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#### Abstract

A total of 24 data sets was available from April until September 2018 for developing a sampling plan to monitor the Panonychus ulmi population. Action threshold of 2.31 mobile forms / sample unit was determined using the empirical model, which based on the presence or absence of one or more mite per sample unit. The mean-variance relationship was established, and the threshold mite density was used for developing Green's model at various precision levels ( $\mathrm{D}=$ $0.20,0.25,0.30,0.35$ and 0.50 ), the sequential probability ratio test of Wald, and Iwao's model. All established sequential plans were thereafter evaluated through 500 Monte Carlo iterations. Result of Green's plan showed that the mean sample numbers decrease significantly with increasing precision. For Wald's model, the presence of a common k ( $k_{c}$ ) of negative binomial distribution was verified, and its value is calculated according to Bliss and Owen's method. A minimum of 19 sample units is required to detect the presence of harmless populations of P. ulmi, comparatively to 4 units obtained by Iwao's sequential sampling plan, which is an alternative to overcoming $\alpha$ and $\beta$ error values and does not need any mathematical distribution of P. ulmi. The terminology and methodology of each plan were clarified, and the results are discussed. Developing a reliable sampling method for monitoring P. ulmi is essential for implementing integrated pest management measures.


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## I. INTRODUCTION

The European red spider mite, Panonychus ulmi (Koch, 1836) (Acari: Tetranychidae), is a significant damaging mite pest of apple at lower population densities. The damage closely resembles magnesium deficiency, bright-yellow patches develop on the leaflets, and gradually coalesce until the whole leaf withers and die (Foster and Barker, 1978).

Development and reproduction s of $P$. ulmi have been studied under controlled conditions. The previously published results reported the effect of temperature on development and life history parameters (Northcraft and Watson, 1987; Qui and Li, 1988; Zhou et al., 2006; Zou et al., 2018), this species develops and reproduces rapidly at high temperatures and can close a considerable number of generations during the growing season. In front of severing attacks of P. ulmi, acaricides are sprayed frequently during the growing season to limit decreasing the phytosanitary status of crops (Plaut and Cohen, 1967).

Panonychus ulmi can develop resistance to miticides and cross-resistance to other pesticides when it has developed resistance to one pesticide (Dağlı and Tunç, 2001). It is recommended often to use integrated pest management programs
wherever possible (Mollet and Sevacherian, 1984; Çakmak et al., 2005). The spray program for other pests and diseases should be designed to have the minimum disruptive effect on predatory mites in biological control cases (Sarwar, 2013).

An aspect of any pest management program is having a sampling procedure that allows determination of when a pest population is so abundant, as to require control. Most of these protocols consist of at least two items: a threshold pest density, which, when exceeded by a local population, dictates management intervention (usually a pesticide application), and a reliable and efficient method for estimating or classifying pest abundance. Much work devoted to devising and analyzing sampling methods that-maximize precision while minimizing sampling costs (Kuno, 1991; Binns \& Nyrop, 1992). Counting all mites on a single sample unit often takes an hour or more. Consequently, few researchers or pest control advisors are willing to commit the time necessary for whole sampling plants for mites (Wilson and Room, 1983).

In this document, we develop binomial and sequential sampling plans that can be used to efficiently monitor $P$. ulmi through time, and how the performance of any plan was assessed by the use of resampling for validation of sequential plan program (RVSP). We describe the methods by specifying in a general way what it is designed to do for establishing the constituents of each plan. Then we discuss the results and compare the efficiency of each monitoring protocol of $P$. ulmi with the potential effect of some parameters on the performance of each procedure.

## II. MATERIAL AND METHODS

### 2.1 Sampling data

The data have been collected on Jeromine variety. The area of the sampled plot is 1.5 ha. Fifty sampling positions were selected, and 100 apple leaves are harvested weekly from April to September 2018. A total of 24 data sets was available for statistical and modelling analysis.

Field data were recorded as a proportion of the infested sample unit and mean density of Panonychus ulmi during each sampling occasion.

### 2.2 Fixed-sample-size plan:

The binomial sampling plan based on data of the presence or absence of one or more P. ulmi per sample unit has been developed. This empirical model is practically simple, and convenient for monitoring the mite pest populations without the need of any mathematical distribution. The density- infestation level relationship established by using Nachman's function (Nachman, 1984), help in determining theoretically the economic threshold corresponding to mite density considered as an intervention limit. The value of the economic threshold will be used to develop sequential sampling plans.

