Mosquito Larvae diversity and abundance relation to land cover in coastal area in Trang Province Thailand

Miss Piyapat Pethin, Miss Kanyakorn Tongsongsom, Mr.Surayut Kongjun

School: Princess Chulabhorn Science High College Trang
Teachers: Mrs. Patchara Pongmanawut
Scientists: Dr. Anantanit Chumsri and Dr. Fahmida Wazed Tina
Email: ppat1994@hotmail.com
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Abstract

This study investigated how types of land cover in coastal areas affecting mosquito species diversity and its abundance. Ovitraps were used to conduct this experiment. An experiment was conducted in each land cover with an area of 10 * 50 square meters. The coastal area was separated in 4 land cover including beach forest area, Melaleuca forest area, mangrove forest area, and forest near residential area with 20 ovitraps with total 80 ovitraps. Before adding the water to the containers, water was filtered through two layers of a fine nylon mesh (intermesh gap = 0.5 mm) to remove any existing macroinvertebrates. Mosquito larvae were collected from each ovitraps from January to February, 2020. We collected mosquito larvae from ovitraps and identified up to genus and/or species level.

Our results, we found 3 types of mosquito larvae coastal area including *Ae.aegypti*, *Ae.albopictus* and *Culex* spp. The number of *Ae.aegypti and Ae.albopictus* were highest in Mangrove forest area (*Ae.aegypti* 88.89%, *Ae.albopictus* 43.34%) . The number of *Culex* spp. was highest in Forest near residential area (*Culex* spp. 60%) and pupa was highest in Forest near residential (*Pupa* 42.99%). The number of *Ae. albopictus* and total mosquito larvae positive correlated with water temperature, Electrical conductivity and water volume but *Culex* spp. larvae positive correlated with Electrical conductivity. The highest abundance of *Aedes aegypti* was found in mangrove forest area and forest near residential area, the highest abundance of *Ae. albopictus* larvae was found in Mangrove forest area and the highest abundance of *Culex* larvae was found in Melaleuca forest area. The highest coexistence rate of *Ae. aegypti* and *Ae. albopictus* was found in Mangrove forest area and forest near residential area (coexistence in 0.67% of positive containers). The highest coexistence rate of *Ae*. albopictus and *Culex* larvae was found in Forest near residential area (coexistence in 5.34% of positive containers). *Ae. aegypti* and *Culex* larvae was not found coexistence all area and three types of larvae were not found coexistence all area. These results provided important information with regards to mosquito vectors in coastal habitats of Trang province, Thailand. It also provided information on species diversity, distribution and factors associated with breeding habitat, preference for surveillance and control to prevent the spread of mosquito-borne diseases to the population of the coastal communities. These findings fulfil knowledge of mosquito ecology and support mosquito control strategies that can be applied in coastal areas of Thailand in the future.

Keywords: ovitrap, beach forest area, Melaleuca forest area, mangrove forest area, forest near residential area, Thailand

Introduction

Mosquitoes are found around the world, especially throughout the tropics and in warmer regions. They thrive in a variety of habitats, including freshwater and brackish water (Clear and polluting) (Rueda, 2008). Mosquitoes are commonly known to be a major threat to public health. Biological diversity of mosquitoes is manifested by the fact that many of the genera have spread around the world and some of them are endemic. Mosquitoes are responsible for various diseases in human beings, such as malaria, chikungunya, elephantiasis, dengue fever and Japanese encephalitis (Gratz, 2004; Carrieri et al., 2011; Kaliwa et al., 2011). Almost 700 million people suffer from mosquito-borne diseases every year (Caraballo et al., 2014)

The Andaman coast is an important source for learning and tourism attraction in Trang province, Thailand. There are many people and students coming to use the service each year because the Andaman coast is an area with diverse ecosystems and beautiful beaches.

The researcher wants to study about the different forest areas of the Andaman coast, such as the beach forest area, Melaleuca forest area, mangrove forest area, forest near residential area effect on the types and numbers of mosquito larvae. Which is a place where people come to use the service at risk of dengue fever? Therefore, studying the abundance and diversity of the species of mosquito larvae be used as a database for surveillance and protection for communities and tourists.

Research questions

1. Do the differences of forest in coast areas (beach forest area, Melaleuca forest area, mangrove forest area, forest near residential area) affect the mosquito larvae species and numbers?

