Effects of Breeding of Mosquito Larvae on High and Low Tourist Temples in Krabi, Thailand using GLOBE Observer Habitat Mapping App.

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Abstract

This study investigated (1) How breeding sites, diversity, and abundance affect high and low tourist temples, (2) mosquito species numbers at high and low tourist temples, and (3) correlation between water pH, temperature and mosquito larvae numbers. We examined the number of *Aedes aegypti*, *Ae. albopictus*, *Anopheles*, and *Toxorhynchites* larvae differed in various types of water containers. Water containers were categorized into the following groups: indoor/outdoor containers, artificial/natural containers, earthen/plastic containers, and containers with/without lids. Samples were collected from a total of 6 temples from 3 high tourist temples and 3 low tourist temples using GLOBE Observer: Mosquito Habitat Mapper App. Our results showed that *Aedes* larvae were found in 682 out of 151 water containers in high and low tourist temples. *Aedes* females laid eggs in different container types depending on temple based communities. *Aedes* larvae were found in highest numbers in plastic buckets in both communities. The number of positive containers was higher in outdoor containers than indoor containers, artificial containers than natural containers, plastic containers than earthen containers, container without lid than container with lid, and higher tourist temples than low tourist temples. Our results showed the number of tourists and others affected by the temple.

Keywords: Water containers, *Aedes* species**,** water qualities, mosquito larval index

Introduction

Dengue fever is caused by dengue viruses of the family Flaviviridae, transmitted principally by *Aedes aegypti*, and *Ae. albopictus*, in the tropical and subtropical regions of the world. WHO (2017) reported that 70% of dengue infected persons were from South East Asia and Western Pacific. Dengue has become a global threat and spreads its authority in urban and semi urban areas with the tropical and the subtropical climates in many countries. Accurate mapping of the spatial distribution of mosquito breeding habitats is essential for cost-effective deployment of control practices.

Mosquito-borne illnesses such as dengue and Zika are spread primarily through the bite of infected female Aedes mosquitoes. Dengue is one of the most significant mosquito-borne illnesses in the tropics and subtropics. More than a third of the world's population is at risk for contracting dengue virus, and as many as 400 million people are infected annually. Belize has an ongoing risk of dengue transmission, with 2,958 cases of dengue being confirmed in Belize in 2017 (PHAO, 2018), and dengue is a leading cause of febrile illness among travelers returning from the Caribbean (CDC, 2018). Zika was designated a public health emergency of international concern in 2016. In addition to being transmitted by mosquitoes, Zika virus can be transmitted sexually, and it can be passed from a pregnant woman to her fetus. Fetal infection with Zika virus can result in severe birth defects, including microcephaly. Cases of Zika, which have also been linked to Guillain-Barre´ syndrome, a serious neurological disorder, are currently occurring in many countries in the Americas, including Belize (CDC, 2018; WHO, 2018). *Aede*s mosquitoes are the dominant disease vector and transmitter of dengue fever, Zika, and Chikunkunya (Preechaporn et al., 2006; Chumsri et al., 2015, 2018). In southern Thailand, the primary vector species is *Ae. aegypti* (Preechaporn et al., 2006; Chumsri et al., 2015, 2018).

Mosquitoes utilise a wide variety of larval habitat types. Artificial containers are a major source of breeding habitats for mosquitoes worldwide. These artificial containers include tyres, plastic bottles, metal boxes, and cans (Wongkoon et al., 2005; Lester & Pike, 2003, Preechaporn et al., 2006). There are several examples of how each mosquito species prefers different breeding sites. *Ae. aegypti* breeds in a wide assortment of domestic containers, whereas *Ae. albopictus* tends to be found in natural containers, such as bamboo stumps and coconut shells, or in artificial containers outside the houses such as tyres, opened cans and plastic bottles (Wongkoon et al., 2005; Lester & Pike, 2003, Preechaporn et al., 2006).