### 2.3 Fixed-precision sampling plan

The sequential sampling plan of Green (1970) was developed from the model of Kuno (Kuno, 1969). The mean-variance relationship is fitted by using Taylor's power law: $s^{2}=a m^{b}$ (Taylor, 1961), a and b , respectively are the regression intercept and regression slope. Both parameters were expressed under logarithmic in the formulae given by Southwood (1978).

$$
\left(s^{2}\right)=\log \log (a)-b \log (m)
$$

In this plan, a decision to take additional sample units should be terminated when the defined precision level was achieved. The fixed-precision stop line was calculated as:

$$
\log \left(T_{n}\right)=\left(\frac{\log _{10}\left(\frac{D^{2}}{a}\right)}{b-2}\right)+\left(\frac{b-1}{b-2}\right) * \log _{10}(n)
$$

Where $T_{n}$ is the cumulative density of $P$. ulmi, a and $b$ are respectively, Taylor's regression parameters. $D$ is the fixed-precision, and n is the sample size. $D$ was expressed as the ratio of the standard error of the mean to mean ( $D=$ SEM/mean) at precision levels of $0.20,0.25$, 0.30, 0.35, and 0.50.
2.4 Sequential probability ratio test (SPRT) of Wald (1945)

The Wald's SPRT test (Wald, 1945) is based on two hypotheses instead to three hypothesis procedure developed by Nyrop et al. (1994) for tripartite classification of mite pest density, the null hypothesis ( $H_{0}: x=x_{0}$ ) against the alternative hypothesis $\left.\left(H_{1}: x=x_{1}\right)\left(x_{o}<x_{t}<x_{t}\right\}\right)$, where $x_{t}$ is the economic threshold estimated from fixed-size sampling plan, $x_{o}$ and $x_{t}$ are parameters that are used to define an acceptable SPRT (Binns and Nyrop, 1992). Stop lines for sequential sampling plan can be computed by using predetermined lower and upper limits of economic thresholds ( $x_{o}$ and $x_{1}$ ) and risk levels for making type I ( $\alpha$ ) (probability of accepting the alternative hypothesis when the null hypothesis is correct) and type II ( $\beta$ ) errors (probability of accepting the null hypothesis when the alternative hypothesis is correct). Wald's sequential sampling plan is defined by its operating characteristic ( $O C$ ) and average sample number (ASN) of observations needed to make a classification given any mean value. The $O C$ and $A S N$ functions were computed from 500 Monte Carlo simulations and the dispersion parameter $k$ of the negative binomial distribution calculated at economic threshold according to the method of Bliss and Owen (1958).

### 2.5 Iwao sequential sampling plan

Iwao's method based on the relationship of Lloyd's (1967) index of mean crowding ( $x^{*}$ ) to $m$. Iwao (1968) showed that the regression of $\left(x^{*}\right)$ on $m$ is linear and described by:
$x^{*}=\alpha+\beta m$. $\alpha$ (intercept) is termed the index of basic contagion and describes the component of the population. $\beta$ (slope) is termed the density-contagiousness coefficient related to the distribution pattern of these components within the habitat, with values of $\beta>1$ representing an aggregated distribution, $\beta=1$, and $\beta<1$ corresponding to $a$ random, and regular distribution, respectively. The stabilized variance $\left(s^{2}\right)$ was given by Iwao and Kuno (1968):
$s^{2}=(\alpha+1) m+(\beta-1) m^{2}$. Where $m=$ mean, while $\alpha$ and $\beta$ are parameters of the relationship. $\alpha$ and $\beta$ were calculated by plotting mean crowding ( $x^{*}$ ) on mean densities ( $m$ ).

The method developed by Iwao, 1968 (1975) for distributions exhibiting a linear relationship for the regression of $x^{*}$ on $m$ should be used. The upper $\left(T_{U}\right)$ and lower $\left(T_{L}\right)$ limits of the plan were calculated from:

$$
\begin{aligned}
& T_{U}=N * E T+t \sqrt{N(\alpha+1) E T+(\beta-1) E T^{2}} \\
& T_{L}=N * E T-t \sqrt{N(\alpha+1) E T+(\beta-1) E T^{2}}
\end{aligned}
$$

Where $N=$ number of samples taken; $E T=$ Economic Threshold; $t=$ Student's t value at the chosen level of significance of a test bilateral for an infinite number of degrees of freedom; $\alpha=$ contagion index (intercepted) and $\beta=$ (slope) or distribution coefficient.