2. Do the water quality (surface temperature, water temperature, pH, Electrical conductivity) and volume affect the species and numbers of mosquito larvae?
3. Do the temporal variations (beach forest area, Melaleuca forest area, mangrove forest area, forest near residential area) effect on the coexistence of *Aedes aegypti*, *Aedes albopictus*, and *Culex* spp. larvae?

4. Do we find any co-exist of mosquito larvae in the same breeding container?

Hypothesis

1. The differences of forest in coast areas (beach forest area, Melaleuca forest area, mangrove forest area, forest near residential area) affect the mosquito larvae species and numbers.
2. The water quality (surface temperature, water temperature, pH, Electrical conductivity) and volume affect the species and numbers of mosquito larvae.
3. The temporal variations (beach forest area, Melaleuca forest area, mangrove forest area, forest near residential area) effect on the coexistence of *Aedes aegypti*, *Aedes albopictus*, and *Culex* spp. Larvae.
4. Same genus of Aedes (ie. *Ae. ag*gypti & *Ae. alboptictus*) are expected to be found in the same breeding container.

Materials and methods

1. Study area

This research was conducted at Rajamongala beach, Sikao district, Trang Provinces southern of Thailand. It located on the latitude 7.5282 N longitude 99.3079E as shown in Figure1.

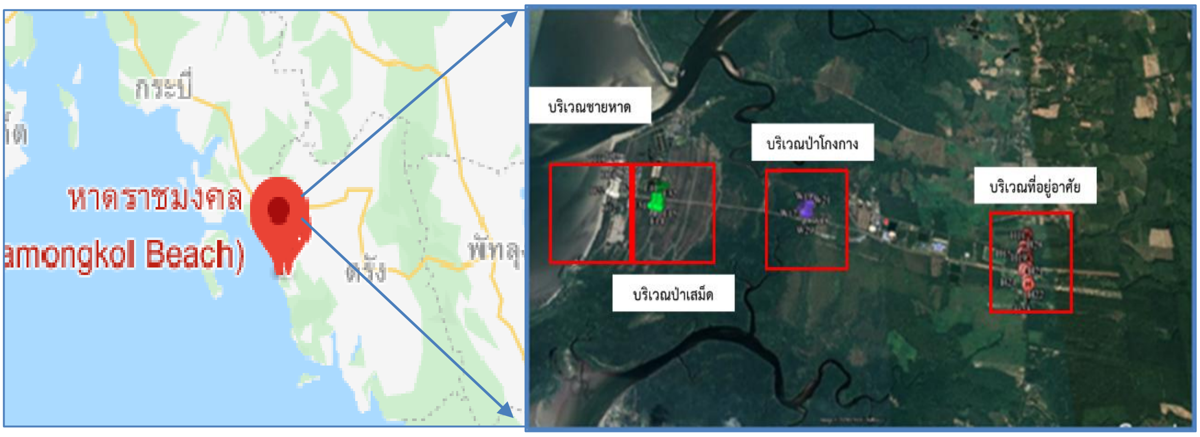


Figure 1 Study area at Rajamongala beach, Sikao district, Trang province

2. Study design

2.1. Time duration and sampling point setting

For sampling, the researchers collected mosquito larvae and water samples at Pak Meng beach during January to February 2020. Dividing the study into 4 areas such as beach forest area, Melaleuca forest area, mangrove forest area, and forest near residential area, as shown in Figure 2.

(a) (b) (c) (d)

Figure 2 Study area (a) beach forest area, (b) Melaleuca forest area, (c) mangrove forest area, and (d) forest near residential area.

3. Equipment for collected mosquito larvae

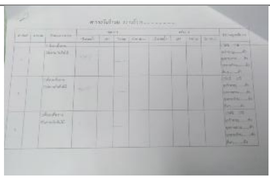
3.1 Equipment for setting up Ovitrap

Rubber cup	Wire	Rope
		

3.2 Equipment for measuring water qualities

PC Test 35	Conductivity Meter	Infrared Thermometer
		

3.3 Equipment for collection mosquito larvae

Bucket		Permanent pen	
Aquarium net		**Rubber band**	
Data sheet		**Plastic bag**	

3.4 Equipment for preserving and identifying mosquito larvae

Small plastic bowl		Dropper	
Alcohol 70%		**Forceps**	
Distilled water		**Microscope**	

4. Installation of ovitraps in study sites

1) Ovitraps were used to conduct this experiment. An experiment was conducted in each land cover with an area of 10 * 50 square meters. The coastal area was separated in 4 land cover including beach forest area, Melaleuca forest area, mangrove forest area, and forest near residential area with 20 ovitraps with total 80 ovitraps. Before adding the water to the containers, water was filtered through two layers of a fine nylon mesh (intermesh gap = 0.5 mm) to remove any existing macroinvertebrates. Mosquito larvae were collected from each ovitraps from January to February, 2020 as show figure 3.

figure 3 Position of ovitraps (a) beach forest area (b) Melaleuca forest area (c) mangrove forest area (d) forest near residential area.

