

Larval Occurrence and Climatic Factors Affecting DHF Incidence in Samui Islands, Thailand

S. Wongkoon, M. Jaroensutasinee, K. Jaroensutasinee, W. Preechaporn, and S. Chumkiew

Abstract—This study investigated the number of *Aedes* larvae, the key breeding sites of *Aedes* sp., and the relationship between climatic factors and the incidence of DHF in Samui Islands. We conducted our questionnaire and larval surveys from randomly selected 105 households in Samui Islands in July-September 2006. Pearson's correlation coefficient was used to explore the primary association between the DHF incidence and all climatic factors. Multiple stepwise regression technique was then used to fit the statistical model. The results showed that the positive indoor containers were small jars, cement tanks, and plastic tanks. The positive outdoor containers were small jars, cement tanks, plastic tanks, used cans, tires, plastic bottles, discarded objects, pot saucers, plant pots, and areca husks. All *Ae. albopictus* larval indices (i.e., CI, HI, and BI) were higher than *Ae. aegypti* larval indices in this area. These larval indices were higher than WHO standard. This indicated a high risk of DHF transmission at Samui Islands. The multiple stepwise regression model was $y = -288.80 + 11.024x_{\text{mean temp}}$. The mean temperature was positively associated with the DHF incidence in this area.

Keywords—Dengue vectors, *Aedes aegypti*, *Aedes albopictus*, Container Index, House Index, Breteau Index, *Aedes* indices, Climatic factors, Temperature.

I. INTRODUCTION

DENGUE fever is caused by dengue viruses of the family Flaviviridae, transmitted principally by *Aedes aegypti* and possible *Ae. albopictus* in the tropical and subtropical regions of the world [1], [2]. These two clinical features namely Dengue Haemorrhagic Fever (DHF) and Dengue

Shock Syndrome (DSS), if not properly managed, may lead to death [3], [4]. No effective vaccine or chemotherapy is currently available for the prevention or treatment of dengue fever; therefore, prevention and control of the disease depend on vector surveillance and control measures [5]. Transmission cycles of dengue virus depend on the interrelationship between the virus and its mosquito vector, which is influenced by environmental conditions [6]. Adult female *Aedes* mosquitoes acquire the dengue virus by biting infected humans during the viremic phase, which usually lasts for 4–5 days, but it may last up to 12 days. The virus is transmitted to other persons via bites from infected mosquitoes [7]. The mosquitoes that adversely affect people in Southern Thailand are primarily *Ae. aegypti* L. and *Ae. albopictus* Skuse [8]–[12].

An epidemic of DHF occurred in Samui Islands in 1966 and 1967 [13] where *Ae. aegypti* and *Ae. albopictus* were abundant, and responsible for transmission of dengue virus [14]–[16]. *Ae. aegypti* breeds in a wide assortment of domestic containers, whereas *Ae. albopictus* is more likely to be found in natural containers, such as bamboo stumps and coconut shells, or in artificial containers outside the houses such as tires, opened cans, and plastic bottles [17]. In Thailand, *Ae. albopictus* has been found in forested habitats ranging in elevation from 450 to 1,800 m as well as in a variety of other habitats in rural and suburban areas [8], [18]–[20]. Since most Thai households store water for cooking and bathing in a variety of jars and cisterns, *Ae. aegypti* is more important threat for DHF. *Ae. aegypti* feeds more readily on humans than does *Ae. albopictus* [7], [21]. *Ae. albopictus* is encountered in the peripheral areas of towns where it replaces *Ae. aegypti* populations [22].

