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Research Article

Study of Diversity and Abundance of Anopheline Mosquitoes in Meghalaya, India

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Abstract

Malaria is anopheline vector-borne disease of serious worry in Southeast Asia. Understanding the spatial distribution of mosquitoes should contribute to the design of malaria control. The diversity, distribution and relative abundance of Anophelines were surveyed using a sampling method for a period of four years from April 2008 to March 2012 in several biotopes of the varying climatic region of Meghalaya. Meghalaya State is situated in highly malaria endemic North-eastern region of India. The biodiversity of Anopheline was examined and divided into alpha and beta components with the aim of comparing its distribution and abundance in all the seven districts of Meghalaya. A total of 37,026 Anopheline mosquitoes belonging to 33 species were collected. During pre-monsoon, monsoon, and post monsoon 9,345 (25.2%), 23,507 (63.5%) and 4,174 (11.3%) mosquitoes respectively were recorded. The most common species were *An. maculatus (21.7%), An. vagus (15.2%), An. annularis (12.91%), An. philippinensis (9.9%), An. nigerimus (9.81%) and An. minimus (8.7%).* The result of the study shows significant differences in species richness between districts. Biodiversity indices indicate that species diversity was highest in West Garo Hills and lowest in West Khasi Hills districts. It is suggested that greater variation in the species composition could be due to temperature differences among the different districts of Meghalaya.

Keywords: Anopheles; Abundance biodiversity; Meghalaya; Principle component analysis; Survey

Abbreviations

WHO: World Health Organization; NVBDCP: National Vector Borne Disease Control Programme

Introduction

Malaria, a major human health threat, occurs globally in tropical and subtropical regions. It is a worrying disease of Africa, South-east Asia and South America. World Health Organization (WHO) has estimated that there are 106 countries in the world where malaria is endemic and India is one of them. About 36% of the world population (i.e., 2020 million) living in these countries are at risk, with fatal rates being extremely high among young children below 5 year of age [1]. As per WHO report concerning South-east Asian region, out of 1.4 billion people living in 11 countries of South-east Asia, 1.2 billion (about 87%) are exposed to the risk of malaria and most of them live in India [2-4]. The disease primarily affects poor population in tropical and subtropical areas, where the temperature and rainfall are suitable for the development of vectors and parasites [5,6].

Meghalaya (in Sanskrit, Megh = clouds, Alaya = house)" is an important North-eastern State of India famous for the place of highest rainfall in the world. Its geographical territory lies between latitude 25°09'30" N to 26°01'42" N and longitudes 89°51'25 E to 92°50'37 E. The physical features and particular tribal dominance have divided Meghalaya into three zones i.e. Khasi Hills, Jaintia Hills and, Garo Hills. The districts in these zones are East Khasi Hills (EKH), Ri-Bhoi (RB), West Khasi Hills (WKH), Jaintia Hills (JH), East Garo Hills (EGH), West Garo Hills (WGH) and South Garo Hills (SGH) (Roy & Tomar, 2001). In Meghalaya, incidence of malaria has been reported to increase significantly from the year 2001 [7]. The annual average prevalence of malaria in India is 106 per 100,000 populations, whereas in Meghalaya it is 920 per 100,000 populations, which is about 8.6 times more than the national average [7]. The prevalence rate of malaria in Meghalaya is highest in the North-eastern States and second in India [7]. Climatic condition of low land areas of the State is warm and humid, but highland areas are cold. The occurrence of malaria has been reported to be prevalent in foothills and valleys of Meghalaya, but now it has been noted to gradually spread in the highland areas also.

Malaria is a vector-borne disease, which is transmitted by female anopheline mosquitoes. Understanding the spatial distribution of mosquitoes should significantly contribute to the design of malaria control strategies. Earlier studies carried out in Meghalaya started from Shortt (1934) [8] to Prakash *et al.*, (1998) [9] revealed that number of Anopheline species ranged from 8 to 34. These studies were based on the survey mainly done from Ri-Bhoi (RB), East Khasi Hills (EKH), West Khasi Hills (WKH) and Jaintia Hills (JH) districts. However, there are no reports on the survey and distribution of Anopheline mosquitoes in highly malaria-affected areas of East Garo Hills (EGH), West Garo Hills (WGH), and South Garo Hills (SGH) districts of Meghalaya. Further, earlier reports on Anopheline mosquitoes records show the rich mosquito diversity in EKH, WKH, JH and RB, but there is no record about species richness and its composition in these regions.

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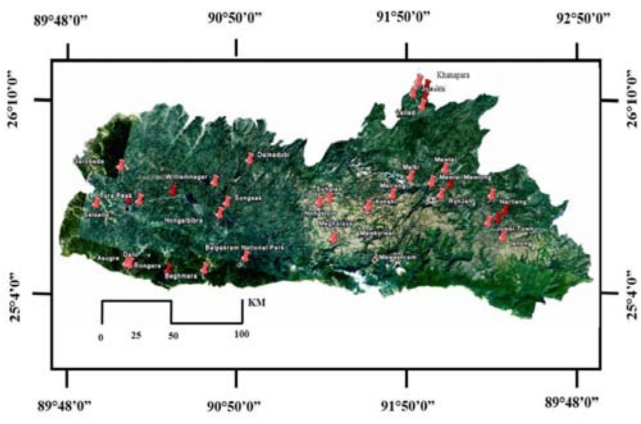

Figure 1: A schematic map of Meghalaya showing the sampling sites (board pins) for Anopheline mosquito in different districts. The mosquito samples were obtained from 28 selected sampling sites. Source: Google Earth. http://www.google.com/earth/download/ge/agree.html

Therefore, the present study was undertaken to update the status of Anopheline mosquito's species in all the above-mentioned seven districts of Meghalaya and to determine the species richness, composition, and abundance in view of specific environmental conditions. This is perhaps the first study of its kind in Meghalaya that documents the information on the Anopheline species distribution.