2) Installed ovitraps with tree at 1 meter height and avoid the sun and the wind as in Figure 7.

Figure 4 Position for Installed ovitrap equipment

3) In each ovitrap input 300 ml of water and measuring the pH water, Electrical conductivity, surface temperature, and water temperature of each ovitraps.

5. **Mosquito larvae collection, preservation and identification**

1) Collected mosquito larvae once a week with total 7 weeks and and collected mosquito larvae based on the GLOBE mosquito protocols and input data with Mosquito

Habitat Mapper App. The latitude, longitude and elevations of ovitraps were recorded using smartphone App.

2) Measured the amount of water of ovitrap and measured the quality of water such as pH, electrical conductivity, Water surface temperature, and water temperature in each ovitraps

3) Collected mosquito larvae in each ovitraps in four study sites. All mosquito larvae from each ovitraps were placed in a plastic bag and tied the bag with a rubber band (Chumsri et al. 2015). We preserved mosquito larvae in 90% alcohol in the laboratory and identified them up to genus level using Rattanarithikul and Panthusiri's keys (Rattanarithikul et al. 1994).

6. Data analysis

Pearson correlations were used to test Relationship among numbers of mosquito larvae, water quality and water volume. The significant tests were one-tailed with significant level at $P<0.05$.

Results

1. Types and numbers of mosquito larvae in coastal area at Trang province

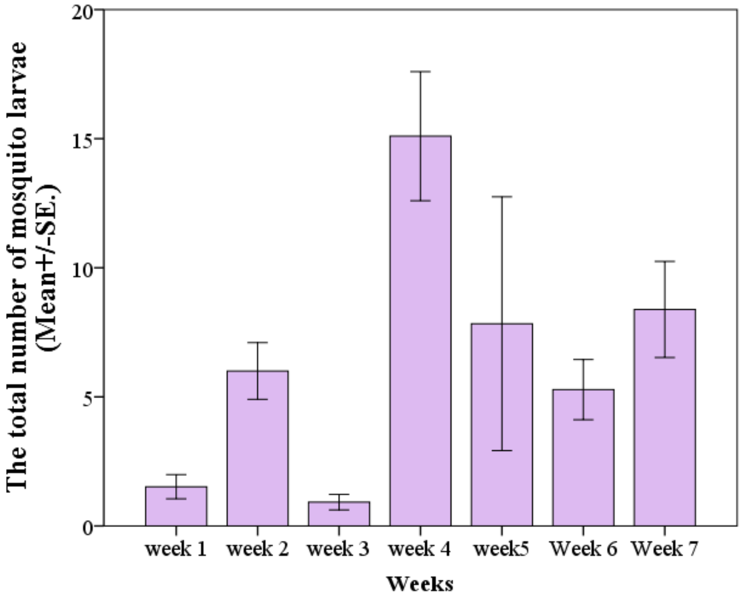
We found 3 types of mosquito larvae coastal area including *Ae.aegypti*, *Ae.albopictus* and *Culex* spp. (Table 1). The number of *Ae.aegypti and Ae.albopictus* were highest in Mangrove forest area (*Ae.aegypti* 88.89%, *Ae.albopictus* 43.34%) . The number of *Culex* spp. was highest in Forest near residential area (*Culex* spp. 60%) and pupa was highest in Forest near residential (*Pupa* 42.99%) (Table 1).

Table 1 Types and numbers of mosquito larvae in coastal area at Trang province.

Types of mosquito larvae	The numbers of mosquito larvae				Total
	Beach forest area	Melaleuca forest area	Mangrove forest area	Forest near residential area	
Ae.aegypti	0 (0)	0 (0)	**8 (88.89)**	1 (11.11)	9 (100)
Ae.albopictus	185 (11.91)	140 (9.01)	**673 (43.34)**	555 (35.74)	1553 (100.00)
Culex spp.	52 (18.90)	29 (10.55)	29 (10.55)	**165 (60.00)**	275 (100.00)
Pupa	19 (17.76)	23 (21.50)	19 (17.76)	**46 (42.99)**	107 (100.00)
Total	256 (13.17)	192 (9.88)	729 (37.50)	**767 (39.40)**	1944 (100.00)

2. The number of mosquito larvae found in each week at Coastal area at Trang province

We found the number of mosquito larvae highest on week 4 (Figure 5).



