

SPATIAL DISTRIBUTION OF *Chrysodeixis includens* EGGS (WALKER, 1858) (LEPIDOPTERA: NOCTUIDAE) IN SOYBEAN CROP

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ABSTRACT: Among pest insects that attack soybean crops, *Chrysodeixis includens* (Walker, 1858) (Lepidoptera: Noctuidae) looper caterpillar deserves attention due to its key pest status in soybean crops. The spatial distribution of *C. includens* eggs should be investigated in order to understand the behavior of this species in the area. The aim of this study was to investigate the spatial distribution of *C. includens* eggs in soybean crops. The experiment was conducted with SYN 9070 RR soybean variety in an experimental area of the Teaching, Research and Extension Farm (FEPE) of FCAV/ UNESP, Jaboticabal, SP, Brazil. The area of 0.6 ha was divided into 60 equidistant plots of 100 m² each. For the study of the spatial distribution of *P. includens* eggs in the area, the following dispersion indexes were used: variance / mean ratio (I), Morisita index (I_d), Green coefficient (C_x), k exponent of the negative binomial distribution for each sampling. Regarding the probabilistic models that describe the spatial distribution of a given variable, the data adjustment to the Poisson and negative binomial distributions was tested. According to values obtained for dispersion indexes, egg distribution occurred in an aggregate way, and the negative binomial distribution was the most appropriate probabilistic model to represent the distribution of *C. includens* eggs in the study area.

KEYWORDS: *Glycine max*, looper caterpillar, oviposition, dispersion, negative binomial.

DISTRIBUIÇÃO ESPACIAL DE OVOS DE *Chrysodeixis includens* (WALKER, 1858) (LEPIDOPTERA: NOCTUIDAE) NA CULTURA DA SOJA

RESUMO: Dentre os insetos-praga que atacam a cultura da soja, a lagarta-falsa-medideira *Chrysodeixis includens* (Walker, 1858) (Lepidoptera: Noctuidae) merece destaque pelo *status* de praga-chave da cultura. Sendo fundamental realizar o estudo de distribuição espacial de ovos de *C. includens* para compreender o comportamento desta espécie na área. Objetivou-se estudar a distribuição espacial dos ovos de *C. includens* na cultura da soja. O experimento foi conduzido com a variedade SYN 9070 RR em uma área experimental da Fazenda de Ensino, Pesquisa e Extensão (FEPE) da FCAV/UNESP, Jaboticabal, SP, Brasil. A área de 0,6 ha foi dividida em 60 parcelas equidistantes de 100 m² cada. Para o estudo da distribuição espacial dos ovos de *P. includens* na área, foram utilizados os seguintes índices de dispersão: razão variância/média (I), índice de Morisita (I_d), coeficiente de Green (C_x), expoente k da distribuição binomial negativa para cada amostragem. Em relação aos modelos probabilísticos que descrevem a distribuição espacial de uma determinada variável, foram testados os ajustes dos dados às distribuições de Poisson e binomial negativa. De acordo com os valores obtidos para os índices de dispersão, a distribuição dos ovos ocorreu de forma agregada, sendo a distribuição binomial negativa o modelo probabilístico mais adequado para representar a distribuição dos ovos de *C. includens* na área em estudo.

PALAVRAS CHAVE: *Glycine max*, lagarta-falsa-medideira, oviposição, dispersão, binomial negativa.

INTRODUCTION

Among insect pests that attack soybean crop, looper caterpillar, *Chrysodeixis includens* (Walker, 1858) (Lepidoptera: Noctuidae) stands out, which consumes about 64 to 200 cm² of the leaf area and causes high losses of the photosynthetic area (Bueno et al., 2011).

The species has globular eggs with approximate diameter of 0.5 mm. Egg color varies from light cream when they are oviposited to light brown when close to the hatching of caterpillars (Peterson, 1964). The oviposition period is approximately 4.5 days, with mean of 318.4 eggs oviposited, ranging from 155 to 449 eggs per female (Barrionuevo et al., 2012).

Over a period of time, *C. includens* was considered a secondary soybean pest because populations were controlled by entomopathogenic fungi and parasitoids (Sosa-Gómez et al., 2003). However, due to changes in the crop production system since the 2000s mainly related to the indiscriminate use of insecticides and fungicides for the control of Asian rust, *C. includens* has become a serious phytosanitary problem, gaining status of key pest in various regions of the country (Bueno et al., 2007).

