

## **MOSQUITO VECTOR INCRIMINATION STUDIES IN KERALA, INDIA: A REVIEW**

By

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### **Abstract**

*Kerala is home to six mosquito-borne diseases, viz., Lymphatic filariasis, Malaria, Japanese Encephalitis, Dengue, Chikungunya and West Nile Virus. Since, 1934 several vector incrimination studies have been done in different species of mosquitoes. Filarial parasites were detected from as many as 16 mosquito species, the important ones being Culex quinquefasciatus, Mansonia annularia and Mansonia uniformis. Malaria parasites were detected from Anopheles culicifacies, An. fluviatilis, An. jeyporiensis and An. varuna. Japanese Encephalitis virus was detected from Culex tritaeniorhynchus, Cx. gelidus, Mansonia uniformis, Ma. indiana, Ma.annulifera and Anopheles subpictus. Both dengue and chikungunya viruses were detected from Aedes albopictus. West Nile Virus has not been detected from any mosquito species so far.*

**Keywords:** *mosquito, vector, incrimination, lymphatic filariasis, dengue, chikungunya, virus.*

### **Introduction**

Mosquito borne diseases are a major public health problem in Kerala, the south Indian state of India. Six mosquito borne diseases viz., Malaria, Lymphatic Filariasis, Dengue, Chikungunya, Japanese Encephalitis and West Nile Virus are prevalent in the state. The state had been haunted by malaria in its highlands and lymphatic filariasis in the coastal belt from pre-historic times. The prevalence of sickle cell anemia among the tribes of Wayanad and Attappadi is

a solid proof for the antiquity of malaria in the state (Kaur et al, 1997 and Feroze & Aravindan, 2001). In the pre-independent era, malaria was a major disease burden causing significant rates of morbidity and mortality in the state (Covell & Harbhagwan, 1939). Documentary evidence for the presence of Lymphatic Filariasis in Kerala goes back to 1709 when Clarke called elephantiasis-legs in Cochin as Malabar legs (Raghavan, 1957). Currently the state is endemic to bancroftian and brugian forms of

lymphatic filariasis and ranks second in India in terms of endemicity. 15.7% of the total cases are reported from the state (Agarwal and Sashindran, 2006).

Japanese encephalitis staged its first appearance in the form of sporadic outbreaks in 1996 in Kottayam and Alappuzha districts (Dhanda et al; 1997 and John, 2006). The first outbreak of dengue was reported from Kottayam district in 1997 with 14 cases and 4 deaths, which was followed by a bigger outbreak with 67 cases and 13 deaths in 1998 in the same district. However, antibodies of dengue viruses were detected from human sera collected from various districts in the state as early as 1973 (Banerjee and Desai, 1973). The first outbreak of Chikungunya in the state was during June-July 2006 along the coastal areas of Alleppey, Quilon, and Trivandrum districts (Manju and Sushamabai, 2009, Kannan et al, 2009). Finally, West Nile Virus was reported for the first time in 2011 (Anukumar et al., 2014).

All mosquitoes are not disease vectors. One of the broadest definitions of a vector is any organism (vertebrate or invertebrate) that functions as a

carrier of an infectious agent between organisms of a different species (Kuno and Chang, 2005). Vector incrimination of mosquito-borne diseases is the identification of a mosquito species involved in the transmission of a disease in nature. It is done primarily by the detection of the disease organisms in a mosquito species. However, if such mosquitoes do not bite humans or are not prevalent in sufficient densities, they need not necessarily be vectors (Scherer et al., 1971). Incrimination of vectors is an important and significant step in the direction of control of any mosquito-borne disease. The discoveries of vectors of Lymphatic Filariasis, Malaria and Yellow Fever by Manson (1878), Ross (1897) and Reed et al. (1901) respectively, helped in controlling these diseases in different parts of the world.

The present paper is intended to compile the vector incrimination studies on mosquitoes in Kerala. The studies are discussed under the respective diseases.

### **Lymphatic Filariasis**

In Kerala, two species of nematode parasites viz., *Wuchereria bancrofti* and *Brugia malayi* cause Lymphatic Filariasis. Between July 1912 and June

1913 Cruickshank and Wright (1914) visited Cochin and did experiments to find out the development of filarial worms in mosquitoes. They experimented with *Culex fatigans* (*Culex quinquefasciatus*), *Cx. sitiens* and *Anopheles rossii* (*An. stephensi*) and *Stegomyia scutellaris*. Of these they found *Culex fatigans* to be a very effective host. The remaining mosquitoes were also found to support the development of *Filaria* nematode to some extent. They used patients of *Wuchereria bancrofti* (then *Filaria bancrofti*) to feed the mosquitoes.

