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PREVALENCE OF DENGUE VIRUS ANTIGEN IN AEDES MOSQUITOES AND LINKING OF DENGUE CASE INCIDENCE IN GIS MAPPING OF TAMILNADU STATE.

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Abstract

INTRODUCTION : Dengue is a Vector borne Disease which is caused by a virus and transmitted by Aedes mosquitoes. A Geographic Information System (GIS) was used as an analytical tool to study the spatial distribution of dengue virus-infection among Aedes mosquitos in Tamilnadu. Databases are available with village wise code. The dengue infection among field caught Aedes female mosquito were subjected through reverse transcriptase - polymerase chain reaction (RT-PCR) assay and its description in GIS mapping of Tamilnadu was the main motto of this study. In laboratory experiments, all assays showed sufficient sensitively to detect viraemia among the infected mosquito through RT-PCR. The mosquito pools for this study were collected from all the Health unit districts including urban areas and segregated into positive and negative pools affected villages respectively and the same has been depicted in GIS mapping so as to study the area or village with positive case incidence as well as positive in mosquito pools. Thus, the geographic distribution of the virus infected Aedes mosquitos, village wise locations were depicted in GIS linked with the human cases. Test results out of 13220 Aedes mosquito pools collected, 514 places found positive along with dengue cases incidence are depicted and linked in GIS mapping and there is a linkage with positive correlation between dengue positive and mosquito pool positive (r = 0.3443,) The development of a village wise RT-PCR laboratory-based dengue virus infection among Aedes mosquitoes with the reporting of dengue cases can successfully serve as an epidemiological tools in an early warning signal for dengue epidemics and simultaneously to initiate the control measures in anticipation.

INTRODUCTION

Dengue is a major public health concern for many tropical and sub-tropical regions of the world, causing periodic or annual outbreaks which eventually results in high mortality esp., in children. Dengue is endemic in Tamil Nadu also. Dengue is reported in all the districts of Tamil Nadu every year. The reporting of cases varies year to year depending upon the environmental situation, climatic condition of rain fall, humidity & temperature which increases the breeding potentiality of the Aedes mosquitoes. The Department of Public health & Preventive Medicine has been taken all necessary prevention & control measures under the guidance of Govt., of Tamil Nadu such as strengthening the fever surveillance and vector control activities. Dengue viruses are transmitted to humans through the bite of infective Aedes mosquitos, which breeds in stagnant water in all forms of receptacles in both urban and rural areas. The present study mainly envisaged to detect dengue viral antigen in Aedes vector mosquitoes throughout Tamil Nadu state. Detection of dengue viral antigen in vector mosquitoes will be helpful in finding out the vector infection rate in an area that determine the transmission potential of dengue virus. This will be useful in mapping positive areas and thereby preventing dengue epidemics. Forecasting dengue epidemics will pave a way to prevent possible outbreaks and dengue deaths.

METHODOLOGY

Geographic Information System (GIS) databases have been recently used to monitor the presence of vireamia among vector mosquitoes plays a vital role in disease transmission. This could be an important tool to assess the field situation of dengue cases epidemic. (Sithiprasasna et al, 1997, 2003) therefore it is noteworthy to identify the hot spot or vulnerable area or villages prone to epidemic and the same to be mapped in GIS so as to curtail the epidemic well in advance in the district as a precautionary measures.

i. Field collection of mosquitos :

Adult Aedes mosquitos were collected at weekly intervals from Jan to December 2023 from both indoors and outdoors in dengue-endemic areas both in urban & rural pockets from all 46 Health Unit Districts. At each indoor station, Aedes mosquitos were captured by suction tubes by district level Entomologist, Junior Entomologist, Field Assistant or insect collectors. At each outdoor station, Aedes mosquitos were collected by an aspirators for 10-15 minutes.

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ii.Mosquito collection from the following area

- Clustering of fever cases
- Dengue - Other Viral fever cases reported area
- Suspected fever death
- High vector Indices
- Dengue free blocks

iii. Frequency of Collection and Dispatch for Analysis

Collection were made on weekly basis and sent to State Public Health Lab (SPHL) and Institute of Vector Control & Zoonoses of this Directorate of Public Health for analysis.

