May      | 41.1    | 25.8    | 36.5 |
| June     | 38.0    | 25.3    | 32.3 |
| July     | 35.7    | 24.5    | 30.1 |
| August   | 32.1    | 22.7    | 27.4 |
| September| 34.8    | 22.6    | 28.6 |
| October  | 37.6    | 31.4    | 30.5 |
| November | 36.6    | 18.6    | 28.1 |
| December | 40.5    | 15.5    | 30.5 |

Table 12. Mean Relative Humidity (%) and Evaporation in El Obeid City during the year 2015.

| Month  | Relative Humidity | Evaporation |
|--------|-------------------|-------------|
| January| 30                | 7.6         |
| February| 21               | 7.2         |
| March  | 18                | 15.6        |
| April  | 16                | 6.3         |
| May    | 37                | 15.6        |
| June   | 56                | 22.2        |
| July   | 67                | 23.0        |
| August | 82                | 25.7        |
| September | 72               | 23.4        |
| October| 40                | 13.6        |
| November| 31               | 11.1        |
| December| 36               | 10.1        |

Table 13. Rainfall (mm) in El Obeid City during the year 2015.

| Month    | Total Rain | Rainy Days |
|----------|------------|------------|
| January  | Nil        | 0          |
| February | Nil        | 0          |
| March    | Nil        | 0          |
| April    | Nil        | 0          |
| May      | 1.25       | 1          |
| June     | 27.6       | 5          |
| July     | 88.2       | 10         |
| August   | 229.9      | 11         |
| September| 31.6       | 5          |
| October  | 4.0        | 2          |
| November | Nil        | 0          |
| December | Nil        | 0          |

Table 14. Rains amount (mm) with Rainy days in El Obeid City during the year 2015.

| Month | Date | Amount mm | Total | Rainy days |
|-------|------|-----------|-------|------------|
| May   | 26/5 | 4.0       |       |            |
|       | 30/5 | 2.3       | 6.3   | 2          |
|       | 6/6  | 9.4       |       |            |
|       | 7/6  | 4.7       |       |            |
| June  | 13/6 | 5.1       |       |            |
|       | 24/6 | 1.7       |       |            |
|       | 23/6 | 6.7       | 27.6  | 5          |
|       | 5/7  | 1.7       |       |            |
|       | 12/7 | 1.7       |       |            |
|       | 14/7 | 14.3      |       |            |
|       | 15/7 | 21.0      |       |            |
| July  | 16/7 | 7.6       |       |            |
|       | 17/7 | 6.2       |       |            |
|       | 24/7 | 6.8       |       |            |
|       | 28/7 | 7.3       |       |            |
|       | 29/7 | 0.8       |       |            |
|       | 31/7 | 20.8      | 88.2  | 10         |
|       | 5/8  | 0.7       |       |            |
|       | 6/8  | 43.0      |       |            |
|       | 7/8  | 47.0      |       |            |
|       | 8/8  | 3.0       |       |            |
|       | 10/8 | 24.5      |       |            |
|       | 11/8 | 9.7       |       |            |
|       | 15/8 | 1.0       |       |            |
|       | 16/8 | 7.6       |       |            |
|       | 23/8 | 0.3       |       |            |
|       | 25/8 | 43.7      |       |            |
|       | 29/8 | 49.7      |       |            |
|       | 1/9  | 3.0       |       |            |
| August| 5/9  | 2.0       |       |            |
|       | 10/9 | Trace     |       |            |
|       | 17/9 | 11.4      |       |            |
Table 15. Temperature (°C) of indoor and outdoor resting habitat in El Obeid City during 2015.

| Area            | Indoor | Outdoor |
|-----------------|--------|---------|
| Alrahma west    | 24.5   | 37.0    |
| Almodereua      | 23.5   | 36.0    |
| Almatar         | 28.0   | 39.0    |
| Alseka Hadied   | 27.0   | 38.0    |
| Arafat          | 26.5   | 37.5    |

Table 16. Relative Humidity (%) of indoor and outdoor resting habitat in El Obeid City during 2015.

| Area            | Indoor | Outdoor |
|-----------------|--------|---------|
| Alrahma west    | 72     | 31      |
| Almodereua      | 82     | 40      |
| Almatar         | 37     | 16      |
| Alseka Hadied   | 56     | 18      |
| Arafat          | 67     | 21      |

Table 17. Temperature (°C) in positive larval breeding site waters and in normal waters in El Obeid City during 2015.

| Area            | Temperature in positive larval breeding site waters | Temperature in normal waters |
|-----------------|----------------------------------------------------|-------------------------------|
| Alrahma west    | 29.0                                               | 24.5                          |
| Almodereua      | 30.0                                               | 24.0                          |
| Almatar         | 27.0                                               | 28.0                          |
| Alseka Hadied   | 28.3                                               | 27.0                          |
| Arafat          | 28.5                                               | 26.5                          |

Table 18. pH in positive larval breeding site waters and in normal waters in El Obeid City during 2015.

| Area            | pH in positive larval breeding site waters | pH in normal waters |
|-----------------|------------------------------------------|---------------------|
| Alrahma west    | 6.50                                     | 6.86                |
| Almodereua      | 6.77                                     | 7.29                |
| Almatar         | 6.61                                     | 6.55                |
| Alseka Hadied   | 6.89                                     | 6.60                |
| Arafat          | 7.22                                     | 7.10                |

Table 19. Salinity in positive larval breeding site waters and in normal waters in El Obeid City during 2015.

