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Seasonal variation of *Anopheles* mosquitoes with special reference to malarial vectors in selected areas of Thiruvananthapuram District, Kerala, India

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Abstract
Malaria is a life threatening tropical endemic disease transmitted by *Anopheles* mosquitoes. *Anopheles stephensi* is the notorious urban vector among the known malaria vectors. According to DHS Kerala data on communicable diseases, 439 malaria cases were reported in the year 2022 and among these 39 were from Thiruvananthapuram district. The present study was undertaken to find out the seasonal variation in the density of *Anopheles* mosquitoes, especially malarial vectors, in 3 areas: Vizhinjam (coastal), Vithura (hilly) and Pattom (urban) of Thiruvananthapuram district. Adult mosquitoes collected from cattle sheds were identified and man hour densities (MHD) from each area were calculated. In the present study 9 species of *Anopheles* mosquitoes were collected and 2 species, *Anopheles stephensi* and *Anopheles varuna* are known malaria vectors. Among these, the urban vector *Anopheles stephensi* showed higher MHD in coastal and urban areas *(*Vizhinjam, Pattom*)* but was not detected in hilly area (Vithura). In the urban area, higher density of *Anopheles stephensi* was observed in all seasons. *Anopheles varuna* was detected from all three areas (hilly, urban and coastal) with lower density and showed no seasonality in this study. The excessive influx of migrant workers from other states especially in Vizhinjam harbour area may be the cause of increase in malaria cases. The presence of vectors indicates the risk of local transmission. Hence, vector control activities should be intensified to prevent the occurrence of indigenous cases.

Keywords: *Anopheles stephensi*, malarial vector, seasonal variation, man hour density, Thiruvananthapuram

Introduction
Malaria is a life-threatening tropical disease transmitted by *Anopheles* mosquitoes. According to the latest WHO world malaria report there were 249 million malarial cases globally with 6,08000 deaths. WHO estimates that India has 15 million cases of malaria with 19500-20000 deaths annually [1]. According to Directorate of Health services (DHS) Kerala data on communicable diseases, 439 malarial cases were reported from Kerala, among these 39 from Thiruvananthapuram district [2]. In 2015 an outbreak of malaria was reported at Vizhinjam coast of Thiruvananthapuram district [3]. A study by Nidhish et.al., indicates migrants and immigrant people were the major contributors of new malarial cases in Trissur district [4]. Unhygienic and overcrowded shelters for migrant labours may become a cause for disease transmission. Another study reveals two problems related to imported malaria. One is the potential of imported malaria to become source of infection and the second problem is delayed diagnosis or misdiagnosis [5]. There are 39 species of *Anopheles* mosquitoes recorded from Kerala [6]. *An. stephensi*, *An. culicifacies*, *An. fluviatilis*, *An. dirus*, *An. minimus*, *An. philippinensis*, *An. annularis and An. varuna* are the reported malaria vectors [7]. *Anopheles stephensi* is the notorious urban vector among the known malaria vectors. A study on mosquito density will help the authorities to take suitable actions at the right time to prevent the occurrence of indigenous cases.

Materials and method
This study was conducted from February 2022 to January 2023 in 3 selected areas of Thiruvananthapuram district *viz*., Vithura, Vizhinjam and Pattom (fig. 1). Vithura is a hilly village area located 36 kilometres from Thiruvananthapuram city. Vizhinjam is a coastal area belonging to Thiruvananthapuram Municipal Corporation. Pattom is an urban area located in the heart of Thiruvananthapuram city. 5 houses with cattle sheds were selected from each area for mosquito collection. Adult mosquitoes were collected using aspirator tube and torch light between 6.00 pm and 10.00 pm at fortnightly intervals. Mosquitoes collected were transferred to the laboratory and identified using standard taxonomic key [8,9,10] and man hour densities (MHD) were calculated from the monthly data. Data on communicable diseases were collected from the DHS Kerala website.

