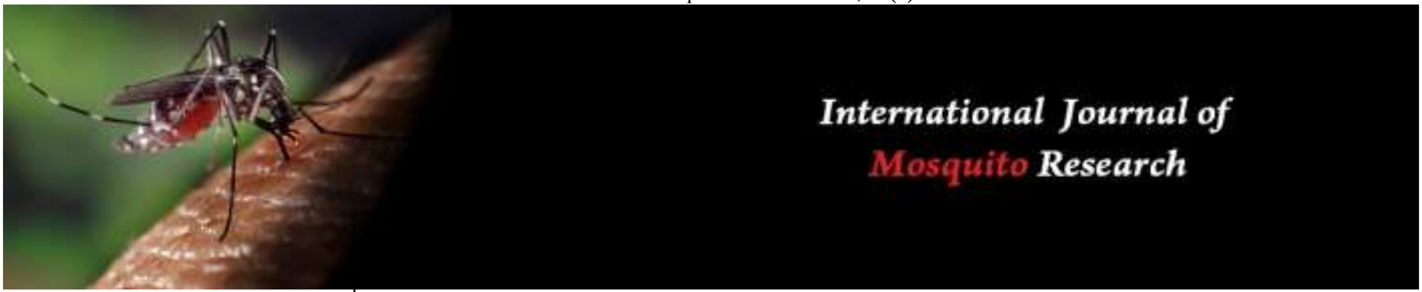

ISSN: **2348-5906**
CODEN: **IJMRK2**
IJMR 2023; 10(5): 14-18

https://www.dipterajournal.com
Received: 20-05-2023
Accepted: 15-06-2023

Venancio A Okic
Department of Medical Entomology, Faculty of Public and Environmental Health - Upper Nile University, Malakal, Republic of South Sudan

Nabil HH Bashir
Department of Medical Entomology and Vector Control, Blue Nile National Institute for Communicable Diseases, University of Gezira, Wad, Sudan

Yousif OH Assad
Department of Pesticides and Toxicology, Faculty of Agricultural Sciences, University of Gezira, Wad Medani, Sudan

Corresponding Author:
Nabil HH Bashir
Department of Medical Entomology and Vector Control, Blue Nile National Institute for Communicable Diseases, University of Gezira, Wad, Sudan

Anopheles species (Diptera: Culicidae) in kodok locality, upper Nile state, Republic of South Sudan

Venancio A Okic, Nabil HH Bashir and Yousif OH Assad

DOI: https://doi.org/10.22271/23487941.2023.v10.i5a.691

Abstract
Anopheline mosquitoes (Diptera: Culicidae) consist of a large number of species. The distribution (Mapping) of malaria vectors is important for strategic planning and control strategies. Malaria risk within communities is usually associated with exposure to its vectors inside households (HHs) and necessarily with the existing larval habitats near the HHs. Studies were conducted from April 2020 to March 2022 in 4 areas/Hai of the Kodok locality (KL), Upper Nile State (UNS), Republic of South Sudan (RSS), to determine the indoor resting density and its seasonal changes, in addition to the number of *Anopheles* and other mosquitoes in a given room by adopting the pyrethrum spray sheet (PSS) collection technique for adults (80 HHs) and the dipping method for larval collection from 8 potential breeding sites. A total of 63,644 adults were collected and identified as follows: *A. gambiae* S.S. (68%), *A. arabiensis* (26%) and other mosquitoes (6%). The density / HH in the rainy- season (April -Oct.) varied between HHs; *viz. A. gambiae s.s.* (52-67) and *A. arabiensis* (20-28). In the dry -season (Nov. to March) the numbers were 2-12 and 0-5, respectively. A total of 7,704 larvae was collected, of which *A. gambiae* formed 88% and the other mosquitoes formed 12%, in the inspected breeding sites (384 visits). It is concluded that several *Anopheles* species are present in KL. *A. gambiae* is the dominant species during both dry -and wet –seasons. Densities were higher in the wet- than the dry-season. This information must be seriously considered during the vector control (VC) programs.