Krabi is one of the most tourist attraction places in Thailand where mosquito breeding sites could affect tourism. The objectives of this study are to investigate (1) how tourist (i.e. high and low tourist temple) affecting water container types, mosquito species, and their numbers, (2) relationship between mosquito numbers and water pH and temperature, and (3) correlation between container numbers and mosquito larvae numbers.

Materials and methods

Study sites

The study was conducted at high - low tourist temples in Krabi, Thailand. The high tourist temples are the following: Kaew Korawaram Temple (8°3'44.14"N, 98°54'45.99"E), Sai-Thai Temple (8°4'54.97"N, 98°52'7.84"E), and Tham Suea (Tiger Cave Temple) (8°7'26.34"N, 98°55'30.42"E). The low tourist temples on the other hand are: Khuan Sabai Temple (8°6'9.59"N, 98°54'22.34"E), Phokha Juthamat Temple (8°5'53.15"N, 98°54'33.60"E), and Panurat Temple (8°4'42.58"N, 98°54'44.94"E) (Figure 1) from 28 February, 2022 to 1 March, 2022.

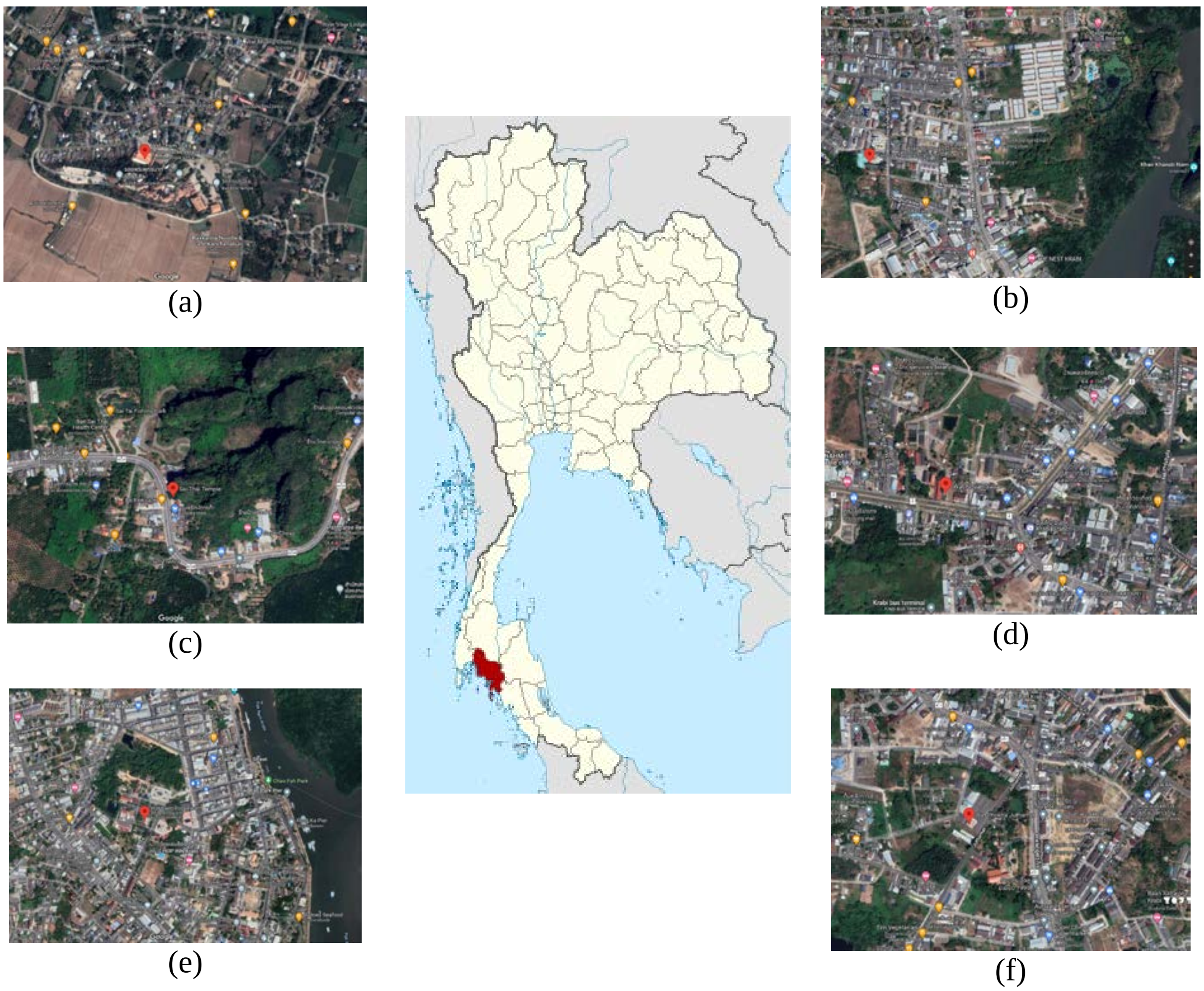

Figure1 Map of Thailand and study site at Krabi Province, Thailand. (a) Tham Suea (Tiger Cave Temple), (c) Sai-Thai Temple, (e) Kaew Korawaram Temple for high tourist temples. (b) Panurat Temple, (d) Khuan Sabai Temple , (f) Phokha Juthamat Temple for low tourist temples.

Data Collection

Mosquito larval survey was conducted in Krabi province, Southern Thailand located at 8°3'32.24"N latitude and 98°55'2.86"E longitude in February 2022. We studied high - low tourist temples: The high tourist temples are the following: Tham Suea (Tiger Cave Temple) (Figure 1a), Sai-Thai Temple (Figure 1c), and Kaew Korawaram Temple (Figure 1e). The low tourist temples are: Phokha Juthamat Temple (Figure 1b), Khuan Sabai Temple (Figure 1d), and Panurat Temple (Figure 1f).

Entomological Studies

