

Distribution patterns of *Diaphorina citri* Kuwayama (Hemiptera: Psyllidae) eggs, nymphs and adults in a Malaysian citrus orchard

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ABSTRACT

Spatial distribution of eggs, nymphs and adults of *Diaphorina citri* (Hemiptera: Psyllidae) was studied in a commercial orchard in SW Sarawak, Malaysia, using mean–variance test, the index of dispersion, and the negative binomial distribution. To further ascertain and confirm the distribution pattern in the population of the psyllid, dispersion indices (index of mean crowding, Lloyd's index of patchiness, Taylor's power law and Iwao's patchiness regression) were calculated. Measurable tests showed that distribution of eggs and nymphs in naturally occurring psyllid populations was highly aggregated, resulting from initially aggregated migration of adults and a contagious dispersion of them on flushes as the population density increased.

KEYWORDS: Asian citrus psyllid, *Diaphorina citri*, honey tangerine, *Citrus aurantium*, spatial distribution, statistical analysis.

INTRODUCTION

The Asian citrus psyllid (ACP), *Diaphorina citri* Kuwayama, 1908 (Hemiptera: Psyllidae), is infamously known as a key insect pest of citrus because it transmits “*Ca. Liberibacter asiaticus*”, Gram-negative bacteria causing greening disease or huanglongbing (HLB) (Bové 2006). Beattie and Holford (2008) stated that HLB detected in the year after 1970 posed a destructive threat to the Malaysian citrus industry. Study of the spatial distribution of *D. citri* within Malaysian citrus plantations is critical for understanding biology and ecology of the psyllid, as well as for development of the pest management plans.

Control of *Diaphorina citri* with pesticides relies on studies of relationships between population density and spatial distribution pattern of the vector in major citrus growing countries, and dispersion indices calculated for eggs, nymphs and adults in *D. citri* populations underlie effective sampling plans for the pest management. Investigations of this kind were undertaken for various citrus species in China (Yang 2006), Taiwan (Wang 1981), India (Dharajothi *et al.* 1989; Aruna &

Jagginavar 2017), USA (Sétamou *et al.* 2008), Malaysia (Soemargono *et al.* 2008) and Brazil (Costa *et al.* 2010; Beloti *et al.* 2013), with no published information on dispersal of *D. citri* on honey tangerine (*Citrus aurantium*) in Asia. Thus, results of the present endeavour dealing with the spatial distribution pattern of *D. citri* in the Malaysian citrus are quite novel, and while the study is linked to other investigations elsewhere in Asia, our methodological approach is radically different and the results are influential.

Several parameters—including the mean–variance test, the index of dispersion (ID), and *K* value of the negative binomial distribution—were used to evaluate and outline the dispersion trend of *D. citri* eggs, nymphs and adults. To further ascertain and confirm the distribution pattern in a population, dispersion indices were used, and these included the index of mean crowding, Lloyd's index of patchiness, Taylor's power law and Iwao's patchiness regression (Taylor 1961, 1984; Lloyd 1967; Iwao 1977a; Southwood 1978).

The objective of our study was to elucidate seasonality in the spatial distribution pattern of *D. citri* populations in the field.

MATERIALS AND METHODS

Experimental sites

The study was carried out between April 1998 and December 1999 in a 1 ha commercial citrus orchard at Jemukan (1°33'N 110°41'E), Kota Samarahan, Sarawak, Malaysia. The plot housed 200 grafted non-bearing, PCR-certified disease-free honey tangerine (*C. aurantium*) seedlings planted in 1996 in 25 rows (N-S) in 8 columns on a 5.3×5.3 m matrix. The citrus trees developed quickly and reached 2–3 m height with relatively open canopies at the commencement of the study. The trees received sprays containing 0.35% nC24 D-C-Tron Plus and when necessary mancozeb M80 @ 0.15% a.i. during the study period to control mite infestation and fungal disease on the foliage. All other practices were in accordance with the farmer's normal schedule. The experimental site was divided into 10 blocks with 20 trees each.

Sampling and assessments

Stratified random sampling was adopted in this study and counts of adults, eggs and nymphs were made at weekly intervals. In this study, flush (immature shoot) was selected as the sample unit and 100 random samples of young flushes were collected weekly from the 200 trees for 79 consecutive weeks. On each occasion, ten young flushes were sampled randomly from each block. Each flush was about 10–15 mm long with five selected young leaves. Eggs, nymphs and adults present on each flush were counted with the aid of a 10× hand lens.

The data were subjected to the analysis of dispersion parameters and also to different dispersion indices. *Diaphorina citri* counts obtained through weekly sampling between April 1998 and December 1999. The GENSTAT 5 computer package was used for the analysis of the spatial dispersal of *D. citri* populations.

The following statistical approaches were used in the present study.

(1) The index of dispersion was calculated as $ID = s^2/x$, where s^2 is the sample variance and x equals the mean number of nymphs or adults per test sample. Aggregated populations would have $ID > 1$ (Elliott 1977).

(2) The negative binomial distribution $K = x^2/s^2 - x$ is an extension of the Poisson series and is defined by two parameters as the arithmetic mean (x) and a positive exponent K , which was used as an index of aggregation for populations by Waters and Henson (1959): K approaches infinity with increasing randomness, and zero as the negative binomial approaches the logarithmic series. The smaller the estimation of K , the more prominent the degree of aggregation, whereas estimations of $K \gg 8$ demonstrate a random dispersion trend (Southwood 1978). Most estimations of K are in the region of 2.

(3) The index of mean crowding (m^*) was calculated as $m^* = m + (s^2/m)$ (Lloyd 1967): Random dispersion trend are shown by values in which m^* are close to m , while m^* values approaching $m^* + m/K$ indicate aggregated trends.

(4) Lloyd's index of patchiness was determined as $P = 1 + (s^2 - m)/m^2$. It corresponds to the relative magnitude of spatial, quadrat-to-quadrat, variations of the population density. The insect population's tendency to aggregate was measured by Lloyd's index of patchiness (m^*/m), where m^* is the mean crowding index while m is the mean density of the species.