The maximum number of samples required to decide if the critical density remains between the upper and lower limits, and is given by:
$N_{\text {max }}=\left(\frac{t}{D}\right)^{2}\left((\alpha+1) E T+(\beta-1) E T^{2}\right)$. With $D$ is the precision level.

The performance of the sequential sampling plans must be evaluated through computing probability curves and average sample numbers ASN from 500 Monte Carlo simulations. We have used Resampling for the Validation Sampling plan software Program (RVSP) (Naranjo and Hutchison, 1997), R (3.4.3) program for statistical adjustment tests, and Excel 2013 for graphing.

## III. RESULTS AND DISCUSSION

### 3.1 Variance-mean relationships:

Because the variance increased significantly faster with the mean densities, since it is normal to having the high variance values in aggregative distribution case, the variance was stabilised using Taylor $\left(s^{2}=a m^{b}\right)$ and Iwao models $\left(s^{2}=\right.$ $\left.(\alpha+1) m+(\beta-1) m^{2}\right)$. Means and variances from the
0.22-9.23 P. ulmi per sample unit were fit to a straight line by using Taylor's power law (Figure 2) and Iwao's regression (Figure 8). For both models, the equations respectively are : $\left(\log \left(s^{2}\right)=\right.$ $0.30+1.14 \log (m) ; a=0.30, b=1.14, r^{2}=0.97, \mathrm{dl}=$ 22, $P<0.05$ ) ) and ( $x^{*}=1.10 m+0.9, \alpha=0.007$, $\left.\beta=1.10, r^{2}=0.99, d l=22, P<0.05\right)$. The slope values greater significantly than 1 attesting that the pattern dispersion of $P$. ulmi is aggregative.

Based on the values of $r^{2}$ for both models, such comparison is only valid if the variables and scale transformations are the same, and the number of parameters of the models is the same (Kvålseth,
1985). The same data sets were used to predict variance, and a significant correlation has been observed. Or, even though the $r^{2}$ values were similar, the functional relationship was very different for each, because the dependent variables are not the same and the data scales are different (Kvålseth, 1985; Scott and Wild, 1991).

### 3.2 Fixed-sample-size plan

The theoretical values of mean densities corresponding to infestation levels as well as confidence and prediction intervals were given in Table 1.

Table 1: Relationship between the proportion of infested sample units and densities of P. ulmi according to Nachman function with confidence (CI) and prediction intervals (PI) to 95\%

| Panonychus ulmi |  |  |  |
| :---: | :---: | :---: | :---: |
| Infestation level | Mean density | $\pm \mathrm{CI}$ | $\pm \mathrm{Pl}$ |
| 0.10 | 1.02 | $0.96-1.11$ | $0.37-2.90$ |
| 0.20 | 1.94 | $1.74-2.17$ | $0.68-5.32$ |
| 0.30 | 2.31 | $2.10-2.54$ | $0.82-6.48$ |
| 0.40 | 3.59 | $3.28-4.01$ | $1.28-10.07$ |
| 0.50 | 5.24 | $3.88-4.89$ | $1.55-12.23$ |
| 0.60 | 6.20 | $4.90-6.67$ | $2.39-14.31$ |
| 0.70 | 9.23 | $5.46-7.05$ | $2.21-17.44$ |
| 0.80 |  | $6.82-8.89$ | $3.55-19.23$ |
| 0.90 | $5.41-10.12$ | $5.21-23.44$ |  |

The relationship between $P$. ulmi density and proportion of infested sample units was established through linear regression, the values of statistical parameters are obtained by plotting mean densities on infestation level ( $a=1.64, b=$ $\left.0.78, r^{2}=0.91, d 1=22 . F=63.142, P<0.05\right)$ (Figure 1).


Figure 1: Binomial sampling stop line plan and theoretical relationship between the proportion of infestation level and the mean density of $P$. ulmi obtained by using Nachman's model.

Based on 24 data sets, the Fixed-sample-size sampling plan or positive binomial was developed first without considering a theoretical distribution of mite (Poisson, binomial, and negative binomial distributions). The Fixed-sample-size sampling plan is an empirical model overcoming the calculation of common $k \quad\left(k_{c}\right)$ when the distribution of mites conforms to negative binomial law (Anscombe, 1950). The relationship between densities of $P$. ulmi and the proportion of infested sample units was established using Nachman function, allowing to grant theoretically at each infestation level a density value within confidence and prediction intervals at-risk level of 5\% (Nyrop et al., 1989). Empirical model using Nachman's function corrects the variations related to the proportion of infested sample units and mean density changes due to interactions with potential zoological and natural systems in the presence of natural enemies, for example.