The transmission of dengue viruses is climatic sensitive for several reasons. First, temperature changes affect vector-borne disease transmission and epidemic potential by altering the vector's reproductive rate, biting rate, the extrinsic incubation period of the pathogen, by shifting a vector's geographical range or distribution and increasing or decreasing vector-pathogen-host interaction and thereby affecting host susceptibility [23]. Second, precipitation affects adult female mosquito density. An increase in the amount of rainfall leads to an increase in available breeding sites which, in turn, leads to an increase in the number of mosquitoes. An increase in the number of adult female mosquitoes increases the odds of a mosquito obtaining a pathogen and transmitting it to a second

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sensitive host [24]. Third, a distinct seasonal pattern in DHF outbreaks is evident in most places. In tropical regions where monsoon weather patterns predominate, DHF hospitalization rates increase during the rainy season and decrease several months after the cessation of the rains [25], [26]. The impact of climatic factors on DHF in Thailand is probably the least understood [27]–[30].

Although there are measurements to control dengue vector every months around Samui Islands [31], dengue vector is a major problem in Samui Islands. In 2002, DHF rate in Samui Islands is 1621.66 cases per 100,000 people and the fatality rate was 7.72 (630 DHF cases reported from 38,849 populations). In 2005, DHF incidence rate in Samui Islands is 790.79 cases per 100,000 people and the fatality rate was 0. In 2006, DHF incidence rate is 111.41 cases per 100,000 people and the fatality rate was 2.18 [31].

In the past two decades, there has been a dramatic increase in the development of infrastructure, accommodations, and facilities for tourism purposes, such as hotels, resorts or bungalows, and associated services as well as residential units in various areas around the island. It is believed that these developments have had an impact on the abundance of *Aedes* mosquitoes by providing more habitats for these mosquitoes and thus leading to an increase in the abundance of dengue vectors [10]. This study investigated the number of mosquito larvae (i.e., *Ae. aegypti* and *Ae. albopictus*), the major sources of larval breeding sites indoor and outdoor containers, and the relationship between climatic factors and the incidence of DHF in Samui Islands.

II. MATERIALS AND METHODS

A. Study Site

Samui Islands is the largest of a group of several dozen islands in the Gulf of Thailand, located at 9° 32' 29.69" N latitude and 99° 56' 42.48" E longitude. Samui Islands is one of the districts of the Surat Thani province in southern Thailand, with an area of 227.250 km². Fifty four % of Samui Islands was covered with mountains. The island is divided into seven administrative sub-districts, consisting of 39 villages with a local population of 45,777 and a density of 201.44 people/km². Normally each year, two tropical monsoons (i.e., southwest and northeast monsoon) dominate the climate of Samui Islands. The onset of the first monsoon starts in May whereas that of the second begins in November. As a result of these monsoons, the annual average rainfall for Samui Islands is over 1000 mm each year [32].

B. Data Collection

105 representative households at Samui Islands were selected randomly to carry out the questionnaire and larval surveys in July–September 2006 (Fig. 1).

C. Entomological Studies

Larval surveys were conducted in the study areas using fishnets. Very small containers were emptied through the

fishnet. Larger containers were sampled by dipping the net in the water, starting at the top of the container and continuing to the bottom in a swirling motion that sampled all edges of the container [33]. Mosquito larvae were placed in plastic bags and transported to the laboratory for identification up to species level using Rattanaarithikul and Panthusiri's [34] keys.

Three larval indices: House Index (HI), Container Index (CI), and Breteau Index (BI) were worked out as per standard WHO guidelines. Breeding places were sampled both indoors and outdoors within 15 m of the houses [10], [35]. The jars were classified into two categories: small jars (<100 L) and large jars (≥100 L) [36].