Materials and Methods

Seasonal features of Meghalaya

The Meghalaya plateau lies in the monsoonic region and is directly influenced by the southwest monsoon and the northeastern winter winds. It has four well-defined seasons: spring (March-April), rainy-summer (May - September), autumn (October - November) and winter (December - February). The spring season (March - April) is characterized by moderate temperature, occasional thunderstorms, and high velocity wind. Rainy-summer season is the wettest period of the year and about three-fourth of the annual rainfall is received during this period. The winter season is the coldest period of the year [10].

For the purpose of survey in present study, the year was divided into three phases, i.e. pre-monsoon (February - May), monsoon (June - October) and post-monsoon (November - January). In each village, one thermometer and relative humidity data loggers (Onset Computer Corporation, Bourne, MA, USA) were placed and one person was appointed to record temperature and humidity daily. Rainfall data were collected from the Meteorological department of India, Shillong.

Mosquitoes collection and identification

The mosquitoes were collected from the 35 sampling sites comprising five sites per district in Meghalaya from April 2008 to March 2012. These sampling sites are as shown in (Figure 1).

All catches were conducted by standard techniques of WHO [11]. All possible habitats of mosquitoes situated within 5 Km radius were searched, for obtaining the maximum number of specimen from every district. Sampling of mosquitoes in each selected area was done at least once approximately at the mid of a phase during a year. In each village, 10 houses were normally examined and the worst ventilated room was selected for sampling as these surroundings usually contain a large number of breeding grounds for mosquitoes. Special attention was paid to the sleeping areas and bathrooms. The collection was done during early morning at about 6 to 8 am. Immature forms of mosquitoes were collected by standard dipping technique as described by Reuben [12] and Service [13]. All collected larvae and pupae were kept in a rearing tray for the emergence of adults. The emerged and collected adults were preserved in glass and plastic vials. Adult and larval forms of mosquitoes were morphologically identified using catalogues of Christopher's [14] Gillie's and Coetzee [15], Das *et al.*, [16] and Nag pal & Sharma [17].

Data analysis

Mosquito community structure was analyzed using following ecological parameters like population abundance, species richness, species evenness (Pileou's index), diversity of species (Simpson index, Shannon - Wiener index), wealth of mosquito (Margalef index) and species dominance (Barger - Parker index) in each district. Population abundance at each district was defined as the sum of individuals of a particular species, counted at each site during study [18]. The number of species found at each study site during the study period expresses species richness. The indices of diversity were calculated using the software PAST 2.16 [19]. The similarity between habitats based on number of species was estimated by Jaccard and Whittaker index. Cluster analysis and Principal Component Analysis (PCA) were used to group the sampling sites by similarity of Anopheline abundance. The Jolliffe cut-off var-covar was also calculated for PCA to know the degree of reliability of the classification system used. Nonparametric abundance estimates were used to verify sampling sufficiency to assess the richness directly related to the number of rare species in the samples. All results are presented as mean ± SE.

Results

Environmental setting of study sites

The average season wise data on different environmental parameters such as rainfall, humidity and temperature, water temperature and pH are shown in Table 1. Analysis of data revealed the presence of higher temperature in East, West and South Garo hills during the month of May and June and maximum rainfall occurred in EKH while minimum rainfall was recorded in Ri-Bhoi district (Table 1). It was noted that during July and August there was more rain fall in study areas. It was also noted that the water pH in the sampling sites was acidic (Table 1).

Sample based rarefaction curve

Rarefaction curve provides measures of species diversity involving the expected number of sampling against the number of individuals (Figure 2). The rise in the curve denotes quick increase in the number of species during sampling, while the flattening of the graph at the later stages may denote the repetition of similar species. This may infer that a reasonable number of individual samples have been taken. More intensive sampling may yield only few additional species.

Species composition and relative abundance

A total of 37,026 Anopheline mosquitoes (larvae: 23,614 and adults: 13,412) were collected during the survey from 35 sampling sites of all seven districts of Meghalaya. Analysis of data revealed the presence of 33 species of two subgenera as: *Anopheles (An.*

Table 1: Environmental analysis of sampling in different districts of Meghalaya. The results are shown as Mean±SD. Sampling districts were surveyed 3 times.