A significant correlation was observed between mean densities and infestation level ( $r^{2}=0.91, P<$ 0.05 ). The proportion of infested sample units increased slowly with an increase in mean densities (Figure 1). The densities of less than two mites do not exceed a proportion of $25 \%$, while about $90 \%$ of leaves were infested at an average density of $9.23 P$. ulmi / sample unit. The mean
density of $2.31 P$. ulmi/ sample unit relative to 30 $\%$ of infestation and $\alpha$ value of 0.05 was used to develop and to valid sequential sampling plans.

Our results can be compared with those of several previous studies using empirical models (Croft et al., 1976; Gerrard \& Chaing, 1970; Wilson \& Room, 1983; Hilbert \& Logan, 1983; Opit et al., 2003). Although the predatory mite, Typhlodromus (T.) setubali (Dosse, 1961), is present in the sampled plot, we found that the aggregation of the $P$. ulmi population was not greatly affected. Or, in biological control of mite pests, the mean densities and associated variances and the $k_{c}$ value can be varied during the season according to their spatial distribution (Anscombe, 1950; Shaw et al., 1998; Giles et al., 2000; Nyrop et al., 1994).

### 3.3 Fixed-precision sampling plan

Based on Taylor's power law, the log variance against log means regression showed that the variance increased significantly with mean density ( $r^{2}=.97, P<0.05$ ) (Figure 2). Intercept value ( $a$ ) was 0.30. The slope (b) value of 1.14 greater than one, indicating that the dispersion pattern of the $P$. ulmi population is aggregative ( $P<0.05$ ).

[^0]

Figure 2: Mean-variance relationship using Taylor power law. Taylor's model was estimated by using

$$
\log \left(s^{2}\right)=\log (a)+b \log (m)
$$

Green's fixed-precision stop lines for estimating the density at $D=0.20 ; 0.25 ; 0.30 ; 0.35$ and 0.50 are given in Figure 3. These stop lines show that fewer samples were needed for estimating high densities, and more samples were needed for estimating low densities. RVSP simulation with field data showed that densities $>2 P$. ulmi/leaf at a fixed precision of 0.20 could be estimated with 20 sample units (Table 2). A different trend is observed at a fixed precision of $0.25,0.30,0.35$, and 0.50 , where the number of sample units needed decreases respectively to 13,11 , and 10 leaves. However, the density of 2.31 mite/leaf could be estimated with 20 sample units, while more than 20 leaves are needed for estimating densities < 2.31 at fixed precision of 0.20 and much more for low densities. Generally, the average sample number decreases inversely with fixed-precision values. The range of $P$. ulmi densities that can be estimated with 20 sampling units changes according to the level of precision chosen. At the fixed- precision of 0.50 and 0.35 , 20 sample units are enough to estimate densities ranging from 0.7 to 9.29 mites/leaf and 1.03 to 9.29, respectively, whereas at $0.20,0.25$ and 0.30 , more sample units were needed for estimating densities less than 1.87 mite pest/ leaf.

The observed precision in estimating $P$. ulmi densities of 0.22-9.29 mites was $\leq 0.20$ when $D=$
0.20 for 21 of 24 data sets. At the fixed precision of $0.25,0.30$, and 0.35 , the observed $D$ was $\leq$ these precision levels for 22 of 24 data sets. Finally, at the fixed precision of 0.50 , the observed precision was $\leq 0.50$ for all data sets. Observed precision was at or below the fixed precision in cases where the sample variance for the field data sets was equal to or less than the variance predicted by Taylor's power law because this law did not predict sample variance of field data at low mean densities.


Figure 3: Sequential sampling graph for $P$. ulmi. Both $T_{n}$ (cumulative densities) and n (average sample number) are logarithmic scales. Stop lines are calculated for precision levels ( $D$ ) of 0.20, 0.25, 0.30, 0.35 and 0.50 .

