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Fixed-Precision Sequential Sampling Plan of *Syringopais temperatella* (Lep., Gelechiidae) in Wheat Fields of Iran

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ABSTRACT

Cereal leaf miner, *Syringopais temperatella* Led. (Lep., Gelechiidae), is an important wheat pest in many regions of the world, including Iran. Fixed precision sequential sampling plan, a cost-efficient method for estimating pest population density, has been used for developing a successful IPM program. In this study, the fixed precision sequential sampling plan of *S. temperatella* larvae was developed on wheat, cultivar Verinac®, during 2017-2019 growing seasons in Iran. For this purpose, first, spatial distribution of the larvae on wheat leaves was determined using Taylor's power law and Iwao's patchiness regression. The spatial distribution of the larvae was aggregative on the wheat leaves. Taylor's power law provided a better fit for the data than Iwao's patchiness regression. Therefore, Green's model was used for developing the fixed precision sequential sampling plan. The optimum sample sizes of the larvae ranged from 5- 68 plants and 12- 189 plants according to the average of larval density at precision levels D= 0.25 and D= 0.1, respectively. Estimated stop lines showed that the sampling must be continued until the cumulative number of the pest larvae reaches 3.45 (D= 0.25) and 83.76 (D= 0.1) per plant. Accuracy of the sampling plan was validated by RVSP software.

Keywords: Cereal leaf miner, Green model, Integrated pest management, Taylor's power law.

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INTRODUCTION

Wheat, *Triticum aestivum* L., is the dominant crop in temperate countries, including Iran, being used for human food and livestock feed (Shewry, 2009). The cereal leaf miner, *Syringopais temperatella* Led. (Lep., Gelechiidae), in one of the economically important pests in many countries, especially in the Middle East (Al-Zyoud *et al.*, 2009). The pest is considered as an important wheat pest in west, southwest, and south provinces of Iran (Jemsi *et al.*, 2002a; Jemsi, 2006). The pest has one generation in southwest of Iran and its activity was recorded during 4.5 to 5 months on various cereals in Khuzestan Province, southwest Iran (Jemsi, 2002a). The pest larvae damage plants by mining into the leaves, feeding on cells of the internal tissues of the blades and leaving the epidermis transparent. Infested leaves are conspicuous by their light brown color (Yaman and Jarjes, 1971).

Fixed precision sequential sampling plan of some leaf miners were studied. For instance, fixed sequential sampling of *Liriomyza huidobrensis* Blanchard (Dip., Agromyzidae) on celery (Heinz and Chaney, 1995) and Lettuce (Burgio *et al.*, 2005), *Phyllocnistis citrella* Stainton (Lep., Gracillaridae) on lime (Pena and Schaffer, 1997), *L. sativa* Blanchard on cucumber (Namvar *et al.*, 2012), *L. trifolii* Burgess on tomato (Lee *et al.*, 2005), *Cameraria ohridella* Deschka & Dimic (Lepidoptera: Gracillariidae) on horse chestnut tree (Ferracini and Alma, 2007),

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Tuta absoluta Meyreck (Lep., Gelechiidae) on tomato (Cocco *et al.*, 2015), and *Phthorimaea operculella* Zeller (Lep. Gelechidae) on potato (Shahbi and Rajabpour, 2017) were studied.

There has not been any effort to develop a fixed precision sequential sampling plan in the case of *S. temperatella* on wheat (cultivar Verinac®). Therefore, the objective of this study was to develop fixed-precision sequential sampling plan of the pest larvae in wheat fields.

MATERIALS AND METHODS

Experimental Design

The experiments were performed during two growing seasons, 2017/2019, in an experimental wheat field, two hectares, in Masjed Soleiman District, Khuzestan Province, southwest of Iran (31° 45' 03.3" N 49° 28' 04.8" E). Seeds of a commercial wheat cultivar, Verinac®, were cultivated (≈80 plants per m^2) in the experimental field. Cultural practices were carried out according to practical advisements of Khuzestan Agricultural Organization and no insecticides were applied during the study.