Figures 5 The number of mosquito larvae found on week 1-week 7 at Coastal area at Trang province

3. The quality of water in ovitraps with and without mosquito larvae at Coastal area at Trang province

Surface temperature was higher in ovitraps with mosquito larvae than in ovitraps without mosquito larvae in Melaleuca forest area but Surface temperature was higher in ovitraps without mosquito larvae than in ovitraps with mosquito larvae in Beach forest area and Forest near residential area and Surface temperature was not difference between ovitraps without mosquito larvae and in ovitraps with mosquito larvae in Mangrove forest area (Table3).

Water temperature and Electrical conductivity were higher in ovitraps with mosquito larvae than in ovitraps without mosquito larvae in Melaleuca forest area, mangrove forest area, and forest near residential area but water temperature and Electrical conductivity were higher in ovitraps without mosquito larvae than in ovitraps with mosquito larvae in Beach forest area (Table 3).

pH water was higher in ovitraps with mosquito larvae than in ovitraps without mosquito larvae in beach forest area, Melaleuca forest area, and forest near residential area but pH water was higher in ovitraps without mosquito larvae than in ovitraps with mosquito larvae in Mangrove forest area (Figure 6).

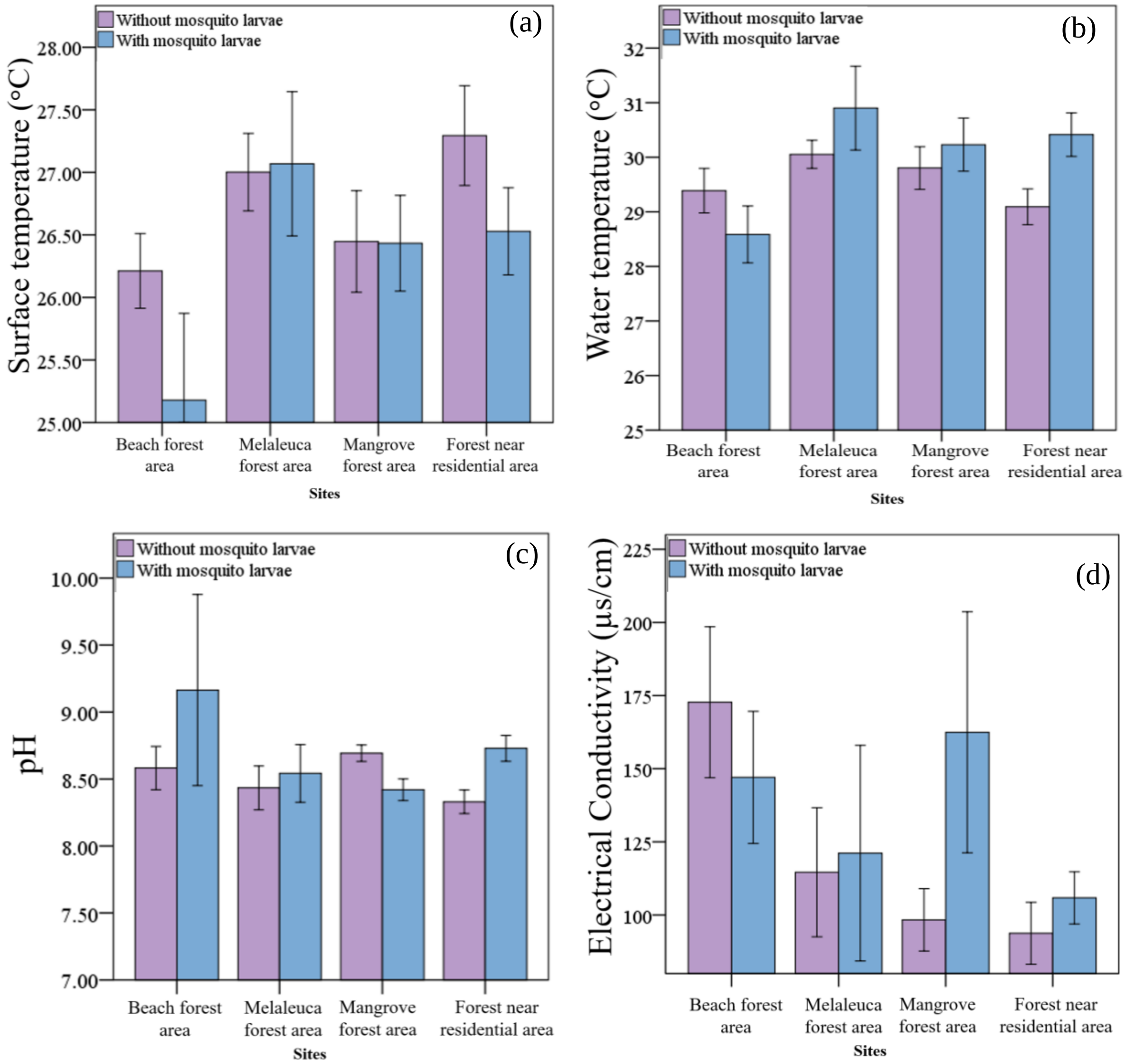


Figure 6 The quality of water (a) surface temperature, (b) water temperature, (c) pH, and (d) Electrical Conductivity in ovitraps with and without mosquito larvae at Coastal area at Trang province

4. Relationship among numbers of mosquito larvae, water quality and water volume

The number of *Ae. albopictus* and total mosquito larvae positive correlated with water temperature, Electrical conductivity and water volume but *Culex* spp. larvae positive correlated with Electrical conductivity (Table 2).

Table 2 The Relationship among numbers of mosquito larvae, water quality and water volume

Types of mosquito larvae	Spearman's rho	Surface Temperature (°C)	Water Temperature (°C)	pH	Electrical Conductivity (μs/cm)	Water volume