Table 2: Performance of Green's fixed-precision sequential sampling plan for estimating mean densities of $P$. ulmi at fixed-precision levels of $D=0.20,0.25,0.30,0.35$, and 0.50

| Data set | Mean ${ }^{\text {A }}$ | Variance ${ }^{\text {B }}$ | Predicted variance ${ }^{\mathrm{c}}$ | Fixed precision D |  |  |  |  |  |  |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  |  |  | $\mathrm{D}=0.20$ |  | $\mathrm{D}=0.25$ |  | $\mathrm{D}=0.30$ |  | $\mathrm{D}=0,35$ |  | $\mathrm{D}=0,50$ |  |
|  |  |  |  | $\mathrm{N}^{\text {D }}$ | $\mathrm{D}^{\mathrm{E}}$ | $\mathrm{N}^{\text {D }}$ | $\mathrm{D}^{\mathrm{E}}$ | $\mathrm{N}^{\text {D }}$ | $\mathrm{D}^{\mathrm{E}}$ | $\mathrm{N}^{\text {D }}$ | $\mathrm{D}^{\mathrm{E}}$ | $\mathrm{N}^{\text {D }}$ | $\mathrm{D}^{\mathrm{E}}$ |
| 1 | 0,22 | 0,430 | 0,353 | 187 | 0,21 | 122 | 0,26 | 85 | 0,30 | 64 | 0,35 | 33 | 0,48 |
| 2 | 0,71 | 1,279 | 1,351 | 69 | 0,19 | 44 | 0,24 | 32 | 0,28 | 24 | 0,33 | 13 | 0,44 |
| 3 | 0,77 | 1,755 | 1,483 | 65 | 0,21 | 42 | 0,26 | 29 | 0,31 | 22 | 0,36 | 13 | 0,47 |
| 4 | 1,03 | 2,110 | 2,069 | 50 | 0,20 | 32 | 0,24 | 23 | 0,29 | 18 | 0,34 | 11 | 0,42 |
| 5 | 1,87 | 2,556 | 4,098 | 30 | 0,11 | 19 | 0,14 | 14 | 0,17 | 11 | 0,19 | 10 | 0,20 |
| 6 | 2,95 | 7,555 | 6,908 | 20 | 0,17 | 13 | 0,21 | 11 | 0,24 | 10 | 0,24 | 10 | 0,24 |
| 7 | 3,86 | 9,988 | 9,400 | 17 | 0,19 | 11 | 0,22 | 10 | 0,23 | 10 | 0,23 | 10 | 0,23 |
| 8 | 4,53 | 12,965 | 11,292 | 14 | 0,15 | 10 | 0,18 | 10 | 0,19 | 10 | 0,19 | 10 | 0,19 |
| 9 | 5,65 | 13,271 | 14,544 | 12 | 0,12 | 10 | 0,13 | 10 | 0,13 | 10 | 0,13 | 10 | 0,14 |
| 10 | 6,23 | 16,878 | 16,267 | 11 | 0,10 | 10 | 0,11 | 10 | 0,11 | 10 | 0,10 | 10 | 0,11 |
| 11 | 5,89 | 14,668 | 15,254 | 11 | 0,15 | 10 | 0,16 | 10 | 0,16 | 10 | 0,16 | 10 | 0,16 |
| 12 | 7,51 | 21,254 | 20,150 | 10 | 0,16 | 10 | 0,17 | 10 | 0,16 | 10 | 0,16 | 10 | 0,17 |
| 13 | 6,92 | 19,233 | 18,347 | 11 | 0,15 | 10 | 0,16 | 10 | 0,16 | 10 | 0,16 | 10 | 0,16 |

[^1]| 14 | 7,07 | 20,456 | 18,804 | 10 | 0,09 | 10 | 0,10 | 10 | 0,10 | 10 | 0,10 | 10 | 0,10 |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 15 | 7,49 | 19,265 | 20,089 | 10 | 0,13 | 10 | 0,13 | 10 | 0,13 | 10 | 0,13 | 10 | 0,13 |
| 16 | 9,29 | 33,784 | 25,710 | 10 | 0,16 | 10 | 0,16 | 10 | 0,16 | 10 | 0,16 | 10 | 0,16 |
| 17 | 7,72 | 25,658 | 20,797 | 10 | 0,14 | 10 | 0,15 | 10 | 0,15 | 10 | 0,14 | 10 | 0,15 |
| 18 | 6,55 | 14,145 | 17,228 | 11 | 0,15 | 10 | 0,15 | 10 | 0,15 | 10 | 0,15 | 10 | 0,14 |
| 19 | 6,17 | 16,564 | 16,088 | 11 | 0,16 | 10 | 0,16 | 10 | 0,16 | 10 | 0,17 | 10 | 0,16 |
| 20 | 6,73 | 18,541 | 17,771 | 11 | 0,18 | 10 | 0,19 | 10 | 0,19 | 10 | 0,19 | 10 | 0,19 |
| 21 | 6,62 | 16,666 | 17,439 | 11 | 0,17 | 10 | 0,16 | 10 | 0,17 | 10 | 0,17 | 10 | 0,17 |
| 22 | 4,79 | 14,887 | 12,038 | 14 | 0,19 | 10 | 0,22 | 10 | 0,23 | 10 | 0,23 | 10 | 0,23 |
| 23 | 3,93 | 6,422 | 9,596 | 16 | 0,15 | 11 | 0,18 | 10 | 0,19 | 10 | 0,20 | 10 | 0,20 |
| 24 | 3,37 | 5,856 | 8,046 | 18 | 0,14 | 12 | 0,17 | 10 | 0,19 | 10 | 0,19 | 10 | 0,19 |