Sampling

The first sampling was started when the first moth of *S. temperatella* was trapped by solar energy-based pest trap (Raha Andish Kavan Company, Tehran, Iran) (Sermsri and Torasa, 2015). For this purpose, two traps were randomly placed in the experimental field. Samplings were usually carried out at weekly intervals. At each sampling date, twenty plants were randomly selected by walking in an X-shaped pattern through the field. From each selected plant, three leaves from top, middle, and bottom tillers of the plant were chosen and number of larvae was recorded.

Spatial Distribution

Taylor's power law and Iwao's patchiness regression were used to evaluate spatial distribution of *S. temperatella* larvae on wheat. Taylor's power law describes the regression between logarithm $_{(10)}$ of population variance and logarithm $_{(10)}$ of population mean according to the following equation:

$$Log(s^2) = a + bLog(\overline{X}) \quad (1)$$

Where, S^2 is the larval population variance, $\overline{X}$ is larval population mean, *a* is the Y-intercept, and *b* is the slope of regression line. The regression slope "*b*" is an index of species spatial pattern. When $b<1$, $b=1$, and $b>1$ spatial distribution pattern of the larvae are uniform, random, and aggregated, respectively (Southwood, 1978, Rajabpour and Yarahmadi, 2012; Kafeshani *et al.*, 2018). Goodness of fir of Taylor's power law was obtained by calculating regression coefficient. Two-tailed *t*-test at $n-2$ degrees of freedom was performed to determine if slope and regression coefficient values of the regression relation were significantly different from 1 and 0, respectively (Snedecor and Cochran, 1980).

Iwao's patchiness regression was used to quantify the relationship between mean crowding index (X^*) and mean $\overline{X}$ by using the following equations:

$$X^* = \alpha + \beta\overline{X} \quad (2)$$

Where,

$$X^* = \overline{X} + (\frac{S^2}{\overline{X}}) - 1 \quad (3)$$

Different linear regressions were tested for heterogeneity of slopes (Sokal and Rohlf, 1995) by Analysis Of Covariance (ANCOVA) of data collected from different growing years using SPSS software (Version 16.0).

Student's *t*-test can be used to determine if the colonies are randomly dispersed:

In Taylor's power law: $t = (b-1)/SE_b$, $b = 1$ (4)

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In Iwao's patchiness regression*:* $t=(\beta-1)/SE_{\beta}$, $\beta= 1$ (5)

Where, SE_b and SE_{β} are the Standard Errors of the slope for Taylor's power law and Iwao's patchiness regression models, respectively. Calculated value of t is compared with tabulated value of t with $n-2$ degrees of freedom. 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 or b< 1, the spatial distribution would be aggregated or uniform, respectively (Kafeshani *et al.*, 2018).

Fixed-Precision Sequential Sampling Plan

The optimum sample size (n) needed to estimate *S. temperatella* density at two levels of fixed precision, 0.25 and 0.1, was calculated using the following equation:

$$n = \frac{a\bar{X}^{\,b-2}}{D^2}, \qquad (6)$$

Where, D is a fixed precision level, and a and b are coefficients obtained from the regression of Taylor's power law (Buntin, 1994). Precision levels of 0.25 and 0.1 are generally acceptable for sampling in IPM and research purposes, respectively (Southwood, 1978).

Due to fitted data with Taylor's mean-variance model, the Green's method was used to calculate stop lines of fixed-precision sequential sampling (Naranjo and Hutchison, 1997). The stop lines of *S. temperatella* in wheat fields were estimated as:

$$T_n \geq ((an^{1-b} / D^2)^{(1/(2-b))}) \qquad (7)$$

Where, T_n is the insect cumulative number in n samples, and a and b are the Taylor coefficients, D is the Desired precision level.

Validation of Sampling Plan

The sequential sampling plan was validated by RVSP (Resampling for Validation of Sampling Plans) software based on Naranjo and Hutchison (1997) method. The software requires independent data sets to serve as validation data sets (Shahbi and Rajabpour, 2017; Kafeshani *et al.*, 2018). Hence, ten independent data sets with a range of low, medium, and high density levels were randomly selected from a total of 35 data sets, which were collected in two growing seasons. The mean densities of these data sets for the pest larvae ranged from 0.02 to 3.33 larvae per plant. The sample size of each data set consisted of 20 plants. The data was not used in Taylor's power law regression. Simulations were carried out using 500 re-samplings without replacement.