(5) Subsequently finding the distribution trend from the dispersion indices, spatial conveyance trends were compared by applying Taylor's (1961) power law $s^2 = ax^b$ (where x is mean, s^2 is variance, the constant a represents a sampling factor, and the constant b represents the true index of aggregation characteristic of a species) and Iwao's patchiness regression (Iwao 1968).

(6) For each data set (one fitted to a Poisson distribution and the other fitted to a negative binomial distribution), separate regressions were calculated for 1998 and 1999 years. The regression of $\log s^2$ on $\log x$ showed Taylor's power law parameter and the value b from the equation $\log s^2 = \log a + b \log x$ (Taylor 1961). The index b is specific and only a is affected by sampling procedure. Values of $b > 1$ indicate aggregated distributions of populations. Random distribution is indicated by insignificant deviation from 1.

(7) The outcomes were then compared with those of Iwao's patchiness regression. Iwao's patchiness model parameters α and β were obtained by linear m^*/m regression ($m^* = \beta m + \alpha$; Iwao (1968)). The parameter β is the index of aggregation in the *D. citri* population; the conveyance is contagious if $\beta > 1$, and the essential parts of the dispersion are randomly circulated if $\beta = 1$ (Iwao 1977b).

(8) The mean number of individuals in aggregations was determined by applying Arbous and Kerrich's (1951) formula $\lambda = (x/2k)v$, where x is mean, v is function with χ^2 distribution with $2k$ degrees of freedom, and λ is the quantity of individuals in an accumulation for the probability level apportioned to v , based on use of the mean size of the 'aggregate' value at the 0.5 probability. When $\lambda < 2$, clumping is

likely to be caused by ecological components, whereas $\lambda > 2$ indicates that either factor might be the reason (Southwood 1978).

RESULTS

Spatial distribution of eggs

Changes of the egg distribution patterns are reflected in the changes of the mean–variance relationship. The variance surpassed the mean for every data sets, Lloyd's mean crowding values (m^*) exceeded the mean, and the *ID* values were greater than 1 in 1998 (Table 1). The highest average values of 1.04 and 1.44 eggs per flush were recorded during September 1998 and February 1999, respectively. The egg population counts therefore followed a contagious distribution meaning that the population was aggregated (Table 1). Lloyd's mean crowding values (m^*) surpassed the mean density and therefore Lloyd's index of patchiness (m^*/m) turned out to be significantly more than unity in every case, which also confirmed the aggregation pattern (Table 1).

The causes of aggregation for the population related by the negative binomial dispersion were described by the mean clump size (λ). Among the data sets, the clump size varied both below and above 2 with average values of 2.12, and 3.87 for 1998 and 1999, respectively. Thirteen out of 20 data sets for egg populations had $\lambda > 2$ (Table 1). Taylor's power law was not well fitted for the egg distribution in view of the linear relationship amongst variance and mean density and the value of $b = -29.3$ for the 1998–1999 seasons (Fig. 1). To understand the dispersion further, Iwao's patchiness regression ($m^* = \beta m + \alpha$) was fitted, followed by linear relationship among the mean crowding index and mean density, over a range of

Table 1. Aggregation and spatial distribution of Asiatic citrus psyllid eggs. Abbreviations: *m* – mean, *Var* – variance, *K* – estimation by maximum likelihood method (Bliss & Fisher 1953), *ID* – index of dispersion, m^* – mean crowding, m^*/m – Lloyd's index of patchiness, λ – clump size.

| Month | <i>m</i> | <i>Var</i> | <i>K</i> | <i>ID</i> | m^* | m^*/m | λ |
|-------|----------|------------|----------|-----------|-------|---------|-----------|
| 1998 | | | | | | | |
| Apr | 0.62 | 3.12 | 0.16 | 10.23 | 8.85 | 28.60 | 1.82 |
| Aug | 0.40 | 4.53 | 0.04 | 11.33 | 10.50 | 26.26 | 2.25 |
| Sept | 1.64 | 30.90 | 0.09 | 48.17 | 48.17 | 113.84 | 11.43 |
| Oct | 1.12 | 9.30 | 0.16 | 22.07 | 19.89 | 71.23 | 3.57 |
| 1999 | | | | | | | |
| Jan | 0.54 | 8.98 | 0.04 | 14.99 | 25.00 | 90.36 | 6.07 |
| Feb | 1.44 | 72.80 | 0.03 | 50.56 | 51.00 | 34.42 | 10.80 |
| Jun | 0.68 | 8.32 | 0.09 | 14.80 | 13.51 | 44.14 | 3.11 |
| Jul | 0.26 | 3.38 | 0.02 | 12.99 | 12.00 | 46.15 | 2.93 |
| Aug | 0.34 | 5.78 | 0.02 | 16.99 | 16.33 | 47.06 | 3.83 |
| Sept | 1.04 | 21.20 | 0.06 | 32.95 | 31.53 | 71.58 | 6.62 |
| Oct | 0.40 | 8.10 | 0.02 | 20.39 | 19.00 | 4.15 | 4.50 |
| Nov | 0.36 | 6.48 | 0.02 | 18.00 | 20.00 | 47.62 | 4.73 |

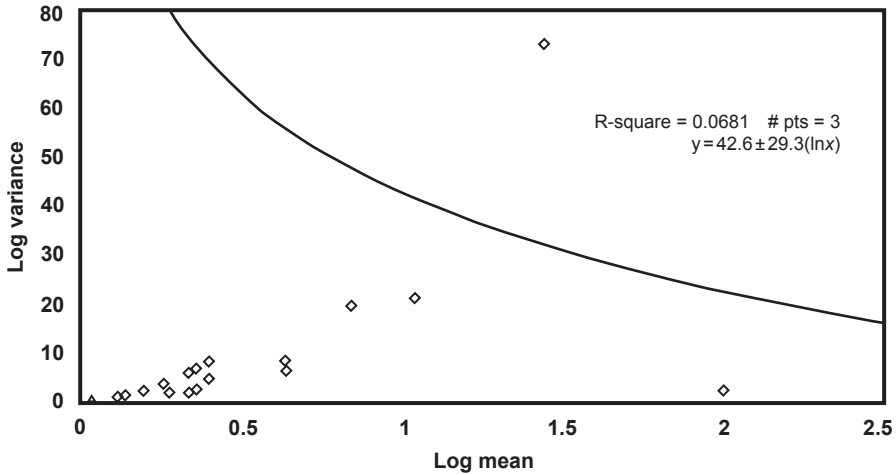


Fig. 1: Taylor's Power Law. Log variance / log mean of *D. citri* egg frequency distribution in 1998–1999 season with pooled value of $b = -29.3$.

