Open Access

International journal of Horticulture, Agriculture and Food science(IJHAF)
Vol-4, Issue-3, May-Jun, 2020
https://dx.doi.org/10.22161/ijhaf.4.3.4
ISSN: 2456-8635

Examining the spatial distribution pattern and optimum sample size for monitoring of the white mango scale insect, *Aulacaspis tubercularis* (Newstead) (Hemiptera: Diaspididae) on certain mango cultivars

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Abstract— *The white mango scale insect, Aulacaspis tubercularis (Newstead) (Hemiptera: Diaspididae) is one of the most destructive pests of mango trees in Egypt. The main objective of the present work is to estimate the spatial distribution pattern and minimum sample size for monitoring populations of A. tubercularis on six different cultivars of mango through the two successive years of 2017/2018 and 2018/2019 at Esna district, Luxor Governorate, Egypt. Data on the indices of distribution and Taylor's and Iwao's regression analyses indicate significant aggregation behaviour during each year in all the tested cultivars of mango trees, that may be caused by environmental heterogeneity. The regression models of Taylor's power law (b) and Iwao's patchiness (β) were both significantly >1, indicating that A. tubercularis had an aggregation distribution with a negative binomial distribution during each year in all the tested mango cultivars. The Iwao regression coefficients were used to determine the optimum sample size required to estimate populations at three fixed precision levels. The optimum size decreased with increased density in all levels of precision (5, 10 and 15%) in all tested mango cultivars. These can be deployed to develop a sampling plan to estimate the population density accurately. Results suggesting that the optimum sample size was flexible and the precision levels of 5 and 10% were suitable for ecological or insect behavioral studies of A. tubercularis where a higher level of precision is required, whereas, for pest management programs, a 15% level would be acceptable. Furthermore, the distribution, different mango cultivars, and sampling protocol presented here could be used as a tool for future research on pest management methods for this pest.*

Keywords**—Aulacaspis tubercularis, population density, spatial distribution, sample size, mango cultivars.**

I. INTRODUCTION

Mango trees (*Mangifera indica* L.) are subjected to infestation by different pests. Among several pests, infesting mango trees (*Mangifera indica* L.), the white mango scale insect, *Aulacaspis tubercularis* (Newstead) (Hemiptera: Diaspididae) is one of the most destructive pests of mango trees in Egypt. This pest injures the shoots, twigs, leaves,

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branches, and fruits by sucking the plant sap with the mouthparts, causing thereafter deformations, defoliation, drying up of young twigs, dieback, poor blossoming, death of twig by the action of the toxic saliva and so affecting the commercial value of fruits and their export potential especially to late cultivars where it causes conspicuous pink blemishes around the feeding sites **(Nabil *et al*., 2012 and Sayed, 2012).** In nurseries, severe early infestation retards growth. Young trees are particularly vulnerable to excessive leaf loss and death of twigs, during hot dry weather **(El-Metwally *et al*., 2011).** The heavily infested premature fruits dropping and the mature fruits became small in size with lacking juice and were rotted and unfit for commercial use, in addition to reductions in quality and quantity of produced fruits **(Bakr *et al*., 2009 and Abo-Shanab, 2012).**

Spatial distribution is one of the most characteristic properties of insect populations; in most cases, it allows us to define them and is an important characteristic of ecological communities **(Debouzie and Thioulouse 1986).** No field sampling can be efficient without understanding the underlying spatial distribution of the population **(Taylor 1984).** An understanding of the spatial distribution (*i.e.* regular, random, or aggregated) of populations provides useful information, not only for theoretical population biology but also for field monitoring programs, especially sequential sampling **(Feng *et al*., 1993 and Binns *et al*., 2000).** A reliable sampling program for estimating the population density should include a proper sampling time (date of sampling), sampling unit, and the number of samplings in which the determination of spatial distribution is crucial **(Pedigo, 1994; Southwood and Henderson, 2000).**

The regression models of **Taylor (1961) and Iwao (Iwao and Kuno, 1968)** depend on the relationship between the sample mean and variance of insect numbers per sampling unit through time, and can provide a stable relationship from one year to another, based only on the observed sampling mean **(Bisseleua *et al.*, 2011).**

Knowledge of the spatial distribution of an insect is central to the design of any management program, important for understanding the bioecology of species, and forms the basis for developing a sampling protocol **(Wearing, 1988; Binns *et al.*, 2000 and Cho *et al.*, 2001).**

No information is available in the literature regarding the spatial distribution of *A. tubercularis*. Therefore, the present study was undertaken to estimate the optimum sample size for this pest on mango trees. The results of this research can be used to draft monitoring methods for this pest and ultimately to establish pest management program strategies for *A. tubercularis*.

II. MATERIALS AND METHODS

Study area

This study was conducted in a private mango orchard, *Mangifera indica* L., located in Esna district, Luxor Governorate, Egypt. The mango cultivars were Goleck, Balady, Ewaise, Zebda, Sediek and Hindi Bisinnara.The orchard was at an altitude of 99 m a.s.l., a latitude and longitude of 25.67° N and 32.71° E, respectively, and was sampled twice monthly from the beginning of March 2017 until mid-February 2019.

Four mango trees from each cultivar, almost uniform and similar in age (10 years-old), size, height, vegetative growth, received the same horticultural practices (*i.e.* irrigation, fertilisation, and pruning), were selected and labeled. These randomly chosen mango trees did not receive any pesticidal control before and during the period of the study. Regular bimonthly samples of 20 leaves per tree were randomly picked from the terminal shoots of tree. Every sample was placed in a polyethylene bag, all samples were transferred to the laboratory for inspection using a stereo-microscope.

The total numbers of live insects (nymphs and adult females) on the upper and lower surfaces of the mango tree leaves were counted and recorded, linked to the inspection date, and presented as mean number of individuals per leaf ± standard error (SE), to express the population size of pest.

General sampling method:

All sampling was conducted from 23040 leaves on 48 dates over 2-year period, *i.e.* 4 trees × 20 leaves × 6 cultivars × 48 dates. Samples were frozen to preserve them for subsequent processing.

The data obtained were statistically analyzed using the analysis of variance. The means were compared according to Duncan's Multiple Range Test **(Duncan, 1955) and Least Significant Difference test (LSD)** at the 5% level was used to determine the significance among means of cultivars, was carried out by computer **(MSTATC Program software, 1980).**

Spatial distribution:

The spatial distribution among the sample units was determined using twenty two distribution indices and two regression methodologies, namely **Taylor's(1961)** and **Iwao's (Iwao and Kuno, 1968).**

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Distribution indices:

Several estimates are based on sample means and variances, such as index of dispersion, clumping, crowding and Green's index **(Green, 1966).**

- Mean ($\bar{X}$): the mean number of individuals as a general average per leaf during the whole year.

- Range of means of a population: The difference between the maximum mean number of a population and the minimum for the whole year was calculated by applying the following equation:

- Range of Density (R) = Population density maximum – Population density minimum during the entire year.

- Variance (S^2), standard deviation (S), standard error (SE) and median (Me) for samples were determined.

- Coefficient of variance (*C.V.*): To assess the fidelity of sampling, the coefficient of variation values for the studied years were compared.

$$C.V. = \frac{S}{\bar{X}} \times 100$$

Where, *S* is the standard deviation of the mean and $\bar{X}$ is the mean of population.