Ae.aegypti	Correlation Coefficient	0.100	0.026	-0.078	0.029	0.574
	Sig. (2-tailed)	0.076	0.649	0.166	0.610	0.201
Ae.albopictus	Correlation Coefficient	0.019	0.233*	-0.034	0.306***	0.238**
	Sig. (2-tailed)	0.830	0.007	0.697	0.000	0.0045
Culex	Correlation Coefficient	-0.007	0.080	-0.002	0.125*	0.151
	Sig. (2-tailed)	0.937	0.356	0.985	0.026	0.081
Pupa	Correlation Coefficient	-0.011	0.062	0.008	0.001	0.456
	Sig. (2-tailed)	0.845	0.268	0.883	0.979	0.125
Total mosquito larvae	Correlation Coefficient	0.016	0.247**	-0.032	0.282**	0.172*
	Sig. (2-tailed)	0.856	0.004	0.710	0.001	0.045
	N	316	316	316	316	316

*Correlation is significant at the 0.05 level (2-tailed)

5. Occurrence and abundance: spatial patterns of mosquito larvae

The highest abundance of *Aedes aegypti* was found in Mangrove forest area and Forest near residential area, the highest abundance of *Ae. albopictus* larvae was found in Mangrove forest area and the highest abundance of *Culex* larvae was found in Melaleuca forest area (Table 3).

The highest coexistence rate of *Ae. aegypti* and *Ae. albopictus* was found in Mangrove forest area and forest near residential area (coexistence in 0.67% (1/149) of positive containers). The highest coexistence rate of *Ae*. albopictus and *Culex* larvae was found in Forest near residential area (coexistence in 5.34 (8/149) of positive containers). *Ae. aegypti* and *Culex* larvae was not found coexistence all area and three types of larvae were not found coexistence all area (Table 3).

Table 3 Numbers and proportions of positive containers occupied by *Ae. aegypti* (AEG), *Ae. albopictus* (ALB), and *Culex* (CX) in four sites.

Sites	Number of Ovitrap	Positive Ovitrap	AEG only	ALB only	CX only	AEG+ALB	AEG+CX	ALB+CX	AEG+ALB +CX	None	Proportion AEG	Proportion ALB	Proportion CX
Beach forest area	140	29	0	18	1	0	0	6	0	111	0.00	0.83	0.24
Melaleuca forest area	140	22	0	13	4	0	0	3	0	118	0.00	0.73	0.32
Mangrove forest area	140	42	0	35	1	1	0	4	0	98	0.02	0.95	0.12
Forest near residential area	140	56	0	39	4	1	0	8	0	84	0.02	0.86	0.21