iv. Procedure for the Collection

1. Dengue vector mosquitoes resting indoor such as dark corners, under tables etc. near breeding sources were collected between 8 am to 11 am and 3 pm to 5 pm and then transported to laboratory for identification and further processing.
2. In the laboratory, mosquitoes were segregated into males and females and the species identified and separated.
3. Only fed and gravid mosquitoes are needed to carry out antigen detection test. These mosquitoes were packed into pools of 25 each for testing the dengue antigen.
4. The desiccated mosquito pools were packed in individual Eppendroff tubes, wrapped in cotton to avoid breakage during transportation and packed in small cartoon boxes. Relevant details such as Date of Collection, Area of Collection including Name of Villages, Primary Health Centre and Block etc were enclosed.
5. Immature were also collected and kept for adult emergence. Hatched out adults were also used for detecting viral antigen. Hence the vector mosquitoes collected from the emergence were also sent for analysis by Reverse Transcriptase-Polymerase Chain Reaction (RTPCR) a standard testing for DENV detection described by Morita et al (1991).

v. Dengue hotspot

A hotspot can be defined as an area that has higher concentration of events compared to the expected number given a random distribution of events. In Dengue the hotspot is a place where there is a repetition of cases occurring for successive years.

RESULTS

In this study, we report the results of dengue virus detected in Aedes mosquitoes collected during 2023 with the help of RT-PCR .

The presence of dengue viraemia among the Aedes mosquitoes has provided important information on vector infection with dengue virus that helps in precise estimation of vector infection rate within a particular geographical area. An extensive study on virus detection in field collected Aedes mosquitoes was hence undertaken to measure the village-wise infection rate and to identify the potential risk areas of dengue infection in the respective HUDs of Tamilnadu. In the present study , a total of 13,220 Aedes mosquitoes pools were collected from all 46 health unit districts in Tamilnadu and subjected the mosquito pools to RT-PCR against DENV virus. Out of these 514 mosquitoes pools were found positive for dengue. The data were entered in MS Excel-2010 and statistical analysis was done to see if any Pearson's correlation (r) is calculated between mosquito pool positivity and dengue case incidence. Since r = 0.3443, (table -1 &2) there is a positive correlation between dengue positive and mosquito pool positive. i.e., if mosquito pool positivity increases dengue positivity will also increases. Vice-versa.

Table 1 : Dengue & mosquito pools Sample Taken & Positive (Man Vs Vector) Comparison Report - 2023

S. No	Name of the Health Unit District	Dengue cases		Mosquito Pool		
		Sample Taken	Positive	Mosquito pool Collected	Positive	Negative
1	Chennai Corp	63880	3121	253	21	232
2	Coimbatore	18330	2810	287	22	265
3	Tiruvallur	7145	2135	319	6	313
4	Madurai	19606	2119	637	17	620
5	Chengalpattu	27436	1851	348	15	333
6	Cuddalore	11185	1697	416	11	405
7	Kancheepuram	8172	1369	241	10	231
8	Tenkasi	3489	1074	287	11	276
9	Thanjavur	9243	1071	394	10	384
10	Poonamallee	3741	1042	180	20	160
11	Tirunelveli	2072	1034	340	14	326
12	Trichy	5577	1010	544	42	502
13	Tirupur	6041	871	558	9	549
14	Sivagangai	3825	855	332	11	321
15	Kanyakumari	13608	788	233	9	224
16	Vellore	73005	779	285	3	282
17	Tiruvannamalai	9876	759	313	5	308
18	Ramnad	5418	738	191	7	184
19	Aranthangi	6464	711	76	3	73
20	Namakkal	3794	634	411	16	395
21	Ranipet	6025	606	234	11	223
22	Perambalur	4225	581	196	3	193
23	Thoothukudi	2089	564	414	18	396
24	Salem	14918	560	591	35	556
25	Pudukottai	5171	556	281	16	265
26	Erode	3231	542	412	26	386
27	Paramakudi	2913	529	274	17	257
28	Dindigal	9904	523	250	7	243
29	Cheyyar	1962	491	318	9	309
30	Theni	3231	489	251	6	245
31	Krishnagiri	5235	481	206	3	203
32	Villupuram	6717	457	303	7	296
33	Ariyalur	6958	456	146	3	143
34	Tiruvarur	6925	394	283	12	271
35	Virudhunagar	2463	382	277	13	264
36	Kallakurichi	4708	362	200	3	197
37	Dharmapuri	3996	329	221	11	210
38	Mayiladuthurai	1518	324	156	2	154
39	Thirupathur	2672	316	315	10	305
40	Karur	5327	298	316	13	303
41	Sivakasi	1261	287	177	6	171
42	Palani	4955	215	180	2	178
43	Attur	1039	213	237	8	229
44	Kovilpatti	472	202	135	4	131
45	Nagapattinam	2487	169	168	6	162
46	Nilgiris	4709	116	32	1	31
	TOTAL	**417018**	**36910**	**13220**	**514**	**12706**

*36910 suspected cases from various sources.