- Relative Variation (*R.V.*) is employed to compare the efficiency of various sampling methods **(Hillhouse and Pitre, 1974).** The relative variation for the studied years was calculated as follows:

$$R.V. = (SE / \bar{X}) \times 100$$

Where, *SE* is the standard error of the mean and $\bar{X}$ is the mean of population.

- Variance to mean ratio ($S^2/\bar{X}$):

The simplest approach used for determining the insect distribution was variance to mean ratio suggested by **Patil and Stiteler (1974).** The value of variance-to-mean is one for 'Poisson' distribution, less than one for positive binomial and more than one for negative binomial distribution. Dispersion of a population can be classified through a calculation of the variance-to-mean ratio; namely: $S^2/\bar{X}$ = 1 random distribution, < 1 regular distribution, and > 1 aggregated distribution (where, S^2 = sample variance; $\bar{X}$ = mean of population).

- Index of Lewis (I_L):

Lewis index was also calculated as per the formula given hereunder to determine the dispersion of *P. oleae*

$$I_L = \sqrt{S^2 / \bar{X}}$$

The value of this index revealed >1 contagious; <1: regular and =1 random distribution.

- Cassie index (*Ca*):

$$Ca = (S^2 - \bar{X}) / \bar{X}^2$$

The spatial distribution pattern is aggregative, random and uniform when *Ca*>0, *Ca* = 0 and *Ca*<0, respectively **(Cassie, 1962).**

- The *K* value of negative binomial distribution:

The parameter *K* of the negative binomial distribution is one measure of aggregation that can be used for insect species having clumped or aggregated spatial pattern. When *K* values are low and positive (*K*< 2), they indicate a highly aggregated population; *K* values ranging from 2 to 8 indicate moderate aggregation; and values higher than 8 (*K*> 8) indicate a random population **(Southwood, 1995).** The *K* values were calculated by the moment's method **(Costa *et al.*, 2010),** and given by:

$$K = \bar{X}^2 / (S^2 - \bar{X})$$

Departure from a random distribution can be tested by calculating the index of dispersion (I_D), where, n: denotes the number of samples:

$$I_D = (n-1)S^2 / \bar{X}$$

I_D is approximately distributed as $x^2$with *n*-1 degrees of freedom. Values of I_D which fall outside a confidence interval bounded with *n*-1 degrees of freedom and selected probability levels of 0.95 and 0.05, for instance, would indicate a significant departure from a random distribution.

This index can be tested by *Z* value as follows:

$$Z = \sqrt{2I_D} - \sqrt{(2\nu - 1)}$$

v = n - 1

If 1.96 ≥ *Z* ≥ -1.96, the spatial distribution would be random, but if *Z* < -1.96 or *Z* > 1.96, it would be uniform and aggregated, respectively **(Patil and Stiteler, 1974).**

- Index of mean clumping (I_{DM}) **(David and Moore, 1954):**

$$(I_{DM}) = (S^2 / \bar{X}) - 1$$

The David and Moore index of clumping values increase with increasing aggregation. If the index value = 0, the distribution is random, positive value for negative binomial

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(aggregated) and negative value for positive binomial (regular).

- Lloyd's mean crowding ($\overset{*}{X}$):

Mean crowding ($\overset{*}{X}$) was proposed by Lloyd to indicate the possible effect of mutual interference or competition among individuals. Theoretically, mean crowding is the mean number of other individuals per individual in the same quadrate:

$$\overset{*}{X} = \overline{X} + [(S^2 / \overline{X}) - 1]$$

As an index, mean crowding is highly dependent upon both the degree of clumping and population density. To remove the effect of changes in density, Lloyd introduced the index of patchiness, expressed as the ratio of mean crowding to the mean. As with the variance-to-mean ratio, the index of patchiness is dependent upon quadrate size **(Lloyd, 1967).**

- Index of patchiness (I_P): is dependent upon quadrate size.

$$I_P = (\overset{*}{X} / \overline{X})$$

If *IP* = 1 random, < 1 regular and > 1 aggregated

- *Green's index* (*GI*):

$$GI = [(S^2 / \overline{X}) - 1] / (n - 1)$$

This index is a modification of the index of cluster size that is independent of n **(Green, 1966).**

If GI > 0 or positive values are indicative of aggregation dispersion, GI < 0 or negative values indicative of uniformity or regular dispersion, and GI = 0 or negative values closer to 0 indicate randomness.

- To evaluate temporal changes in spatial pattern of *P. oleae* population during the studied years, an aggregation index (1/*k*) **(Southwood and Henderson, 2000)** was used.

It was calculated using the following formula:

$$1/k = (\overset{*}{X} / \overline{X}) - 1$$

where: 1/k is aggregation index or Cassie's index C and ($\overset{*}{X} / \overline{X}$) is Lloyd's patchiness index. The values of 1/*k*< 0, = 0, and > 0 represent regularity, randomness, and aggregation of the population in spatial pattern, respectively **(Feng and Nowierski, 1992).**

- Analysis of causes of aggregation:

The population aggregations mean (λ) **(Blackith, 1961)** was used to analysis the causes for the insect population being in an aggregated state, and was calculated as follows:

$$\lambda = m / 2k \times \gamma$$

Where, y equals to $X^2_{0.5}$ when the value of the degree of freedom is 2*K*. The aggregation of insect individuals is caused by environmental factors when λ < 2; on the other hand, if λ >2, the phenomenon is caused by aggregation behavior or the aggregation behavior works in combination with the environment.

***Regression methods*:**

Taylor's power law

The sampling distributions for the total live stages of *A. tubercularis* were modeled using **Taylor's power law (TPL) (1961).** A power law function can be used to model the relationship between mean and variance as:

$$S^2 = a\overline{X}^b$$

where, S^2 is the variance, $\overline{X}$ is the sample mean, and a is the scaling factor related to sample size. The b measures the species aggregation, and when b = 1, b<1, and b > 1, the distribution is random, regular, and aggregated, respectively.

Using a log transformation, one can estimate the coefficients with linear regression as:

$$\log(s^2) = \log(a) + b\log(\overline{X})$$

where, a and b are the parameters of the model, estimated by linearising the equation after a log–log transformation **(Taylor, 1984).**

Iwao's patchiness regression (IPR):

Iwao's regression method was used to quantify the relationship between the mean crowding index ($\overset{*}{X}$) and mean population density ($\overline{X}$) **Lloyd's (1967)** and using the following equation:

$$\overset{*}{X} = a + \beta\overline{X}$$

where, α is the index of basic contagion and indicates the tendency of crowding (positive) or repulsion (negative), ifa = 0 indicates single individuals, a colony (a > 0) or a negative association of individuals and (0 > a > -1)is the basic component of the distribution. The slope (β) reflects the distribution of the population in space and is interpreted in the same manner as b of the TPL **(Iwao and Kuno, 1968).**

The goodness of fit for both models was evaluated using the coefficients of determination (R^2) and multiple correlations.

A Student's *t*-test can be used to determine whether the colony is composed of single individuals and whether the colonies are dispersed randomly **(Sedaratian *et al.*, 2010 and Moradi-Vajargah *et al.*, 2011).**

Test b = 1, $t = (b - 1)/SE_b$ and Test β = 1, $t = (\beta - 1)/SE_\beta$

where, SE_b and SE_β are the standard errors of the slope for the TPL and Iwao's model, respectively.