While investigating filariasis in all the endemic districts of Kerala, Iyengar (1938) dissected 75 species of mosquitoes for the presence of the nematodes and found 16 species naturally infected with either *Wuchereria bancrofti* or *Brugia malayi*. In areas where *Wuchereria bancrofti* was prevalent, the species found infected were *Culex fatigans* (*Culex quinquefasciatus*), *Cx. gelidus*, *Cx. vishnui*, *Cx. sitiens*, *Cx. bitaeniorhynchus*, *Anopheles subpictus*, *An. vagus* and *An. varuna*. However, full-grown larvae were found mainly in *Cx. fatigans*. In areas where *Brugia malayi* was prevalent, *Mansonia* (*Mansonioides*) *annulifera*, *Ma. (M.)*

*uniformis*, *Ma. (M.) indiana*, *Armigeres obturbans* (*Ar. subalbatus*), *Cx. gelidus*, *Cx. vishnui*, *Cx. sitiens*, *Cx. bitaeniorhynchus*, *Lutzia fusca*, *Culex pallidothorax*, *Anopheles barbirostris*, *An. hyrcanus* var. *nigerrimus*, *An. subpictus* and *An. varuna* showed appreciable infection. The rate of infection in the first two were 19.2 and 6.5% respectively; hence considered as major vectors.

Raghavan (1957) compiled the data on the incrimination studies till 1957. He reported the results of dissection communicated by Director of Public Health services, Madras in 1951. Accordingly, *Wuchereria bancrofti* was detected from *Culex fatigans* (*Culex quinquefasciatus*) from Malabar.

### Malaria

Malaria is caused by two species of *Plasmodium* viz., *Plasmodium vivax* and *Plasmodium falciparum* in Kerala. Covell (1931) compiled the data on vector incrimination studies all over the world including India till then. However, no such studies were conducted from regions which now come under Kerala state, though many areas of Kerala were highly endemic to Malaria. Sporozoites from salivary glands and oocysts from guts of *Anopheles* mosquitoes were recorded

for the first time in Kerala by Iyengar in 1934. Examination of mosquitoes caught in villages in an intensely malarious region of Travancore from February 1932 to September 1933 revealed malaria parasites in *Anopheles jeyporiensis* (3 gut infections among 1988 examples dissected), *An. varuna* (1 gut infection among 59), *An. fluviatilis* (1 salivary gland and 4 gut infections among 132) and *An. culicifacies* (1 gland infection among 984).

Mathew (1939) also reported the presence of sporozoites and oocysts in *An. fluviatilis* (Out of 2602 dissected 23.7% had oocysts and 13% had sporozoites) *An. varuna* (Out of 429 dissected 2.3% had oocysts and 1.6% had sporozoites), and *An. culicifacies* (Out of 1131 dissected 0.5% had oocysts and 0.3% had sporozoites).

During an investigation on malaria by Covell and Singh (1939) between January 1938 and May 1939 in Wayanad, 11, 930 Anophelines mosquitoes were dissected and four *Anopheles fluviatilis* were found infected with malaria parasites.

### Japanese Encephalitis (JE)

As discussed earlier, Japanese Encephalitis is a recently emerged

disease in Kerala. In 1996 when the first outbreak of Japanese Encephalitis occurred in the southern districts of Kerala, Dhanda et al. (1996) investigated the presence of its virus in mosquitoes. Mosquito collections were done between 23-25 February; 15-17 March and 10-11 April 1996 in Kavalam and Pallimukku villages of Alleppey district. A total of 5357 mosquitoes were collected in 163 pools. The presence of virus was detected by using antigen-capturing ELISA and an insect bioassay. 12 Japanese Encephalitis viral isolates were detected. Out of these 7 were from *Culex tritaeniorhynchus*; 3 from *Mansonia uniformis* and 1 each from *Ma. indiana* and *Anopheles subpictus*.

During 1999-2000, Arunachalam et al (2004) collected 146560 mosquitoes under 5 genera and 18 species from Kuttanad area of Alappuzha district in Kerala. These mosquitoes were pooled into 3374 pools. Presence of JE virus detected by two procedures; antigen capture ELISA and Insect Bioassay involving *Toxorhynchus splendens* (Toxo-IFA). Viral infection was detected by ELISA in 64 pools of *Culex tritaeniorhynchus*, 10 pools of *Culex gelidus*, 12 pools of *Mansonia indiana*; 5 pools of *Ma. uniformis* and 3 pools of *Ma. annulifera*. However, viral

presence could be confirmed using Toxo-IFA only in *Cx. tritaeniorhynchus* and *Mansoniaindiana*. Presence of virus was detected in *Cx. tritaeniorhynchus* in the months of January to April and November in 1999 and January to March and September to December in 2000. In the case of *Mansoniaindiana* it was in May, July and August in 1999 and May, August and October in 2000.