RESULTS

Spatial Distribution

Spatial distribution patterns and parameters of *S. temperatella* according to Taylor's power law and Iwao's patchiness regression on wheat are presented in Table 1.

Taylor's power law provided a significant relationship between variance and mean density of the pest on wheat. No significant relationship was observed between mean crowding and mean density of *S. temperatella* on wheat according to Iwao's patchiness regression. The aggregation incidence (b) of Taylor's power law was significantly more than 1, which indicates aggregative dispersions of *S. temperatella* larvae on wheat.

Fixed-Precision Sequential Sampling Plan

The optimum sample size of *S. temperatella* at fixed precision levels of 0.25 and 0.1 in wheat fields are shown in Figure 1. With increasing larval densities, the optimum sample size dramatically decreased. Also, at precision level of 0.25, the optimum sample size was always lower than 0.1. The optimum sample size to estimate *S. temperatella* larval densities on wheat ranged from 5-68 plants and 12-189 plants at precision levels of 0.25 and 0.1, respectively.

Table 1. Spatial distribution statistics of *Syringopais temperatella* on wheat using Taylor's power law and Iwao's patchiness regression analyses.

Model/Statistics	N	Intercept±SE	Slope±SE	R^2	Spatial distribution	F	$P_{regressio}$
Taylor's power law	20	0.627±0.157	1.425±0.182	0.774	clumped	61.492	<0.0001
Iwao's patchiness regression	20	381.67±662.09	-628.2±553.2	0.017	clumped	0.332	<0.571

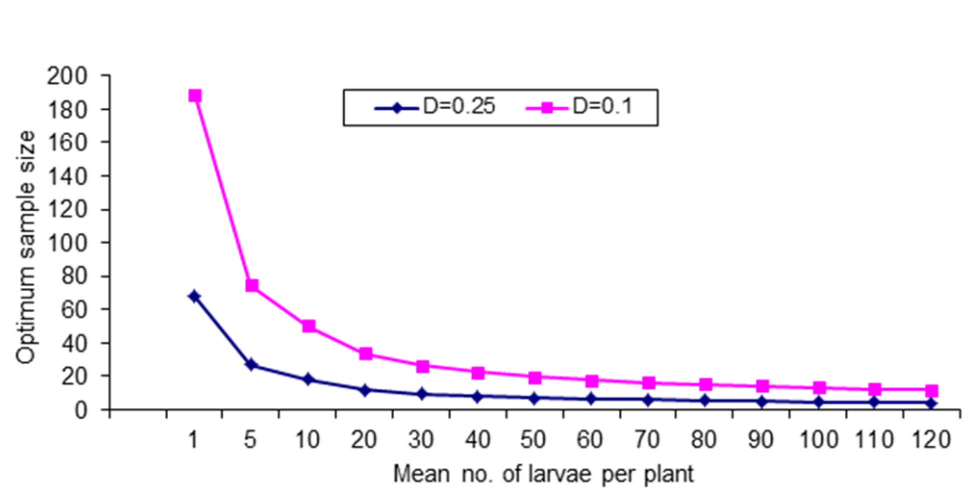


Figure 1. Optimum sample size of *Syringopais temperatella* on wheat at precision levels of 0.25 and 0.1.

Estimated stop lines using Green's model for *S. temperatella* in wheat fields are presented in Figure 2. Based on the estimated stop lines, numbers of required sampled plants to cross the stop lines are significantly changed. The results indicated that the sampling of the pest must be continued until the cumulative number of larvae on wheat plant reaches 3.45 and 83.76 larvae per plant at precision levels of 0.25 and 0.1, respectively.

Validation of Developed Sampling Plan

Estimated sample sizes of *S. temperatella* on wheat according to resampling analysis using RVSP software are shown in Figure 3. The means of sample sizes for *S. temperatella* on wheat were 140.1 and 389.1 plants at precision levels of 0.25 and 0.1, respectively.