Calculated values are compared with tabulated (*t*) values with *n*-1 degrees of freedom **(Feng and Nowierski, 1992).** If the calculated t (t_c)<t-table (t_t), the null hypothesis (b = 1) would be accepted and the spatial distribution would be random. If $t_c > t_t$, the null hypothesis would be rejected and if b > 1 and b < 1, the spatial distribution would be aggregated and uniform, respectively **(Naeimamini *et al.*, 2014).**

The annual data was pooled between the two years and the overall distribution coefficients were used.

The presence or absence of differences between years was calculated based on the following formulas:

$$t_{slope} = -\frac{\beta_1 - \beta_2}{\sqrt{(SE\,\beta_1^{\,2} + SE\,\beta_2)^2}}$$

$$t_{slope} = -\frac{b_1 - b_2}{\sqrt{(SEb_1^{\,2} + SEb_2)^2}}$$

where, b_1 and b_2 were Taylor's coefficient for the two years, β_1 and β_2 were Iwao's coefficient for the two years, and SE_1 and SE_2 were their standard errors with ($n_1 + n_2$)-2 degrees of freedom **(Feng and Nowierski, 1992).** The data of the two years were integrated and a total distribution coefficient was estimated only when the difference between the coefficients of the two years was not significant.

Development of sampling plan (minimum sample size number).

The coefficient from the IPR model was used to determine the minimum sample size requirements for estimating population means for each year with fixed levels of precision. Precision (*D*) was defined as follows:

$$D = \frac{S_{\bar{X}}}{\bar{X}}$$

Where, $S_{\bar{X}}$ is the standard error of the mean and D is a fixed proportion of the absolute mean of the population involved. This is also known as the allowable error, or fixed precision level, with which the mean is measured **(Lindblade *et al.*, 2000).** Estimators with standard errors of 5%, 10%, and 15% at 0.05 probabilities were chosen for the present study.

The number of samples required to estimate the mean with fixed precision was determined by solving the following:

$$\mathrm{n} = \left[(\alpha + 1)/\bar{\mathrm{X}} + (\beta - 1)\right]/C^2$$

Where, a and b are coefficients obtained from IPR. Because Iwao's regression coefficients provided the best explanation for the data, sampling recommendations were developed based on **Kuno's (1969)** formula.

All obtained data were depicted graphically by Microsoft Excel 2010.

III. RESULTS AND DISCUSSION

1- Population density of A. tubercularis on certain mango cultivars:

Data represented in Table (1), revealed that the statistical analysis of data indicated that, there was a highly significant difference among mango cultivars to infestation by *A. tubercularis*. It is clear from the results that the mean maximum population of the pest was observed on Goleck cultivar with an average of 92.51 ± 5.47 and 85.51 ± 4.28 individuals per leaf during the two years, respectively as compared with the other tested cultivars. But, the cultivars of Ewaise, Zebda, Hindi Bisinnara and Sediek were moderately population of the pest with an average of 78.23 ± 4.63, 69.29 ± 3.57; 75.58 ± 4.01, 65.70 ± 2.92; 64.07 ± 3.44, 62.39 ± 3.01 and 59.08 ± 2.89, 51.00 ± 2.17 individuals per leaf during the two years, respectively. However, the minimum individuals of the population were recorded on Balady cultivar with an average of 45.01 ± 2.60 and 37.72 ± 1.82 individuals per leaf through the two years, respectively.

Statistically, highly significant differences among the six tested cultivars regarding the level of infestation were obtained (L.S.D. values were 6.12, 4.32 and 4.89) through the first, second year, and between the two years, respectively. As well, highly significant differences between the first and the second years for *A. tubercularis* population (L.S.D. was 3.39).It is clear from the results that the mean numbers were higher in the first year (69.08 ± 1.84 per leaf) than that of the second year (61.98 ± 1.53 per leaf).

In general, it could be concluded that Goleck mango cultivar was the most preference to white mango scale insect, *A. tubercularis*, followed by Ewaise, then by Zebda and Hindi Bisinnara, while the Balady cultivar was less preferable

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https://dx.doi.org/10.22161/ijhaf.4.3.4
ISSN: 2456-8635

cultivar for this insect.

It is clear that the differences in the population densities of *A. tubercularis* on different mango cultivars which may be due to the differences not only in the environmental conditions (such as temperature, relative humidity) but also there are numerous other factors such as the leaf structure (density of stomata, softness of tissues and size of leaves) as well as growth features (such as growing period) for the tested cultivars of mango.

We concluded that the host plant affects the development of pest and that the choice of the most appropriate cultivar can help to reduce pest infestation, and is therefore an additional component to be included in the integrated pest management of mango.

The degree of infestation on different mango cultivars varied according to insect species. In this respect, **Selim (2002)** in Egypt, however with different insect species, also studied the susceptibility of five mango cultivars to infestation with two armoured scale insect pest, *Insulaspis pallidula* and *Aonidella aurantii* and recorded that *I. pallidula* infested all cultivars more than *A. aurantii*. On the other hand, Hindy cultivar was the most susceptible cultivar to infestation with both scale insects followed by Mabrouka, then by Kobania and Taimour, while the least susceptible cultivar was Dabsha. **Bakry (2009)** in Egypt, however with different insect species, also studied the variability among four mango cultivars in the levels of infestation with two armoured scale insects, *I. pallidula* and *A. aurantii*. He recorded that the population density of *A. aurantii* was greater than that of *I. pallidula*. On the other hand, grafted Balady was the most infested cultivar with the two scale insects followed by Hindy, then by Goleck, while the least infested cultivar was seedy Balady cultivar for two-scale insects.

2. Sampling program:

The obtained values in Table (2) showed that the relative variation (R.V.%) for the primary sampling data of *A. tubercularis* indicated that the population densities of pest ranging from (4.88 to 5.92%) and (4.26 to 5.15%)in the all different mango cultivars through the first and second years, respectively. As well, the *R.V.* (%) for the primary sampling data of *A. tubercularis*indicated that the mean population densities were 5.49, 4.69, and 4.78% during the first and second years, and for the two years combined, respectively (Table, 3). The values of R.V.% were very appropriate for a sampling program. However, with different insect species and different hosts, ***Naeimamini et al.* (2014)** stated that the relative variation for the primary sampling data of different stages of *Pulvinaria floccifera* (Hemiptera: Coccidae) was less than 25% and were acceptable. **Bakry (2018)** reported that the relative variation for the primary sampling data of total populations of *Waxiella mimosae* (Signoret) (Coccomorpha: Coccidae) on sunt trees ranged from 8.52 to 19.79% in all seasons, as well as over the entire year.

Bakry (2020) recorded that the *R.V.* (%) for the primary sampling data of *Parlatoria oleae* on mango trees indicated that the total population density was 2.41, 2.35 and 1.73% during the first and second years, and for the two years combined, respectively. **Bakry and Arbab (2020a)** reported that the relative variation for the primary sampling data of *Icerya seychellarum* on guava trees indicated that total population density was 4.07 (2017- 2018), 5.62 (2018-2019) and 3.55% (pooled). **Bakry and Shakal (2020b)** recorded that the relative variation (R.V.%) for the primary sampling data of *S. graminum* indicated that the population densities of pest ranging from (8.90 to 11.56%) and (7.21 to 10.28%)in the all different cultivars and lines of wheat through the two growing seasons, respectively. As well, the *R.V.* (%) for the primary sampling data of *S. graminum* indicated that the mean population densities was 9.78 and 8.04% during the first and second growing seasons, respectively.

