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ORIGINAL PAPER

The spatio-temporal characteristics of potential dengue risk assessed by *Aedes aegypti* and *Aedes albopictus* in high-epidemic areas

Chia-Hsien Lin · Tzai-Hung Wen · Hwa-Jen Teng · Niann-Tai Chang

Abstract *Aedes aegypti* (AE) and *Aedes albopictus* (AA) are the two major dengue vectors in the world. To control dengue, monitoring vectors is essential. The abundance and distribution of mosquitoes are usually considered as a proxy for dengue outbreaks. Most of the researches catch the abundance and distribution of vectors by using traps, but this kind of information only can reflect partial relationship between the mosquitoes and dengue since human factors were not taken into account. To have a better proxy for dengue risk, this research evaluated the spatiotemporal distribution of dengue vectors by a human population-stratified ovitrap survey. We demonstrated the potential intensity of dengue transmission spatio-temporally in the high epidemic areas. We analyzed the temporal patterns of vector abundances, compared the levels of vector aggregation indoors with outdoors, and assess the vector concentration tendency around residential centers and administration edges form June 2010 to June 2011. We found significantly more AA outdoors than indoors, and no matter outdoor or indoor settings were, the abundances of AA were descending from the pre-epidemic period to post-epidemic period. AE also had the descending pattern, but there were no significant differences detected between outdoor and indoor settings. AA hotspots appeared almost the same locations from the pre-epidemic period to post-epidemic period, but no obvious repetitive AE hotspots were shown. We also found that AA tended to aggregate around the residential centers while AE likely clustered around the district boundaries. Our study concluded these spatio-temporal characteristics of AE and AA addressed from a population adjusted ovitrap method implied the risks of dengue in different times and locations. To more efficiently control dengue, except for routine unspecific control interventions, the authorities should realize different environmental management strategies were needed for AE and AA.

Keywords Dengue vector · Transmission risk · Urban settings · Spatial epidemiology · Taiwan

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1 Introduction

Dengue is a rapidly spreading mosquito-borne infectious disease with around 50 million infections each year in the world (WHO 2009). *Aedes aegypti* (AE) and *Aedes albopictus* (AA) are the two major dengue vectors. AE is generally recognized as the urban and domestic dengue virus mosquito as it typically lives around human dwellings and tends to breed in the artificial containers (Rudnick 1986; WHO 2009). In contrast, AA is more likely to develop in the natural habitats (Hawley 1988), but was recently found adapted to the indoor artificial water containers (Rudnick, 1986; Dieng et al. 2010). To restrain dengue, instead of eradicating vectors, most control programs nowadays focus on reducing vector abundances to a certain threshold of transmission (Focks et al. 2000; Focks 2004; WHO 2009). Capturing the spatio-temporal characteristics of vectors such

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as the changes of distribution and abundance, in reflection of potential dengue risk, will efficiently improve human and financial resources allocation.

Different sampling techniques and designs of vector surveillance provide different information (Focks 2004; Ritchie 1984). To detect the existence of AE, ovitrap is the most suitable method and usually less expensive than lar-vae/adult approaches (Furlow and Young 1970). Ovitrap researches from Malaysia, Trinidad, and Brazil have described seasonal and spatial dynamics of AA and AE population (Rozilawati et al. 2007; Chadee and Corbet 1987; Morato et al. 2005). However, these patterns can only present the ecological changes of vectors, but not the potential dengue transmission since they did not put the factor of human population into study designs (Van-wambeke et al. 2011). Recently, the aggregation level of hosts has shown to be negatively associated with the fre-quency of mosquito bites in the U.S. (Foppa et al. 2011). The vector-to-host ratio was used for determining the level of risk of vector-bone diseases (Rogers et al. 2002) and further applied to estimate the risk of exposure for dengue transmission (Vanwambeke et al. 2011). These studies suggested that to assess the potential risks of disease transmission, host factors such as aggregation level and density should be considered in the study.