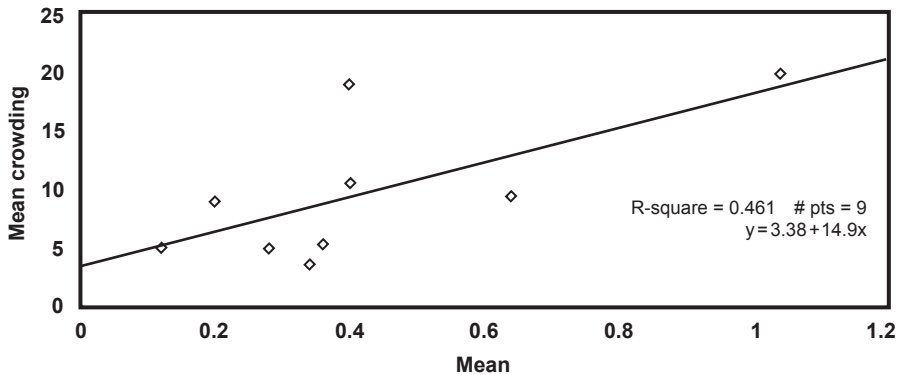


Fig. 2: Iwao's patchiness regression. Series m^* - m relationship of *D. citri* egg in field in 1998 with the value of $\alpha = 3.38$ and $\beta = 14.9$.

different densities; $m^* = 14.9m + 3.38$ in 1998, and $m^* = 30.5m + 3.29$ in 1999. The α values for the egg populations in the field in both 1998 and 1999 were fairly close to each other but greater than one. This confirmed the aggregation among egg populations with stronger aggregation tendency (Figs 2, 3).

Spatial distribution of nymphs

The variance and mean crowding values (m^*) were greater than the mean for all data sets (Table 2). Similarly, the exponent K ranged from 0.04–1.69. These values indicated that nymph populations followed a contagious distribution. The values of Iwao's patchiness regression, $m^* = 8.3m + 2.04$ in 1998 and $m^* = 2.88m + 7.72$ in

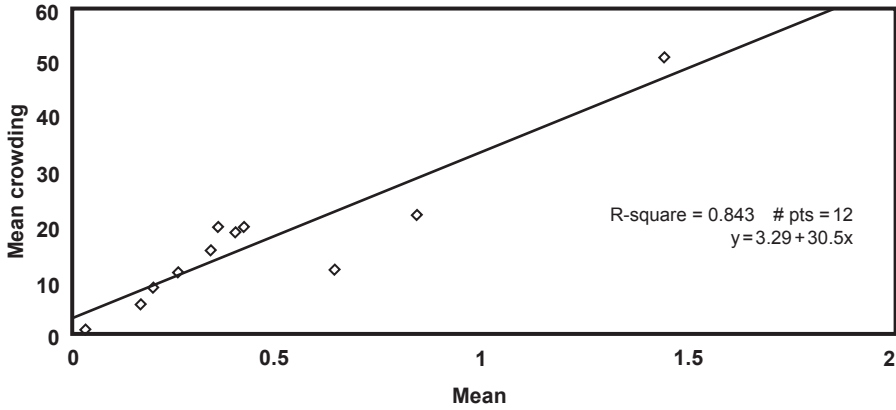


Fig. 3: Iwao’s patchiness regression. Series m^* - m relationship of *D. citri* egg in field in 1999 with the value of $\alpha=3.29$ and $\beta=30.5$.

1999 (Figs 4, 5), further confirmed the aggregated distribution. The α value of 7.72 in 1999 was greater than that of 2.04 in 1998. In 28 out of 70 (40%) data sets, λ for nymph population was <2 (Table 2). For Taylor’s power law, the b was 2.15 and 2.6 in 1998 and 1999, respectively (Figs 6, 7). This outcome also confirmed aggregated distributions for nymphs, as the values for b were greater than 1.

Table 2. Aggregation and spatial distribution of Asiatic citrus psyllid nymphs. Abbreviations: m – mean, Var – variance, K – estimation by maximum likelihood method (Bliss & Fisher 1953), ID – index of dispersion, m^* – mean crowding, m^*/m – Lloyd’s index of patchiness, λ – clump size.

