A survey of anopheline mosquitoes and malarial parasite in commuters in a rural and an urban area in West Bengal, India

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Malaria and other mosquito borne diseases are common in Kolkata, India. Among the 34 species of mosquitoes recorded from Kolkata, 13 are anophelines, of that *Anopheles annularis* Van der Wulp, *An. culicifacies* Giles, and *An. stephensi* Liston are established vectors of malaria^{1–3}. The abundance of these vector mosquitoes varies with the season and available habitats^{2–4}. Mobility of human hosts between different regions influence the transmission process too^{5,6}. In this regard, assessment of the density and diversity of anopheline mosquitoes in space and time can help to monitor the possibility of malaria in a region apart from formulating the controlling strategies.

Keeping these in view, the present survey was aimed at evaluation of the relative abundance of anopheline mosquitoes and an association malarial parasite in commuters. The survey considered two distinct regions—one urban and one rural area that are expected to provide different larval mosquito habitats and completely different landscape. Since the migrating human population is expected to be susceptible to mosquito bites from both the regions, an estimate of malarial parasites in the commuters from this region will help to evaluate their possible role in spreading the disease vis-avis the abundance of anopheline mosquitoes.

Survey was carried out between March 2003 and February 2006 in two districts of West Bengal,

Kolkata, an urban area and three villages of Amta, Howrah. Commuters frequenting between these places were considered as potential hosts of malarial parasites.

Adult anopheline mosquitoes were collected from two habitats-cattlesheds and human dwellings between 0630 and 0730 hrs by a single man using an aspirator. At least three sites of each habitat were considered for sampling. The temperature and relative humidity were noted during collection. The collected mosquitoes were identified using appropriate keys⁷. The numbers of each species of the anopheline mosquitoes were recorded with respect to the habitats as well as urban (Kolkata) and rural (Amta, Howrah) areas concerned. Records of the blood test performed in a selected clinical laboratory of about 400 people suspected of malaria were considered. These people commute between Amta, Howrah and Kolkata-at least once a week. Positive cases of Plasmodium vivax and P. falciparum were noted for each month in a year.

Correlation coefficients and regression analysis were performed^{8,9} with respect to the abundance of anophelines and the malaria positive cases in respect to months. Also, seasonal [summer (March –May), monsoon (June–August), post-monsoon (September–November) and winter (December– February)] variation of anophelines was subjected to correlation analysis. Factorial analysis was made in respect to the data obtained on the species diversity of anophelines in respect to the urban and rural areas irrespective of human dwellings and cattlesheds.

Four species of anopheline mosquitoes were recorded from the rural sectors namely An. annularis, An. culicifacies, An. subpictus Grassi, and An. vagus Donitz. In addition to these, An. stephensi was recorded in the urban sector, making a total of five mosquito species. The abundance pattern of these mosquitoes varied with the months in a year, and between urban and rural areas. In rural as well as urban areas An. subpictus was found to be the dominant species in terms of its abundance. The relative and absolute number of mosquitoes, irrespective of species was greater in cattlesheds as compared to the human dwellings. The post-monsoon season was found to be the most favourable period for the abundance of the mosquitoes, irrespective of areas and habitats surveyed followed by monsoon, summer and winter (Fig. 1). During the study period, the vector mosquitoes constituted 25.97, 19.94, 20.19 and 0.85% of the total in rural area (Amta) and 38.19, 33.32, 37.39 and 2.29% of the total in urban area (Kolkata) in the summer, monsoon, post-monsoon and winter seasons respectively (Table 1).

Of the total blood samples tested, the positive cases for *P. vivax* ranged from 1.16 to 7.95% during the whole study period. Contrast to this, *P. falciparum* cases ranged from 0 to 1.13%. There was a complete absence of *P. falciparum* in the year 2005–06. None of the blood slides was found positive for both the parasites. Also, in winter season, there was no record of positive cases of malaria. The coefficient of correlation (r) between occurrence of malarial parasite and abundance of known anopheline mosquitoes are + 0.85, +0.955 and +0.986 in the years of 2003–04, 2004–05 and 2005–06 respectively. The 'r' values for anopheline abundance and positive cases of malaria among the commuters for the same years were +0.601, +0.831 and +0.866 respectively.