3-Spatial distribution:

3.1- Distribution indices:

The results in Tables (2 and 3) showed that the spatial distribution among the sample units was determined by twenty two indices of distribution. The distribution results using the variance of *A. tubercularis* population on mango trees was greater than the general average of the population densities by the pest, and thus the variance-to-mean ratio (S^2/m) was greater than one were recorded in the all tested mango cultivars. Therefore, the spatial distribution of *A. tubercularis* population was aggregated over the entire year and for the two cumulative years.

The Lewis index of the pest was significantly greater than the index of contagious dispersion. Similar conclusions were made from the results of the Cassie index. The mean population of the pest distribution was greater than zero; therefore, *A. tubercularis*on all tested mango cultivars had an aggregated distribution. The *K* values of the negative binomial distribution of *A. tubercularis*population ranged from 2 to 8 in all mango cultivars during the first year, thus indicating moderate aggregation, and the higher than 8 (*K*> 8) indicates a

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https://dx.doi.org/10.22161/ijhaf.4.3.4
ISSN: 2456-8635

random population during the second year in the all tested cultivars and on the cumulative analysis.

The Index values of mean clumping (I_{DM}) of the pest in all mango cultivars were positive for the negative binomial. The Z-test values were greater than 1.96. The index of patchiness was greater than one and Green's index was greater than zero. All these indices showed an aggregated distribution for the population of *A. tubercularis* in all the different mango cultivars during the two years and when the data were pooled.

The temporal changes in the spatial distribution pattern of *A. tubercularis*population during each year were evaluated using 1/*k* (the aggregation index). The value was greater than one, thus indicating an aggregated pattern that became more dispersed with time.

Also, the highest values for the variance to- mean, the index of mean clumping, Z-test, patchiness index and green's index on the population density of *A. tubercularis* were (15.51, 14.51, 28.54, 1.16 and 0.31) for the first year (2017/2018) and (10.23, 9.23, 21.36, 1.11 and 0.20) during the second year (2018/2019), were recorded on Goleck mango cultivar as compared with the other tested mango cultivars during the two years, respectively. This cultivar was suffered the highest population density for this pest as mentioned previously (Table, 2).

In contrary, the lowest values for the distribution *i.e.*(S^2/m, I_{DM}, Z test, x*/m, and GI) on the population of *A. tubercularis* were (6.76, 5.76, 15.57, 1.10 and 0.12) were recorded on Sediek mango cultivar than those the tested mango cultivars during the first year of study (2017/2018), respectively.

While, during the second year (2018/2019), the lowest values for these indices (S^2/m, $I_{D\ M}$, Z test and GI) were recorded on Balady mango cultivar as compared with the other mango cultivars was (4.22, 3.22, 10.26 and 0.07), respectively. This cultivar was exhibited the lowest population density of *A. tubercularis* as mentioned previously. However, the lowest value of the index of patchiness (x*/m) by 1.07 was recorded on Sediek mango cultivar (Table, 2).

The values of population aggregations (λ) were all less than 2 in all tested mango cultivars over the entire year and for the two cumulative years, however, indicating that the aggregation phenomenon may be caused by environment variations (Table, 2). A similar conclusion was found to occur in distribution of *Parapoynx crisonalis* (Lepidoptera: Crambidae) on water chestnuts plant (**Li *et al*., 2017).**

The results in Table (3) show that the values of distribution indices of mean population density of *A. tubercularis* were higher in the first year (2017/2018) as compared to the second year (2018/2019). This evidence may be due to the general average of population density *A. tubercularis* was (69.08 ± 1.84 per leaf) during the first year was higher than in the second one (61.98 ± 1.53 per leaf).

It is clear that the mango cultivars affect the population density and spatial distribution of A. tubercularis. Therefore, the spatial distribution for the population of *A. tubercularis* using twenty two distribution indices indicated an aggregated distribution in all different mango cultivars in the two successive years and for the two cumulative years, that due to the effects of environmental factors (Table, 2 and 3).

However, there is no study in the literature regarding the distribution patterns of *A. tubercularis*. Studying different insect species and different hosts, **Chellappan *et al*. (2013)** reported that the value of mean crowding increased with an increase in the mean population density of *Paracoccus marginatus* (Hemiptera: Pseudococcidae). **Li *et al.* (2017)** recorded that the K value of the negative binomial distribution, aggregation index, and Cassie index were all higher than zero during May. This would indicate that *Parapoynx crisonalis* (Lepidoptera: Crambidae) larvae were in an aggregated distribution. **Bala and Kumar (2018)** recorded that the values of the Lewis index for all sampling dates of the bug, *Chauliops fallax* (Hemiptera: Malcidae) population on soybean were also found to be more than one, thus indicating that the distribution of the bug population was aggregated.

Bakry (2018) studied the spatial distribution of *W. mimosae* on sunt trees using 14 dispersion indices and recorded that all the models exhibited an aggregated distribution and followed a negative binomial distribution pattern all the different life stages and for the total population of *W. mimosae* over all seasons and for the pooled data over the two years of study (2016 to 2018). **Bakry (2020)** recorded that the spatial distribution of *P. oleae* on mango trees using twenty one distribution indices and stated that all indices of distribution indicated significant aggregated behaviour in each year, except, the K values of the negative binomial distribution of the total population ranged about 15-17 for each year during the two successive years, indicating random behavior. **Bakry and Arbab (2020a)** studied that the spatial distribution of *I. seychellarum* on guava trees using distribution indices, indicated aggregated behaviour over the entire year. **Bakry and Shakal (2020b)** recorded that the spatial distribution pattern of *Schizaphis graminum* (Hemiptera: Aphididae) on

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Vol-4, Issue-3, May-Jun, 2020
https://dx.doi.org/10.22161/ijhaf.4.3.4
ISSN: 2456-8635

some wheat cultivars and lines. They found that all distribution indices indicated a significant aggregated behaviour during each growing season in all the tested wheat cultivars and lines.

3.2- Regression methods:

Taylor's power law regression showed highly significant positive relationships between the log (mean of the population) and log (variance) for the population densities of *A. tubercularis* in the all different mango cultivars during the first and second years of study (Table, 4 and Fig. 1).

The calculated regression coefficient from Taylor's method indicated that an increase of one degree in the log (mean of the population) would increase the log of variance by approximately 1.84 to 2.43 for the population of *A. tubercularis* in the all tested mango cultivars during the first and second years, and on the pooled data (Table, 4).

The regression coefficient values from TPL were significant and greater than one. The values were (2.02, 1.87 and 1.95) during the first and second years and during the cumulative years, respectively, and the values of *t*-calculated of the slope (t_c) >*t*-table (t_t) in all mango cultivars during the two years, respectively, indicated an aggregated distribution for the population densities of *A. tubercularis*.

The relationship between these factors gave a good fit for Taylor's model and the R^2 was 94.44, 95.63 and 93.14 for the population of *A. tubercularis* in all mango cultivars during the first and second years and on the cumulative years. The R^2 value showed that the increase in values of population variance occurred due to the increase in mean population density.