Kaohsiung has been the epicenter of dengue outbreaks in Taiwan for many years and been invaded by both dengue vectors, AA and AE (Taiwan Centers for Disease Control (Taiwan CDC) 2009; Lin et al. 2012). The routine larval surveillance data from Kaohsiung city showed there were one to three months time lag between the vector abundance and the number of dengue cases, but no detailed spatial correlation was discussed (Chen et al. 2010). Yu et al. developed a climate-based dengue model, claiming that by using weekly dengue incidences and climate data in the southern Taiwan, dengue incidences in the preceding weeks could be predicted (Yu et al.2011). Pai et al. found more vectors in the dengue positive households than the negative households (2005). The study of oviposition showed more dengue vectors outdoors than indoors and also found open space, such as parks and empty houses, had higher vector density than space close to households in Kaohsiung (Pai and Lu 2009). These researches pointed out the possible environmental risks for dengue via vector distribution, but they all did not consider human factors when assessing the risks. Without adjusting human factors, the collected vector information has no connection with hosts so was hardly to explain dengue risks. This study aimed to assess the poten-tial environmental risks of dengue transmission, spatially and temporally, in Kaohsiung by conducting a human pop-ulation adjusted ovitrap survey. Three dimensions of com-parisons between AA and AE were designed: the temporal patterns of vector abundances, the levels of vector
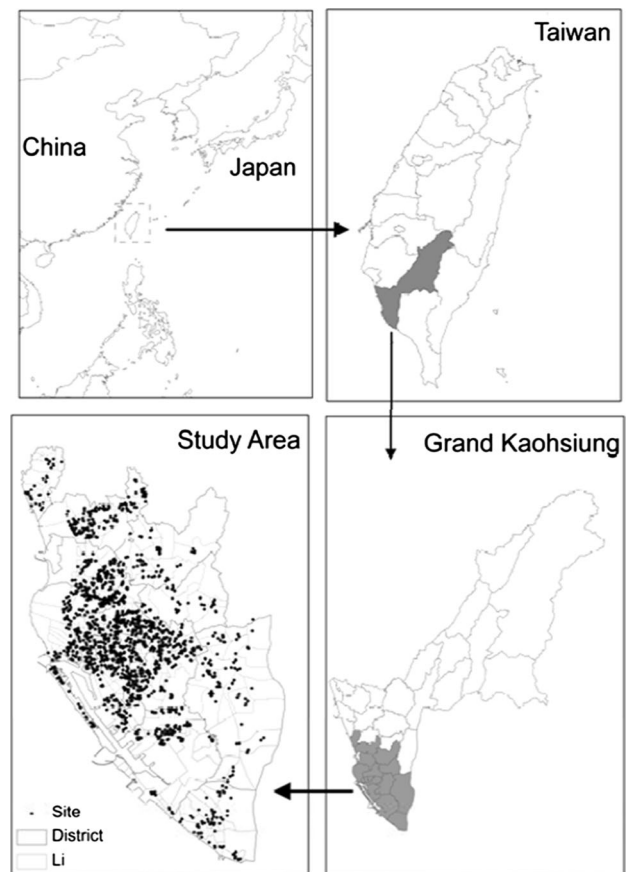

Fig. 1 Map of the study area in Grand Kaohsiung, Taiwan as well as the distribution of study sites. A total of 1,727 sites were selected (3,454 ovitraps) in 17 districts. Li is the smallest administration unit in Taiwan

aggregation indoors and outdoors, and the vector concen-tration tendency around residential centers and administra-tion edges. By differentiating the patterns of spatio-temporal changes of potential dengue risks, the authorities could more specifically design the strategies for epidemics when the human and financial resources are limited.

2 Data and methods

2.1 Study area

The Grand Kaohsiung area is located in the southwestern of Taiwan where the climate is governed by the tropical monsoon (Chen et al. 2004). It has 2.7 million populations with 2,950 km^2 and suffered the most dengue epidemics in Taiwan for several decades (Lin et al. 2012). In the Grand Kaohsiung, only districts fit the criteria of 1) the population density is higher than 1,500 population/km^2, and 2) the ratio of adult AE to *Ae. albopictus* is higher than 1/5, based on available routine vector surveillance data provided by