Correlation coefficient, $r = \frac{\sum XY - \sum X \sum Y}{\sqrt{\sum X^2 - (\sum X)^2}\sqrt{\sum Y^2 - (\sum Y)^2}}$

$= \frac{(18971740) - (36910)(514)}{\sqrt{(1362348100) - (\sum 36910)^2}\sqrt{(264196) - (514)^2}}$

r = 0.344

Correlation coefficient between Mosquito pool positive and Dengue case positive,

Table-2 : Dengue Positive and Mosquito pool positive Report -2023

Sl. No	Name of the Health Unit District	Dengue Positive (X)	Mosquito Pool Positive (Y)	X^2	Y^2	XY
1	Chennai Corp	3121	21	9740641	441	65541
2	Coimbatore	2810	22	7896100	484	61820
3	Tiruvallur	2135	6	4558225	36	12810
4	Madurai	2119	17	4490161	289	36023
5	Chengalpattu	1851	15	3426201	225	27765
6	Cuddalore	1697	11	2879809	121	18667
7	Kancheepuram	1369	10	1874161	100	13690
8	Tenkasi	1074	11	1153476	121	11814
9	Thanjavur	1071	10	1147041	100	10710
10	Poonamallee	1042	20	1085764	400	20840
11	Tirunelveli	1034	14	1069156	196	14476
12	Trichy	1010	42	1020100	1764	42420
13	Tirupur	871	9	758641	81	7839
14	Sivagangai	855	11	731025	121	9405
15	Kanyakumari	788	9	620944	81	7092
16	Vellore	779	3	606841	9	2337
17	Tiruvannamalai	759	5	576081	25	3795
18	Ramnad	738	7	544644	49	5166
19	Aranthangi	711	3	505521	9	2133
20	Namakkal	634	16	401956	256	10144
21	Ranipet	606	11	367236	121	6666
22	Perambalur	581	3	337561	9	1743
23	Thoothukudi	564	18	318096	324	10152
24	Salem	560	35	313600	1225	19600
25	Pudukottai	556	16	309136	256	8896
26	Erode	542	26	293764	676	14092
27	Paramakudi	529	17	279841	289	8993
28	Dindigal	523	7	273529	49	3661
29	Cheyyar	491	9	241081	81	4419
30	Theni	489	6	239121	36	2934
31	Krishnagiri	481	3	231361	9	1443
32	Villupuram	457	7	208849	49	3199
33	Ariyalur	456	3	207936	9	1368
34	Tiruvarur	394	12	155236	144	4728
35	Virudhunagar	382	13	145924	169	4966
36	Kallakurichi	362	3	131044	9	1086
37	Dharmapuri	329	11	108241	121	3619
38	Mayiladuthurai	324	2	104976	4	648
39	Thirupathur	316	10	99856	100	3160
40	Karur	298	13	88804	169	3874
41	Sivakasi	287	6	82369	36	1722
42	Palani	215	2	46225	4	430
43	Attur	213	8	45369	64	1704
44	Kovilpatti	202	4	40804	16	808
45	Nagapattinam	169	6	28561	36	1014
46	Nilgiris	116	1	13456	1	116
	TOTAL	**36910**	**514**	**1362348100**	**264196**	**18971740**

r = 0.3443,

*36910 suspected cases from various sources.

DISCUSSIONS

Tamilnadu is the only state in conducting Aedes mosquito pool analysis against dengue control which is unique nowhere found in India. Dengue cases occurring every year in all the districts. Previously only few districts reported dengue, perhaps currently all the districts are reporting dengue cases every year and the infection has become a perennial problem. Tamilnadu lies with subtropical climatic conditions , partial south-west monsoon rain shower during June to August towards the southern districts and fully north-east rainfall every year starting in the month of Sep to December and occasionally with unusual rain during summer. The dengue cases incidence from 2012 to 2023 is given below in table-3. During 2017, a massive outbreak occurred 23,294 cases with 65 death reported in Tamilnadu. The present control strategy includes fever surveillance and mosquito control. These control activities are mainly carried out only after reporting of cases or epidemic in an area. In the absence of a safe and effective mass vaccination, the prevention and control of dengue outbreaks depends on the surveillance of cases and mosquito vectors (WHO, 1986; Khundsen and Sloff, 1992; Gubler and Clarke, 1994). Vector surveillance allows timely implementation of emergency mosquito control measures, such as insecticidal fogging of adult mosquitos and destruction of breeding places, to limit an impending outbreak. There is a need for a proper scientific tool to forecast dengue incidence. Hence, there is a need to opt for a new innovative tool to predict such incidence in future so as to prevent the occurrence of epidemic. So, detecting dengue virus in the field caught mosquitoes and initiating preventive measures in anticipation was required.