In this model, the heterogeneity of the regression model indicated that the slope of TPL did not differ significantly between the two years (TPL: slope = 2.02, SE_b = 0.15, and n_1 = 12 for the first year and the slope = 1.87, SE_b = 0.13, and n_2 = 12 for the second year). The values of *t*-calculated (t_c) = 0.77 <*t*-table (t_t) = 2.07 when the df = [(n_1+n_2)-2] = 22 for the total pest population between the two years.

Taylor (1984) showed that the slope (b) is an index of the spatial distribution characteristic of the species; however, other studies have shown that b is not species specific and varies among environments and developmental stages **(Downing, 1986).**

The regression method of Iwao described the relationship between population mean ($\overline{X}$) and mean crowding index $\overset{*}{X}$ for the population of *A. tubercularis*in all tested mango cultivars for the first and second years and during the two cumulative years. The regression coefficient (β) values were significantly greater than one, was 1.02, the values of t-calculated of slope (t_c) > t-table (t_t) for the population of pest in the all tested mango cultivars and all years and on the two cumulative years, indicated an aggregated distribution for the population of *A. tubercularis* (Table, 4).

The calculated regression coefficient of Iwao's method indicated that an increase of one degree in the population mean would increase the mean crowding index approximately one degree for the population in all tested years.

Additionally, the intercept values (α) or the index of basic contagion were negative and < zero and > -1. The values of α ranged from -0.83 to -2.32 for the pest population in both years as well as for the cumulative years (Table, 4). The negative values indicated that aggregation was from individuals rather than from colonies and the smaller than zero values indicated that of the pest population, the basic component of the population tended to be a single individual in both years and for the two cumulative years.

The relationship between the population mean and mean crowding index had a better fit. R^2 was 99.9% for the pest population in all tested years as well as the pooled data. R^2 showed that the increase in the mean crowding index occurred due to the increase in the population mean (Table, 4 and Fig., 1).

The heterogeneity of the regression model showed that the slope of the IPR did not differ significantly between the two years: IPR: slope = 1.02, SE_b = 0.004, and n_1 = 12 for the first year and slope = 1.02, SE_b = 0.002, and n_2 = 12 for the second year. The *t*-calculated (t_c) values = 1.36 <*t*-table (t_t) = 2.07 when the df = [(n_1+n_2)-2] = 22 for the total pest population between the two studied years. These findings are in agreement with those reported by **Tonhasca *et al.* (1996)** who recorded that patchiness regression is not subject to the stabilizing effect of a log transformation.

The present results revealed that the regression coefficient (b) values of Taylor and (β) values of Iwao were both significantly greater than one and the *t*-calculated slope values (t_c) >*t*-table (t_t). Therefore, the population of *A. tubercularis* tended to have an aggregated distribution.

Generally, the regression models of TPL and Iwao are used to estimate the spatial distribution of a pest. The results from the present study showed an aggregated pattern and a negative binomial distribution pattern for the population density of *A. tubercularis*. Factors that may influence the

https://dx.doi.org/10.22161/ijhaf.4.3.4
ISSN: 2456-8635

spatial distribution of pests include the level of infestation, environmental condition, or behavioural characteristics of the insects. In addition, these two regression methods were more accurate and produced different results than the other distribution indices used in the present study. These models were based on sample means and population variances.

The present results are generally in agreement with those of **Hsu *et al*. (2001)** however with the same genus of insect and different host, they studied that the dispersion pattern of varied stages generated by Iwao's patchiness regression and Taylor's power law for *A. yabunikkei* in different sampling units of camphor trees, *Cinnamomum camphora* (L.), were compared. Taylor's power law provided a consistently good fit to the data, whereas the fit of Iwao's patchiness regression was erratic, and the values of aggregation index of Taylor's power law (1.76 to 2.65) were narrower than those of Iwao's (1.26 to 11.83), but both indices ($b > 1.7$ and $\beta > 1.2$) indicate a clumped distribution pattern in all sampling units. Also, **Lee *et al*. (2005)** however with the different insect species and different hosts, reported an aggregated distribution pattern for leaf miner in tomato greenhouses, because all b values from Taylor's power law regressions were significantly greater than one.Furthermore, aggregated distribution patterns of other pests using these aggregation indices have been reported previously **(Naseri *et al*., 2009; Payandeh *et al*., 2010; Beltra *et al*., 2013; Nematollahi *et al*., 2014; Arbab, 2014; Arbab and McNeill, 2014 and Bakry and Arbab 2020a).**

4- Optimum number of sample size:

The optimal sample size number was calculated using Iwao's regression coefficient, and the relationship between the optimum sample size number and mean numbers of *A. tubercularis*population with levels of precision of 5%, 10, and 15% were calculated during the first and second years and for the two cumulative years (Table, 5 and Fig. 2).

The minimum number of samples that were required were shown to decrease rapidly with an increase in mean population density of *A. tubercularis* (inverse relationship) at all precision levels (5%, 10%, and 15%, Fig. 2).

These results agree with those obtained by **Arbab and McNeill (2014)**; however, they used different insect species and host and reported that the optimum sample size number decreased rapidly as *Sitona humeralis* weevil density increased at the 10% and 15% precision levels. **Bakry and Arbab (2020a)** recorded that minimum numbers of sample required decreased rapidly with increased mean total population density of *I. seychellarum* at levels of precision of (5, 10 and 15%).

The optimum sample size fluctuated during each year of study and depended upon the population density of *A. tubercularis*and the desired precision level. At the 15% precision level, the mean number of samples required for estimating the population density of *A. tubercularis*was approximately three leaves in each of the years, which would be suitable for integrated pest management purposes. However, the values used for population ecology studies require a precision of 10%, thus the sample size was increased and was approximately 6 leaves during each year for the two successive years (Table, 5 and Fig. 2). However, for a more accurate estimate (*i.e.* 5%), the required sample size of mango leaves increased to approximately 22 leaves in the first and second years, respectively (Table, 5 and Fig. 2).

The results showed that the sample size required to achieve a desired level of precision was similar from one year to another. This could be explained by the fact that the population pest density from one year to another was similar and may be due to the environmental conditions, which were suitable during both years of study.

The different precision levels (5%, 10%, and 15%) adopted in the present study could be chosen for ecological or insect behavioural studies. This study showed that precision levels of 5% and 10% are suitable for ecological or insect behavioural studies where a higher level of precision is required, whereas for pest management programmes, a 15% level is acceptable. However, even though such a number of samples is acceptable for research purposes, it is not practicable for agronomic or pest management.