Based on the above two studies it can be concluded that *Cx. tritaeniorhynchus* and *Ma.indiana* are the major vectors of Japanese Encephalitis in Kerala.

### Dengue

Although the first outbreak of dengue in the state was reported in 1997, the first isolation of its virus from mosquitoes was successful only in 2004 (Dhanda et al., 2004). They had collected adult *Aedes albopictus* mosquitoes from a residential area near Calicut airport in north Kerala. Dengue virus was detected from one of the pools of 10 mosquitoes.

Subsequently, in 2007, Thenmozhi et al reported the presence of dengue virus in male and female *Aedes albopictus* adults collected in the wild and females emerged from larvae. The

collections were made from March 2002 to June 2005. Out of a total of 1,445 field collected *Ae. albopictus* male mosquitoes in 101 pools and 1,817 field-collected *Ae. albopictus* female mosquitoes in 160 pools screened for dengue virus antigen by ELISA, 1 and 4 of the male and female field-collected adult pools were found to be positive for dengue virus antigen, respectively. From March 2002 to August 2003, out of 77 pools (1,472 specimens) of reared males and 76 pools (1,485 specimens) of reared females screened, only 1 pool of female was found positive. The results of this study also supported the hypothesis of transovarial transmission of dengue virus.

### Chikungunya (CHIK)

Though Chikungunya started appearing in Kerala since 2006, incrimination of a vector was done only in 2009 (Niyas et al. 2010). CHIKV was detected in *Aedes albopictus* collected from Olavanna, Chaliyam and Bepore villages of Kozhikode district in North Kerala from May to September 2009. For virus detection in mosquitoes, households of CHIK patients, whose serum samples were confirmed in the laboratory by RT-PCR, were subsequently visited and

larval sampling was done from May to September 2009 (Niyas et al. 2010). Stagnant water collected in discarded articles like coconut shells, broken earthen-wares, plastic bottles and damaged drains were searched for *Ae. albopictus* larvae. Third and fourth instar larvae and pupae were phenotypically known in place victimisation normal keys and these were collected and transferred to containers with fresh water. Four households each in Olavanna and Chaliyem, and three households in Beypore were surveyed. Larvae and pupae collected from every location were created into a single pool. All the three pools turned out to be positive for CHIKV. Earlier, Kumar et al (2008) reported the A226V mutation in the glycoprotein envelope 1 (E1) gene of the virus among isolates collected from the three worst-affected districts of the state. This mutation had already been steered to be directly accountable for a big increase in CHIKV infectivity in *Aedes albopictus*. This mutation was detected in the CHIKV virus isolated in the above study. The presence of viruses in the adults reared from the larvae indicated the possibility of

transovarial transmission of the virus similar to dengue virus.

### **West Nile Virus (WNV)**

As discussed earlier West Nile Virus was reported from Kerala for the first time in 2014. However, this virus has not been detected from any species of mosquitoes in Kerala so far. However, WNV was isolated from *Culex vishnui* and *Cx. quinquefasciatus* collected from Tamil Nadu and Andhra Pradesh. Besides, experimental infection was observed in *Cx. tritaeniorhynchus*, *Cx. Bitaenior hynchus* and *Cx. univittatus*. In Kerala it is not yet clear which species is the vector of WNV. This information is vital in tackling future outbreaks of this virus in the disease.

### **Conclusion**

Kerala has a long history of mosquito-borne diseases. However, vector incrimination studies in the state are scanty and unsatisfactory. In the case of lymphatic filariasis and malaria there were no studies after independence. Incrimination studies related to dengue and chikungunya are centred on a single species, viz., *Aedes albopictus*. There is no data related to *Aedes aegypti*, the primary

vector of both the diseases elsewhere in the world and also *Aedes vittatus* which is a suspected vector of Dengue. The status of incrimination studies related to Japanese Encephalitis is also not better. In recent years Japanese Encephalitis has started to emerge in northern districts of the state but there have been no incrimination studies. In the case of West Nile Virus which is also an emerging disease in

some parts of north Kerala, there is no information on the vectors involved. The present review has amply exposed the gaping lacunae in this vital knowledge repository.

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