We collected all mosquito larvae from both indoor and outdoor containers using GLOBE Observer: Mosquito Habitat Mapper App (GO MHM). We collected water pH and water temperature of each water container using the GLOBE Hydrosphere protocol. We collected mosquito larvae from each outside (natural and artificial) water container by using fishnets with 0.55 mm mesh size nets. All mosquito larvae from each water container were placed in a plastic bag and tied to the bag with a rubber band (Chumsri et al., 2015). We preserved mosquito larvae in 70% alcohol and identified them up to genus level using Rattanarithikul and Panthusiri's keys (Rattanarithikul et al., 1994).

Data Analysis

All variables were tested for normality using the Kromogorov-Smirnov test. The equality of variances was evaluated using Levene's test. Descriptive statistics of the data were analysed. Independent sample-t tests were used to test the differences in numbers of water containers, container types, numbers of *Aedes* larvae between high and low tourist temples. Correlation was used to find some association between water pH and water temperature and the number of *Aedes* larvae.

Results

Water temperature and pH

Water temperature in containers at Kaew Korawaram Temple was higher than Sai-Thai Temple, Tham Suea (Tiger Cave Temple), Khuan Sabai Temple, Phokha Juthamat Temple, and Panurat Temple (Table 1) but water pH did not differ between these high-low tourist temples.

Numbers of Mosquito larvae

There were only *Ae. aegypti*, *Ae. albopictus, Culex, and Toxorhynchites* larvae found at high tourist temples but *Toxorhynchites* larvae were found at Sai Thai temple for low tourist temples. The number of *Ae. aegypti* and *Ae. albopictus* larvae were significantly higher at high tourist temples than at low tourist temples (Table 1).

Container types

Container types and numbers differed significantly among the sites. High tourist temples had the highest number of containers, those were mostly buckets. On the other hand, low tourist temples had the lowest number of containers (Table 1).