For the 10 independent data sets covering different densities, the average precision levels for *S. temperatella* larvae on wheat plant were 0.26 and 0.108 at precision levels of 0.25 and 0.1, respectively, which were close to the desired precision (Figure 4).

DISCUSSION

In our experiments, Taylor's power law provided a better fit to the data than Iwao's patchiness regression. Similar results were obtained for some other Gelechiid leaf miners including *T. absoluta* on greenhouse cucumber (Cocco *et al.*, 2015), and *P. operculella* on potato (Shahbi and Rajabpour, 2017). Spatial distribution of *S. teperatella* on wheat leaves was aggregative. Similarly, spatial distributions of some lepidopteran leaf miners were reported as aggregative, including *C. ohridella* on horse chestnut (Ferracini and Alma, 2007), *P. citrella* on lemon (Liu *et al.*, 2008), and *T. absoluta* on tomato (Cocco *et al.*, 2015; Ghaderi *et al.*, 2018). However, spatial

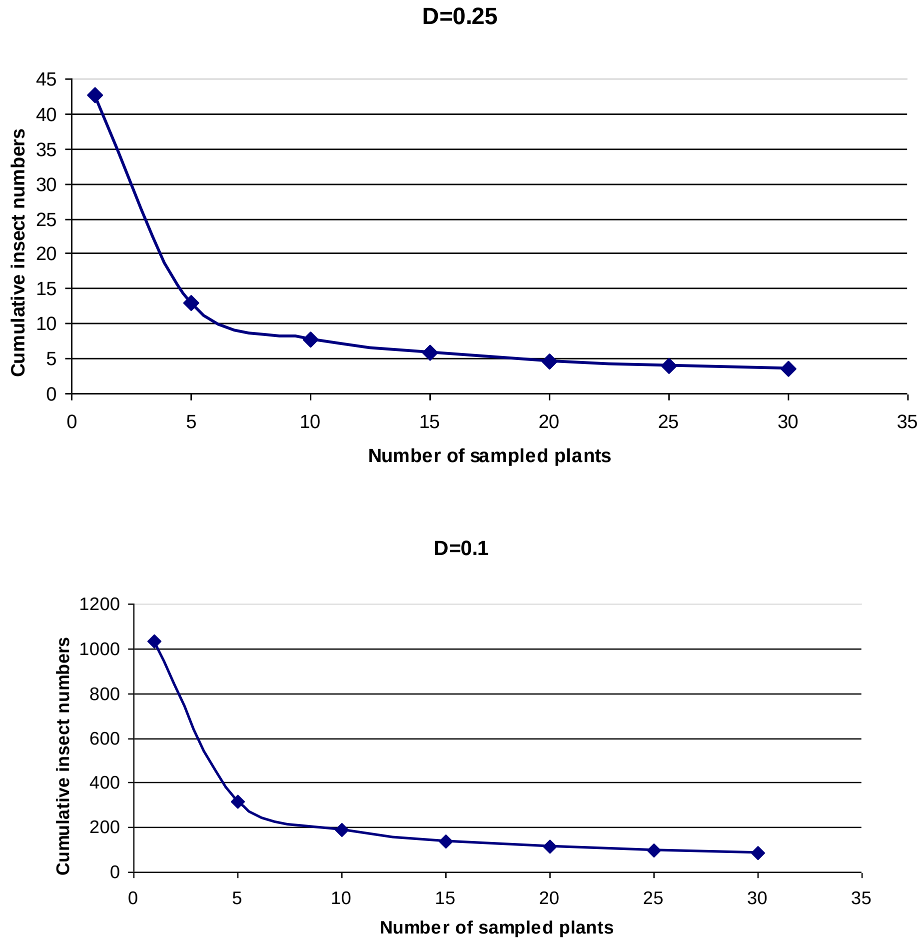
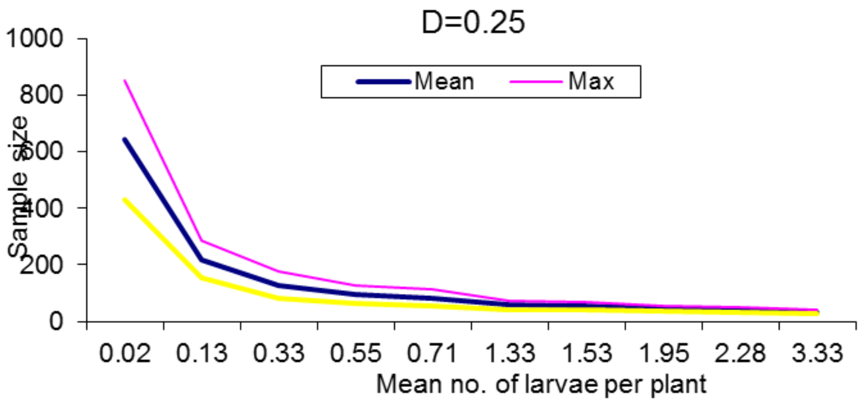