Results
In the present study 16 species of mosquitoes were collected (Table-1). The mosquitoes composed of 9 species of *Anopheles*, 2 species of *Culex*, 2 species of *Mansonia*, 2 species of *Aedes* and 1 species of *Armigeres*.
In Pattom, 8 species of *Anopheles* mosquitoes were detected. Among these *An. stephensi* and *An. varuna* are known malarial vectors. From Vizhinjam 6 species of *Anopheles* mosquitoes were found, 2 of them were malarial vectors namely, *An. stephensi* and *An. varuna*. In Vithura, 4 species of *Anopheles* mosquitoes were collected and *An. varuna* was the only malarial vector found there.
The Mean Man Hour Densities (MHD) of mosquitoes from each site Pattom, Vizhinjam and Vithura are shown in table 2, table 3 and table 4 respectively.
In pre-monsoon season there was no significant difference (p =.070) between the Man hour densities of *An. stephensi*, *An. varuna* and other non-vector *Anopheles* mosquitoes in Pattom area. In Vizhinjam (p=0.004) and Vithura (p=0.003) there was a significant difference between the densities of vector mosquitoes. In monsoon season, the density of mosquitoes showed significant difference in all 3 areas. p

values of Pattom, Vizhinjam and Vithura were 0.038, 0.003. and 0.038 respectively. In post monsoon also all three areas showed significant difference in MHD (p=0.009, p= 0.0001, and p=0.006).

The density of An. stephensi showed no significant difference between pre-monsoon, monsoon and post monsoon seasons (p = 0.436), in Pattom area. But *An. varuna* was detected only in pre-monsoon season in that area. Other non-vector mosquitoes *viz*., *An. jamesi, An. vagus, An. nigerrimus, An. subpictus, An. tessellatus, and An. barbirostris* showed medium level of densities (table. 2).

In Vizhinjam also density of *An. stephensi* (p = 0.528) and *An. varuna* (p = 0.40) showed no significant difference between pre-monsoon, monsoon and post monsoon *seasons*, but showed a slightly higher density in pre-monsoon season. *An. varuna* was detected only in post monsoon season from this area. A higher density of other non-vector *Anopheles* mosquitoes, i.e, *An. jamesi*, *An. nigerimus* and *An. karwari* were observed from this area (table 3).

In Vithura, *An. varuna* showed a slightly higher density in pre-monsoon season but no significant difference (p=0.569) between seasons were observed. *An. stephensi* was not recorded from this locality. A higher density of other non-vector *Anopheles* mosquitoes like, *An. jamesi*, *An. nigerimus* and *An. karwari* were observed (table 4).

Table 5 shows annual incidences of malaria cases in Kerala state and Thiruvananthapuram district from 2000 to 2002. There was an increase in malaria cases in the state from the year 2020 to 2022. The status of malaria cases in Thiruvananthapuram district also showed an increasing trend. Number of malaria cases in Thiruvananthapuram was 29 in 2020 and was increased to 39 cases in 2022. Among the 39 cases reported from Thiruvananthapuram district, 24 cases were caused due to *Plasmodium vivax*, 14 were due to *P. falciparum and 1 fr*om *P. malaria*. All of these cases were categorised as imported cases. Most of the patients were migrants from different states like Andra Pradesh, Bihar, Chhattisgarh, West Bengal, Utter Pradesh and Maharashtra. Besides, 4 of them were reported to get infections from Africa and 2 persons from Democratic republic of Congo (DRC).

D iscussion

The present study was carried out in three geographically different areas of Thiruvananthapuram district of Kerala state *viz*., hilly, coastal and urban areas. The presence of malaria vectors was recorded in all 3 areas. However, the notorious urban vector *An. stephensi* was recorded only from the coastal and urban areas. The presence of *An. stephensi* in these areas is a matter of concern. Since all the malaria cases reported from Kerala from 2020 to 2022 were imported cases and many of the cases were detected from migrant workers, the excessive influx of migrant workers from other states especially in Vizhinjam harbour area could trigger malaria outbreaks in the presence of this vector. The construction work of an international seaport was in progress at the time of the study. According to WHO world malaria report, the Indian states of Odisha, Chhattisgarh, Jharkhand, Meghalaya and Madhya Pradesh disproportionately accounted for nearly 45.47 percent (1,53,909 cases out of India's 3,38,494 cases) of malaria cases and 70.54 percent (1,10,708 cases out of India's 1,56,940 cases) of P. falciparum Malaria cases in 2019 [11].