| Area            | Salinity in positive larval breeding site waters | Salinity in normal waters |
|-----------------|-----------------------------------------------|---------------------------|
| Alrahma west    | 1.60                                          | 1.73                      |
| Almodereua      | 1.65                                          | 1.77                      |
| Almatar         | 1.90                                          | 1.95                      |
| Alseka Hadied   | 1.70                                          | 1.99                      |
| Arafat          | 1.80                                          | 1.88                      |

Table 20. Number of Anopheles Spp. larvae and adults which correlated with Malaria cases in El Obeid City during 2015.

| Area            | Number of Collected Larvae | Number of Collected Adults | Malaria cases |
|-----------------|---------------------------|----------------------------|---------------|
| Alrahma west    | 68                        | 74                         | 123           |
| Almodereua      | 122                       | 46                         | 130           |
| Almatar         | 0                         | 45                         | 87            |
| Alseka Hadied   | 60                        | 0                          | 103           |
| Arafat          | 0                         | 0                          | 67            |

4. Discussion

Sudan is one of the developing countries that falls under the risk of diseases transmitted by Anopheles mosquitoes such as Malaria and other diseases [11]. Malaria contributed considerably to maternal mortality which is 509/100,000 in Sudan [12]. It accounted for 37.2% of all maternal deaths in Sudan at hospital level. It was also found to be a cause of 18.1% of low birth weight. Malaria in pregnancy (MIP) is another dilemma, 94% of clinicians interviewed in a situation analysis of MIP stated that it was a problem [13]. Before any control measures, classification of mosquitoes which transmitted malaria disease is crucial. However, no previous study over the past thirty years was conducted to identify Anopheles species in El Obeid City [6]. Anopheles arabiensis is the principal vector in Sudan. It is the most widespread member of the Anopheles gambiae, abundant throughout most of the Afro-Tropical region, extending northwards along the River Nile to ≈ 20 ° N in Sudan [14]. These findings did not agree with the results of this study, which indicated that An. squamosus is entirely distributed in El Obeid City. There is no other Anopheles species recorded in El Obeid City during the period of this study. It seems that Anopheles mosquito breeds in ponds water. Females and males rested indoor, according to the finding of white sheets method of survey conducted during the study. In Sudan, Anopheles symesi was reported also by [15] in Lake Jur in Faras area {South Darfur State}. This finding did not agree with the results obtained in this study.
Anopheles squamosus was recorded also by Musa [2013] in shallow pond in the northern west part of {El Obeid City}. This finding agreed with the results obtained in this study. Anopheles squamosus larval stages were only collected from Alrahma west, Almodereua and Alseka hadied area because this type of habitats may probably be the best and the available tap water for oviposition of the indoor resting An. squamosus. The {1.284} per room and {1.570} per dip mean density of An. squamosus was recorded in all locations {Alrahma west, Almodereua, Almatar and Alseka Hadied and Arafat} The highest density of An. squamosus indicate that this may be probably be agreed with the similar temperature during the study period which was recorded and ranged between, {15.5 - 41.1°C} so as the humidity {°%} of indoor resting habitat in El Obeid City during the study period which was recorded and ranged between {37% - 82%} with a difference of {45%}, while that of outdoor recorded and ranged between {16% - 40%} with a difference of {45%} or so as the rainfall rate which were recorded and ranged between {0 and –229.9 mm} and with rainy days of {24%} or so as the rainfall rate which were recorded and ranged between, {27 - 30°C} with a difference of {3°C}, while that of normal waters ranged between ( 24 - 28°C ) with a difference of {4°C}. so as pH in positive larval breeding site waters ranged between {1.73-1.99}with a difference of {1.6 - 1.9} with a difference of {0.3,} while that of normal waters ranged between {1.65 - 7.29} with a difference of {0.74}, so as the salinity in positive larval breeding site waters ranged between {1.6 - 1.9} with a difference of {0.3,} while that of normal waters ranged between {1.73-1.99} with a difference of {0.26}. The study was conducted during all seasons.

Jar, as a habitats recorded high density of anophele immature instars, this is because most of the people tend to keep water in open jars, therefore amass program of health education messages is urgently needed to raise the awareness of people regarding vector control [4]. These findings did not agree with the results of this study. The present study revealed that is predominant by An. arabiensis followed by An. rufipes and An. pharoensis. These findings [16] These findings did not agree with the results of this study. The highest density of An. squamosus larvae was correlated with Malaria cases which was record {0.92043}.

5. Conclusions

An. squamosus was identified according to MES Key 1960 and confirmed by Medical Entomology Department, the Sudan National laboratory for Public Health. Biodiversity of larvae and adults of An mosquitoes in El Obeid City, during the study period {April, 2014 – April, 2016}, was An. squamosus.

The density of adult stage by using white sheets spray method was 0.916 per room, and this finding indicated that, An. squamosus resting indoors.

The obtained larval adult stage density indicated that, the inhabitants of area were at risk of mosquito-borne diseases including malaria, which is one of the greatest causes of morbidity and mortality in this area.

Acknowledgements

I am extremely grateful to. Grateful thanks are due to: Prof. EL Nour Elamin A/Rahman. and Ustaz. Faisal Altaub Hassan. From Academic Health Sciences in Wad Madane town for their advises, Staff faculty of health and environmental Sciences University of Gezira, the staff of the Blue Nile Institute for Communicable Diseases, the staff of the Prof El Gadall Center for training and Malaria Research, Staff faculty of Public and Environmental Health West Kordofan University for their help and facilities Thanks are also due to, Director of Malaria Control Program

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