Keywords: Kodok locality, Upper Nile State, South Sudan, *Anopheles gambiae s.s*, *A. arabiensis,* population density, seasonal changes

1. Introduction

Malaria is a major global public health problem and a leading cause of morbidity and mortality in many countries. Most of the people at risk of malaria in Africa live in areas where transmission is relatively intense and continuous [1]. Globally, mortality was estimated as 429, 000 deaths from malaria, most of it (92%) occurred in the African Region [2, 3]. The peak period of transmission is during the rainy –season. *Plasmodium falciparum* is the dominant species of parasite and responsible for >90% of the cases in the RSS [4]. The parasites are transmitted by infected female mosquitoes of the genus *Anophele*s. The species are anthropophilic, bite in the morning or evening; others feed during the day indoors (endophagic) or outdoors (exophagic) [5, 6]. After emerging from pupae, mating takes place around dusk, when the males form swarms to which females are attracted [7]. The flight ranges of *A. gambiae* is within 1-2 km from suitable breeding sites [8]. The population density of this species is expected to vary according to the season in relationship to rainfall. This density in several countries increases quickly with the first rains and the maximum density is reached at the end of the rainy-season [9, 10]. Malaria transmission rates can differ depending on local factors, *e.g*. Rain-fall patterns, the proximity of mosquito breeding sites to people, and *Anopheles* mosquito species resting indoor in the area. Some regions have a fairly constant number of cases round the year. These countries are termed "malaria endemic"; in other areas there are malaria seasons, usually coinciding with the rainy -season [13, 14].

Studies were conducted for 2 year in 4 areas (Hai) of the Kodok locality (KL), Upper Nile State (UNS), Republic of South Sudan (RSS), to study the habitats, breeding sites, the indoor resting density and its seasonal changes, in addition to the number of *Anopheles* and other mosquitoes in a given room.

2. Materials and methods

2.1 Study area

KL (Fig.1) is located in UNS (90 53’ N and 32^0 07’ E), the RSS. The locality lies in western bank of the White Nile River, 306m above sea level; 49% R.H., and a population of *ca*. 7,709 (National Census 2012); the natural vegetation consisted of grasses in swamps, and different tree species, mainly *Acacia spp*.

Fig 1: Upper Nile State map and its Localities including Fashoda (Kodok)

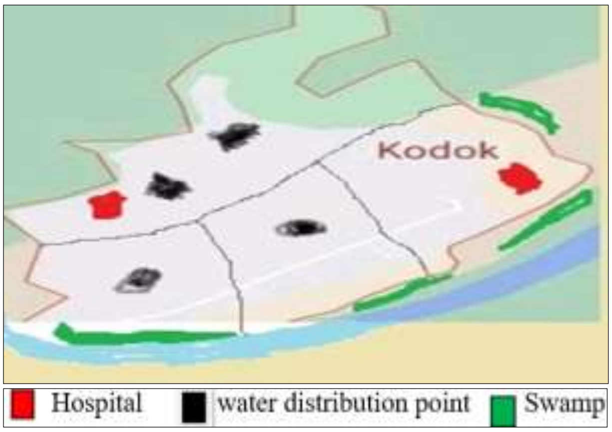

Fig 1: Map of Kodok town and locality.

2.2 Study design

This study was designed as a cross- sectional; counts were conducted bi-weekly using pyrethrum spray sheet (PSS) for mosquito collection from 80 households (HHs) in the 4 study areas (locally called Hai). These areas (Hai, sections) of Kodok were Bilpam, Ochugi, Nevasha and Salam. The study was conducted over 24 month period (April 2020 to March 2022) in the 4 areas, during the 1st and 3rd wk of each month in both the HHs and the breeding sites.

2.3. HHs

HHs [20] were examined /day/hai; overall 80 HHs/KL. These HHs were randomly selected from a random list, choosing a 1st block, and within the block 3 HHs were included.

2.4 Breeding habitats

Larval survey was carried out in 8 breeding habitats/sites in the abovementioned 4 areas. Larvae were collected from 2 breeding sites / area; 10 dips were taken / breeding site. Two visits / month were achieved in both the permanent and the seasonal habitats (swamps, S; and wastewater; WW); overall 348 visits.

2.5 Entomological sampling method

Pyrethrum spray sheet (PSS) collection method was adopted for indoor resting adults. Collection was from 7:30 to 11:00 am after consent of HH occupant. All mosquitoes collected were preserved in labelled paper cups covered with mesh netting for each HH and each room. The larval survey was achieved using the dipping technique in the swamps (S) and man-made ponds/habitats (WW); specimens were sorted out (by species and area) and subjected to identification adopting the taxonomic keys [5, 6], using light microscope and lens.

2.6 Data analysis

The data were analyzed using SPSS software Program, ANOVA (Ver.16). The descriptive analysis was used. The data was analyzed step by step, and tabulated to explain the possible relationships between the variables (seasonality, dry- *vs.* rainy- season, HH density, and spatial distribution).