In addition, soybean cultivation has been undergoing major changes, among which the increasing use of genetically modified plants, which can directly or indirectly affect the behavior of insect pest populations in the agroecosystem (Rodrigues et al., 2010). In addition to the factors above, cultivation in extensive areas impairs the sampling of pest insects (Bueno et al., 2012).

The behavior of individuals in a population in their habitat is of utmost importance in pest management decision-making (Rodrigues et al., 2010). Information related to the distribution of insect populations may be useful for the development of effective sampling programs, since distribution directly affects sampling efficiency and accuracy (Davis, 1994).

The types of spatial distribution of pests in cultivated areas can be: regular (uniform), random and aggregate (contagious), and the probabilistic models that describe these distribution forms are called positive binomial, Poisson and negative binomial, respectively (Perecin and Barbosa, 1992).

In order to contribute to studies related to pest management strategies, and given the importance of *C. includens* to soybean crop, the aim of this work was to

investigate the spatial distribution pattern of eggs of this species in soybean crop.

MATERIAL AND METHODS

The experiment was conducted in the 2012/13 agricultural year at the Teaching, Research and Extension Farm (FEPE) - Faculty of Agricultural and Veterinary Sciences (FCAV/UNESP), Campus of Jaboticabal, state of São Paulo (21°15'S and 48°16'W, mean altitude of 615 m a.s.l.). According to the Köppen classification, the climate of the region is CWc, characterized as hot summer and dry winter, with mean temperature of 28 °C.

An experimental area of 6.000 m² (0.6 ha) was selected and divided into 60 equidistant sampling units of 100 m² (10 x 10 m) each. The soybean variety used was SYN 9070 RR (transgenic with glyphosate tolerance). Sowing was performed on November 15, 2012, and plant emergence occurred on November 20, 2012 (VE). Sowing was performed according to technical recommendations for the region.

Evaluations were weekly performed from the last vegetative stages (Vn) to the reproductive stage of formed grains (R6), counting *C. includens* eggs in one randomly chosen plant per plot. Both abaxial and adaxial leaf surfaces as well as leaf petiole were evaluated.

The dispersion indexes used to verify the degree of aggregation of *C. includens* eggs were calculated using the Microsoft Excel[®] software.

Variance/mean ratio (I). It is the most common index, also called dispersion index. It is the relationship between variance (s²) and mean ($I = s^2/m$) used to measure the deviation of an arrangement of randomness conditions (Rabinovich, 1980).

The deviation from randomness can be tested by the chi-square test with n-1 degrees of freedom, $\chi^2 = (n-1) s^2/m$ (Elliott, 1979).

Morisita Index (Id). According to Morisita (1962), the index is given by the following formula:

$$Id = n \frac{\sum [x(x-1)]}{\sum x(\sum x - 1)} = n \frac{\sum x^2 - \sum x}{(\sum x)^2 - \sum x}$$

Where: n = number of sampling units; x = number of eggs per plot and $\sum x$ = sum of individuals present in the sampling units.

Green coefficient (Cx). In this index, negative values indicate uniform distribution pattern, while positive values indicate aggregated pattern (Green, 1966). It is based on the variance / mean distribution ratio and is given by:

$$C_x = \frac{(s^2 / \hat{m}) - 1}{\sum_{i=1}^n x_i - 1}$$

Where: s^2 = sample variance; \hat{m} = sampling mean; x_i = number of eggs per plot.

Exponent k of the negative binomial distribution (k). The initial estimation of k values was performed by the moment method:

$$k = \frac{\hat{m}^2}{s^2 - \hat{m}}$$

And later, by the maximum likelihood method, because it is more accurate (Elliott, 1979):

$$N \ln\left(1 + \frac{\hat{m}}{\hat{k}}\right) = \sum_{i=1}^{nc} \left(\frac{A(x_i)}{\hat{k} + x_i} \right)$$

Where: N = number of sampling units, A (x_i) = sum of frequencies greater than x, nc = number of frequency distribution classes and, x_i = number of eggs per plot; ln = neperian logarithm.

The probabilistic model was obtained in each sampling, where adjustments to the Poisson and negative binomial distribution were tested, because variances were equal to or greater than the mean, respectively. In this case, there is no need to test the positive binomial distribution that is characterized by presenting less-than-mean variance.

Poisson distribution. Distribution that best represents the random spatial distribution of insects and is characterized by presenting variance equal to the mean ($\sigma^2 = m$) (Southwood, 1978).