| Month | m | Var | K | ID | m^* | m^*/m | λ |
|-------|-------|--------|------|-------|--------|---------|-----------|
| 1998 | | | | | | | |
| Apr | 2.48 | 15.51 | 0.61 | 26.19 | 19.36 | 30.89 | 5.51 |
| May | 0.64 | 1.50 | 0.04 | 17.97 | 17.61 | 26.95 | 3.60 |
| Jun | 1.42 | 8.69 | 0.39 | 26.41 | 18.33 | 73.35 | 4.86 |
| Jul | 0.08 | 0.61 | 0.11 | 7.64 | 4.67 | 15.16 | 1.28 |
| Dec | 1.64 | 17.77 | 0.20 | 38.66 | 35.53 | 90.48 | 7.44 |
| 1999 | | | | | | | |
| Jan | 1.22 | 14.46 | 0.14 | 25.87 | 22.58 | 90.48 | 7.44 |
| Feb | 0.64 | 5.83 | 0.08 | 9.10 | 8.56 | 13.37 | 1.80 |
| Mar | 5.20 | 99.94 | 0.40 | 79.89 | 78.50 | 90.99 | 18.85 |
| Apr | 1.72 | 12.63 | 0.27 | 7.35 | 7.92 | 4.60 | 1.43 |
| May | 14.42 | 362.13 | 0.62 | 95.38 | 102.91 | 89.39 | 40.17 |
| Jun | 9.54 | 211.68 | 1.02 | 66.42 | 70.91 | 24.98 | 84.59 |
| Jul | 10.96 | 303.04 | 0.44 | 56.44 | 63.25 | 29.01 | 17.42 |
| Aug | 26.06 | 576.76 | 1.69 | 95.36 | 115.11 | 76.41 | 24.07 |
| Sept | 5.24 | 39.52 | 0.83 | 30.78 | 31.42 | 29.67 | 6.16 |
| Oct | 2.74 | 17.71 | 0.64 | 20.58 | 18.93 | 40.27 | 5.03 |
| Nov | 1.04 | 7.03 | 0.39 | 26.49 | 22.04 | 92.87 | 4.30 |
| Dec | 2.18 | 25.59 | 0.34 | 43.54 | 40.86 | 80.22 | 9.11 |

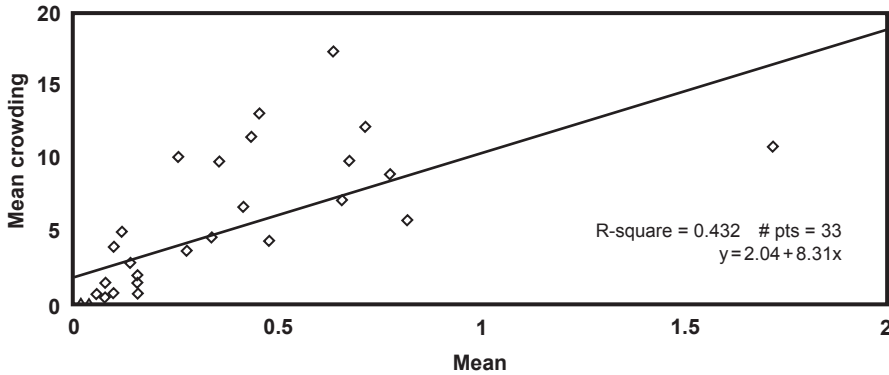


Fig. 4: Iwao's patchiness regression. Series m^* - m relationship of *D. citri* nymphs in 1998 with the value of $\alpha=2.04$ and $\beta=8.31$.

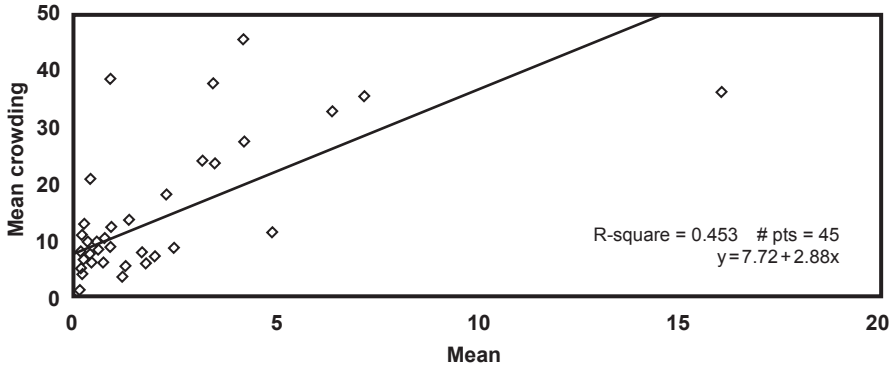


Fig. 5: Iwao's patchiness regression. Series m^* - m relationship of *D. citri* nymphs in 1999 with the value of $\alpha=7.72$ and $\beta=2.88$.

Spatial distribution of adults

The variance s^2 was greater than the mean x in 65% and 87% of samples in 1998 and 1999, respectively (Table 3). This indicated that the distributions were mostly aggregated and was confirmed by the K values that ranged from 0.10–0.17 in 1998, and 0.01–0.33 in 1999; $ID > 1$ and mean crowding values m^* exceeding mean density in 83% of data sets. This suggests that the pest was more active in 1999. The Iwao's patchiness regression was calculated with $m^* = 23m - 0.74$ and $m^* = 11.7m + 2.52$ in 1998 and 1999, respectively (Figs 8, 9). The value of the index of basic contagion (α) was -0.74 (i.e. < 0) in 1998 and 2.52 (i.e. > 0) in 1999. The coefficient of density-contagiousness for 1999 was greater than 1, and this confirmed aggregation among adult colonies. The b value was 7.5 and 0.81 for 1998 and 1999 respectively in applying Taylor's power law (Figs 10, 11). These values showed an initial aggregated distribution for 1998 and contagious distribution of adults on flushes for 1999 as population densities increased.

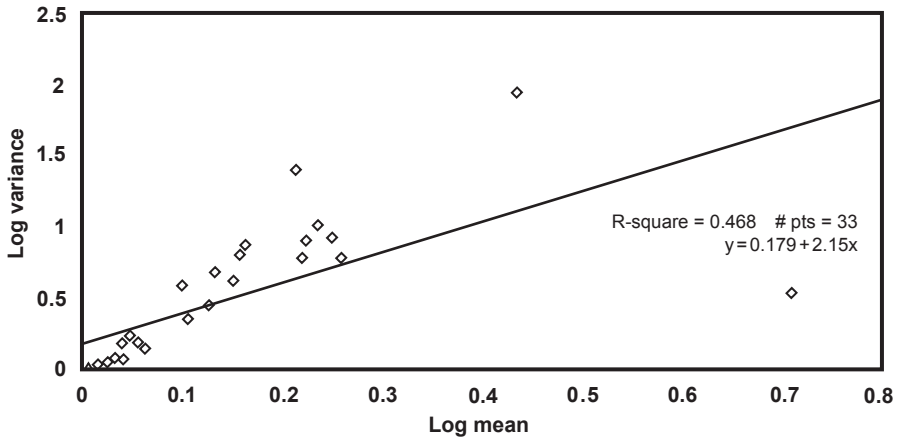


Fig. 6: Taylor's Power Law. Log variance / log mean of *D. citri* nymphs frequency distribution in 1998 with the value of $b=2.15$.