Fig. 2 The precipitation, temperature and the activities of confirmed dengue cases cumulated based on onset by month in the 17 study districts in the Grand Kaohsiung, Taiwan for the period of June 2009–June 2010. Pre-epidemic is the period of June–September 2009, epidemic is the period of October 2009–January 2010 and post-epidemic is the period of February–June 2010. The extreme rainfall in August was due to typhoon
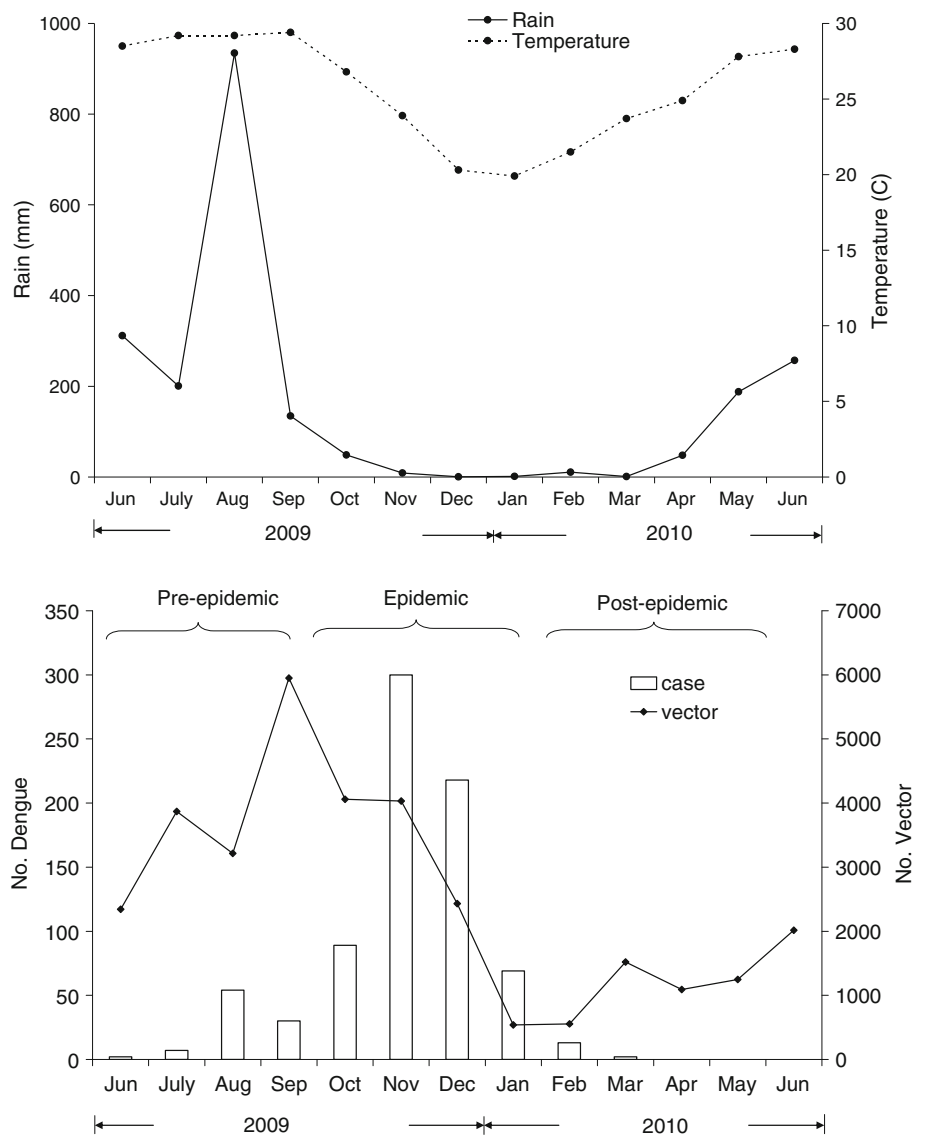
Taiwan Centers for Disease Control (Taiwan CDC) would be selected as study area. Overall, there were 17 out of 38 districts selected. The area range of selected districts was from 1.45 km^2 to 71.85 km^2 and on average each district had a population of 129,000 in 21.77 km^2. The distribution of 17 districts showed in Fig. 1.

Taiwan CDC recorded a total of 784 laboratory confirmed indigenous dengue cases in the study districts from June 2009 to June 2010. The number of cases increased monthly with epidemic apex in November and December 2009, while all activity ceased at the end of the study period (Fig. 2). The inter-month variations in outbreak scale were considerable, ranging from 0 confirmed patients (April 2009–June 2009) to 300 patients (November 2009). We divide this outbreak into three stages to discuss the vector distribution in different epidemic stages. They were pre-epidemic period (June–September 2009), epidemic period (October 2009–January

2010), and post-epidemic period (February–June 2010) (Fig. 2). Spatial distribution of dengue incidence hotspots in three periods showed in Fig. 3.

2.2 Vector surveillance

A human population-stratified ovitrap survey was conducted. In each Li, the smallest administrative unit in Taiwan, a study site was taken every 400 resident based on 2008 population data. Each site contained one indoor and one outdoor ovitraps. The indoor ovitraps were placed in the low (<2 m high) and dark corners of living room while the outdoor ovitraps were set up in the low and shadow area outside of the houses. A total of 1,727 sites were selected (3,454 ovitraps), covered area of 3,150 km^2 as shown in Fig. 1. More detailed information can be found in Wu et al. 2013.