Figure 2. Fixed precision sequential sampling stop lines for *Syringopais temperatella* on wheat at precision levels of 0.25 and 0.1.

distributions of some leaf miner larvae, e.g., *Lyonetia speculella* Clemens (Lepidoptera: Lyontiidae) on apple (Brown, 1989) and *Erionota thrax* L. (Lepidoptera, Hesperiidae) (Okolle *et al.*, 2006) were random. The difference in results may be due to the differences in host plant. Moreover, many factors related to host plant, including morphological difference among cultivars (Shahbi and Rajabpour, 2017) and weed status (Dinarvand *et al.*, 2019), and/or factors related to pest including oviposition completion patterns (Damos, 2018) affect spatial distribution parameters. Jemsi *et al.* (2002b) reported spatial distribution of the pest larvae as random in wheat fields. In our study, spatial distribution of larvae on wheat leaf was determined as aggregative. Therefore, different sample universe is the main reason for the different results.

Optimum sample size to estimate *S. temperatella* larval densities strongly depended on the desired precision level and the pest larval density (from 5-68 plants and 12-189 plants according to the average of larval density at precision levels of 0.25 and 0.1, respectively). The number of samples required to attain a certain precision was a strong function of density; higher sample size was required at the lower pest density. This was due to the relationship between the mean and the variance of the pest densities as expressed by the slope of Taylor's regression (Kapatos *et al.*, 1998).



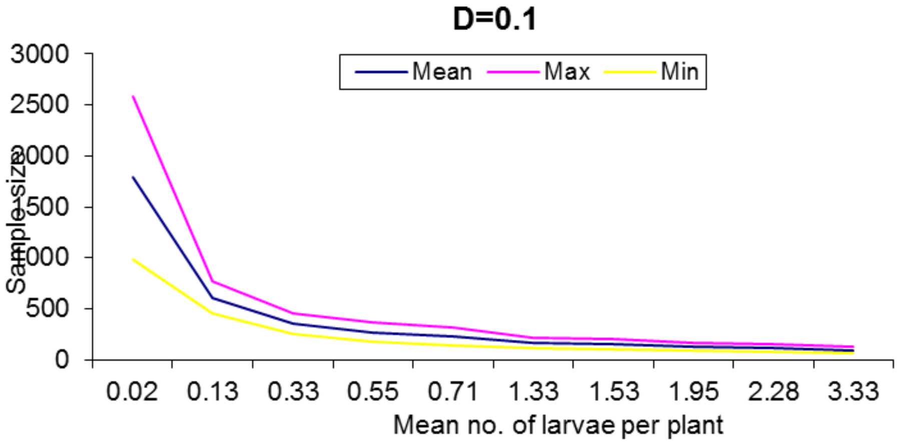


Figure 3. Summary of re-sampling validation analysis using 10 independent data sets of *Syringopais temperatella* on wheat showing the calculated sample size means (±SE) for Green's sequential sampling plan at precision levels of 0.25 and 0.1.