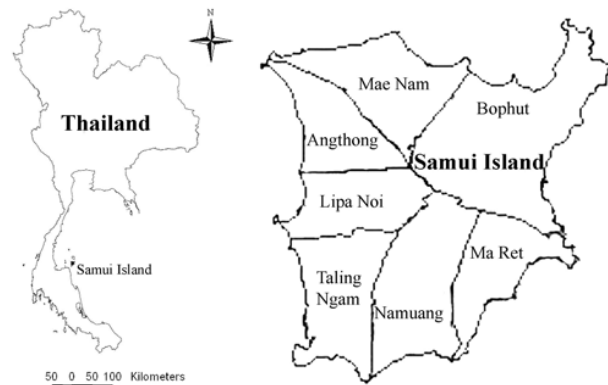


Fig. 1 Location of study area on the Samui Islands

D. Climatic Factors on DHF incidence in Samui Islands

Climatic data of Samui Islands from 1999–2006 were provided by the Climatology Division of Thailand Meteorological Department. The monthly DHF data over the same period were collected by the Center of Epidemiological Information at Samui Islands, Ministry of Public Health. Monthly climatic data was comprised of 11 factors: the amount of rainfall, the number of rainy days, daily max rainfall, mean pressure, mean/max/min relative humidity, mean/max/min temperature, and max wind speed.

E. Statistical Analysis

Descriptive statistics of the data were analyzed. The number of *Ae. aegypti* and *Ae. albopictus* larvae in different types of water containers both indoor and outdoor containers were analyzed using independent sample *t*-tests. Pearson's correlation coefficient test was employed to detect primary association between DHF incidence and climatic factors. The significantly variables correlative of the DHF incidence were then submitted to multiple stepwise regression analysis.

III. RESULTS AND DISCUSSIONS

A. Positive Containers and *Aedes* larvae

We collected the total of 8 types of indoor containers and 16 types of outdoor containers. Three types of indoor containers (i.e., small jars, cement tanks, and plastic tanks) were infested with *Aedes* larvae and 10 types of outdoor

containers (i.e., small jars, cement tanks, plastic tanks, used cans, tires, plastic bottles, discarded objects, pot saucers, plant pots, and areca husks) were infested with *Aedes* larvae (Table I).

A total of 746 containers from 105 houses in Samui Islands were inspected for *Aedes* larvae. Of these containers, a total of 36 containers situated in and around 15 houses were infested with *Ae. aegypti* larvae. A total of 53 containers situated in and around 24 houses were infested with *Ae. albopictus* larvae. The average number of all types of containers positive for *Aedes* larvae found per house was ranging from 0.01–0.17 containers. A total of 1,139 mosquito larvae were collected from all containers. 147 mosquito larvae were *Ae. aegypti* and 759 mosquito larvae were *Ae. albopictus*. Thavara *et al.* [10] showed that the average number of all types of containers positive for *Aedes* larvae found per house in 1996, 1997, and 1998 was 1.6, 2.3, and 3.0, respectively. Previous study [37] found a greater number of *Aedes* larvae than this study. These results show a decrease in the number of positive containers per household and the number *Aedes* larvae. This could be due to the fact that there is a strong campaign on vector control in Samui Islands since 2005. The Center of Epidemiological Information at Samui Islands, Ministry of Public Health sprayed insecticide monthly in all sub-districts in Samui Islands.