Fig. 3 The spatial distribution of dengue incidence hotspots in the Grand Kaohsiung, Taiwan. Pre-epidemic is the period of June–September 2009, epidemic is the period of October 2009–January 2010 and post-epidemic is the period of February–June 2010. Hotspots were identified by G_i^* statistic with 0.05 significant level
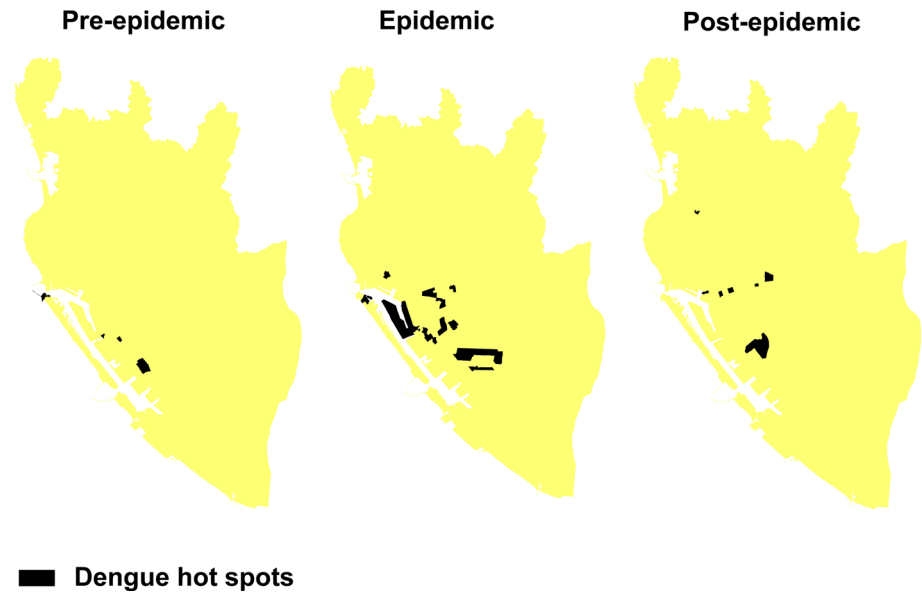
The ovitrap was a black plastic cylindrical container, a radius of 6.5 cm and a height of 10–13.5 cm, with a gray umbrella cover on the top for avoiding the influence of rain and dusts. It contained a wood paddle (1.8 cm wide and 15 cm long) and 400 ml tap water inside. All wooden paddles were collected, monitored and replaced weekly; the water level was checked and refilled by tap water every week. All eggs collected from wooden paddles were hatched to 3–4 instars larvae for species discrimination by trained personnel. The number of successfully hatched larvae and their species were documented. This ovitraps sampling program was carried out from June 2009 through June 2010, while ova were collected for 52 weeks (Wu et al. 2013).

2.3 Identify spatial patterns of dengue vectors

To realize the spatial distribution of AE and AA, we created the interpolation maps and identified the hotspots of vectors for all three periods. Interpolation technique is used for assessing the value of unsampled locations by producing the continuous surface grids for the whole study area. In this study, inverse distance weighting (IDW) was carried out, which was based on the assumption that the value of interpolated points would be influenced more by the nearby observations than the more distant observed samples (Isaaks and Srivastava 1989). In other words, the method is based on the concept of distance-decay principle, which means the relationship between observations declines as the distance between them increase. More sophisticated spatial interpolation methods, such as Kriging and Bayesian maximum entropy (BME), are based the statistical functions of the semivariogram which are also another forms of measuring similarity of distance-decay

relationships (Yu et al. 2009). Mapping continuous surfaces through spatial interpolation can show smoothed spatial patterns more clearly than point samples for further exploration and analysis (Getis and Ord 1992). IDW is widely used and accepted for estimating the spatial distribution of infectious diseases (Moise and Kalipeni 2012; Naish et al. 2011; Bhunia et al. 2013), especially in dengue studies, for example, mapping spatial patterns of dengue vectors (Lozano-Fuentes et al. 2009), assessing the suitable habitats of mosquitoes (Xu et al. 2010; Thammapalo et al. 2008), and distribution of dengue cases (Naish et al. 2014). IDW is defined as: $\hat{z}(x_0) = \sum_{i=1}^{n} \lambda_i \cdot z(x_i)$, where $\hat{z}(x_0)$ is the estimated number of Ae. aegypti or Ae. albopictus; n is the number of ovitrap; $z(x_i)$ is the number of vectors hatched from eggs in each ovitrap. λ_i is the weight function with the form of $\lambda_i = \frac{d_{i0}^{-p}}{\sum_{i=1}^{n} d_{i0}^{-p}}$, where d_{i0} is the distance between estimated point and ovitrap i, and the power parameter p is set as 1 which assumed that distance is inverse proportional to the estimated number of dengue vector, Ae. aegypti or Ae. albopictus.

After creating the interpolation surfaces, we used the local G-statistic (G_i^*) for hotspots identification. G_i^* was the measurement of spatial concentration, which mean it can identify clusters of high or low values. It is defined as: $G_i^*(d) = \frac{\sum_j w_{ij}(d)x_j}{\sum_j x_j}, i \neq j$, where i and j represent different ovitraps; x_j is the number of vectors in neighbor ovitrap j of the target ovitrap i; w_{ij} is a contiguity spatial weights matrix used for defining the neighbor relationships by using the function of inverse distance d between ovitrap i and j. A

Fig. 4 The buffering zones of
district boundaries and
residential centers for the 17
districts in the Grand
Kaohsiung, Taiwan

group of significant positive G_i^* represented a concentration
of high vector abundances which also called a hotspot (Ord
and Getis 1995; Anselin 1992). In this study, 0.05 signifi-
cant level was used.