Similar to the optimum sample sizes, the estimated stop lines of the larvae were different based on the desired precision level (3.45 and 83.76 cumulative larvae numbers per plant at precision levels of 0.25 and 0.1, respectively). Totally, the optimum sample sizes and sampling stop lines were increased by increasing the desired precision level (from 0.25 to 0.1). Similarly, Afshari *et al*. (2009) stated that the optimum sample size was flexible and depended upon the aphid density and desired level of precision.

The developed sequential sampling could not be compared with other studies because there were no previous studies to develop a fixed-precision sequential sampling of *S. temperatella* on wheat or other host plants. However, some studies were done to develop the sampling plan for estimating population densities of other pests belonging to Gelechiidae. For instance, the developed fixed precision sequential sampling plan for estimating population of *P. operculella* on different cultivars showed that the required optimum sample size ranged from 149 to 1,054 leaves based on precision level, 0.1 or 0.25, larval population density, and potato cultivar (Shahbi and Rajabpour, 2017). Moreover, the optimum sample size for population monitoring of *Anarsia lineatella*

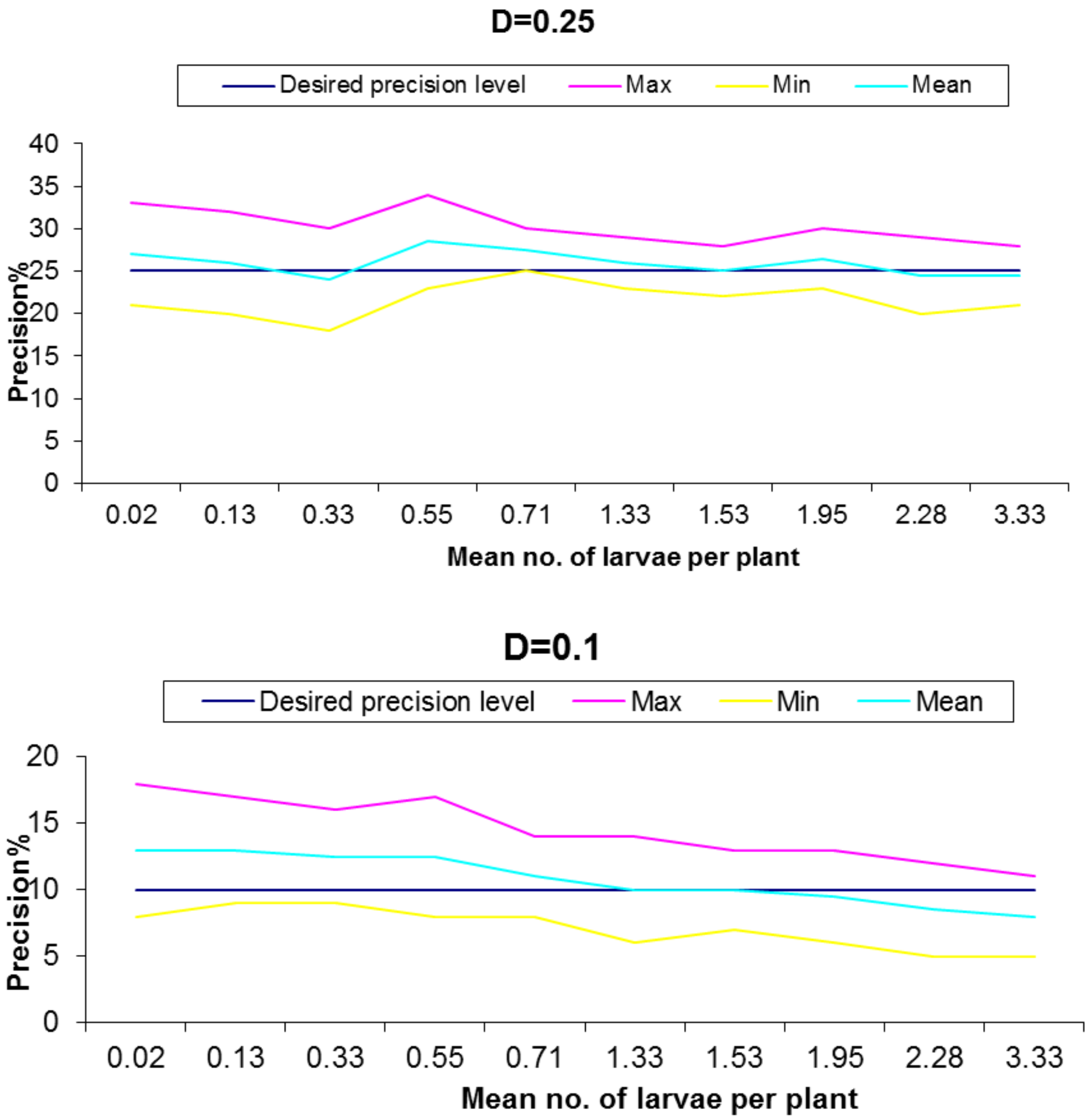


Figure 4. Summary of re-sampling validation analysis using 10 independent data sets of *Syringopais temperatella* on wheat showing the calculated precision level means (±SE) for Green's sequential sampling plan at precision levels of 0.25 and 0.1.

Zeller at the precision level of 0.2 varies from 3 to 10 samples according to the larval density (Damos, 2018).

Some studies were performed to develop fixed precision sequential sampling of wheat pests (Boeve and Weiss, 1998; Elliott, 2003; Parker *et al.*, 2002; Fathi and Bakhshizadeh, 2014). All of these studies are not comparable with our studies since all fixed precision sequential sampling models, Green's or Kuno's model, basically depend on spatial distribution parameters of each pest species. Spatial distribution is one of the most characteristic ecological properties of species. Therefore, the parameters are different according to pest species. Moreover, many factors related to host plant, environmental conditions, competitions, etc. can affect the spatial distribution parameters (Taylor, 1984).