These results agree with those obtained by **Arbab and McNeill (2014).** One attribute of the optimum sample size is that, at a given density, the required sample size increases as the target population becomes more aggregated **(Wipfli *et al*., 1992).** Successful management of *A. tubercularis*strongly depends on the development of an appropriate sampling plan (*i.e.* easy to implement, suitable for rapid decision-making processes). In sampling programmes, precision and cost-effectiveness are two of the most important factors that must be considered **(Pedigo, 1994).** The development of a sequential sampling scheme with a fixed statistical precision, therefore, may be useful for estimating the density of *A. tubercularis*in mango orchards. Such an estimation, in turn, would be valuable for ecological and pest management studies. A sampling programme can be used in ecological

International journal of Horticulture, Agriculture and Food science(IJHAF)
Vol-4, Issue-3, May-Jun, 2020
https://dx.doi.org/10.22161/ijhaf.4.3.4
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investigations **(Faleiro *et al*. 2002)** and when detecting pest levels that lead to a justification of control measures **(Arnaldo and Torres 2005).**

Table.1: Mean numbers of A. tubercularis population on different mango cultivars during the two successive years of (2017/2018 and 2018/2019):

Cultivars	Average no. of individuals of insect per leaf ± S.E		
	First year 2017/2018	Second year 2018/2019	Average of both years
Goleck	92.51 ± 5.47 a	85.81 ± 4.28 a	89.16 ± 4.61 a
Balady	45.01 ± 2.60 d	37.72 ± 1.82 e	41.36 ± 2.09 e
Ewaise	78.23 ± 4.63 b	69.29 ± 3.57 b	73.76 ± 3.88 b
Zebda	75.58 ± 4.01 b	65.70 ± 2.92 bc	70.64 ± 3.19 b
Sediek	59.08 ± 2.89 c	51.00 ± 2.17 d	55.04 ± 2.32 d
Hindi Bisinnara	64.07 ± 3.44 c	62.39 ± 3.01 c	63.23 ± 3.02 c
Mean	69.08 ± 1.84 a	61.98 ± 1.53 b	65.53 ± 1.60
L.S.D. at 0.05 between cultivars	6.12**	4.32**	4.89**
L.S.D. at 0.05 between two years	3.39**		

Means followed by the same letter (s), in each column, are not significantly different at 0.05 level probability, by Duncan's multiple range test (DRMT).

Table.2: Estimated parameters for spatial distribution of population density of A. tubercularis on different mango cultivars during the two successive years (2017/2018 and 2018/2019).

Parameters	First year (2017/2018)						Second year (2018/2019)					
	Goleck	Balady	Ewaise	Zebda	Sediek	Hindi Bisinnara	Goleck	Balady	Ewaise	Zebda	Sediek	Hindi Bisinnara
Max.	153.2	74.7	130.8	118.5	87.8	101.6	147.7	66.5	118.3	106.3	74.1	108.2
Min.	34.10	16.50	29.52	32.49	23.41	25.30	39.12	17.13	31.69	35.71	24.19	31.94
Mean	92.51	45.01	78.23	75.58	59.08	64.07	85.81	37.72	69.29	65.70	51.00	62.39
Range of mean	119.06	58.15	101.24	85.98	64.37	76.35	108.59	49.38	86.58	70.58	49.86	76.22
Median	88.50	43.00	72.27	72.24	58.90	63.02	84.31	36.68	67.72	62.32	49.01	60.43
S^2	1435.2	325.2	1030.1	773.7	399.5	568.9	877.6	159.0	611.0	409.3	226.7	434.3
S	37.88	18.03	32.09	27.82	19.99	23.85	29.62	12.61	24.72	20.23	15.06	20.84
S.E.	5.47	2.60	4.63	4.01	2.89	3.44	4.28	1.82	3.57	2.92	2.17	3.01
C.V.	40.95	40.07	41.03	36.80	33.83	37.23	34.53	33.43	35.67	30.79	29.52	33.41
R.V.	5.91	5.78	5.92	5.31	4.88	5.37	4.98	4.83	5.15	4.44	4.26	4.82
S^2/m	15.51	7.22	13.17	10.24	6.76	8.88	10.23	4.22	8.82	6.23	4.45	6.96
Lewis Index	3.94	2.69	3.63	3.20	2.60	2.98	3.20	2.05	2.97	2.50	2.11	2.64
Cassie index	0.16	0.14	0.16	0.12	0.10	0.12	0.11	0.09	0.11	0.08	0.07	0.10
K	6.37	7.23	6.43	8.18	10.25	8.13	9.30	11.73	8.86	12.56	14.80	10.46
I_D	729.2	339.6	618.9	481.1	317.9	417.4	480.7	198.2	414.5	292.8	208.9	327.2
Z value	28.54	16.42	25.54	21.38	15.57	19.25	21.36	10.26	19.15	14.56	10.80	15.94
I_{DM}	14.51	6.22	12.17	9.24	5.76	7.88	9.23	3.22	7.82	5.23	3.45	5.96
X*	107.02	51.23	90.40	84.82	64.84	71.95	95.03	40.94	77.11	70.92	54.44	68.35
X*/m	1.16	1.14	1.16	1.12	1.10	1.12	1.11	1.09	1.11	1.08	1.07	1.10
GI	0.31	0.13	0.26	0.20	0.12	0.17	0.20	0.07	0.17	0.11	0.07	0.13
1/*k*	0.16	0.14	0.16	0.12	0.10	0.12	0.11	0.09	0.11	0.08	0.07	0.10
λ	0.32	0.13	0.27	0.18	0.09	0.15	0.21	0.07	0.17	0.10	0.05	0.11

International journal of Horticulture, Agriculture and Food science(IJHAF)
Vol-4, Issue-3, May-Jun, 2020
https://dx.doi.org/10.22161/ijhaf.4.3.4
ISSN: 2456-8635

Therefore, the distribution pattern of the insect is essential for the management of *A. tubercularis*. The next steps are to develop an efficient scouting programme and establish threshold densities for action that will inform growers on when the pest is active and when interventions (e.g. insecticides) can be applied. The results from the present study are indicative of populations present in the field and provide information on the relative change over time. Thus, the spatial distribution parameters of this species can be employed to estimate the population density of *A. tubercularis*. For the monitoring, sampling, and population density estimation of *A. tubercularis*, the spatial distribution pattern should be considered because the minimal sample size is dependent on the spatial pattern of the sampled population.

Table.3: Estimated parameters for spatial distribution of mean population density of A. tubercularis during the two successive years (2017-2019).

Parameters	During the First year (2017/2018)	During the Second year (2018/2019)	Combined the two years
Max.	111.08	103.50	104.86
Min.	26.89	29.96	36.86
Mean	69.08	61.98	65.53
Range of mean	84.19	73.54	68.00
Median	66.01	59.77	63.04
S^2	689.18	406.00	471.12
S	26.25	20.15	21.71
S.E.	3.79	2.91	3.13
C.V.	38.00	32.51	33.12
R.V.	5.49	4.69	4.78
S^2/m	9.98	6.55	7.19
Lewis Index	3.16	2.56	2.68
Cassie index	0.13	0.09	0.09
K	7.69	11.17	10.59
I_D	468.92	307.86	337.90
Z value	20.98	15.17	16.35
I_{DM}	8.98	5.55	6.19
X*	78.05	67.53	71.72
X*/m	1.13	1.09	1.09
GI	0.19	0.12	0.13
1/*k*	0.13	0.09	0.09
λ	0.18	0.11	0.12

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International journal of Horticulture, Agriculture and Food science(IJHAF)
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Table.4: Parameters estimation for spatial distribution of A. tubercularis population on different mango cultivars derived from different regression methods during the two successive years (2017/2018 and 2018/2019).