TABLE I
THE MEAN (\pm S.D.) NUMBER OF CONTAINERS AND POSITIVE CONTAINERS PER HOUSEHOLD AT SAMUI ISLANDS

| Container type | No. of Container | No. of Positive Container |
|------------------------|------------------|---------------------------|
| Indoor containers | | |
| Small jars | 0.07 \pm 0.29 | 0.02 \pm 0.14 |
| Large jars | 0.02 \pm 0.14 | 0.00 \pm 0.00 |
| Cement tanks | 0.33 \pm 0.58 | 0.08 \pm 0.33 |
| Plastic tanks | 0.70 \pm 0.86 | 0.10 \pm 0.34 |
| Ant guards | 0.09 \pm 0.44 | 0.00 \pm 0.00 |
| Flower vases | 0.08 \pm 0.43 | 0.00 \pm 0.00 |
| Pludang bottles | 0.01 \pm 0.10 | 0.00 \pm 0.00 |
| Refrigerator drainages | 0.03 \pm 0.17 | 0.00 \pm 0.00 |
| Outdoor containers | | |
| Small jars | 0.62 \pm 1.27 | 0.17 \pm 0.66 |
| Large jars | 0.27 \pm 0.71 | 0.00 \pm 0.00 |
| Cement tanks | 0.18 \pm 0.52 | 0.02 \pm 0.14 |
| Plastic tanks | 0.56 \pm 0.99 | 0.07 \pm 0.25 |
| Used cans | 0.34 \pm 1.36 | 0.02 \pm 0.14 |
| Tires | 0.29 \pm 0.97 | 0.01 \pm 0.10 |
| Plastic bottles | 0.21 \pm 0.82 | 0.01 \pm 0.10 |
| Discarded objects | 0.29 \pm 0.68 | 0.05 \pm 0.21 |
| Pot saucers | 0.10 \pm 0.54 | 0.01 \pm 0.10 |
| Plant pots | 0.32 \pm 1.86 | 0.02 \pm 0.14 |
| Animal pans | 0.12 \pm 0.39 | 0.00 \pm 0.00 |
| Areca husks | 0.15 \pm 0.82 | 0.01 \pm 0.10 |
| Banana clumps | 0.47 \pm 1.80 | 0.00 \pm 0.00 |
| Coconut shells | 1.21 \pm 7.33 | 0.00 \pm 0.00 |
| Tree holes | 0.10 \pm 0.56 | 0.00 \pm 0.00 |
| Bamboo clumps | 0.10 \pm 0.55 | 0.00 \pm 0.00 |

The result shows that, *Ae. albopictus* was not found in small jars and areca husks. *Ae. aegypti* was not found in cement tanks, used cans, tires, plastic bottles, and pot saucers. However, there were not significantly different between *Ae. albopictus* and *Ae. aegypti* in these containers (Table II).

Most number of *Ae. aegypti* larvae were found in the outdoor containers and *Ae. albopictus* larvae were found most in the indoor containers. The number of *Ae. aegypti* and *Ae. albopictus* larvae were not significantly different between indoor and outdoor containers (*Ae. aegypti*: $t_{326} = -0.658$, *ns*; *Ae. albopictus*: $t_{326} = 1.379$, *ns*). The results confirm the previous finding [38] that *Ae. aegypti* larvae were found most in the outdoor containers and *Ae. albopictus* larvae were found most in the indoor containers. It is because the houses in Samui Islands were located in and around coconut plantations. Therefore, *Ae. albopictus* was very likely to be able to lay eggs inside the houses.

TABLE II
THE MEAN (\pm S.D.) NUMBER OF *AE. AEGYPTI* AND *AE. ALBOPICTUS* INDOOR AND OUTDOOR CONTAINERS AT SAMUI ISLANDS

| Container type | <i>Ae. aegypti</i> | <i>Ae. albopictus</i> | <i>t</i> -test |
|--------------------|--------------------|-----------------------|-------------------|
| Indoor containers | | | |
| Small jars | 0.17 \pm 0.41 | 0.00 \pm 0.00 | $t_5 = 1.000$ |
| Cement tanks | 0.59 \pm 2.61 | 15.24 \pm 54.79 | $t_{28} = -1.439$ |
| Plastic tanks | 0.27 \pm 1.44 | 1.69 \pm 7.92 | $t_{54} = -1.275$ |
| Outdoor containers | | | |
| Small jars | 0.54 \pm 1.77 | 1.97 \pm 7.18 | $t_{38} = -1.143$ |
| Cement tanks | 0.00 \pm 0.00 | 0.21 \pm 0.80 | $t_{13} = -1.000$ |
| Plastic tanks | 1.24 \pm 4.34 | 2.79 \pm 14.81 | $t_{64} = -0.575$ |
| Used cans | 0.00 \pm 0.00 | 1.07 \pm 4.01 | $t_{13} = -1.000$ |
| Tires | 0.00 \pm 0.00 | 0.07 \pm 0.26 | $t_{14} = -1.000$ |
| Plastic bottles | 0.00 \pm 0.00 | 1.10 \pm 3.48 | $t_9 = -1.000$ |
| Discarded objects | 1.32 \pm 6.18 | 0.82 \pm 1.82 | $t_{42} = 0.364$ |
| Pot saucers | 0.00 \pm 0.00 | 2.00 \pm 5.29 | $t_6 = -1.000$ |
| Plant pots | 0.64 \pm 2.11 | 0.55 \pm 1.51 | $t_{20} = 0.116$ |
| Areca husks | 3.17 \pm 7.76 | 0.00 \pm 0.00 | $t_5 = 1.000$ |