Districts	Parameters	2010-11			2011-12		
		Pre-Monsoon	Monsoon	Post-Monsoon	Pre-Monsoon	Monsoon	Post-Monsoon
East Khasi Hills	Temperature	11.5±0.6	22.5±2.8	13.5±2.1	19.2±2.8	21.2 ±0.9	9.6±2.5
	Rain fall	33.7±35.7	265.4 ±116.4	7.9 ±0.8	20.0 ±18.3	230.8 ±137.2	32.7±19.4
	pH	6.3±0.2	6.6 ±0.2	6.5±0.2	6.5 ±0.4	6.6 ±0.2	6.6±0.3
	Humidity	50 ±3.4	90±4.3	70±5.3	57±5.5	89±6.7	60±.5
West Khasi Hills	Temperature	12.7±1.6	20.5 ±1.8	30.5±0.1	19.2 ±2.8	21.2±0.9	4.6 ±0.5
	Rain fall	133.7±35.7	274.5 ±66.4	12.5 ±0.8	50.0 ±18.3	254.8 ±137.2	32.7±11.2
	pH	6.4±0.2	6.3 ±0.2	6.6±0.2	6.3±0.4	6.7±0.7	6.6±0.5
	Humidity	66±3.4	90±4.3	60-70±5.3	50-70±5.5	70-90±6.7	60-70±.5
Jaintia Hills	Water Temp.	20.7±1.65	29.3±4.9	16.2 ±1.83	26.67 ±2.48	29.4±0.93	15.57±1.42
	Rain fall	44.7±27.1	291.7 ±138.2	9.47 ±0.65	45.0 ±9.82	324.4±62.9	13.47±2.17
	pH	7.7±.25	6.92 ±1.3	6.9 ±0.51	6.8 ±0.70	6.7±0.39	6.2±.21
	Humidity	50 ±2.6	80±3.1	60±3.3	55±3.6	82±3.1	64±2.4
Ri-Bhoi	Temperature	23.27 ±1.65	32.3 ±4.9	19.2 ±1.83	26.67±2.48	31.4±0.93	17.5±1.42
	Rain fall	49.27±37.1	251.5 ±138.2	11.47±0.78	25.0 ±10.86	244.4 ±67.9	9.47 ±2.97
	pH	7.67± .25	6.97±0.3	7.2 ±0.21	7.8 ±.70	7.3 ±0.40	7.2 ±.26
	Humidity	64±3.2	88±4.5	68±3.1	60±2.8	85±5.4	66±3.9
East Garo Hills	Temperature	27.3±5.7	30.8±1.9	19.1±26.6	26.7±4.32	33.8±3.1	16.0±3.7
	Rain fall	170.0±17.51	209.5±33.6	21.9±3.2	173.0 ±17.5	6.6±0.57	22.0±2.9
	pH	72.0±5.8	73.3±4.1	65,3±12.2	70±5.8	76.0±4.1	66.0±12.2
	Humidity	50±4.6	80±5.1	62±2.8	68±2.6	80±3.7	66±3.7
West Garo Hills	Temperature	26.8±5.14	32.8±3.5	19.4±6.2	27.3±5.7	33.8±3.0	20.1±5.6
	Rain fall	31.0 ±22.7	356.1±122.8	73.4±82.7	170.0 ±170.51	315.4±99.9	15.1±24.3
	pH	7.1 ±0.15	6.7±0.2	6.9 ±0.17	6.9 ±0.17	6.8 ±0.05	6.9 ±0.2
	Humidity	62±2.8	88±2.6	70±3.7	64±3.3	84±4.4	70±4.1
South Garo Hills	Temperature	29.3 ±1.8	30.1 ±2.4	17.9 ±3.5	26.7±2.0	32.7±2.9	18.7 ±2.1
	Rain fall	92.0 ±143.2	278.0 ±129.1	12.4 ±3.5	96.1±108.1	301.4 ±139.6	1.3 ±1.1
	pH	7.6 ±0.3	7.2 ±0.2	7.3 ±0.1	7.0 ±0.1	7.1 ±0.3	7.3 ±0.20
	Humidity	50±2.9	84±5.4	67±4.8	55±3.4	82±5.4	67±3.4

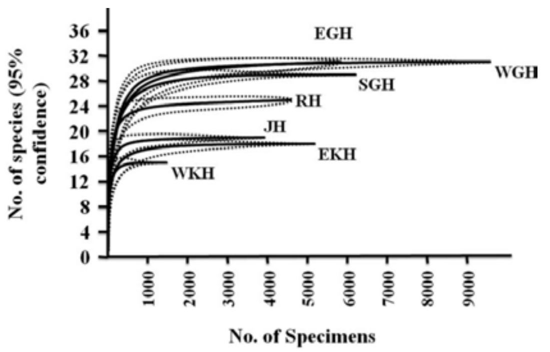

Figure 2: Sample based individual rare fraction curve showing number of individuals against number of species recorded in different districts. Solid lines show rare fraction curves of Anopheline mosquito communities and dotted lines show 95% confidence limits of related solid lines. EGH and WGH rare fraction curve are overlaps. The curve steepness is a function of the community taxon evenness, while its height indicates the taxon richness. EKH: East Khasi Hills district; WKH: West Khasi Hills district; RB: Ri-Bhoi district; JH: Jaintia Hills district; EGH: East Garo Hills districts; WGH: West Garo Hills District and SGH: South Garo Hills district

aitkenii, An. ahomi, An. barbirostris, An. gigas, An. lindesayiand An. maculates) and Cellia (An. aconitus, An. annularis, An. balabacensis, An. culicifacies, An. crawfordi, An. fluviatilis An. dirus, An. jamesii, An. jeyporiensis, An. karwari, An. kochi, An. maculates, An. minimus, , An. maculates, An. majidi, An. nivipes, An. philippinensis, An. peditaeniatus, An. pallidus, An. pseudojamesi, An. splendidus, An. subpictus, An. stephensi, An. tessellates, An. vagus An. varuna and An. willmorei,) (Table 2).

The average density of Anopheline was about 5289 ± 931.15 per district. Out of the total 37,026 mosquitoes, the most common species were *An. maculatus* (21.7%), An. vagus (15.2%), *An. annularis* (12.91%), *An. philippinensis* (9.9%), *An. nigerimus* (9.81%) and *An. minimus* (8.7%). These form nearly 78.2% ($p \leq 0.01$) of the total mosquitoes caught. The relative abundance of subgenera *Cellia* (86.4%) was higher than subgenera *Anopheles* (13.6%). The number of specimens of potential malaria vector species, i.e. *An. minimus* Theobald, 1901, *An. philippinensis* Ludlow, 1902, *An. annularis* Vander wulp, 1884 and *An. culicifacies* Giles, 1901 was 33.4% of total individual caught. A total 71.4% ($p \leq 0.01$) of *Anopheles* were

Table 2: Anopheline species collected from different districts of Meghalaya. Species are listed in descending order of the total number of each species collected.