The formulas for calculating the probability series are given by:

$$P_{(x)} = \frac{\lambda^x e^{-\lambda}}{x!}$$

Where $P_{(x)}$ is the probability of x individuals occurring in the sampling unit, is the distribution parameter ($\lambda = \mu = \sigma^2$), and e is the basis of the Neperian (Natural) logarithm = 2.71828.

Negative binomial distribution. This type of distribution has variance greater than the mean ($\sigma^2 > \mu$), and has two parameters, the mean (m) and k exponent ($k > 0$) (Taylor, 1984).

The probability series can be calculated for a sample using the recurring formula given by:

$$P(x) = \frac{P(x-1) R(k+x-1)}{x}, x = 1, 2, 3, \dots$$

Where,

$$P(0) = \left(1 + \frac{\hat{m}}{k}\right)^{-k}$$

$$R = \frac{\hat{m}}{k + \hat{m}}$$

And \hat{m} is the sampling mean, k is the estimate of the negative binomial k exponent, P(x) is the probability of x individuals occurring in the sampling unit.

The model presents good adjustment to original data when observed and expected frequencies are close. This proximity was tested by the chi-square test, given by:

$$X^2 = \sum_{i=1}^{n_c} \frac{(FO_i - FE_i)^2}{FE_i}$$

Where FO_i = Frequency observed in class i; FE_i = Expected frequency in class i; n_c = number of classes in the sample.

The number of degrees of freedom of χ^2 is given by v = number of classes - estimated number of parameters in the sample - 1, i.e.: $v = n_c - n_p - 1$.

If k value is too high ($k \rightarrow \infty$), the negative binomial distribution approaches the Poisson series and when k value tends to zero, the negative binomial distribution tends to the logarithmic series (Southwood, 1978).

RESULTS AND DISCUSSION

According to the dispersion index results, the mean number of *C. includens* eggs found in soybean plants was 0.23 ± 0.07 in the first evaluation, increasing gradually until reaching a peak of 5.63 ± 0.46 at 72 days after emergence (DAE) (Table 1).

Table 1. Dispersal indexes of *Chrysodeixis includens* eggs (Walker, 1858) (Lepidoptera: Noctuidae) in soybean crop. Jaboticabal - SP, Brazil 2012/13.

Sampling dates	Indexes						
	(m ± EP)	s ²	I	Id	Cx	k mom	k max.ver
24 DAE	0.23 ± 0.07	0.28	1.22	1.98	0.0166	1.08	0.85
30 DAE	0.38 ± 0.08	0.41	1.07	1.19	0.0032	5.53	5.53
37 DAE	0.50 ± 0.10	0.59	1.19	1.40	0.0064	2.68	2.68
44 DAE	0.57 ± 0.11	0.79	1.40	1.71	0.0120	1.42	1.22
49 DAE	0.92 ± 0.18	1.84	2.01	2.10	0.0187	0.91	0.77
57 DAE	0.62 ± 0.13	1.05	1.71	2.16	0.0197	0.87	0.6
65 DAE	2.88 ± 0.31	5.66	1.96	1.33	0.0056	2.99	3.12
72 DAE	5.63 ± 0.46	12.57	2.23	1.22	0.0037	4.57	4.25
78 DAE	2.40 ± 0.36	7.63	3.18	1.90	0.0152	1.1	0.94
84 DAE	0.95 ± 0.17	1.67	1.76	1.80	0.0136	1.24	0.97
93 DAE	0.88 ± 0.15	1.29	1.46	1.52	0.0089	1.91	1.33

m = sampling mean; EP = standard error of the mean; s² = sample variance; I = variance / mean ratio; Id = Morisite Index; Cx = Green's coefficient; k_{mom} = k by the moment method; k max.ver. = k by the maximum likelihood method; DAE = days after plant emergence.

After the population peak at 72 DAE, a reduction in the number of *C. includens* eggs was observed, thus, this fact can be understood by the occurrence of phenological changes in soybean plants (Jost and Pitre, 2002). In this context, according to Serpa (2018), throughout the reproductive period of soybean, there was a decline in the population of *C. includens* caterpillars, especially at the end of the crop cycle, since the population is smaller due to the low supply of food in the senescence period of plants.