Discussion

The results of the study we found 3 types of mosquito larvae coastal area including *Ae.aegypti, Ae.albopictus* and *Culex* spp. The number of *Ae.aegypti and Ae.albopictus* were highest in Mangrove forest area (*Ae.aegypti* 88.89%, *Ae.albopictus* 43.34%) . The number of *Culex* spp. was highest in Forest near residential area (*Culex* spp. 60%) and pupa was highest in Forest near residential (*Pupa* 42.99%). Chaiphongpachara and Sumruayphol (2017) assessed species diversity and distribution of mosquito vectors in coastal habitats dividing it to three areas according to the distance from the sea species diversity and distribution of mosquito vectors in coastal habitats of Samut Songkhram province, Thailand. They found that a total of 1,764 mosquitoes belonging to 3 genera with 5 species were captured. The most abundant species was *Anopheles epiroticus* accounting for 37.13% followed by *Culex sitiens* (34.92%), *Cx. quinquefasciatus* (27.66%), *Aedes aegypti* (0.23%) and *Cx. gelidus* (0.06%) respectively. *Cx. sitiens* and *An. epiroticus* larvae were collected from available breeding habitats. Pearson correlation showed association the factors associated with breeding habitat preference of mosquito larvae. *Cx.sitiens* larvae were significantly higher in permanent, temporary water resources and mangrove trees

Various physical and chemical factors of larval habitats contribute to mosquito breeding site selection, such as temperature, pH, ammonia, nitrate, sulphate, phosphate and dissolved solids (Oyewole et al, 2009; Oleymi et al, 2010). In our study we found a correlation between factors investigated and larval prevalence. Surface temperature was higher in ovitraps with mosquito larvae than in ovitraps without mosquito larvae in Melaleuca forest area but surface temperature was higher in ovitraps without mosquito larvae than in ovitraps with mosquito larvae in beach forest area and forest near residential area and surface temperature was not difference between ovitraps without mosquito larvae and in ovitraps with mosquito larvae in mangrove forest area.

Water temperature and electrical conductivity were higher in ovitraps with mosquito larvae than in ovitraps without mosquito larvae in Melaleuca forest area, mangrove forest area, and forest near residential area but water temperature and electrical conductivity were higher in ovitraps without mosquito larvae than in ovitraps with mosquito larvae in Beach forest area. pH water was higher in ovitraps with mosquito larvae than in ovitraps without mosquito larvae in beach forest area, Melaleuca forest area, and forest near residential area but pH water was higher in ovitraps without mosquito larvae than in ovitraps with mosquito larvae in Mangrove forest area. The pH of water has an impact on mosquitoes, influencing osmoregulation and oxygen transportation (Umar and Donpedro, 2008). In this study of *Ae. albopictus* breeding, an increase in pH resulted in a significant decrease in larval density. The container sites sampled had a comparable pH level. This demonstrates that *Ae. albopictus* mosquitoes favor a specific pH range for breeding. This could be an important factor in breeding site selection and larval survival. Salinity is another important factor that can have a repelling or attracting effect on oviposition. Navarro et al (2003) found increasing salinity in the laboratory decreased oviposition. However, our findings showed *Ae. albopictus* was tolerant to variations in salinity, possibly leading to further geographical expansion of this mosquito's breeding sites. From their found can support our result about the number of *Ae. albopictus* and total mosquito larvae positive correlated with water temperature, electrical conductivity and water volume but *Culex* spp. larvae positive correlated with electrical conductivity.

The highest abundance of *Aedes aegypti* was found in Mangrove forest area and Forest near residential area, the highest abundance of *Ae. albopictus* larvae was found in mangrove forest area and the highest abundance of *Culex* larvae was found in Melaleuca forest area. The highest coexistence rate of *Ae. aegypti* and *Ae. albopictus* was found in Mangrove forest area and forest near residential area (coexistence in 0.67% (1/149) of positive containers). The highest coexistence rate of *Ae*. albopictus and *Culex* larvae was found in forest near residential area (coexistence in 5.34 (8/149) of positive containers). *Ae. aegypti* and *Culex* larvae was not found coexistence all area and three types of larvae were not found coexistence all area. On the other hand, Muller et al. (2018) found that the physicochemical and microclimatic conditions of potential and actual mosquito breeding sites in Germany offer suitable conditions for a balanced coexistence and frequent competitive superiority of *Cx. pipiens molestus* over *Ae. Albopictus* with regard to larval and pupal survival.

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