Years	Varieties	Taylor's power law									Iwao's patchiness regression								
		a	SE_a	b	SE_b	MR	R^2 x 100	p	t_c	t_t	α	$SE_α$	β	$SE_β$	MR	R^2 x 100	p	t_c	t_t
2017/2018	Goleck	-1.56	0.21	1.93	0.11	0.984	96.87	<0.000	8.5	2.24	-0.83	0.25	1.02	0.002	0.99997	99.994	<0.000	7.44	2.24
	Balady	-1.66	0.19	1.98	0.12	0.983	96.70	<0.000	8.46	2.24	-0.95	0.14	1.02	0.003	0.99996	99.992	<0.000	6.92	2.24
	Ewaise	-1.57	0.21	1.93	0.11	0.984	96.75	<0.000	8.33	2.24	-0.84	0.2	1.02	0.002	0.99997	99.994	<0.000	7.65	2.24
	Zebda	-2.37	0.74	2.43	0.4	0.889	78.95	<0.000	3.6	2.24	-2.32	0.77	1.05	0.010	0.99958	99.915	<0.000	4.97	2.24
	Sediek	-1.6	0.23	1.95	0.13	0.978	95.73	<0.000	7.29	2.24	-0.92	0.2	1.02	0.003	0.99995	99.99	<0.000	5.92	2.24
	Hindi Bisinnara	-1.61	0.22	1.95	0.12	0.981	96.18	<0.000	7.73	2.24	-0.9	0.2	1.02	0.003	0.99996	99.992	<0.000	6.37	2.24
	Mean	-1.71	0.28	2.02	0.15	0.97	94.44	<0.000	6.58	2.24	-1.06	0.261	1.023	0.004	0.99994	99.9879	<0.000	6.37	2.24
2018/2019	Goleck	-1.39	0.26	1.84	0.13	0.974	94.94	<0.000	6.25	2.24	-0.68	0.25	1.02	0.003	0.99996	99.993	<0.000	6.01	2.24
	Balady	-1.55	0.23	1.91	0.15	0.972	94.44	<0.000	6.21	2.24	-0.9	0.13	1.02	0.003	0.99995	99.99	<0.000	5.44	2.24
	Ewaise	-1.42	0.24	1.85	0.13	0.975	95.15	<0.000	6.44	2.24	-0.75	0.19	1.02	0.003	0.99997	99.994	<0.000	6.49	2.24
	Zebda	-1.57	0.22	1.91	0.12	0.98	96.11	<0.000	7.50	2.24	-0.94	0.15	1.02	0.002	0.99998	99.995	<0.000	8.07	2.24
	Sediek	-1.45	0.27	1.86	0.16	0.966	93.31	<0.000	5.45	2.24	-0.83	0.18	1.02	0.004	0.99994	99.988	<0.000	4.86	2.24
	Hindi Bisinnara	-1.46	0.26	1.87	0.15	0.97	94.08	<0.000	5.85	2.24	-0.76	0.19	1.02	0.003	0.99996	99.991	<0.000	5.52	2.24
	Mean	-1.46	0.22	1.87	0.13	0.98	95.63	<0.000	6.86	2.24	-0.81	0.166	1.02	0.003	0.99997	99.9937	<0.000	6.54	2.24
Combined two years		-1.64	0.18	1.97	0.10	0.97	94.61	<0.000	9.69	2.07	-1.01	0.160	1.02	0.002	0.99994	99.9886	<0.000	9.06	2.07
Difference between two years									0.77	2.07								1.36	2.07

Table.5: Maximum, minimum and mean population density for the population density of A. tubercularis and optimum sample size (n) for achieving a fixed precision levels of (D = 0.05, 0.10 and 0.15) using enumerative sampling procedures during the two successive years (2017/2018 and 2018/2019).

Years	Varieties	Population density				Fixed precision levels (D)											
						5%				10%				15%			
		Max.	Min.	Mean	Range	Max.	Min.	Mean	Range	Max.	Min.	Mean	Range	Max.	Min.	Mean	Range
2017/2018	Goleck	153.16	34.10	92.51	119.06	28.82	12.15	16.84	16.67	7.20	3.04	4.21	4.17	3.20	1.35	1.87	1.85
	Balady	74.65	16.50	45.01	58.15	55.17	18.26	28.35	36.91	13.79	4.56	7.09	9.23	6.13	2.03	3.15	4.10
	Ewaise	130.76	29.52	78.23	101.24	32.27	12.95	18.51	19.33	8.07	3.24	4.63	4.83	3.59	1.44	2.06	2.15
	Zebda	118.48	32.49	75.58	85.98	60.09	30.39	39.08	29.70	15.02	7.60	9.77	7.42	6.68	3.38	4.34	3.30
	Sediek	87.78	23.41	59.08	64.37	40.35	16.34	22.41	24.01	10.09	4.08	5.60	6.00	4.48	1.82	2.49	2.67
	Hindi Bisinnara	101.65	25.30	64.07	76.35	37.53	14.99	21.25	22.54	9.38	3.75	5.31	5.64	4.17	1.67	2.36	2.50
	Mean	111.08	26.89	69.08	84.19	39.75	16.50	22.97	23.24	9.94	4.13	5.74	5.81	4.42	1.83	2.55	2.58
2018/2019	Goleck	147.71	39.12	85.81	108.59	23.79	11.15	15.35	12.64	5.95	2.79	3.84	3.16	2.64	1.24	1.71	1.40
	Balady	66.51	17.13	37.72	49.38	51.51	18.54	29.44	32.96	12.88	4.64	7.36	8.24	5.72	2.06	3.27	3.66
	Ewaise	118.27	31.69	69.29	86.58	28.73	12.56	17.97	16.17	7.18	3.14	4.49	4.04	3.19	1.40	2.00	1.80
	Zebda	106.29	35.71	65.70	70.58	28.77	14.35	19.82	14.42	7.19	3.59	4.96	3.60	3.20	1.59	2.20	1.60
	Sediek	74.05	24.19	51.00	49.86	37.12	16.72	22.47	20.41	9.28	4.18	5.62	5.10	4.12	1.86	2.50	2.27
	Hindi Bisinnara	108.16	31.94	62.39	76.22	28.65	13.10	19.01	15.55	7.16	3.27	4.75	3.89	3.18	1.46	2.11	1.73
	Mean	103.50	29.96	61.98	73.54	30.80	13.66	19.50	17.14	7.70	3.42	4.87	4.28	3.42	1.52	2.17	1.90
Combined two years		111.08	26.89	65.53	84.19	38.27	15.63	22.29	22.64	9.57	3.91	5.57	5.66	4.25	1.74	2.48	2.52

International journal of Horticulture, Agriculture and Food science(IJHAF)
Vol-4, Issue-3, May-Jun, 2020
https://dx.doi.org/10.22161/ijhaf.4.3.4
ISSN: 2456-8635