2.4 Comparisons between residential centers and boundaries of districts with hotspots of vectors

District is the smallest management unit of health and
environment affairs. The marginal areas of districts are
usually lack of management due to unclear responsibilities
among adjacent districts. To realize the potential dengue
risks of population centers and district boundaries, we create
buffer zones for assessment. We took 10–30 % of district
area as buffer area for both buffers. 100, 300 and 500 m were
the distances taken for district boundary buffers for districts
<12, 12–30 and >30 km². The residential center buffers
were circles, using the mean center of study sites of each
district as the center, and having the same size of area as the
district boundary buffer in each district (Fig. 4).

2.5 Comparison of patterns of larval abundance

This research investigated the temporal and spatio-temporal
patterns of larval abundance based on population adjusted
ovitrap sampling through three approaches. First was to
assess the temporal distribution of AE and AA. The abun-
dance levels of both vectors and each of them were compared
among the pre-epidemic, epidemic and post-epidemic peri-
ods by two estimators: the median numbers of hatched larvae

per site or per ovitrap and the proportion of positive sites (the
number of sites with hatched larvae out of 1,727). Wilcoxon
signed-rank test is also a non-parametric method, used for
comparisons between paired data. We used Wilcoxon
signed-rank test to evaluate the number of hatched per site
between periods and the number of hatched larvae per site
between indoor and outdoor settings. The linear trend of the
proportion of positive sites across three time periods was
detected through extended Mantel–Haenszel Chi square for
linear trend (Schlesselman 1982). The last was to assess the
cluster tendencies of both vectors between residential centers
and district boundaries. We defined the hotspot ratio is the
ratio of hotspot area overlapping with the buffers of resi-
dential centers (or district boundaries) to those hotspot area
not overlapping with the buffers. We then compared the
hotpot ratios of district boundaries with the ratios of resi-
dential centers among three periods. All statistical analyses
were performed by R-2.11.1 for Windows (http://cran.r-pro
ject.org/bin/windows/base/old/2.11.1/) with a significance
level of 0.0.5. Spatial pattern analyses were done by ESRI
ArcGIS 9.3 (ESRI, Redlands, California, USA).

3 Results

From June 2009 through June 2010, there were totally
153,376 eggs collected and successfully hatched 15,586
Ae. aegypti and 17,283 *Ae. albopictus*. In the total of 3,454
ovitraps, there were 1,466 (42.4 %) samples were *Ae. ae-
gypti* or *Ae. albopictus* positive.

Table 1 The summary of the hatched *Aedes albopictus* (AA) and *Aedes aegypti* (AE) per site in the different time periods in the 17 districts of Grand Kaohsiung, Taiwan. The Wilcoxon signed-rank test was used to evaluate the median differences of larvae per site between different time periods and extended Mantel–Haenszel Chi square test was used for detecting linear trend for positive proportion. Pre-epidemic is the period of June–September 2009, epidemic is the period of October 2009–January 2010 and February–June 2010 is the post-epidemic period

Species	Period	Median	P value[a]	Positive Proportion (%)	P value
AA					
	Pre-epidemic	8	<0.001[a]	46.1	<0.001
	Epidemic	0	<0.001[b]	29.1	
	Post-epidemic	0		21.3	
AE					<0.001
	Pre-epidemic	1	<0.001[a]	29.6	
	Epidemic	0	<0.001[b]	13.2	
	Post-epidemic	0		9.7	
AA + AE					<0.001
	Pre-epidemic	9	<0.001[a]	28.2	
	Epidemic	0	<0.001[b]	22.0	
	Post-epidemic	0		14.7	

[a] The comparison between pre-epidemic and epidemic period

[b] The comparison between epidemic and post-epidemic period

3.1 Temporal distribution

During the whole study period, there were on average 19 larvae/site hatched. The median number of hatched AA per site was significant different among three time periods, with the largest in the pre-epidemic period (n = 8) and smallest in the rest periods (n = 0). AE had the same significant pattern. This temporal decline phenomenon also appeared on the proportion of positive site. AA had 46.1 % positive rate in the pre-epidemic period, decreasing to 29.1 and 21.3 % in the epidemic and the post epidemic periods. AE had 29.6, 13.2 and 9.7 % respectively. The tendency of decline by time was statistically significant at 0.05 level for both vectors (Table 1).