3.1 Results

3.1 Adult collection

3.1.1 *Anopheles* spp. adults collected indoors

The total number of mosquitoes collected during the surveys were 63,644. *A. gambiae S.S.* formed 68%, *A. arabiensis* 26% and the other mosquitoes 6% (Table 1; Fig.2).

Table 1: Number of adults collected resting indoors over 24 months in 4 study areas of KL

Area /Hai	*A. gambiae S.S.*		*A. arabiensis*		*Others		Total
Bilpam	12,405	65%	5,324	28%	1,324	7%	19,053
Nevasha	10,679	68%	4,058	26%	896	6%	15,633
Ochugi	9,165	71%	3,175	24%	624	5%	12,964
Salam	10,835	68%	4,283	27%	876	5%	15,994
Total	43,084	68%	16,840	26%	3,720	6%	63,644

*Others: *A. funestus*, *Culex* and *Aedes*.

3.1.2 The species distribution

The collected mosquitoes were identified as follows /area: Bilpam (*A. gambiae* 65%, *A. arabiensis* 28%); Ochugi (*A. gambiae* 71%, *A. arabiensis* 24%), Salam (*A. gambiae* s.s 68%, *A. arabiensis* 27%), and Nevasha (*A. gambiae* 68%, *A. arabiensis* 26%) (Table 1). Regarding the other mosquitoes species, following the same order of areas, was as follows: 7%, 5%, 5%, and 6%. The highest number of *A. gambiae* was collected from Ochugi (71%), whereas the lowest was that of Bilpam (65%). For *A. arabiensis*, the highest was that of Bilpam (28%), whereas the lowest in Ochugi (24%).

3.1.3 Densities
3.1.3.1 Resting indoors
The density of *A. gambiae* complex /HH (rooms) varied between different HHs according to the season. During the rainy-season, *A. gambiae* ranged between 52- 67 and *A. arabiensis* 20-28. However, in the dry -season, *A. gambiae s.s.* registered only 2- 12 and *A. arabiensis* was 0-5 (Table 2). *A. gambiae* predominated, compared to the total indoor resting mosquito species (Table 1). These species were most abundant in the rainy- season (Figs. 2 and 3). Overall, indoor resting densities of vector species increased from April to September.

Table 2: Densities of resting indoor /HH/day.

Species	No. of rooms	Mean Density/ room	Total
A. gambiae s.s.	35	4.8	169
A. arabiensis	35	1.8	66
Others	35	0.3	11

Table 3: Dry- and rainy- seasons densities of indoor resting mosquito species adults over 24 months.

Species	Season	Mean	SE (±)	C.V.%
A. gambiae s.s.	Dry	4.11	0.21	5.1
	Rainy	20.23	0.55	2.7
A. arabiensis	Dry	2.11	0.12	5.6
	Rainy	8.73	0.22	2.5
Others	Dry	0.4	0.05	12.5
	Rainy	24.8	0.10	0.4

3.1.3.2 Larval density
The density of *A. gambiae* in swamps (S) proved to be higher than the WW (Table 4) and varied significantly between the sampling habitats. The highest number was found in Ochugi (71%), whereas the lowest in Bilpam (65%). However, for *A. arabiensis*, the highest number was reported in Bilpam (28%), while the lowest in Ochugi (24%). The high percentage of *A. gambiae* was found in S + WW (Salam).

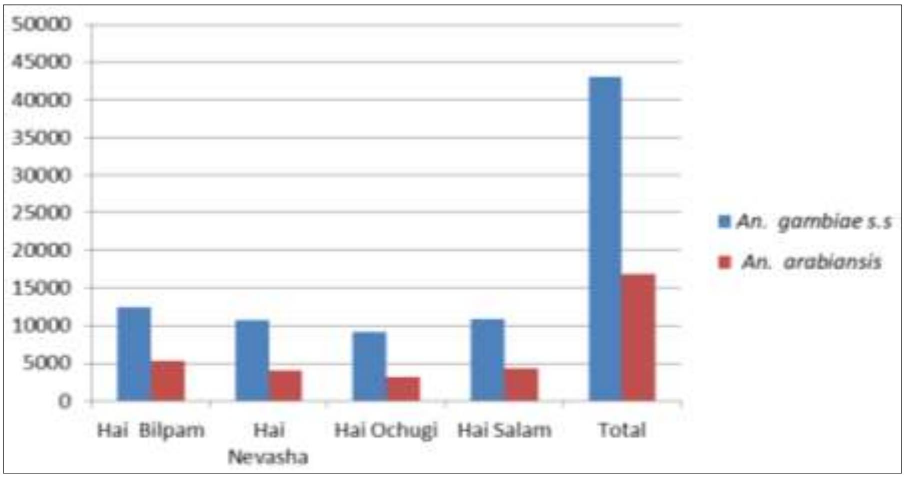

Fig 2: Distribution of *A. gambiae* S.S. and *A. arabiensis* adults at sites.