Subgenera	Species	Adult	Larvae	Total	RA (%)
Anopheles	*An. nigerimus* Giles, 1900	1269	2357	3626	9.79
	An. lindesayi Giles, 1900	304	573	877	2.37
	An. ahomi Chowdhury, 1929	53	95	148	0.40
	An. gigas Giles, 1901	33	85	118	0.32
	An. barbirostris Van der Wulp, 1884	32	78	110	0.30
	An. gigas var baileyiedwards 1929	26	81	107	0.29
	An. aitkenii James 1903	21	51	72	0.19
Total (*Anopheles*)		1738	3320	5058	13.67
Cellia	*An. maculatus* Theobald, 1901	2712	5316	8028	21.68
	An. vagus Donitz, 1902	2089	3535	5624	15.19
	An. annularis Vander wulp, 1884	1688	3087	4775	12.90
	An. philippinensis Ludlow,1902*	1444	2207	3651	9.86
	An. minimus Theobald, 1901*	1083	2136	3219	8.69
	An. varuna Iyengar, 1924	451	937	1388	3.75
	An. balabacensis Baisas, 1936	381	710	1091	2.95
	An. subpictus Grani,1899*	345	664	1009	2.73
	An. culicifacies Giles, 1901	300	403	703	1.90
	An. aconitus Donitz, 1902	146	261	407	1.10
	An. kochi Doenitz, 1901	186	218	404	1.09
	An. stephensi Liston, 1901*	132	211	343	0.93
	An. crawfordi Reid, 1953	173	98	271	0.73
	An. jamesii Theobald, 1901	119	140	259	0.70
	An. jeyporiensis James, 1902	104	71	175	0.47
	An. pseudojamesii Strickland and Chowdhury, 1927	36	78	114	0.31
	An. tessellatus Theobald, 1901	40	69	109	0.29
	An. karwari James, 1903	74	0	74	0.20
	An. Pallidus	0	74	74	0.20
	An. splendidus Koidzumi, 1902	0	55	55	0.15
	An. fluviatilis James, 1902	16	21	37	0.10
	An. willmorei James, 1903	22	13	35	0.09
	An. majidi Young and Majid, 1928	12	21	33	0.09
	An. dirus Peyton and Harrison, 1979*	0	32	32	0.09
	An. nivipes Theobald, 1903	20	11	31	0.08
	An. peditaeniatus Leicester, 1908	27	0	27	0.07
Total (*Cellia*)		11674	20294	31978	86.36
	Grand Total (*Anopheles* and *Cellia*)	**13338**	**23688**	**37026**	**100**

RA: Relative Abundance
* = potential malaria vector species

collected from lowland areas of Meghalaya, out of which 25.9% ($p \leq 0.01$) were collected from WGH, 19.7% ($p \leq 0.01$) from EGH and 16.8% ($p \leq 0.01$) from SGH districts (Table 3).

The district wise species abundance and species richness relationship shows that species abundance was highest in WGH and lowest in WKH (Figure 3).

Seasonal distribution

During pre-monsoon, monsoon, and post-monsoon 9,345 (25.2%), 23,507 (63.5%) and 4,174 (11.3%) mosquitoes respectively were recorded (Figure 4). It was noted that Anopheline species abundance was associated with temperature and rainfalls because the abundance of Anopheline peaked in month of July to October (20840, 56.2%) and fell progressively from November to January (4,174, 11.3%). Gradual increase in the density of *An. annularis*, *An. philippinensis*, *An. minimus* and *An. subpictusstarted* during pre-monsoon, peaked in post-monsoon and declined during winter. *An. peditaeniatus* and *An. pallidus* was found only in border areas of EGH

Table 3: Number of Anopheles species collected from different districts of Meghalaya from April, 2010 to March, 2012.

Species	Districts						
	EKH	WKH	RB	JH	EGH	WGH	SGH
An. aitkenii	0	9	0	16	11	22	14
An. lindesayi	436	56	195	98	32	41	19
An. gigas Giles	0	31	0	49	16	22	0
An. gigas var	0	18	0	29	19	41	0
An. nigerimus	613	149	548	300	555	915	546
An. ahomi	28	0	55	25	2	33	5
An. barbirostris	0	0	55	0	26	29	0
An. balabacensis	357	104	133	0	41	285	171
An. aconitus	56	0	61	0	60	123	107
An. tessellatus	39	0	25	0	13	28	4
An. fluviatilis	16	20	1	0	0	0	0
An. minimus	263	0	686	420	418	864	568
An. varuna	95	0	146	236	227	383	301
An. culicifacies	0	73	161	153	71	141	104
An. subpictus	86	0	104	200	105	458	56
An. vagus	822	164	498	662	849	1706	923
An. annularis	699	216	396	414	1040	1180	830
An. jamesii	23	6	49	60	41	54	26
An. maculatus	1309	595	850	710	1344	1810	1410
An. philippinensis	352	0	379	476	690	1038	716
An. splendidus	0	0	12	0	0	17	26
An. stephensi	0	45	77	54	43	66	58
An. jeyporiensis	10	6	21	14	32	49	43
An. karwari	0	0	0	0	21	19	34
An. willmorei	3	0	10	0	11	2	9
An. majidi	9	0	0	0	3	5	16
An. nivipes	0	0	5	0	1	11	14
An. dirus	0	23	0	0	5	0	4
An. peditaeniatus	0	0	0	0	7	11	9
An. kochi	0	0	112	0	118	70	104
An. crawfordi	0	0	34	35	43	106	53
An. pallidus	0	0	0	0	15	33	26
An. pseudojamesii	0	0	15	3	9	53	34
Total number of Individual	**5216**	**1515**	**4628**	**3954**	**5868**	**9615**	**6230**

EKH: East Khasi Hills, WKH: West Khasi Hills, RB: Ri-Bhoi, JH: Jaintia Hills, EGH: East Garo Hills, WGH: West Garo Hills and SGH: South Garo Hills.

and WGH in the month of July and August. The abundance of all species declined gradually at different rates in winter season (Figure 4).