3.2 Indoor/outdoor settings

Aedes albopictus and AE showed different patterns in the median number of larvae per ovitrap between indoors and outdoors. AA had descending pattern both indoors and outdoors from the pre-epidemic period to the post-epidemic period. More AA per ovitrap was collected outdoors than indoors, significantly in each period with median differences of 1.8, 0.7 and 0.2. The AE per ovitrap showed no difference between outdoor and indoor environment (pre-epidemic period: median difference = 0, p = 0.05; epidemic period: median difference = 0, p = 0.46; post-epidemic period: median difference = 0, p = 0.13). In both indoor and outdoor settings, most AA hotspots locations appeared repeatedly from pre-epidemic to post-

epidemic period, while AE hotspots were less repetitive (Fig. 5).

3.3 District boundary vs. Population centers

Aedes albopictus and AE showed different hotspot patterns around the residential centers and district boundaries in Fig. 6. It shows the AA hotspot ratios of residential centers are higher than the ratios of district boundaries among three periods. It means that AA hotspots were more likely to appear at the residential centers than district boundaries. On the other hand, AE hotspot ratios of district boundaries are higher than the ratios of residential centers among three periods. It means that AE more likely clustered around the district boundaries than residential centers.

4 Discussion

This study presented the potential environmental risks of dengue transmission spatio-temporally through the human population adjusted ovitrap survey. There were four major findings. First, both AA and AE showed significant descending patterns from pre-epidemic to post-epidemic period. Second, more AA per outdoor ovitrap were observed than indoors, while AE showed no differences between outdoors and indoors in all three periods. Third, from the beginning to the end of epidemic, the hotspots of AA occurred almost the same places while less repetitive locations of AE hotspots found. Fourth, AA tended to

Fig. 5 The location shift of *Aedes albopictus* (AA) and *Aedes aegypti* (AE) hotspots in the Grand Kaohsiung, Taiwan. The mosquito hotspots were identified based on the number of collected larvae per ovitrap by G_i^* statistic with 0.05 significant level. Pre-epidemic is the period of June–September 2009, epidemic is the period of October 2009–January 2010 and post-epidemic is the period of February–June 2010
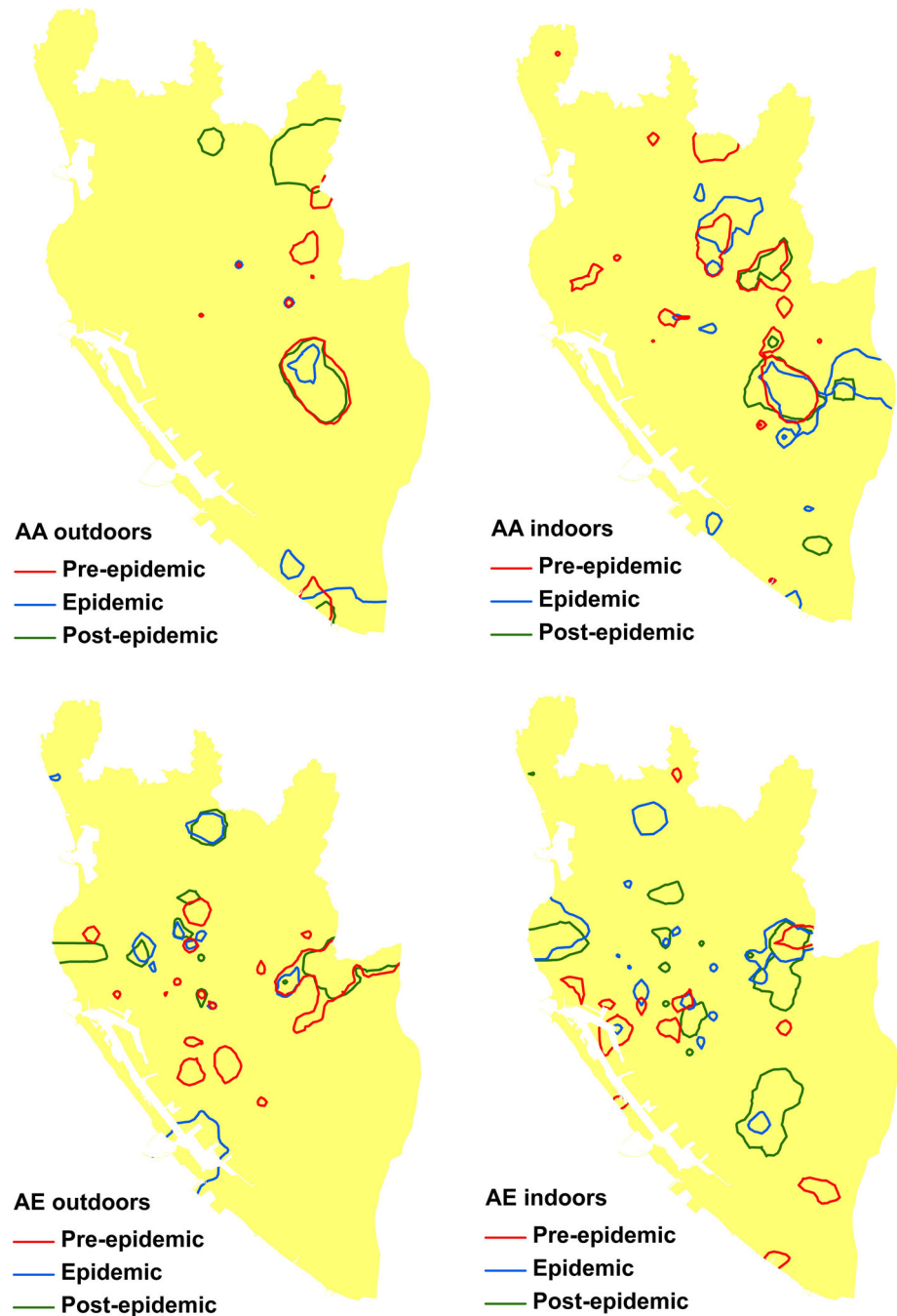
cluster around the residential centers while AE more likely clustered around the district boundaries. Through tracking the spatio-temporal changes of both AA and AE, the areas with hidden or ignored habitats could be identified. The risk of potential transmission could also be reduced through more proper control strategies based on the characteristics of different mosquitoes.