3.4 Larval surveys
3.4.1 Number collected in different areas/Hai
Total number of mosquitoes larvae collected were 7,704. Of these *A. gambiae* formed *88*% and other mosquitoes 12%, following 384 visits to the sites. The number of *A. gambiae* was recorded in the different areas of the study indicated the following: Ochugi, Bilpam, and Nevasha registered 87% of the collected population of the area, whereas Salam registered 89%**.** Accordingly, KL is highly infested with *A. gambiae* 87.59%; the other mosquito species formed only 12.4% (Table 4; Figs. 3 and 4). The high number of *A. gambiae and A. arabiensis* are found indoor in Bilpam.

Table 4: Spatial composition of *An. gambiae* larvae and other mosquitoes over 24 months

Location	Breeding site	*A. gambiae*		*Others		Total
Ochugi	S	1,778	87%	272	13%	2,050
Bilpam	S + WW	1,581	87%	229	13%	1,810
Salam	S + WW	1,942	89%	245	11%	2,187
Nevasha	WW	1,447	87%	210	13%	1,657
Total		6,748	87.59	956	12.4	7,704

*Others: *A. funestus*, *Culex* and *Aedes*.
No. of dips = 96/ area S = swamp
WW = wastewater

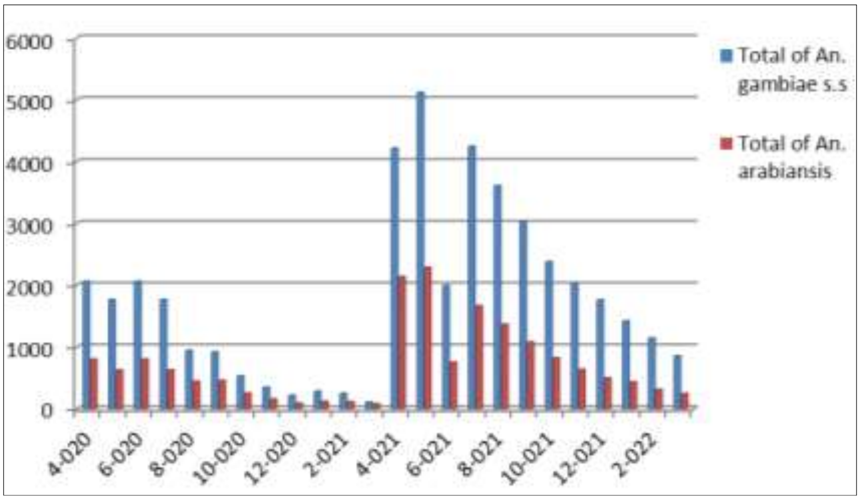

Fig 3: Average of monthly distribution of *A. gambiae s.s.* and *A. arabiensis* adults (April 2020 to Feb. 2022) in the locality.

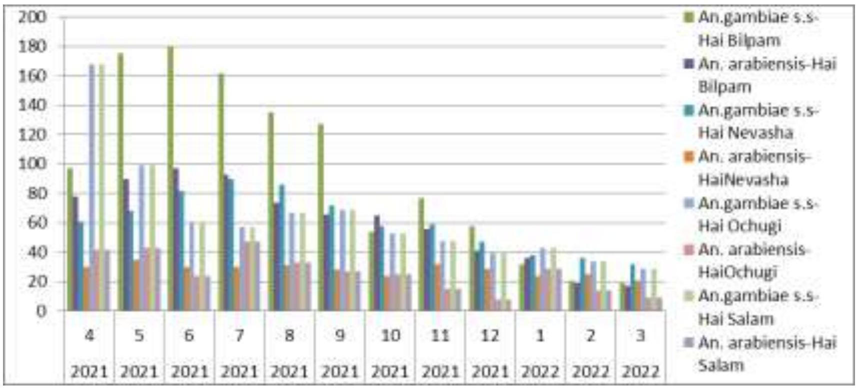

Fig 4: Distribution of *A. gambiae s.s.* and *A. arabiensis* adults resting indoor at different areas/Hai over 12 months.