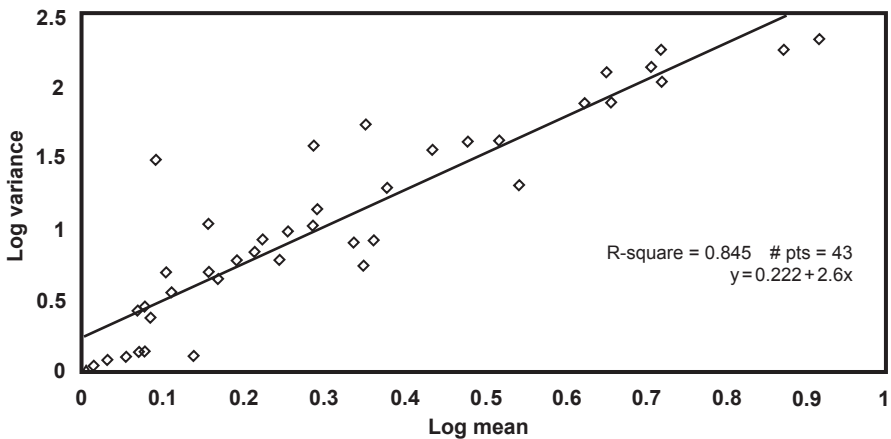


Fig. 7: Taylor's Power Law. Log variance / log mean of *D. citri* nymphs frequency distribution in 1999 with the value of $b=2.6$.

DISCUSSION

Results pertaining to the different dispersion parameters and dispersion indices (Tables 1–3; Figs 1, 2, 4–11) favour the aggregated distribution pattern of the *D. citri* population. This corroborates the aggregative nature of dispersion of *D. citri* population in the field as described by Southwood (1978). This basic test has been used to evaluate the distribution pattern of insect pest populations in different crops (e.g. Wang 1981; Dharajothi *et al.* 1989).

The spatial distribution of eggs and nymphs is an outcome of the movement trends of gravid females to oviposition sites as *D. citri* only disperses in the adult

Table 3. Aggregation and spatial distribution of Asiatic citrus psyllid adults. Abbreviations: m – mean, Var – variance, K – estimation by maximum likelihood method (Bliss & Fisher 1953), ID – index of dispersion, m^* – mean crowding, m^*/m – Lloyd's index of patchiness.

| Month | m | Var | K | ID | m^* | m^*/m |
|-------|------|-------|-------|-------|-------|---------|
| 1998 | | | | | | |
| Apr | 1.00 | 3.52 | 2.13 | 10.40 | 7.53 | 23.22 |
| May | 0.36 | 0.62 | 0.26 | 3.95 | 1.31 | 5.58 |
| Jun | 0.26 | 0.45 | 0.10 | 5.86 | 2.12 | 23.60 |
| Jul | 0.04 | 0.04 | - | 1.98 | 0.02 | 0.38 |
| Aug | 0.15 | 0.39 | 0.34 | 4.27 | 2.51 | 37.82 |
| Sept | 0.54 | 13.52 | 0.02 | 25.04 | 24.58 | 44.57 |
| Oct | 0.42 | 3.11 | 0.07 | 7.40 | 6.67 | 15.88 |
| Nov | 0.20 | 1.06 | 0.05 | 5.30 | 4.40 | 22.00 |
| 1999 | | | | | | |
| Jan | 0.44 | 1.03 | 0.42 | 7.23 | 4.52 | 36.17 |
| Feb | 0.42 | 1.03 | 0.16 | 12.78 | 8.94 | 72.62 |
| Mar | 4.02 | 77.3 | 0.28 | 82.31 | 79.69 | 111.58 |
| Apr | 1.00 | 11.51 | 0.25 | 17.11 | 14.77 | 38.53 |
| May | 7.76 | 49.8 | 1.71 | 30.22 | 32.37 | 27.92 |
| Jun | 4.12 | 40.11 | 1.77 | 28.58 | 31.41 | 24.50 |
| Jul | 4.44 | 22.90 | 1.59 | 13.95 | 15.07 | 9.73 |
| Aug | 7.88 | 26.30 | 11.93 | 17.28 | 19.82 | 13.29 |
| Sept | 3.80 | 4.47 | 10.67 | 5.95 | 4.96 | 7.49 |
| Oct | 1.90 | 5.11 | 1.79 | 13.38 | 10.9 | 35.43 |
| Nov | 0.26 | 0.41 | 0.51 | 7.33 | 2.33 | 36.08 |
| Dec | 0.32 | 0.34 | 0.48 | 3.14 | 0.33 | 2.75 |

stage of its life cycle (Aurambout *et al.* 2009). Our studies on the spatial distribution of *D. citri* populations in the Malaysian honey tangerine orchard showed that both the eggs and nymphs followed a contagious distribution (Tables 1–3; Figs 1–5) on flushes within trees and that the distribution could be expressed as a negative binomial distribution (Bliss & Owen 1958). The dispersion is clarified by two parameters, the mean and the value K which is a measure of clumping. The trend was reflected by the exponent K with values ranging from 0.02–0.16 in the different sets of data. The dispersion parameter K was generally less than 2, which indicated that the egg populations were excessively clumped or aggregated ($K < 1$). The contagious distribution of eggs on flushes indicated that female adults tend to lay eggs on trees on which they land before searching for a new host plant. These data also suggested a preference of females for 4 to 7 day-old (3–10 mm long) flushes for oviposition rather than older and longer flushes (Leong *et al.* 2010). The values of the index of basic contagion (α) were 3.38 and 3.29, respectively for the egg populations in 1998 and 1999 (Figs 2, 3), indicating positive associations between individuals in the colonies. Because the coefficient of density-contagiousness (β) was greater 1, the aggregation patterns among the colonies were confirmed.