The number of *Ae. aegypti* and *Ae. albopictus* larvae were not significantly different between natural and artificial containers (*Ae. aegypti*: $t_{326} = 0.192$, *ns*; *Ae. albopictus*: $t_{326} = -0.832$, *ns*). However, *Ae. aegypti* and *Ae. albopictus* tended to be found most in artificial containers. This could be because Samui Islands is currently overdeveloped with new housing and hotel projects. Lots of artificial containers were highly available in the area at the construction sites and became a major source of breeding sites for mosquitoes.

Ae. aegypti prefers to lay eggs in different containers than *Ae. albopictus* [12], [35]. Phong and Nam [39] studied *Aedes* larval occurrence in Vietnam and found that *Ae. aegypti* and *Ae. albopictus* larvae were mostly found in artificial containers. *Ae. aegypti* larvae were found in drums, jars, concrete tanks, and discarded objects. On the other hand, *Ae. albopictus* larvae were mainly found in jars, discarded objects, drums, and aquariums. Wongkoon *et al.* [12] studied *Aedes* larval occurrence in Nakhon Si Thammarat, Thailand and found *Ae. aegypti* and *Ae. albopictus* larvae in six water storage containers including plant pots, animal pans, tires, small jars, water containers in bathroom, and concrete tanks. They found that from these six containers, there were a higher number of *Ae. aegypti* larvae in artificial containers (i.e., water containers in bathrooms and concrete tanks) than *Ae. albopictus* [12]. Our results supported previous findings and showed that key breeding sites of *Ae. aegypti* were outdoor areca husks and the key breeding site of *Ae. albopictus* were indoor cement tanks.

B. Larval Indices

Aedes larval indices at Samui Islands were shown in Table III. The National Institute of Communicable Diseases [3] defined a high risk of DHF transmission when BI was ≥ 50 , HI was ≥ 10 , and a low risk of transmission when BI was ≤ 5 , HI was ≤ 1 . All larval indices from our study indicated a high risk of DHF transmission. For *Ae. aegypti* larval indices, the House Index (HI) was 14.29%, the Container Index (CI) was 4.83%, and the Breteau Index (BI) was 34.29 infected containers per 100 households. For *Ae. albopictus* larval indices, HI was 22.86%, CI was 7.11%, and BI was 50.48 infected containers per 100 households.

TABLE III
LARVAL ABUNDANCE INDICES OF *AE. AEGYPTI* AND *AE. ALBOPICTUS* IN SAMUI ISLANDS

| Larval Indices | <i>Ae. aegypti</i> | <i>Ae. albopictus</i> |
|----------------------|--------------------|-----------------------|
| Container Index (CI) | 4.83 | 7.11 |
| House Index (HI) | 14.29 | 22.86 |
| Breteau Index (BI) | 34.29 | 50.48 |

All *Ae. albopictus* larval indices (i.e., CI, HI, and BI) were higher than *Ae. aegypti* larval indices in this area. Previous studies [9], [37] also found more *Ae. albopictus* larvae than *Ae. aegypti*. However, after checking for dengue virus by Dig-cDNA probe, it was found that *Ae. aegypti* had a higher percentage of dengue infection than *Ae. albopictus*. This implies that even though there are a higher number of *Ae. albopictus* larvae present, there might not be a high DHF transmission due to a lower susceptibility to dengue virus in *Ae. albopictus*.