4. Discussion

The abundance of *Anopheles* species in KL could be attributed to a number of factors, one of which is that some communities are located along the banks of the White Nile River. This river experiences seasonal flooding that provides favorable temporary and permanent breeding sites for this vector [11] like swamps and mane-made WW. The mean over 24 months, in the different 4 studied areas, revealed that *A. gambiae* proved to be the main existing species, with very high numbers and density resting indoors.

Hai Ochugi (swamps only) took the lead in numbers and density, followed by Salam (S + WW) and Nevasha (WW only); the lowest numbers and density was registered in Hai Bilpam (S+WW). *A. arabiensis* ranked 2nd.. Surprisingly, the highest numbers for the latter species collected resting indoors was registered in Bilpam, followed by Salam, and Nevasha; the lowest was that of Ochugi. This situation requires more research to investigate the reasons behind such phenomenon, where *A. gambiae* exists in higher numbers or density in some habitats or HHs, in the contrary, the other species exists in lower numbers and density. The counts of the other mosquito species in the 4 areas were by far lower than the main 2 species. However, one cannot ignore their role as vectors for this disease. This also requires more intensive incrimination studies for the other existing species.

The seasonal abundance and density of *A. gambiae* species results in KL covered some information gaps and is expected to improve the understanding on this vector and its role in malaria transmission in KL. The results confirmed that density of *A. gambiae* was influenced by rains and increased at the beginning of the rainy- season between April and September, similar to what is happening in the neighboring Sudanese states (*viz*. White Nile, Blue Nile, South Kordofan and South Darfur), other RSS states and the Ethiopian Zoogeographical Region [17, 18]. The density peaked towards the end of the rainy-season. The highest % *A. gambiae* was found in Ochugi 71% and *A. arabiensis* 28%, that is, the density of *A. gambiae* was rising during the 2 climatic seasons (dry- and rainy) in the 4 sites, followed by *A. arabiensis* as reported in 2017 [19].

The spatial distribution of *A. gambiae* was aggregated throughout the year, but the degree of aggregation increased during the rainy- season as reported by some African authors [20]. The high-density HHs persisted throughout the season, however, the high-density HHs values during the dry –season proved to be significantly different from those of the rainy-season. The locations of the hot-spots within each period (rainy- and dry -season) were the same between the 1st and 2nd yr. similar results were obtained in Nigeria [22].

ANOVA results revealed significant differences in counts of *A. gambiae* in KL where it showed that the populations are significantly higher in HHs, especially in areas near to swamps, followed by *A. arabiensis*. The availability of permanent water sources complements vector survival by ensuring species that are best adapted to these kinds of habitats, *e.g*. *A. gambiae*, which is able to breed and sustain malaria transmission [23].

Large permanent habitats with emergent aquatic vegetation are known to favor proliferation of *Anopheles*. The results of the present work showed that KL is heavily infested with *A.*

gambiae larvae (88%), whereas the other mosquito species formed as little as 12% of the collected larval population. *A. gambiae* larvae peaked in April and the lowest densities occurred in October to March. Hai Salam registered the highest percentage, followed by Nevasha, Ochugi and Bilpam. The lowest % of other species (*A. funestus*, *Culex* and *Aedes*) was found in Salam in all breeding sites. The density of *A. gambiae* in the swamps in the 3 areas were higher than WW during the study period, especially in Salam, and *A. gambiae* population density / swamp was higher for all the sites. This point requires more studies about the habitat physical and chemical properties. The larval density in the swamps for sure contributes strongly to the adult population density [24].The present study showed that mosquito larvae were abundant after the rainy- seasons, due to the formation and availability of larval habitats, particularly at the edges of White Nile River, which served to sustain *Anopheles* populations during the dry -seasons and it was observed that habitats that contained growing grass and other vegetation registered more *Anopheles* larvae than habitats without vegetation. This is in line with several authors [25, 26].The late instars were significantly higher in swamps, when compared to WW throughout the year, *i.e*. Swamps long-term contribution to larval productivity seems to be more important, since their water is available for longer period than the other habitats, which are liable to dryness. Other permanent water sources are located outside the study areas, e.g. River Nile located <1 km away. That is well within the flight range [27].