The descending patterns of both dengue vectors from pre-epidemic to post-epidemic period could be driven by the meteorological factors (rainfall and temperature), intervention and the choice of oviposition. The pre-epidemic period

was the summer season, relatively wet and hot in Taiwan (Fig. 2). Studies from Puerto Rico, Trinidad, Thailand and Philippines found more AE eggs and positive containers in the wet season than the dry period (Chadee and Corbet 1987; Moore et al. 1978; Sheppard et al. 1969; Schultz 1993). Entomology data in Taiwan also showed breeding activities of AE and the vector index were higher under hot and wet conditions (Yu et al. 2011; Tsai et al. 2012). More vectors detected in the wet and hot conditions could be because unoccupied outdoor containers or natural plants were more likely to become available habitats in the rainy season and
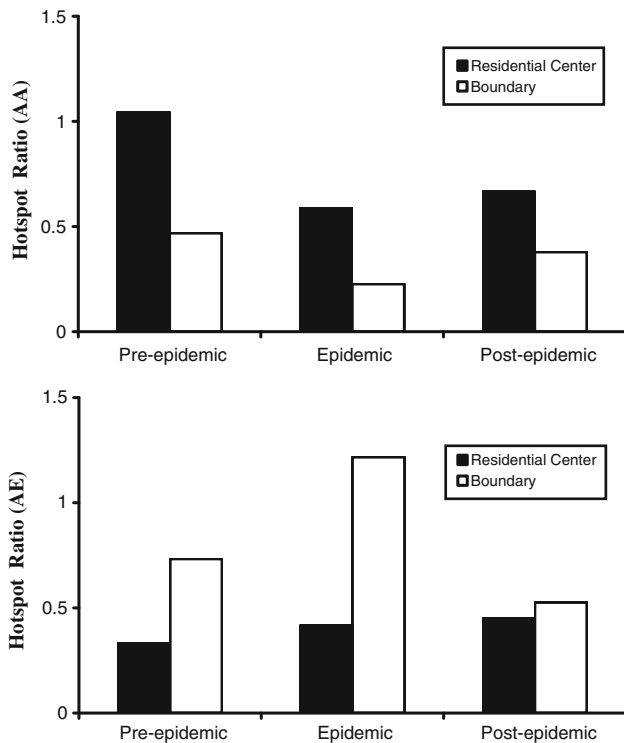
Fig. 6 Comparisons of the clustering tendency between *Aedes albopictus* (AA) and *Aedes aegypti* (AE) around residential centers and district boundaries in different periods. The hotpot ratio is defined as the ratio of hotspot area overlapping with the buffers of residential centers or district boundaries to those hotspot area not overlapping with the buffers. AA and AE were collected from the 17 districts in the Grand Kaohsiung, Taiwan. Pre-epidemic is the period of June–September 2009, epidemic is the period of October 2009–January 2010 and post-epidemic is the period of February–June 2010