Species richness and diversity

Alpha and beta biodiversity indices (mentioned above) were calculated district wise for the entire study area (Table 4). The average Anopheline species richness was about 24 ± 2.5 species per district and ranged from 15 (WKH) to 31 (WGH and SGH). The Welch F test showed that species richness varied significantly among districts

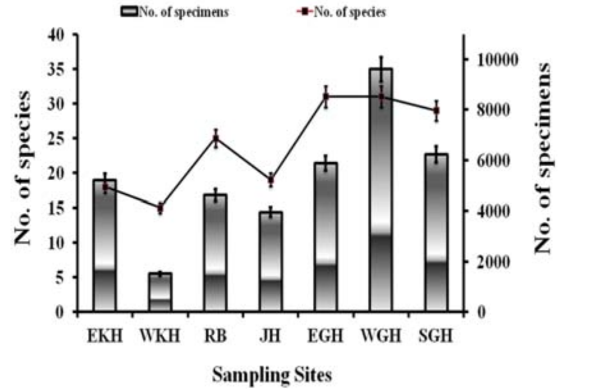

Figure 3: Graph showing species richness and relative abundance of Anopheles genera in all seven districts of Meghalaya.
EKH: East Khasi Hills district; WKH: West Khasi Hills district; RB: Ri-Bhoi district; JH: Jaintia Hills district; EGH: East Garo Hills districts; WGH: West Garo Hills District and SGH: South Garo Hills district

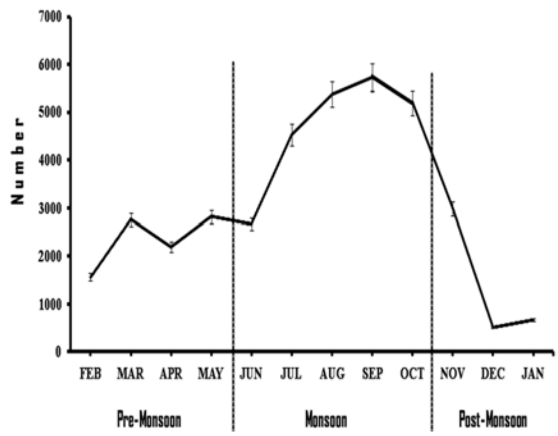

Figure 4: Seasonal distribution of Anopheline mosquitoes.

($F96.47 = 3.092$, $p \le 0.001$). The comparison of alpha diversity using Simpson and Shannon indices showed that RB district is the most diverse ($H'= 2.56$, $\lambda = 0.90$,) and E ranged from 0.33 to 0.58. According to the Margalef richness index, East Garo Hills district was the most diverse ($D_{mg} = 3.46$) and the West Khasi Hills districts showed least diversity ($D_{mg} = 1.91$, $\lambda = 0.79$, $H'= 2.00$).The order of the districts on the basis of higher to lower diversity is noted to be as EGH>WGH>SGH>RB>JH>EKH>WKH.

The β biodiversity analysis showed that EGH and SGH are the closest districts with reference to Anopheles diversity ($IJ= 0.94$) with the specific alternate among them being very low ($\beta_w = 0.03$). There is low similarity of 0.38, 0.38, 0.39, 0.38, and 0.44 found when WKH was compared to the EKH, RB, EGH, WGH and SGH districts (Table 5).

Cluster analysis and principal component analysis

The districts were also classified on the basis of species similarity in richness and abundance as depicted in the form of Bray-Curtis similarity cluster analysis. Cluster analysis produced 3 separate groups, out of which the major group (B) consisted of four districts followed by group A with two districts and group C with one district (Figure 5). Group B is the largest one which is further subdivided into small sub-units. Group C (West Khasi hills district) separated

Table 4: comparision of various community indices among the study sites in seven districts of Meghalaya.

Alpha Diversity	WKH	EKH	RB	JH	EGH	WGH	SGH
Species Richness	15	18	25	19	31	31	29
Individuals	5216	1515	4628	3954	5868	9615	6230
Simpson (λ)	0.79	0.86	0.90	0.89	0.86	0.88	0.88
Shannon (*H'*)	2.00	2.21	2.56	2.40	2.33	2.45	2.40
Evenness (*E''*)	0.49	0.50	0.52	0.58	0.33	0.37	0.38
Margalef (*Dmg*)	1.91	1.99	2.84	2.17	3.46	3.27	3.21
Barger-Parker	0.39	0.25	0.18	0.18	0.23	0.19	0.23

EKH: East Khasi Hills, WKH: West Khasi Hills, RB: Ri-Bhoi, JH: Jaintia Hills, EGH: East Garo Hills, WGH: West Garo Hills and SGH: South Garo Hills.

Table 5: Pair wise Whittaker and Jaccard β comparisons for each district.

Comparison	Jaccard (Ij)	Whittaker (βw)
EKH-WKH	0.38	0.45
EKH- RB	0.65	0.20
EKH-JH	0.48	0.35
EKH-EGH	0.53	0.31
EKH-WGH	0.53	0.31
EKH-SGO	0.57	0.27
WKH-RB	0.38	0.45
WKH-JH	0.55	0.29
WKH-EGH	0.44	0.39
WKH-WGH	0.39	0.43
WKH-SGH	0.38	0.45
RB-JH	0.57	0.27
RB-EGH	0.70	0.18
RB-WGH	0.75	0.14
RB-SGH	0.74	0.15
JH-EGH	0.61	0.24
JH-WGH	0.61	0.24
JH-SGH	0.55	0.29
EGH-WGH	0.94	0.03
EGH-SGH	0.88	0.06
WGH-SGH	0.88	0.07

- The Whittaker average for comparisons of the districts is 0.375.
- EKH: East Khasi Hills; WKH: West Khasi Hills; RB: Ri-Bhoi; JH: Jaintia Hills; EGH: East Garo Hills; WGH: West Garo Hills; SGH: South Garo Hills.

first at a major distance with only 39% similarity with other group, followed by West Garo hills districts (72.5% similarity). Anopheline mosquitoes in WKH were the poorest in terms of richness and abundance where mainly *An. maculatus* dominated with many rare species (relative abundance is less than 1%).The high value of Bray-Curtis cophenitic correlation (r_c= 0.9361) indicates a high correlation between ecological distance observed in our study.

The Principal Component Analysis (PCA) was used to analyze the correlation of all seven districts. The high value of the Jolliffe cut-off (1.035E) indicates a high correlation as noted after cluster analysis. The results of PCA also showed similar result with cluster analysis (Figure 6).