CONCLUSIONS

Spatial distribution of *S. temperatella* larvae on wheat leaves was aggregative. The optimum sample sizes of the larvae ranged from 5-68 plants and 12-189 plants according to the average of larval density at precision levels of 0.25 and 0.1, respectively. The sampling must be continued until the cumulative number of larvae per plant reaches 3.45 (at precision level 0.25) and 83.76 (at precision level 0.1) larvae per plant. Results of this study can be used in integrated pest management program of *S. temperatella* in wheat fields.

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نمونه‌برداری دنباله‌ای با دقت ثابت از *Syringopais temperatella* (Lep., Gelechiidae) در مزارع گندم ایران

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چکیده

مینوز برگ غلات *Syringopais temperatella* Led. (Lep., Gelechiidae) یکی از آفات مهم گندم در بسیاری از مناطق جهان از جمله ایران می‌باشد. نمونه‌برداری دنباله‌ای با دقت ثابت (روشی مقرون به صرفه برای تخمین تراکم جمعیت آفت) برای توسعه برنامه موفق مدیریت تلفیقی آفات مورد استفاده قرار می گیرد. در این مطالعه، برنامه نمونه‌برداری دنباله‌ای با دقت ثابت لاروهای *S. temperatella* روی گندم (رقم وریناک®) در طول فصل‌های زراعی ۱۳۹۸-۱۳۹۷توسعه یافت. برای این منظور در مرحله اول، توزیع فضایی این لاروها روی برگ‌های گندم با استفاده از شاخص تایلور و رگرسیون آیواوو تعیین شد. توزیع فضایی این لارو روی برگ‌های گندم تجمعی بود. بنابراین از مدل گرین برای توسعه این برنامه نمونه-برداری دنباله‌ای با دقت ثابت استفاده شد. تعداد نمونه بهینه لاروی براساس میانگین تراکم لاروی به ترتیب از ۵ تا ۶۸ و از ۱۲ تا ۱۸۹ گیاه به ترتیب در سطوح دقت D=25 و D=0.1 متغیر بود. خطوط توقف محاسبه شده نشان داد که نمونه‌برداری می‌بایست تا زمانی که تعداد تجمعی لاروهای آفت به ۳/۴۵ (D=0.25) و ۸۳/۶۵ (D=0.1) در هر گیاه برسد، ادامه داشته باشد. درستی این برنامه نمونه برداری توسط RVSP مورد تایید قرار گرفت.

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