The overall abundance of anopheline mosquitoes in rural areas was negatively correlated with the air temperature (r = -0.274), while for the urban areas a positive correlation was noted (r = +0.131). With the relative humidity, the r-values for the rural and urban areas were +0.399 and +0.482, respectively. For both the environmental parameters, the values were not significant. However, for the known vectors of malaria, a positive correlation values between +0.12 and +0.22 were noted for the years surveyed irrespective of the rural or urban areas. The regions considered in our survey were not distant and thus at a broader level the air temperature and the relative humidity values did not differ much.

The multivariate analysis (factorial analysis based on principle components) on the abundance of different anopheline species reflected that a positive correlation exists in the abundance pattern throughout the



Fig. 1: Occurrence and abundance of adult anopheline mosquitoes in rural Amta and urban Kolkata in relation to atmoshperic temperature and relative humidity (R — Rural; U — Urban)

Table 1. Abundance (in man hour density—MHD) of adult anopheline mosquitoes of Amta (rural) and Kolkata (urban)
during March 2003 to February 2006 (n = 3 habitats/month/area) and the number of malaria positive cases
[P. vivax, P. falciparum (Total number of commuters sampled)]

Year/Species		Sur (Marci	nmer h–May)	ler Monsoon May) (June–August)		Post-monsoon (September–November)				Winter (December–February)						
	Ar	nta	Kolk	ata	A	mta	Ko	lkata	Am	nta	Kol	kata	Am	ta	Ko	kata
-	CS	HD	CS	HD	CS	HD	CS	HD	CS	HD	CS	HD	CS	HD	CS	HD
2003-04																
An. annularis	2.0	0.66	1.66	1.0	5.33	2.33	1.33	0.66	5.66	2.33	11.0	2.66	0	0	0.33	0
An. culicifacies	8.0	4.0	3.0	2.0	9.0	4.33	5.0	2.66	16.66	8.66	11.66	5.0	0	1	0.66	0
An. stephensi	0	0	6.0	3.33	0	0	20.33	8.33	0	0	30.0	16.66	0	0	1.33	0.33
An. subpictus	25.33	12.66	22.66	11.33	46.33	25.33	38.66	22.66	93.0	43.33	71.33	32.66	59.66	23.66	41.0	15.66
An. vagus	14.0	7.0	3.0	1.66	31.66	15.0	20.66	7.33	32.33	20.33	22.33	18.66	25.0	8.33	15.0	5.33
Malaria +ve case	laria +ve cases $1, 0 (21)$			2, 0 (25)				3, 1 (33)				0, 0 (7)				
2004–05																
An. annularis	7.66	1.33	4.0	0.66	6.66	1.66	2.0	0.66	16.0	3.0	12.0	5.0	0.66	0	0	0
An. culicifacies	13.66	5.33	8.0	2.66	14.0	5.66	10.0	4.33	20.0	6.33	18.0	6.0	0.33	0	1.0	0
An. stephensi	0	0	16.0	6.0	0	0	15.0	4.0	0	0	30.66	11.66	0	0	0.33	0
An. subpictus	34.0	3.0	28.0	6.0	54.0	13.66	38.0	9.0	102.0	31.0	80.0	21.33	60.66	12.66	47.33	9.33
An. vagus	18.66	5.0	11.33	3.0	31.33	8.66	21.0	6.0	41.0	13.33	33.33	11.0	21.66	7.0	15.0	2.33
Malaria +ve case	s	4,0((25)			2,0(28)			7,1((29)			0, 0	(6)	
2005–06																
An. annularis	4.0	1.0	1.33	0.66	8.0	1.0	5.0	0.66	15.0	3.33	13.0	2.66	0	0	0.33	0
An. culicifacies	12.0	4.33	9.0	2.33	15.0	6.0	12.0	3.0	30.66	10.33	24.0	6.0	0.66	0	0.33	0
An. stephensi	0	0	13.0	3.0	0	0	16.0	4.33	0	0	27.33	7.66	0	0	0.66	0
An. subpictus	35.33	9.66	24.33	8.0	45.0	14.33	37.0	10.0	89.66	30.0	71.33	25.0	57.0	18.66	39.66	11.33
An. vagus	14.33	4.0	12.66	3.33	24.0	8.0	15.0	5.33	35.33	14.0	26.0	8.33	24.33	6.33	19.0	4.33
Malaria +ve case	s	3, 0 ((37)			3, 0 ((32)			7,0((36)			0, 0	(8)	

CS = Cattlesheds; HD = Human dwellings.