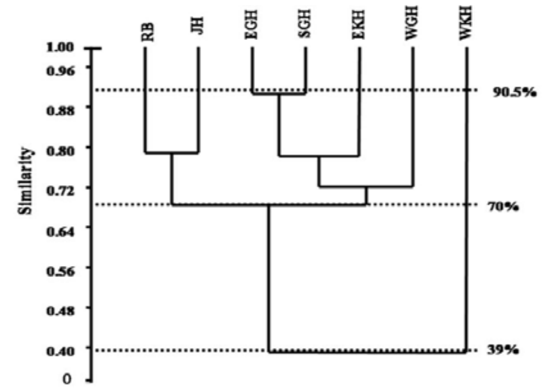

Figure 5: Cluster analysis using Bray Curtis Similarity for all seven districts and Cophenetic correlation (r_c) = 0.9361.
EKH: East Khasi Hills district; WKH: West Khasi Hills district; RB: Ri-Bhoi district; JH: Jaintia Hills district; EGH: East Garo Hills districts; WGH: West Garo Hills District and SGH: South Garo Hills district

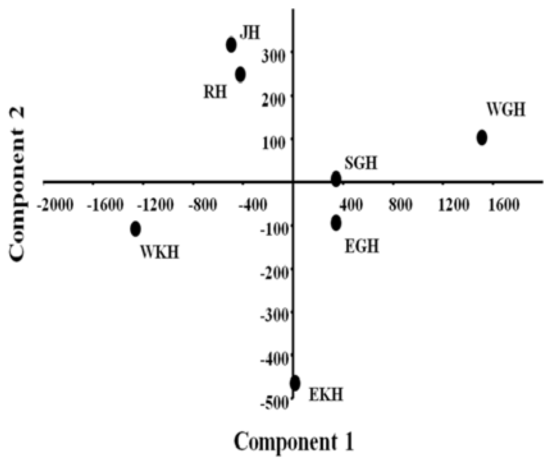

Figure 6: Principal component analysis of different districts based on the Anopheles species richness. The two axes explained 59% of the variation. Dots are showing study sites.
EKH: East Khasi Hills district; WKH: West Khasi Hills district; RB: Ri-Bhoi district; JH: Jaintia Hills district; EGH: East Garo Hills districts; WGH: West Garo Hills District and SGH: South Garo Hills district

Discussion

Meghalaya has a unique landscape of North-eastern India, supporting a varied flora and fauna because of highly humid, tropical, and subtropical climate [20]. The study areas have highly dissected and irregular topography, which includes warmer foothill areas and subalpine, alpine central plateau areas (Figure 1). Thus, it may be expected to find the presence of Anopheline species from two broad bio-geographical zones with unique possibilities for studies on the impact of climate and habitat on mosquito's species richness and biodiversity. Similar correlation has also been proposed in many other highlands [21,22]. Climate variability is widely considered to be a major driver of inter-annual variability of Anopheles species richness and abundance. The relationship between environmental variability and Anopheles abundance was assessed in this study. Climate suitability for Anopheles was defined as the coincidence of precipitation accumulation greater than 100 mm, mean temperature between 18ºC and 32ºC, and relative humidity greater than 60 percent [23-25]. Considering these climate suitability, present findings revealed that the environmental conditions of all the districts should be suitable for Anopheles mosquito survival except East Khasi Hills and West Khasi hills districts (Table 1).

The obtained rare faction curves (Figure 2) showed that the mosquitoes in an environment are randomly distributed. The sample size is sufficiently large and taxonomically similar and all the samples have similar pattern of distribution in each district. The curve is flattened to right (for each district) which suggests very high beta diversity. Rarefaction only works well when no species is extremely rare or common, or when beta diversity is very high. Rarefaction assumes that the number of occurrences of a species reflects the sampling intensity, but if one species is especially common or rare, the number of occurrences will be related to the extremity of the number of individuals of that species, not to the intensity of sampling [26]. In EGH and WGH, the curve has tendency to stabilize with 31 species. According to Magurran [27] geometric models for the abundance curves, more flattened distributions correspond to diverse sample, which is consistent with our present study (Figure 2).

During the present study, 33 species and 2 sub-genera of *Anopheles* were recorded. The number of Anopheline species recorded were more as compared to neighboring States i.e. Nagaland, 22 [28], Assam, 30 [29,30], Manipur, 17 [31], Mizoram 16 [32] and Tripura, 22 [29,33]. EGH (31), WGH (31), SGH (29) and RB (25) are warmer in spring- summer season recording maximum temperature range of 29ºC – 32ºC and wettest districts with humidity ranging from 60% - 80%. These factors may help to increase the larval habitat density and related biodiversity. Most parts of WKH (15) and EKH (18) are situated on central plateau area, thus, only a few species were capable of adapting to this adverse situation [21,34]. Overall species abundance and richness revealed that subgenera *Cellia* (86.4%) were the most dominant in all the districts. Earlier also *An. Maculatus* has been reported as the most dominant species of Meghalaya [35]. Further, our findings reveal that there is abundance of different species in different districts i.e. EKH is dominated by *An. lindesayi* (n =1309), WKH is dominated by *An. annularis* (n = 595) and WGH is dominated by *An. vagus* (n = 1810) although they are found in all seven surveyed districts (Table 2). Some species of Anopheles (*An. willmorei, An. majidi, An. dirus, An. nivipes and An. paditaeniatus*) were found in very low number during the study. The results of this study also support the report of NVBDCP, 2012 regarding the increase of malaria cases in Meghalaya compared to other part of India [36,7]. This could be due to high density of vector Anopheline mosquitoes.

Anopheles mosquitoes' survival and development has been suggested to significantly depend on ambient temperature [37-40]. The present findings on the occurrence of more diverse and abundant Anopheline species in low land and mid range areas of Garo Hills, Khasi Hills and Jaintia Hills, may also suggest the presence of suitable environmental conditions such as high temperature, humidity and acidic pH of aquatic habitat (Table1). Another possible cause of *Anopheles* abundance and richness in central plateau area of Khasi Hills could be anthropogenic land use changes. Normally at high altitude, mosquitoes population is controlled by low temperature and availability of less breeding place. Several studies have suggested that land use changes in any area may increase the Anopheline density and diversity [41-44].