the growth rate and oviposition activity of mosquitoes were enhanced under higher temperature (Giatropoulos et al. 2012; Alto and Juliano 2001). Meanwhile, mosquitoes would ingest higher viral dose from hosts, and increase the biting activities in the higher temperature (Yang et al. 2009). Increased number of mosquitoes combined with their enhanced transmission abilities, could be the trigger of dengue outbreaks. The phenomenon of dengue major outbreak followed the peaks of mosquito density was also reported in Puerto Rico (Barrera et al. 2011). Fewer vectors found in the epidemic period than the pre-epidemic time could be due to the control intervention such as Ultra Low Volume spray, and habitat removal campaigns carried out by the health authorities (Taiwan Centers for Disease Control (Taiwan CDC) 2009). This observation, lower vector densities but high dengue cases, was opposite to studies in Puerto Rico and Trinidad which declaimed the number of female *Aedes* mosquitoes, larvae and oviposition were positively correlated to dengue activities (Barrera et al. 2011; Chadee et al. 2007). The different relationships between the abundances of dengue vectors and the number

of dengue cases could be due to demography, spatial distribution of vector and host, herd immunity, virus strains, and human behaviors in the different areas (Barrera et al. 2011). The post-epidemic period had the fewest vectors could be the result of intensive interventions in the epidemic period. The increased precipitation in the post-epidemic period produced more naturally attractive rain-filled habitats could be another reason of fewer vectors collected in the ovitraps (Bond and Fay 1969).

Aedes albopictus and AE displayed different aggregated patterns indoors and outdoors. It was not surprising that significant more AA larvae collected outdoors in all three periods, since in general AA prefers breeding in the vegetated areas, permeable types of land cover, and natural water-filled habitats (Richards et al. 2006). Two ovitrap researches carried out in Malaysia also supported our observations, which claimed AA laid more eggs in the outdoor settings than indoor environment (Norzahira et al. 2011; Yap 1975). To control AA, authorities should pay attention on outdoor environment management, especially those areas frequently identified as hotspots (Fig. 5), since suitable habitats for the AA offspring may exist persistently in these locations even though the interventions were implemented (Reiter 2007). Contrast to AA, AE was viewed as an indoor mosquito. It was observed in or around the house (Rudnick 1986), spending most of the time resting in the shade area indoors (Reiter 2007). Its female preferred laid eggs in the indoor domestic containers (Wongkoon et al. 2007). A research in Malaysia showed AE larvae collected more indoors (Yap 1975), but an Indonesia ovitrap study came out with opposite results (Syarifah et al. 2008). Like an ovitrap research carried out by Norzahira et al. (2011) in Malaysia, our ovitrap study found no significant difference of hatched AE abundances between indoor and outdoor settings. This finding seemed not to correspond with the knowledge of AE as a domestic indoor vector, but this could exactly reflect the fact that the distribution of breeding sites did not represent the distribution of adult mosquitoes (Rudnick 1986). Instead of laying all eggs in one habitat, AE females would visit a lot of suitable habitats for the entire eggs (Reiter 2007), so the spatial distribution of oviposition could more likely represent the pattern of suitable sites. The result of no difference of AE larvae indoors and outdoors from our research may imply both environments had equally available breeding sites, including indoor containers (Wongkoon et al. 2007), rain-filled containers around the peridomestic (El-Badry and Al-Ali 2010) or habitats under the tree shadow (Russell et al. 2005). Equal strength of intervention and attention should be carried out in both settings.

Our study also found AA tended to cluster around the residential centers while the clusters of AE were observed closer to the district boundaries in all three periods. In

Grand Kaohsiung, the residential center areas are those high-storey modern buildings with gardens and public parks nearby. This characteristic of urban structure could attract AA more than AE for oviposition (Yap, 1975). Combing our observations of AA found more outdoors (Table 1), these results suggest to reduce the potential dengue transmission risk, the authorities need to delicate in discovering the hidden outdoor habitats in the crowded areas (Wen et al. 2012). Unlike AA, our study found AE tended to cluster around district boundaries. Studies from Puerto Rico and Thailand reported AE oviposition would cluster at household level (Harrington et al. 2005). Most of the dwellings in the outer district area are one to two stories houses in Grand Kaohsiung, so we may bring out a theory that areas with low story houses were likely to provide suitable habitats for AE. To reduce the potential indoor oviposition sites, the characteristics of residents living in low story houses are worth investigated (Rudnick 1986). In addition, oviposition clusters around boundaries could also imply the communications and cooperation among authorities of adjacent districts should be enhanced (Yap 1975).

Through the human adjusted ovitrap survey, we found that the abundances of AA were descending from the beginning to the end of outbreak, more AA were outdoors, aggregated around the same areas and tended to clustering around the residential centers. AE had the descending pattern as well. It had no significant differences between indoors and outdoors, and no obvious repetitive clustering areas. AE hotspots likely appeared around the boundaries of adjacent districts. We concluded that the spatio-temporal patterns of AE and AA addressed from a population adjusted ovitrap method implied the risks of dengue in different times and locations. To efficiently control dengue, except for routine control interventions, the authorities should design particular environment management strategies for different vectors.

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