5. References

1. WHO. World malaria report day, Geneva, WHO; c2016.
2. WHO. Eliminating malaria,ttp://www.who.int/malaria/publications/atoz/eliminating-malaria/en, accessed; c2016.
3. Republic of South Sudan. Malaria Indicator Survey (MIS) 2017.In: World Health Organization. World Malaria Report; c2020. https://malariasurveys.org/documents]-
4. Ministry of Health, Government of Southern Sudan. Prevention and treatment guidelines for primary health centers and hospitals; c2006. p.89-90.
5. Gilles MT, Coetzee MA. Supplement to Anophelinae of Africa south of the Sahara. South Africa Inst. Med. Res, Johannesburg; c1987. p.55.
6. Gilles HM, Warrell DA. Bruce-Chwatt's Essential Malariology, Edward Arnold, London; c1996.
7. Takken W, Knols BG. Odor-mediated behavior of Afro tropical malaria mosquitoes. Annual Review of Entomology. 1999;44:131-57.
8. Thomson MC, Connor SJ, Quinones ML. Movement of *Anopheles gambiae* s.l. malaria vectors between villages in The Gambia. Medical and Veterinary Entomology. 1995;9(4):413-419.
9. Precious A, Dzorgbe M, Godfred F, Leonard KA, Memuna MM, Dziedzorm KDS, *et al*. Diversity in breeding sites and distribution of *Anopheles mosquitoes* in selected urban areas of southern Ghana. Parasit Vectors. 2017;10(1):25. doi: 10.1186/s13071-016-1941-3.
10. Gilles HM, Warrell DA. Bruce-Chwatt's Essential Malariology. Edward Arnold, London; c1993.
11. Service M. Medical Entomology for Students, 3rd ed, Cambridge University Press; c2004.
12. Kaufmann C, Briegel H. Flight performance of the malaria vectors *Anopheles gambiae* and *Anopheles atroparvus*. J Vector Ecol. 2004;29(1):140-153.
13. Gilles HM. Epidemiology of malaria In: Bruce Chwatt's Essential Malariology, 3rd edn; c1993.
14. Martens, P. *et al*. Climate change and future populations at risk of malaria. Global Environmental Change. 1999;9:S89–S107.
15. White GB. *Anopheles gambiae* complex and disease transmission in Africa. Transactions of the Royal Society of Tropical Medicine and Hygiene. 1974;68:278-298.
16. White GB, Magayuka SA, Boreham PFL: Comparative studies on sibling species of the *Anopheles gambiae* Giles complex (Dipt. Culicidae): Bionomics and vectorial activity of species A and B at Segera, Tanzania. Bull Entomol Res. 1972;62(2):295-317.
17. Nwabor OF, Nnamonu EI, Martins PE, Odiachi O. Anopheline mosquitoes and the malaria scourge. International Journal of Mosquito Research. 2015;2(3):200-207.
18. Gilles MT, De Meillon, B. The Anophelinae of Africa South of the Sahara (Ethiopian Zoogeographical Region). South African Inst. Med. Res., Johannesburg; c1968. p.54.
19. WHO. World Malaria Report. Republic of South Sudan. Malaria Indicator Survey (MIS); c2017. https://malariasurveys.org/documents]-
20. Gilles MT, Coetzee MA. Supplement to Anophelinae of Africa south of the Sahara. South Africa Inst. Med. Res, Johannesburg; c1987. p.55.
21. Oyerinde OO, Ibidapo AC, Oduola OA, Obansa BJ, Awolola ST. Anthropophilic mosquitoes and malaria transmission in a tropical rainforest area of Nigeria. Journal of Life Physical Sciences. 2005;2(1):6-10.
22. WHO. Malaria Entomology and vector control. Guide for Participants. WHO, Geneva, Switzerland; c2013.
23. Rozendaal JA. Vector Control Method for used by individuals and communities, WHO; c1997. p.398:20.
24. Thomson MC, Connor SJ, Quinones ML *et al*. Movement of *Anopheles gambiae* s.l. malaria vectors between villages in The Gambia. Medical and Veterinary Entomology. 1995;9:413-419.
25. White GB. *Anopheles gambiae* complex and disease transmission in Africa. Transactions of the Royal Society of Tropical Medicine and Hygiene. 1974;68:278-298.
26. Mattah PAD, Futagbi G, Amekudzi LK *et al*. Diversity in breeding sites and distribution of *Anopheles mosquitoes* in selected urban areas of southern Ghana. Parasites Vectors. 2017;10:25. https://doi.org/10.1186/s13071-016-1941-3.
27. Kaufmann C, Briegel H. Flight performance of the malaria vectors *Anopheles gambiae* and *Anopheles atroparvus*. J Vector Ecol. 2004;29:140-153.