Total nine malaria vector species were known in India and all were encountered and captured at varying density at study sites. Similar results have been reported from Sonitpur district of Assam [45,46] from Nagaland [28] and from Mizoram [32].

Alpha and beta biodiversity study is one of the easiest ways to measure the diversity and similarity in pair of study sites. Alpha diversity index like Shannon-Wiener, Simpson, Pileou's, Margalef and Barger-Parker are the most useful tool to measure the species diversity, species evenness, wealth of mosquito and species dominance [18]. The analysis of similarity in Shannon diversity index, Simpson's evenness and Barger–Parker dominance of each district indicated that uneven biodiversity prevailed in the whole study area. The Anopheles communities in the districts of Garo Hills zone showed higher species richness and diversity, and lower dominance index compared to the districts in Khasi Hills and Jaintia Hills zone. The decline in species richness and diversity from foothills to mountain habitat could be due to unfavorable environmental low temperature. Several hypotheses and studies suggested that Land Use and Land Cover (LULC) changes have influenced malaria vector, larval habitat availability, productivity, density and distribution in the world [24,39,47,48].

The observed increase in Anopheline mosquitoes in EKH having mostly plateau areas may be attributed due to human land manipulation and disturbance through urbanization involving LULC. Similar type of observation and suggestion has been given for study areas in Kenya and Africa [40].

Meri & Peydro [34] and Confalonieri &Costa-Neto [49] grouped Anopheline mosquitoes habitats on the basis of population. In present study, the same method was used to construct the dendrogram to group Anopheline mosquito habitat. This cluster analysis showed 90.5% similarity between EGH and SGH which is probably due to the similar environmental conditions. However, the similarity between EKH and Garo hills zone is suggested to be due to land use disturbances. The WKH had very low similarity with another groups (39%), may be due to the low temperature and less land use changes.

The results of the present studies show that biodiversity indices are very useful tool for environmental changes assessments. The similarity in Shannon diversity index, Simpson's evenness and Barger–Parker dominance of each district showed that uneven biodiversity prevail in the whole study area. Thus, based on the present study it may be suggested that the local environment plays the significant role in explaining Anopheline mosquitoes species distributions and indicates that community composition can be sustained under changing land use.