The cause of aggregation for the populations was explained by the mean clump size (λ). As this value fell both below and above 2 (Tables 1–3), it reflected the

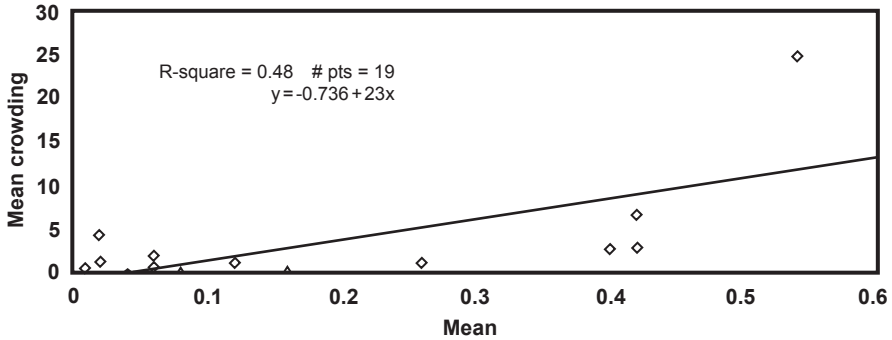


Fig. 8: Iwao's patchiness regression. Series m^*-m relationship of *D. citri* adults in 1998 with the value of $\alpha = -0.74$ and $\beta = 23$.

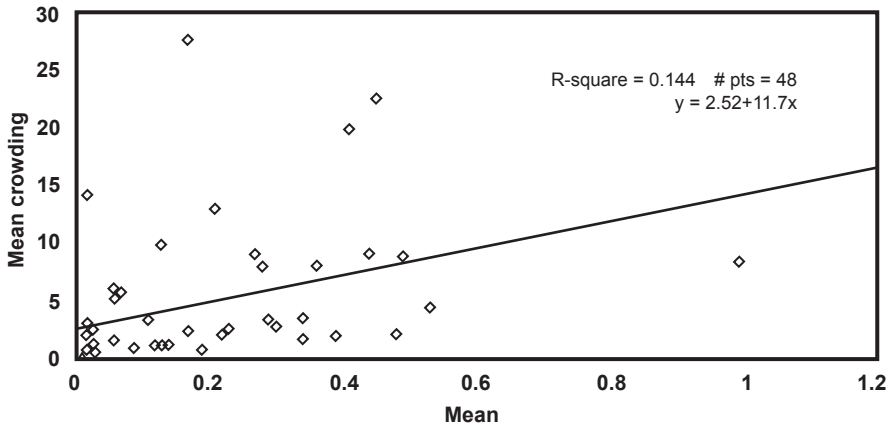


Fig. 9: Iwao's patchiness regression. Series m^*-m relationship of *D. citri* adults in 1999 with the value of $\alpha = 2.52$ and $\beta = 11.7$.

role of both environment and oviposition behaviour in the aggregation pattern of eggs. Changes in distribution of nymphs on flushes associated with fluctuating population must be attributed to the movement of adults over time (nymphs cannot disperse from flush to flush), although females always tended to lay eggs contagiously, as indicated by the aggregated distribution of eggs. However, Martini *et al.* (2015) reported that female psyllids avoided conspecific female cues in a density-dependent manner and the conspecific females were attracted to odours from citrus plants actively damaged by psyllid feeding. They initiated dispersion when population density increased, resulting in a change from the initially contagious to almost random dispersion on infested citrus.

Several indices have been suggested to measure the distribution trend of animal populations obtained based on quadrat counts (Southwood 1978). Although pa-

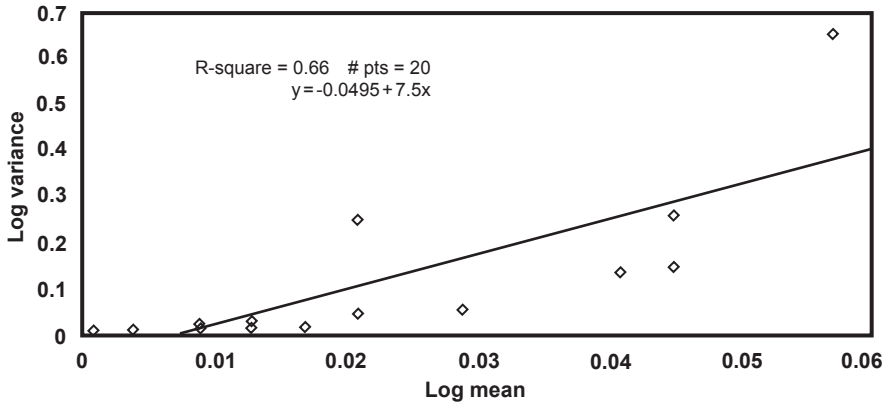


Fig. 10: Taylor's Power Law. Log variance / log mean of *D. citri* adults frequency distribution in 1998 with the value of $b=7.5$.