C. Climatic Factors on DHF Incidence in Samui Islands

The relationship between the mean temperature and the transmission of DHF at Samui Islands between 1999–2006 were shown in Fig. 2 and Fig. 3. The multiple stepwise regression model was $y = -288.803 + 11.024x_{mean\ temp}$ ($R^2 = 0.166$, $F_{1,76} = 15.077$, $P < 0.001$). Promprou *et al.* [40] studied climatic factors affecting DHF incidence in Southern Thailand and found that the significant variables were minimum temperature, the number of rainy days, and relative humidity on the Gulf of Thailand side. The selected regression model was $y = 0.072x_{min\ temp} + 0.015x_{rain} - 0.017x_{relative\ hum}$ ($R^2 = 0.34$, $F_{3,838} = 144.85$, $P < 0.001$).

Warmer temperature can increase the transmission rates of DHF in various ways. First, warmer temperature may allow vectors to survive and reach maturity much faster than at lower temperature [41]. Secondly, warmer temperature may reduce the size of mosquito larvae resulting in smaller adults that have high metabolism rates, require more frequent blood meal, and need to lay eggs more often [28], [42], [43]. Thirdly, environmental temperature has a marked effect on the length and efficiency of the extrinsic incubation periods (EIPs) of arboviruses in their vectors [41], [43]. This means that mosquitoes exposed to higher temperature after ingestion of virus become infectious more rapidly than mosquitoes of the same species which are exposed to lower temperatures [41]. Therefore, the transmission of arboviruses may increase

under warmer conditions as more vector mosquitoes become infectious within their life-span. Higher temperature may reduce the length of viral extrinsic incubation periods (EIPs) in mosquitoes [44]–[46]. At 30 °C, the duration of dengue virus EIPs is 12 days, compared with only 7 days at 32–35 °C [27]. Moreover, a 5-day decrease in the duration of the incubation period can triple the transmission rate of dengue [47]. It was found in this study that the mean temperatures were positively associated with the transmission of DHF at Samui Islands. As the mean temperature increased, the DHF cases also increased. It is possible that most of the physiological functions of vectors in this area are subject to optimal mean temperature.

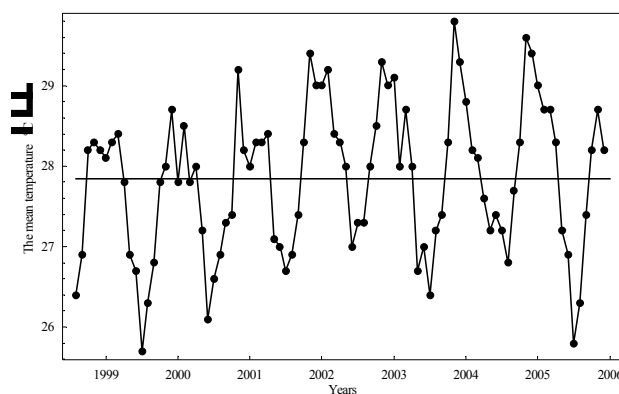


Fig. 2 The mean temperature at Samui Islands from 1999–2006

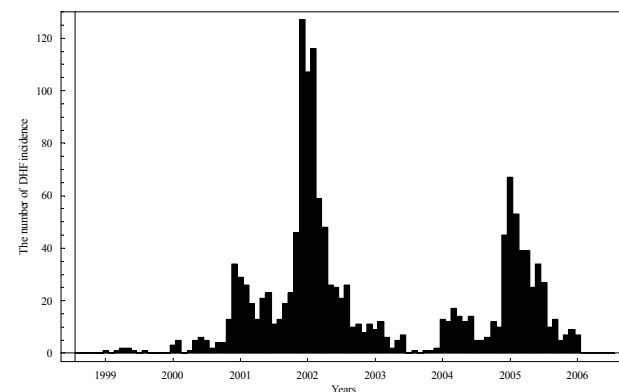


Fig. 3 The number of monthly DHF incidences at Samui Islands from 1999–2006

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