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References

1. WHO: World Malaria Report. 2012; pp 1-249.
2. Narain JP. Malaria in South - East region: Myth and the reality. Indian Journal of Medical Research. 2008; 128: 1-3.
3. Bhattacharjee A, Bhattacharjee DA. Bayesian approach to compare the state wise malaria death counts in India. Healthline. 2011; 2: 54-58.
4. Sinka ME, Bangs MJ, Manguin S, Chareonviriyaphap T, Patil AP, Temperley W, et al. The dominant Anopheles vector of human malaria in the Asia-Pacific region: occurrence data, distribution map and bionomic précis. Parasite and Vectors. 2011; 4: 1-78.
5. Greenwood BM, Fidock DA, Kyle DE, Kappe SH, Alonso PL, Collins FH, et al. Malaria: progress, perils, and prospects for eradication. The Journal of clinical investigation. 2008; 118: 1266-1276.
6. Dash AP. "Estimation of true malaria burden in India: 12. A profile of National Institute of Malaria Research". 2009.
7. Srivastava AK, Kharbuli B, Shira DS, Sood A. Effect of land use and land cover modification on distribution of Anopheline larval habitat in Meghalaya, India. Journal of Vector Borne Diseases. 2013; 50: 121-126.
8. Shortt H E. The occurrence of malaria in hill station. Indian Journal of Medical Research 11. 1925; 767-771.
9. Prakash A, Bhattacharyya DR, Mohapatra PK, Mahanta J. Anopheline fauna of the north-eastern states of India with notes on vectors of Malaria. Proceeding of the Indian National Science Academy. 1998; 68: 217-229.
10. Marak CP. State profile of community forestry: Meghalaya, Ne India. Community forestry international (CFI), USA. 2007; 1-13.
11. WHO: Manual on practical entomology in malaria vector bionomics and organization of antimalaria activities, Part I and Part II. Offset Publication No. 13. World Health Organization, Geneva. 1975.
12. Reuben R. A report on mosquitoes collected in the Krishna-Godavari delta, Andhra Pradesh. Indian Journal of Medical Research. 1978; 68: 603-609.
13. Service MN. Mosquito ecology: Field Sampling Methods. Edition II. London, Kluwer Academic Publishers. 1993; 889.
14. Christophers SR. The fauna of British India, including Ceylon and Burma. Diptera. Family Culicidae, Tribe Anopheline Taylor & Franci, London. 1933; 1-371.
15. Gillies MT, Coetzee M. A supplement to the Anophelinae of Africa South of the Sahara (Afro-tropical region). Johannesburg: South Africa institute of Medical Research. 1987; 55: 1-143.
16. Das BP, Rajagopal R, Akiyama J. Pictorial keys to the species of Indian Anophelines mosquitoes. Zoology: Journal of Pure and Applied Zoology. 1990; 2: 131-162.
17. Nagpal BN, Sharma VP. Indian Anophelines. New Delhi, India, Oxford and IBH Publishing Co Pvt Ltd. 1995; 416.
18. Montes J. [Culicidae fauna of Serra da Cantareira, Sao Paulo, Brazil]. Rev Saude Publica. 2005; 39: 578-584.
19. Hammer DAT, Harper, Ryan PD. Past: paleontological statistics software package for education and data analysis. Paleontologica Electronica. 2001; 4: 1- 9.
20. Roy PS, Tomar S. Landscape cover dynamics pattern in Meghalaya. International Journal of remote sensing. 2001; 22: 3813-3825.
21. Eisen D, Bolling BG, Blair CD, Beaty BJ, Moore CG. Mosquito species richness, composition, and abundance along habitat-climate-elevation gradients in the Northern Colorado Front range. Journal of Medical Entomology. 2008; 45: 800-811.
22. Pinault LL, Hunter FF. New highland distribution records of multiple Anopheles species in the Ecuadorian Andes. Malaria Journal. 2011; 10: 1-11.
23. Koenraadt CJ, Paaijmans KP, Githeko A, Knols B, Takken W. Egg hatching, larval movement and larval survival of the malaria vector Anopheles gambiae in desiccating habitats. Malaria Journals. 2003.
24. Munga S, Minakawa N, Zhou G, Mushinzimana E, Barrack OJ, Githeko AK, et al. Association between land cover and habitat productivity of malaria vectors in western Kenyan Highlands. American Journal of Tropical Medicine and Hygiene. 2006; 74: 69 -75.
25. Afrane YA, Little TJ, Lawson BW, Githko A, Yan G. Deforestation and vectoral capacity of Anopheles gambiae Giles mosquitoes in malaria transmission, Kenya. Emerging Infectious Diseases. 2008; 14: 1533-1538.
26. Bush AM, Markey MJ, Marshall CR. Removing bias from diversity curves: the effects of spatially organized biodiversity on sampling-standardization. Paleobiology. 2004; 30: 666-686.
27. Magurran AE. Measuring Biological Diversity. Malden, USA, Blackwell Publishing Company. 2004; 215.
28. Dutta P, Khan SA, Khan AM, Sharma CK, Mahanta J. Survey of mosquito species in Nagaland, a hilly state of north east region of India. Journal of Environmental Biology. 2010; 31: 781-785.
29. Malhotra PR, Mahanta HC. Check- list of mosquitoes of Northeast India (Diptera: Culicidae). Oriental Insects. 1994; 28: 125-149.
30. Dutta P, Prakash A, Bhattacharyya DR, Khan SA, Gogoi PR, Sharma CK, et al. Mosquito biodiversity of Dibru-Saikhowa biosphere reserve in Assam, India. J Environ Biol. 2010; 31: 695-699.
31. Dutta P, Khan SA, Khan AM, Sharma CK, Mahanta J. Biodiversity of mosquitoes in Manipur State and their medical significance. J Environ Biol. 2005; 26: 531-538.
32. Vanramliana, Lalramnghinglova H. Anopheline diversity in Undivided Aizawl district of Mizoram, India. Science vision. 2013; 13: 35-39.
33. Nagpal BN, Sharma VP. Survey of mosquito fauna of northeastern region of India. Indian J Malariol. 1987; 24: 143-149.
34. Marí RB, Jiménez-Peydró R. Differences in mosquito (Diptera: Culicidae) biodiversity across varying climates and land-use categories in Eastern Spain. Entomologica Fennica. 2011; 22: 190-198.
35. Das SC, Das NG, Baruah I. Mosquito survey in Meghalaya. Indian J Public Health. 1984; 28: 147-151.
36. Delhi India: National Vector Borne Disease Control Programme Ministry of Health and Family Welfare.
37. Leeson HS. Longevity of Anopheles maculipennis race atroparvus, Van Thiel, at controlled temperature and humidity after one blood meal. Bulletin of Entomological Research. 1939; 30: 103-301.
38. DeMeillon, B. Observations on Anopheles funestus and Anopheles gambiae in the Transvaal. South African Institute of Medical Research. 1934; 6: 199-248.
39. Mouchet J, Manguin S, Sircoulon J, Laventure S, Faye O, Onapa AW, et al. Evolution of malaria in Africa for the past 40 years: impact of climatic and human factors. J Am Mosq Control Assoc. 1998; 14: 121-130.
40. Afrane YA, Githeko AK, Yan G. The ecology of Anopheles mosquitoes under climate change: case studies from the effects of deforestation in East African highlands. Ann N Y Acad Sci. 2012; 1249: 204-210.
41. GARNHAM PC. The incidence of malaria at high altitudes. J Natl Malar Soc. 1948; 7: 275-284.
42. Lindblade KA, Walker ED, Onapa AW, Katungu J, Wilson ML. Land use change alters malaria transmission parameters by modifying temperature in

a highland area of Uganda. Tropical Medicine and International Health. 2005; 5: 263-274.

43. Munga S, Minakawa N, Zhou G, Githeko AK, Yan G. Survivorship of immature stages of Anopheles gambiae s.l. (Diptera: Culicidae) in natural habitats in western Kenya highlands. J Med Entomol. 2007; 44: 758-764.

44. Phasomkusolsil S, Lerdthusnee K, Khuntirat B, Kongtak W, Pantuwatana K, Murphy JR. Effect of Temperature on laboratory reared Anopheles dirus Peyton and Harrison and Anopheles sawadwongporni Rattanarithikul and Green. Southeast Asian Journal of Tropical Medicine and Public Health. 2011; 42: 63-70.

45. Baruah I, Das NG, Kalita J. Seasonal prevalence of malaria vectors in Sonitpur district of Assam, India. J Vector Borne Dis. 2007; 44: 149-153.

46. Adhikari ML, Kalita S, Deka TC. Diversity and abundance of malaria vectors in Dhekiajuli Subdivision of Sonitpur District, Assam. Asian Journal of Experimental Biological Sciences. 2013; 14: 103-111.

47. Lindsay SW, Martens WJ. Malaria in the African highlands: past, present and future. Bull World Health Organ. 1998; 76: 33-45.

48. Githeko AK, Lindsay SW, Confalonieri UE, Patz JA. Climate change and vector-borne diseases: a regional analysis. Bull World Health Organ. 2000; 78: 1136-1147.

49. Confalonieri UE, Neto CC. Diversity of mosquito vectors (Diptera: culicidae) in caxiuanã, pará, Brazil. Interdiscip Perspect Infect Dis. 2012; 2012: 741273.

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