Total mosquito larvae, container types at high and low tourist temples: The number of *Aedes aegypti*, *Aedes albopictus*, *Anopheles* spp, and *Toxorhynchites* spp. larvae were higher at high tourist temples than at low tourist temples. The number of *Aedes aegypti*, *Aedes albopictus*, *Anopheles* spp, and *Toxorhynchites* spp. larvae do not differ among container types (Table 2).

Total tourist temples on high and low tourist temples in Krabi: The number of tourists, monks, novice, buddhist nuns, dogs and cats were higher at high tourist temples than at low tourist temples (Table 3).

Discussion

Among different sites at high and low tourist temples. Chumsri et al. (2018) found that container types and their numbers were different among sub districts in Nakhon Si Thammarat province. Furthermore, we observed that mosquito larvae numbers increased with increasing container numbers in all sub-districts. To our knowledge, no study has shown the direct relationship between container numbers and mosquito larvae numbers. Snow & Medlock (2006) suggested that increasing numbers of water butts (containers used to collect rainwater) might increase the numbers of mosquitoes that breed in them. The results of our study show that the number of tourists also has an effect on the number of mosquito larvae, along with container types and seasons.

We also observed that *Ae*. *aegypti* preferred to breed both in outdoor water containers, but *Ae*. *albopictus* preferred to breed mostly in outdoor containers. Previous studies found similar results, Chareonviriyaphap et al. (2003) and Wongkoon et al. (2007) found *Ae*. *aegypti* larvae both in indoor and outdoor containers but *Ae*. *albopictus* larvae mostly in outdoor containers. However, our results showed higher numbers of *Ae*. *aegypti* and *Ae*. *albopictus* larvae in mangrove forest area than the previous study (Wongkoon et al., 2007). The possible reason could be because we collected our samples in Thasala district. *Ae*. *aegypti* larvae mostly prefer to breed in drums, jars, concrete tanks and discarded objects (Phong and Nam, 1999), *Ae*. *albopictus* prefers to breed outside in open spaces with shaded vegetation (Wongkoon et al., 2007; Saleeza et al., 2011). This study uses the number of tourists affected to observe breeding sites of mosquito larvae and use Globe observers. Number of tourists can be used for mosquito breeding and water containers compared with ground trothing, a high number of tourist temples have the effect of allowing more mosquitoes to breed and encounter more mosquitoes than low tourist temples.

In this study *Anopheles* spp. and *Toxorhynchites* spp. were found only in outdoor containers. Previous studies also observed that *Culex* spp. larvae were mostly found in outside containers (Preechaporn et al., 2007) such as in plant plates, used pots, plastic cement mixer tubes, and used bowls (Chumsri et al., 2015), those were situated outside of the households.

Through this study, we have been informed which containers are used as breeding sites for *Aedes* spp. *Ae. aegypti*, *Ae. albopictus*, *Anopheles* spp. and *Toxorhynchites* spp. the most important mosquito vectors of dengue fever viruses (Knudsen, 1995; Mousson et al., 2005). If we can suggest to the fishery households to destroy those *Aedes* spp. holding containers, may be it is possible to control dengue fever, as destruction of breeding habitats is an important strategy to reduce the *Aedes* mosquito population; as well to reduce the larval development and adult mosquito population growth (Li et al., 2014).

2022 IVSS: Badges

1. **I Am A Collaborator:** Because our group is working together with experts on mosquitoes habitat mapper application. Mr. Krisanadej Jaroensutasinee, Mrs. Mullica Jaroensutasinee, Ms. Sirirat Somchuea, and Ms Benjamas Khunpan which makes the work more efficient.
2. **I Am A STEM Professional:** In this section, our group works in a systematic process, problem solving, and systematic thinking. There is also attached information to support the project with a STEM Professional.
3. **I Am a Data Scientist:** We went to collect data in Krabi province by ourselves. We use a helper to collect data. It is possible in more detail with the application of the GLOBE Observer app, which can collect MHM data for analysis of human behavior.

Acknowledgements

We thank Assoc. Prof. Dr. Krisanadej Jaroensutasinee, Assoc. and Prof. Dr. Mullica Jaroensutasinee for helping with experimental design, data analysis and manuscript preparation. This work was supported in participation by Samsenwittayalai and Center of Excellence for Ecoinformatics, Walailak University.