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rrelation matrix of multivariate factorial analysis of abundance of anopheline mosquitoes in rural Amta and urban Kolks	rs of the survey carried out between 2003 and 2006 (n = 36 months). In rural area An. stephensi was completely absent
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Correlation Matrix

Species		Amta	_				Kolkata		
	An. annularis	An. culicifacies A	In. subpictus	An. vagus Ar	ı. annularis An.	culicifacies	An. stephensi	An. subpictus	An. vagus
Rural									
An. annularis	1.0	0.883	0.544	0.635	0.830	0.927	0.810	0.609	0.642
An. culicifacies	0.883	1.0	0.465	0.532	0.822	0.957	0.833	0.539	0.534
An. subpictus	0.544	0.465	1.0	0.870	0.785	0.570	0.659	0.977	0.909
An. vagus	0.635	0.532	0.870	1.0	0.712	0.605	0.738	0.893	0.922
Urban									
An. annularis	0.830	0.822	0.785	0.712	1.0	0.874	0.859	0.822	0.748
An. culicifacies	0.927	0.957	0.570	0.605	0.874	1.0	0.825	0.614	0.637
An. stephensi	0.810	0.833	0.659	0.738	0.859	0.825	1.0	0.719	0.752
An. subpictus	0.609	0.539	0.977	0.893	0.822	0.614	0.719	1.0	0.897
An. vagus	0.642	0.534	0.909	0.922	0.748	0.637	0.752	0.897	1.0

year in the rural and urban areas (Table 2). In the analysis nine species of anophelines observed in the rural and urban areas were considered as variables. Two factors were extracted as major determinants or components from the correlation matrix, having Eigen values of 6.98 and 1.31; the Eigen value is a measure of the total test variance accounted for by a particular factor when the total variance for each test being unity. These components accounts for approximately 93% of the total variance obtained in the abundance pattern of the anopheline species in the rural and urban areas. Overall the analysis reveals that the abundance pattern was positively correlated for each of the species irrespective of rural or urban habitat and two major components-the mathematical factors explaining the variance in the abundance and distribution of the mosquitoes.

However, contrast to this, the relative number of each of the species of anopheline mosquitoes varied significantly in respect to the rural and urban areas. Paired t-test revealed significant difference in the relative abundance of the anopheline species, in urban and rural areas (for An. annularis, $|\mathbf{x}| = 1.78$, t = 2.929, p < 0.006; for An. culicifacies, $|\mathbf{x}| = 4.89$, t = 7.272, p < 0.0001; for An. subpictus, $|\mathbf{x}| = 18.75$, t = 11.546, p < 0.0001; for An. vagus |x| = 11.64, t = 14.733, p < 0.0001; in all cases df = 35). Thus, it can be inferred from the data obtained that the basic pattern of abundance of anopheline mosquitoes in rural and urban areas was positively correlated, though significant difference in the relative representation by any Anopheles species in the urban and rural areas are prominent at a particular time and space.

The choice of breeding habitats of anopheline mosquitoes in West Bengal is not known specifically, yet the occurrence of *An. stephensi* in the urban areas only is one evidence that the urban and rural areas might provide different breeding habitats. Similar variation was observed between irrigated and nonirrigated rice-fields in Sri Lanka¹⁰ and Mali¹¹ and in rural and urban areas of sub-Saharan Africa¹². In reference to the abundance of the anopheline mosquitoes and the malarial parasites in the human sample, post-monsoon and monsoon seasons were noted to be significant. Similar pattern of seasonal variation of mosquito abundance and malaria cases were noted from Kalsi, Uttaranchal¹³, with the winter being the season for least number of malaria cases.

Several factors are involved in the perpetuation and persistence of malarial parasites in a region, despite controlling anopheline mosquitoes and establishing strict health and hygiene norms. Human mobility from one place to another can change the whole state of precautions against malaria ^{1-3,5,6}. In Kolkata, the health and hygiene measures are relatively strict compared to the rural Amta³. Also, cattlesheds are more common in Amta compared to Kolkata. Our survey as a model study reveals that a positive relationship exists in terms of anopheline density and the presence of malarial parasites in the commuters between the seasons. These preliminary findings can serve as the basis for further studies on the people, mosquito and malaria in the state of West Bengal as a whole and Kolkata in particular.

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