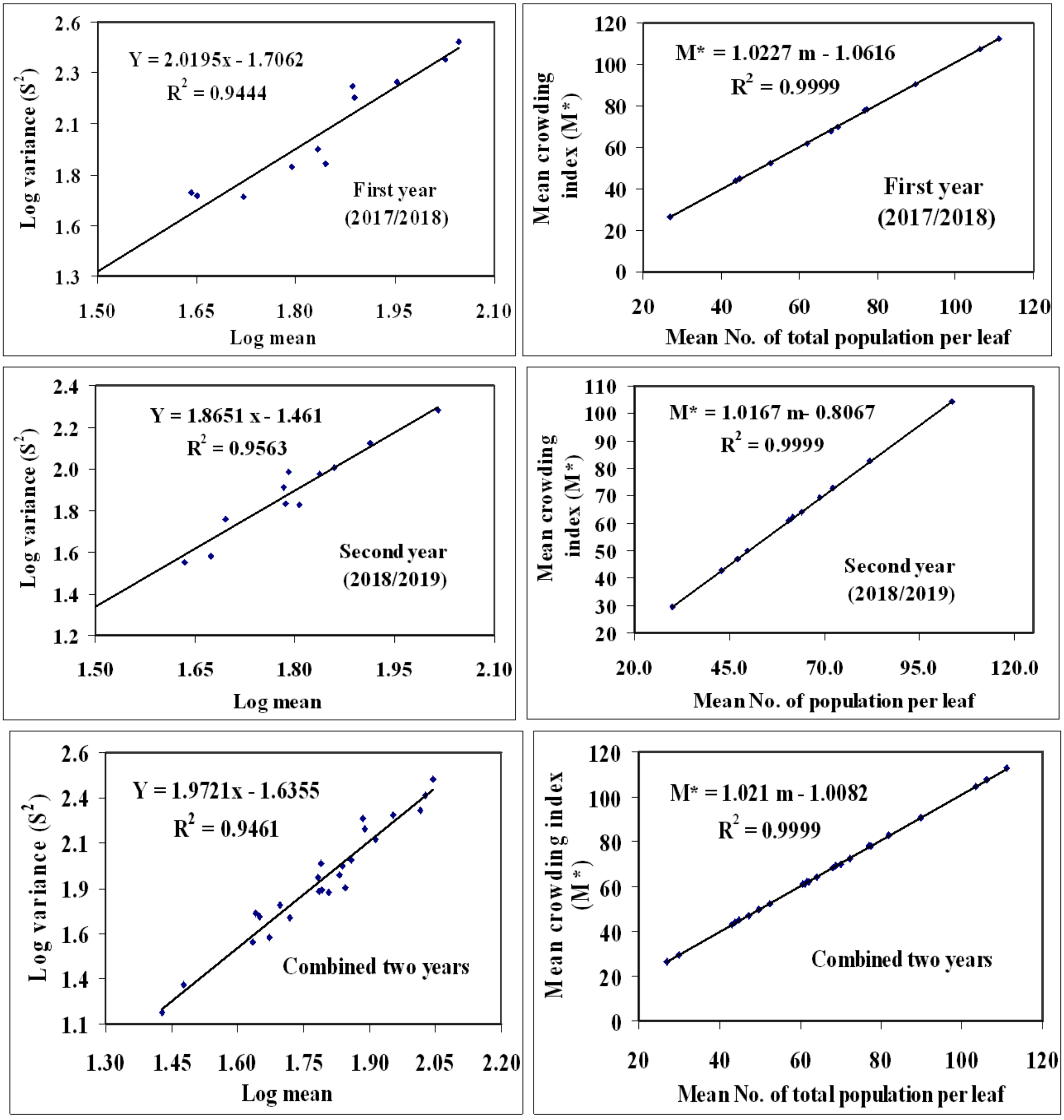

Fig. 1: Regression analyses of Taylor's power law (A) and Iwao's patchiness (B) for A. tubercularis population on mango trees during the two successive years (2017/2018 and 2018/2019).

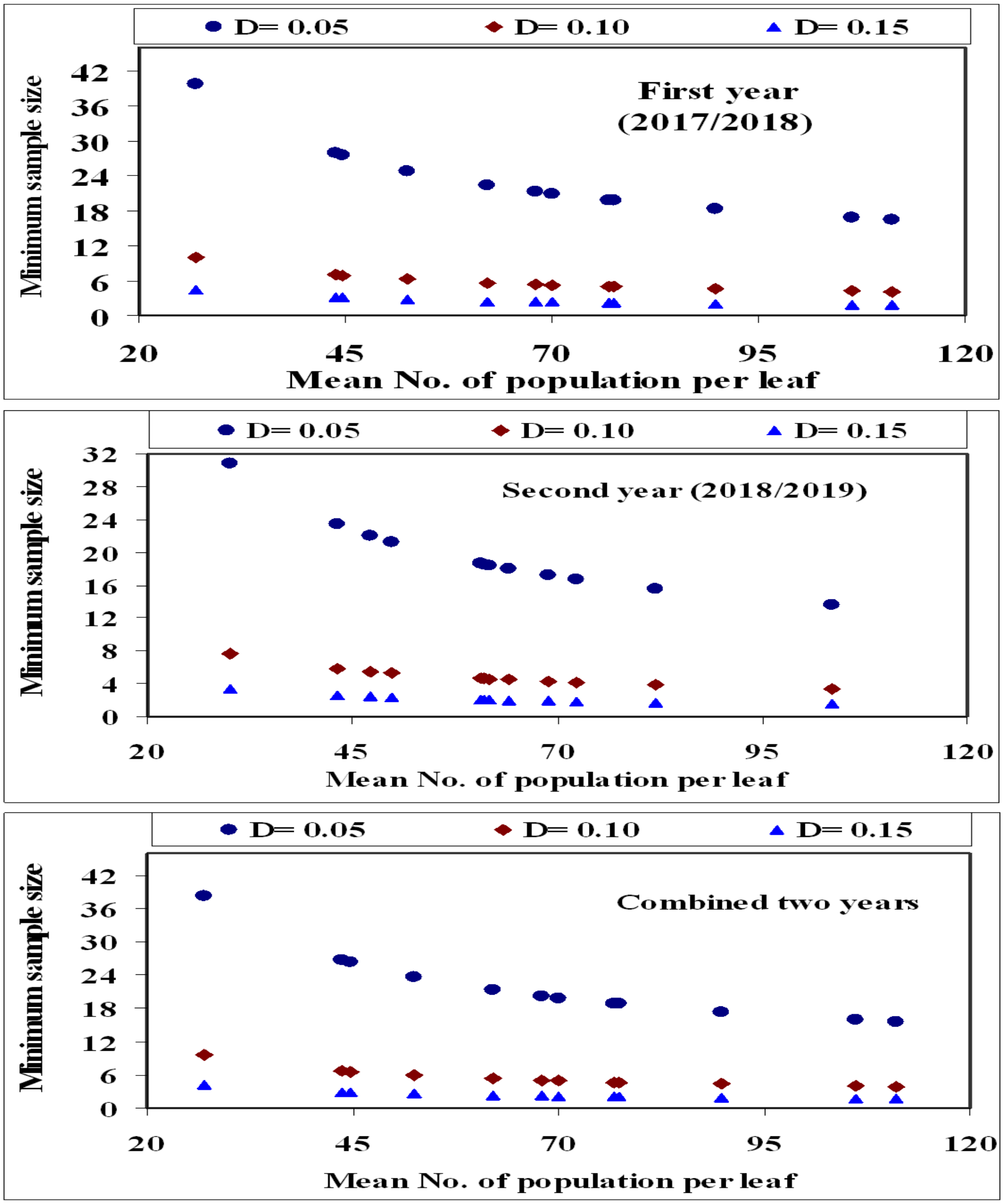

Fig. 2: Relationship between population density for the population density of A. tubercularis and optimum number of sample size (n) for achieving a fixed precision levels of (D = 0.05, 0.10 and 0.15) using enumerative sampling procedures during the two successive years (2017/2018 and 2018/2019).

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International journal of Horticulture, Agriculture and Food science(IJHAF)
Vol-4, Issue-3, May-Jun, 2020
https://dx.doi.org/10.22161/ijhaf.4.3.4
ISSN: 2456-8635

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