The values obtained for the variance / mean ratio (*I*) were higher than the unit in all samples, indicating aggregate distribution of eggs in the study area. The values obtained for the Morisita index (*I_q*) also indicated egg aggregation, since all evaluations obtained values greater than one (Table 1).

The values found for Green coefficient (*Cx*) analyses were greater than zero in all samples, indicating, according to Davis (1993), that the variable under study had aggregate distribution. This type of distribution was confirmed by the negative binomial distribution *k* parameter values by the maximum likelihood method, which were positive and less than two for most evaluations, indicating high egg aggregation level (Elliott, 1979) (Table 1).

However, such behavior was not observed in some studies on the spatial distribution of

lepidopteran eggs. Okolle et al. (2006) while studying the spatial distribution of *Erionota thrax* Linnaeus, 1767 (Lepidoptera: Hesperidae), in banana crops in Malaysia, reported that pest eggs and caterpillars are randomly distributed in the area.

The spatial distribution of insects is generally aggregate, and this behavior is described by the negative binomial distribution, whereas the Poisson distribution may provide a better adjustment to the lower counts of individuals, which may occur in random arrangements (Southwood, 1987; Taylor, 1984).

Initially, the adjustment of data to the Poisson distribution was tested. Subsequently, data adjustment test was performed for the negative binomial distribution, since means were lower than the variances in all samples of *C. includens* eggs. The Chi-square test values were significant at 1% or 5% probability when adjusted for Poisson distribution for most samples, demonstrating that egg distribution is not random.

Regarding the adjustment of data to the negative binomial distribution, values were not significant in 82% of evaluations, confirming that the spatial distribution of *C. includens* eggs was aggregate (Table 2).

Table 2. Chi-square (X^2) test for adjustment of Poisson and negative binomial distributions for *Chrysodeixis includens* eggs (Walker, 1858) (Lepidoptera: Noctuidae) in soybean crop. Jaboticabal - SP, Brazil 2012/13.

Sampling dates	Poisson		Negative Binomial	
	X^2	d.f.	X^2	d.f.
24 DAE	2.7337 ^{NS}	1	1.3958 ^{NS}	1
30 DAE	1.2073 ^{NS}	1	1.9279 ^{NS}	2
37 DAE	0.3999 ^{NS}	1	1.0651 ^{NS}	4
44 DAE	8.8678*	2	4.0509 ^{NS}	2
49 DAE	13.0377**	2	1.4245 ^{NS}	3
57 DAE	16.1143**	2	3.0446 ^{NS}	2
65 DAE	16.2288*	6	1.3617 ^{NS}	4
72 DAE	35.0649**	9	5.6489 ^{NS}	11
78 DAE	55.5077**	5	4.9974 ^{NS}	4
84 DAE	9.5848**	2	8.0293*	3
93 DAE	12.2054**	2	6.1179*	2

X^2 = Chi-square test statistic; d.f. = number of the chi-square degrees of freedom; ** Significant at 1% probability; *Significant at 5% probability; ^{NS}Not significant at 5% probability; DAE = days after plant emergence.

Adjustment duality to Poisson and negative binomial distributions was observed in the first three weeks of sampling, and these results are probably related to the low number of eggs sampled at these dates. This overlap of both distributions was previously reported by Mallampalli and Isaacs (2002), when studying the distribution of lepidopteran eggs in blueberry plants in the United States.

Several authors have reported this behavior for other pest arthropods, such as *Spodoptera frugiperda* (JE Smith) in maize (Farias et al., 2001), and in cotton (Fernandes et al., 2002), *Alabama argillacea* (Hübner) in cotton (Fernandes et al., 2003), *Diaphorina citri* Kuwayama in citrus (Costa et al., 2010) and *Euschistus heros* (F.) in soybean (Souza et al., 2018).

The early detection of eggs is extremely important in a sampling scheme because it is used to direct pest control before damage to plants occurs. The results obtained for the spatial distribution of *C. includens* eggs should be added to previous research data on integrated pest management.

The development of a sampling plan for *C. includens* could contribute to the integrated management of this pest in relation to the decrease in synthetic insecticide applications. Research on *C. includens* oviposition can be valuable tools in predicting the occurrence of this pest in the field. Further studies should be carried out as they may provide crucial information regarding pest control strategies.

The spatial distribution of *C. includens* eggs

was aggregate according to the interpretation of values obtained for the dispersion indexes, and the negative binomial distribution was the most appropriate probabilistic model to represent the distribution of *C. includens* eggs.

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