Tamil Nadu State –Map-1

Figure 1 : Tamil Nadu State –Map-1

Table 1 : Dengue case incidence in Tamil Nadu from 2012-2023

Year	Cases	Deaths
2012	13204	66
2013	6122	0
2014	2804	3
2015	4535	12
2016	2531	5
2017	**23294**	**65**
2018	4486	13
2019	8527	5
2020	2410	0
2021	6039	8
2022	6430	8
2023	9121	12

*9121 confirmed dengue cases

In this study, epidemiological parameters such as the dengue human cases incidence have been linked with spatial distribution of mosquito pools positive and negative status assay by RT-PCR depicted in GIS-Mapping of these positives, linked with the dengue cases incidence hamlet wise were done for the effective control and prevention activities against dengue. The positive villages comes under the districts which includes Coimbatore, Tiruppur, Erode, salem, Tenkasi, Tiruvellore, Madurai, Chengalpattu, Kancheepuram where mosquito positive pools as well as incidence of more number of human dengue cases reported during 2023 which clearly indicate that there is a linkage between mosquito pool and the occurrence of human dengue cases, even the mosquito pool positive as well as the reporting of human cases are more in the southern pockets as clearly shown in the GIS mapping as well the reporting of cases in table-2. If the mosquito pools positive shown more in an area in a district or in a hamlet villages, there is a need to tighten the control measures effectively in controlling the reporting of human cases and thus it becomes the effective forecasting mechanism in preventing the epidemic origin of cases in a particular period of time at a particular geographical locations as actually reported during the year 2017 in Tamilnadu.

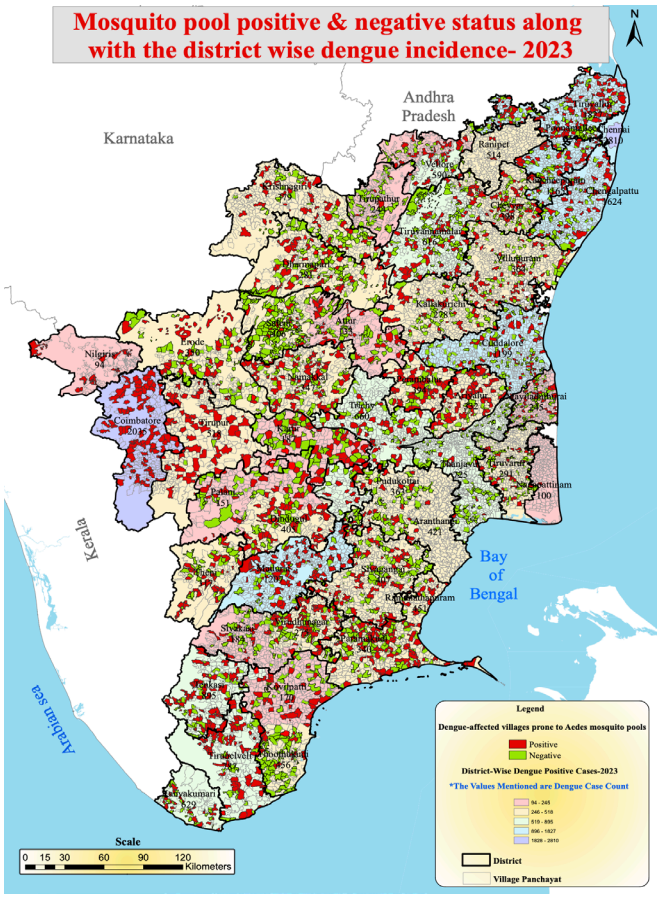

Figure 1 : Linking of Aedes Mosquito pool with case incidence in GIS

CONCLUSION

Based on the above field study and with the help of statistical analysis it seems that there is a linkage between mosquito pool positive with dengue case incidence as shown in the GIS map (1) It is clearly visible in high definition map with matching colours of case incidence range district wise for the year 2023 as shown in table-1 & Map 2.

CONFLICT OF INTEREST

There is no conflict of interest for this study.

RECOMMENDATIONS

Xenodiagnosis among Aedes mosquitoes with positive area-wise linking in GIS mapping should be taken into consideration for initiating vector control measures. The above measures can identify the impending outbreak at the earliest to initiate the Dengue prevention and control activities. It will significantly reduce the mortality and morbidity due to Dengue thereby reducing the burden of disease. Hence, it is highly suggested to adopt Xenodiagnosis and GIS mapping combinedly in all endemic states to prevent major outbreaks of Dengue in the future.

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