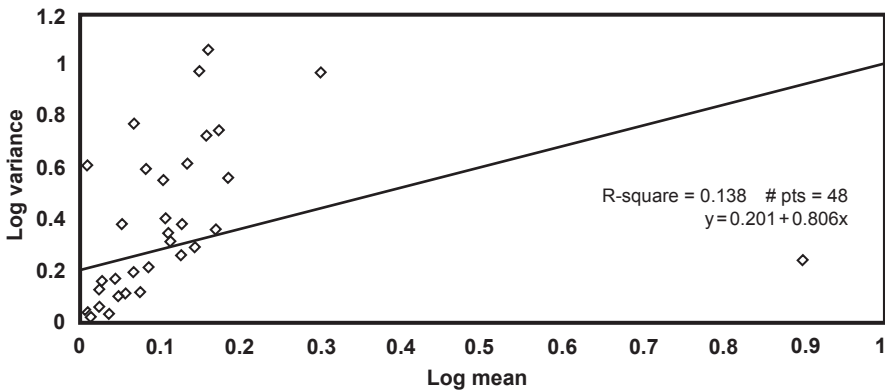


Fig. 11: Taylor's Power Law. Log variance / log mean of *D. citri* adults frequency distribution in 1999 with the value of $b=0.81$.

parameter K and ID are widely used as functional measures of aggregation in animal populations, they are generally inadequate to evaluate the kind and degree of aggregation. However, Iwao's patchiness regression ($m^* = \beta m + \alpha$) can test the dual nature of aggregation, basic contagion (α) values concerned with the clustering habit or aggregation characteristics, and aggregation tendency or distribution pattern of clusters. As regards the index of the aggregation β in the *D. citri* population, the conveyance is contagious when $\beta > 1$, and the essential parts of the dispersion are randomly circulated when $\beta = 1$. The aggregated distribution of infested citrus flushes is diagnosed when both b and β are greater than 1. The intercept α and β of the regression therefore appears to be valuable as an index to depict diverse aspects of the scattering pattern of *D. citri* populations. Although α values (Figs 2–5,

8, 9) were lower for adults than for eggs and nymphs, positive intercepts ($\alpha > 0$) indicated that generally more than one egg, nymph or adult could be found on a flush. This may be related to the behaviour of females, which tend to oviposit on young flushes, especially during flushing cycle, and distributions of eggs being dependent on the densities of flushes within the tree.

The positive value of α suggests a preference for high value resources such as young flushes. It also suggests that individuals are prone to live together in discrete quadrats, even at a low density. Distribution of nymphs per flush were clearly related to the original clumped distribution of eggs distribution, as α and β were 3.38 and 14.9, and 3.29 and 30.5, respectively in 1998 and 1999 (Figs 2, 3). This indicated a strong aggregation tendency.

The observed negative value of α (-0.74) for adults (Fig. 8) may be attributed to the behaviour that allows females either to avoid oviposition on flushes on which they encounter eggs laid earlier by other females during abundant flush growth, or by forcing them to lay eggs on flushes with eggs during non-flushing periods. Martini *et al.* (2015) investigated effects of chemical cues derived from females *D. citri* and their host plants on host acceptance choices by conspecific individuals. They reported that psyllids avoided conspecific female cues in a density-dependent manner and the conspecific females were attracted to odours from citrus plants actively damaged by psyllid feeding. Such behaviour would result in a less uniform egg distributions. The intercept α values or index of basic contagion was lower in the adult populations than in the egg and nymph populations for 1998 and 1999. This suggests that the basic component of the adult population was smaller than that of the egg and nymph populations. The changeable and complex environment in the orchard led to marked fluctuations of egg, nymph and adults populations. This could have been related to a larger migration space and higher mortality of *D. citri* in a natural environment (Wang 1981). The overall psyllid distribution pattern is defined by the dispersal of adults, the initial distribution of eggs, and nymphal mortality (Michaud 2004).

Taylor's power law usually indicates that the degree of aggregation increases with the population density. Adults were observed to have aggregated distribution ($b=7.5$) in 1998 (Fig. 10) but almost random distribution ($b=0.806$) in 1999 (Fig. 11), possibly due to migration or dispersal. The dispersion of adults is density-related. They disperse when the population density increases and aggregate when population density declines (Sétamou *et al.* 2008). The dispersion behaviour minimises intraspecific competition for resources.

Although Lloyd's index of patchiness and index of mean crowding (Tables 1–3) decreased with increasing mean density, they were useful for determining the dispersion pattern of *D. citri* populations. Taylor's power law and Iwao's patchiness regression (Figs 4–7, 10, 11) provided good indications of the relationship amongst variance and mean density of nymphs and adults per mean density of flushes, but Taylor's power law did not work well with the egg populations (Fig. 1). The intercept α and slope β of the Iwao's patchiness regression were more

appropriate for measuring the aggregation tendency in terms of α and β rather than by Lloyd's index of patchiness (m^*). Both α and β were integral in clarifying the scattering pattern of *D. citri* and were useful as indices for clarifying diverse aspects of dispersion trend of the *D. citri* population. Lloyd's index of mean crowding and index of patchiness were easy to use and provided basic information on spatial distribution patterns.