Table 1 (Mean ± SE) of parameter, mosquito larvae and storage containers at the high and low tourist temple ; '**' indicates P < 0.001 and '*' $P < 0.05$.

Parameters	Kaew Korawaram Temple	Tiger Cave Temple	Sai-Thai Temple	Khuan Sabai Temple	Panurat Temple	Phokha Juthamat Temple	*F*-test
1. Water temperature (°c)	28.54 ± 0.35	27.75 ± 0.42	29.20 ± 0.88	28.34 ± 0.56	27.40 ± 0.47	27.47 ± 0.52	$F_{5,148}$= 2.03, *ns*
2. pH	7.85 ± 0.98	7.50 ± 0.10	7.65 ± 0.17	7.08 ± 0.12	7.05 ± 0.14	7.14 ± 0.09	$F_{5,143}$ = 5.94**, P<0.001
3. *Aedes aegypti* larvae	0.00 ± 0.00	7.14 ± 0.16	0.00 ± 0.00	1.48 ± 0.88	0.00 ± 0.00	0.38 ± 0.39	$F_{5,148}$= 0.79, *ns*
4. *Aedes albopictus* larvae	2.74 ± 1.08	5.00 ± 3.02	5.00 ± 0.54	2.41 ± 1.18	2.14 ± 1.22	1.54 ± 1.04	$F_{5,148}$= 0.39, *ns*
5. *Culex* spp. larvae	0.12 ± 0.04	0.18 ± 0.07	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	$F_{5,148}$= 0.48, *ns*

6. *Toxorhynchites* spp.	0.00 ± 0.00	0.00 ± 0.00	0.63 ± 0.15	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	$F_{5,148}$= 1.48, *ns*
7. containers with lid/ without lid	0:42	1:27	0:18	0:27	1:20	0:13	
8. Indoor/ Outdoor container	4:39	10:18	1:17	4:23	2:19	2:11	
9. Natural/Manmade containers	3:36	0:28	0:18	0:27	0:21	0:13	
10. Earthern jar: cement tank: plastic container: metal container: pond	0:21:16:0:5	12:10:6:0:0	3:3:6:3:3	0:4:16:0:7	0:9:10:0:2	0:4:9:0:0	

Table 2 (Mean ± SE) of mosquito larvae at high and low tourist temples, Krabi province.

Mosquito Larvae	Kaew Korawaram Temple	Tiger Cave Temple	Sai-Thai Temple	Khuan Sabai Temple	Panurat Temple	Phokha Juthamat Temple	One-way ANOVA
Aedes aegypti	0.00 ± 0.00	37.79 ± 7.14	0.00 ± 0.00	4.56 ± 0.88	0.00 ± 0.00	1.38 ± 0.38	$F_{5,148}$= 0.56
Aedes albopictus	7.00 ± 1.08	15.98 ± 3.02	9.56 ± 2.25	6.10 ± 1.17	5.60 ± 1.22	3.75 ± 1.04	$F_{5,148}$= 0.86
Culex spp.	0.77 ± 0.11	0.94 ± 0.17	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	$F_{5,148}$= 0.79
Toxorhynchites spp.	0.00 ± 0.00	0.00 ± 0.00	1.18 ± 0.28	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	$F_{5,148}$= 0.20

Table 3 Number of tourist temples at high and low tourist temples.

Study sites	Number of monk	Number of novice	Number of buddhist nun	Number of dog and cat	Number of tourist / Day
High tourist temple					
Kaew Korawaram Temple	25	63	5	40	80
Tiger Cave Temple	59	0	60	30	200
Sai-Thai Temple	6	0	0	25	20
Low tourist temple					
Khuan Sabai Temple	20	0	0	10	15
Panurat Temple	10	0	1	20	15
Phokha Juthamat Temple	13	0	0	15	15

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