The seasonal difference in the densities of *An. stephensi* was found different from earlier studies. In 2014 a study reported the maximum densities of mosquitoes during monsoon season and minimum during pre-monsoon season [12]. Another study in 2006 recorded the highest density of *An. stephensi* in Thiruvananthapuram city during pre-monsoon and monsoon seasons [13]. However, in the present study, there was no considerable difference in MHD of *An. stephensi* with respect to seasons. This may be due to the unseasonal rain falls. The provision of unsafe and unhygienic shelters to migrant workers increases the chances of them being bitten by mosquitoes. A study on mosquito density will surely help the authorities to take suitable action at the right time to prevent the occurrence of indigenous cases. The presence of vectors indicates the risk of local transmission, so vector control activities should be intensified to prevent the occurrence of indigenous cases.

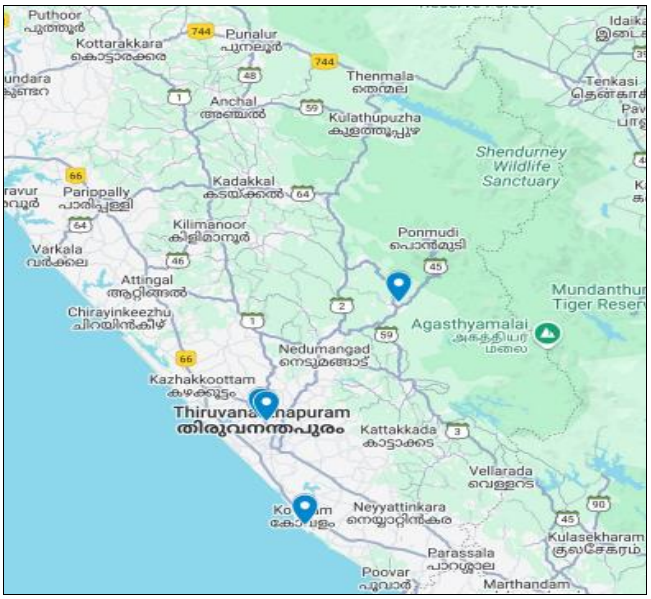

Fig 1: Map showing study areas in Thiruvananthapuram district (Vizhinjam, Vithura and Pattam)

Table 1: Mosquito species collected from the study area

S no	Mosquito species	Pattom	Vizhinjam	Vithura
1	*Anopheles stephensi*	+	+	-
2	*Anopheles varuna*	+	+	+
3	*Anopheles jamesi*	+	+	+
4	*Anopheles vagus*	+	+	-
5	*Anopheles nigerrimus*	+	+	+
6	*Anopheles karwari*	-	-	+
7	*Anopheles subpictus*	+	+	-
8	*Anopheles tesselatus*	+	-	-
9	*Anopheles barbirostris*	+	-	-
10	*Mansonia annulifera*	+	-	+
11	*Mansonia uniformis*	-	+	+
12	*Culex gelidus*	+	+	+
13	*Culex tritaeniorhynchus*	+	+	+
14	*Aedes vittatus*	+	+	-
15	*Aedes vexans*	+	-	-
16	*Armigeres subalbatus*	+	+	+

Table 2: Mean Man Hour Densities (MHD) of mosquitoes in Pattom

Season	*An. stephensi*	*An. varuna*	others
Pre-monsoon	6.75	0.25	2.25
Monsoon	4	0	0.5
Post monsoon	3.75	0	1.75

Table 3: Mean Man Hour Densities (MHD) of mosquitoes in Vizhinjam

Season	An. stephensi	An. varuna	others
pre-monsoon	3.75	0	6
monsoon	2.5	0	0.875
post monsoon	2.75	0.125	0.375

Table 4: Mean Man Hour Densities (MHD) of mosquitoes in Vithura

season	*An. stephensi*	*An. varuna*	others
pre-monsoon	0	0.25	2.75
monsoon	0	0.125	3.75
post monsoon	0	0	2.25

Table 5: Malaria cases according to DHS Kerala data on communicable diseases

Area	2020	2021	2022
Kerala	268	309	439
Thiruvananthapuram	23	39	39

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Declaration of competing interest
The authors affirm that there is no conflict of interest associated with this research

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