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REFERENCES

- ARBOUS, A.G. & KERRICH, J.E. 1951. Accident statistics and the concept of accident-proneness. *Biometrics* **7** (4): 340–432.
<https://doi.org/10.2307/3001656>
- ARUNA, J. & JAGGINAVAR, S.B. 2017. Seasonal abundance of citrus psyllid, *Diaphorina citri* Kuwayama on acid lime. *Journal of Experimental Zoology, India* **20** (Suppl. 1): 1511–1513.
- AURAMBOUT, J.P., FINLAY, K.J., LUCK, J. & BEATTIE, G.A.C. 2009. A concept model to estimate the potential distribution of the Asiatic citrus psyllid (*Diaphorina citri* Kuwayama) in Australia under climate change—A means for assessing biosecurity risk. *Ecological Modelling* **220**: 2512–2524.
<https://doi.org/10.1016/j.ecolmodel.2009.05.010>
- BEATTIE, G.A.C. & HOLFORD, P. 2008. Current HLB situation and industry perspective in Asia. In: *Proceeding of International research conference on huanglongbing*. Orlando, Florida, pp. 94.
- BELOTI, V.H., RUGNO, G.R., FELIPPE, M.R., DO CARMO-UEHARA, A., GARBIM, L.F., GODOY, W.A.C. & YAMAMOTO, P.T. 2013. Population dynamics of *Diaphorina citri* Kuwayama (Hemiptera: Liviidae) in orchards of 'Valencia' orange, 'Ponkan' mandarin and 'Murcott' tangor trees. *Florida Entomologist* **96** (1): 173–179.
<https://doi.org/10.1653/024.096.0123>
- BLISS, C.I. & FISHER, R.A. 1953. Fitting the negative binomial distribution to biological data, and note on the efficient fitting of the negative binomial. *Biometrics* **9** (2): 176–200.
<https://doi.org/10.2307/3001850>
- BLISS, C.I. & OWEN, A.R.G. 1958. Negative binomial distributions with a common K . *Biometrika* **45** (1–2): 37–58.
<https://doi.org/10.1093/biomet/45.1-2.37>
- BOVÉ, J.M. 2006. Huanglongbing: a destructive, newly-emerging, century-old disease of citrus. *Journal of Plant Pathology* **88** (1): 7–37.
<http://www.sipav.org/main/jpp/index.php/jpp/article/view/828> (accessed 18 June 2018)
- COSTA, M.G., BARBOSA, J.C., YAMAMOTO, P.T. & LEAL, R.M. 2010. Spatial distribution of *Diaphorina citri* Kuwayama (Hemiptera: Psyllidae) in citrus orchards. *Scientia Agricola* **67** (5): 546–554.
<http://dx.doi.org/10.1590/S0103-90162010000500008>
- DHARAJOTHI, B., VERGHESE, A. & TANDON, P.L. 1989. Ecological studies on citrus psylla, *Diaphorina citri* Kuwayama (Hemiptera: Psyllidae) with special reference to its spatial distribution and sampling plan. *Entomon* **14** (3–4): 319–324.
<https://swfrec.ifas.ufl.edu/hlb/database/pdf/Dharajothi%20et%20al.%201989.pdf>
- ELLIOTT, J.M. 1977. *Some methods for the statistical analysis of samples of benthic invertebrates*. 2nd Ed. Freshwater Biological Association, Ambleside, England. 160 pp.
- IWAO, S. 1968. A new regression method for analyzing the aggregation pattern of animal populations. *Researches on Population Ecology* **10**: 1–20.

- 1977a. The m^*-m statistics as a comprehensive method for analyzing spatial patterns of biological populations and its application to sampling problems. In: Morisita, M. (Ed.), *Studies on methods of estimating population density, biomass and productivity in terrestrial animals*. Vol. 17. JIBP Synthesis, Tokyo, pp. 21–46.
- 1977b. Analysis of spatial association between two species based on the interspecies mean crowding. *Researches on Population Ecology* **18**: 243–260.
- LEONG, S.C.T., FATIMAH, A., BEATTIE, A., KUEH, J.H. & WONG, S.K. 2010. Influence of host plant species and flush growth stage on the Asian citrus psyllid, *Diaphorina citri* Kuwayama. *American Journal of Agricultural and Biological Sciences* **6** (4): 536–543.
<https://doi.org/10.3844/ajabssp.2011.536.543>
- LLOYD, M. 1967. “Mean crowding”. *Journal of Animal Ecology* **36** (1): 1–30.
<https://doi.org/10.2307/3012>
- MARTINI, X., HOFFMANN, M., COY, M.R., STELINSKI, L.L. & PELZ-STELINSKI, K. 2015. Infection of an insect vector with a bacterial plant pathogen increases its propensity for dispersal. *PLoS One* **10** (6): Art. e0129373.
<https://doi.org/10.1371/journal.pone.0129373>
- MICHAUD, J.P. 2004. Natural mortality of Asian citrus psyllid, *Diaphorina citri* (Homoptera: Psyllidae) in Central Florida. *Biological Control* **29** (2): 260–269.
[https://doi.org/10.1016/S1049-9644\(03\)00161-0](https://doi.org/10.1016/S1049-9644(03)00161-0)
- SÉTAMOU, M., FLORES, D., FRENCH, J.V. & HALL, D.G. 2008. Dispersion patterns and sampling plans for *Diaphorina citri* (Hemiptera: Psyllidae) in citrus. *Journal of Economic Entomology* **101** (4): 1478–1487.
<https://doi.org/10.1093/jee/101.4.1478>
- SOEMARGONO, A., IBRAHIM, Y., IBRAHIM, R. & OSMAN, M.S. 2008. Spatial distribution of the Asian citrus psyllid, *Diaphorina citri* Kuwayama (Homoptera: Psyllidae) on citrus and orange jasmine. *Journal of Biosciences* **19** (2): 9–19.
http://www.tlsr.usm.my/tlsr19022008/19022008_02.pdf (accessed 18 June 2018)
- SOUTHWOOD, T.R.E. 1978. *Ecological methods. With particular reference to the study of insect populations*. 2nd Ed. Chapman and Hall, London, New York. 524 pp.
<https://doi.org/10.1007/978-94-009-1225-0>
- TAYLOR, L.R. 1961. Aggregation, variance and the mean. *Nature* **189**: 732–735.
<https://doi.org/10.1038/189732a0>
- 1984. Assessing and interpreting the spatial distributions of insect populations. *Annual Review of Entomology* **29**: 321–357.
<https://doi.org/10.1146/annurev.en.29.010184.001541>
- WANG, C.L. 1981. Ecological studies of Asiatic citrus psyllid (*Diaphorina citri* K.) with special reference to its spatial distribution. *Journal of Agricultural Research of China* **30** (4): 412–419.
- WATERS, W.E. & HENSON, W.R. 1959. Some sampling attributes of the negative binomial distribution with special reference to forest insects. *Forest Science* **5** (4): 397–412.
<https://academic.oup.com/forestscience/article-abstract/5/4/397/4763992>
- YANG, Y., HUANG, M., BEATTIE, G.A.C., XIA, Y., OUYANG, G. & XIONG, J. 2006. Distribution, biology, ecology and control of the psyllid *Diaphorina citri* Kuwayama, a major pest of citrus: A status report for China. *International Journal of Pest Management* **52** (4): 343–352.
<https://doi.org/10.1080/09670870600872994>