A: Sample means, expressed as the mean number of mobile forms of Panonychus ulmi. Each mean number based on 100 sample units.
B: Sample variance of field data.
C: The predicted sample variance is calculated as $\mathrm{am}^{\mathrm{b}}$, where $a=1.14, b=0.30$, and $m$ is the sample mean.
D: The mean number of sample units required to estimate $m$ as determined by the RVSP software.
E: Observed mean precision level for estimating $m$ as determined by the RVSP software.

The sequential sampling design of Green (1970) for estimating populations at fixed-precision $D=$ $0.20,0.25,0.30,0.35$, and 0.50 was developed and validated by using 500 Monte Carlo simulations (Naranjo and Hutchison, 1997). Virtually, A reasonable $D$ is usually between 0.20 and 0.35 (Southwood, 1978; Hutchison et al., 1988). For all densities observed from April to September, precision levels of $0.30,0.35$, and 0.50 offer the possibility of taking sample sizes below 100 sample units for classifying the densities of $P$. ulmi; this number increases when the precision is 0.20 (Table 2). A similar $A S N$ result was obtained by Mollet et al. (1984) at $D=$ $0.20,0.25$, and 0.30 with significant correlations for both mean-variance relationships ( $r^{2}=0.95$ for Taylor and 0.80 for Iwao's regression).

To further monitor and manage the population of P. ulmi, similar plans have been established for different life stages, including eggs, larvae, nymphs, and adults. This approach shows that,
when data sets were used for validation, the resulting sequential sampling design can be used reliably to ensure that the desired fixed precision levels are achieved (O'Rourke and Hutchison, 2003; Serra et al., 2013).

### 3.4 Wald's sequential probability ratio test (SPRT)

The linear regression equation (plotting $y^{\prime}$ on $x^{\prime}$ ) is: $y^{\prime}=0.63 x^{\prime}+0.10\left(r^{2}=0.98: d l=22, P<0.05\right)$. The intercept value (0.10) is not significantly different from zero ( $P>0.05$ ) (Figure 4A). Therefore, the condition of the negative binomial law is verified. The presence of a common $k\left(k_{c}\right)$ is obvious (Bliss and Owen, 1958).

To calculate the common $k\left(k_{c}\right)$, we plotted on the mean densities of Panonychus ulmi. The linear regression equation is $y=0.09 x+2.64\left(r^{2}=0.88\right.$, $d l=22, P<0.05)$. The regression slope is not significant at the error level of $5 \%$ and shows no trend existing (Figure 4B). The basic postulate of the Wald's procedure is respected, and the
presence of a common $k$ is justified. The calculation of $k_{c}$ value corresponds to an approximate estimate using the formula:
$\frac{1}{k_{c}}=\sum y^{\prime} / \sum x^{\prime}$ which gives a $k_{c}$ value equal to 1.58.

(A)

(B)

Figure 4: Regression estimate of a common $k\left(k_{c}\right)$ for $P$. ulmi (A). Relation of the $1 / k c$ to the mean based on 24 sample data (B).

The lower and upper stop lines for classifying the infestation level with respect to the $E T$ of $2.31 P$. ulmi/ sample unit are shown in Figure 5. The lower stop line intersects the $x$-axis at 19 sample units. Therefore, a minimum of 19 sample units must be examined for classifying the infestation level. The accuracy of classifying the P. ulmi infestation levels relative to an $A T$ was tested by RVSP at tally threshold $T=0$ using the 24
independent data sets. The $O C$ function was near unity when $m<2.14$. At a mean density value of $2.26\left(x_{1}\right)$, the $O C$ function was 0.90. At an $m$ value of $2.36\left(x_{2}\right)$, the $O C$ function was 0.20 . At $m>$ 2.36, the $O C$ function decreased from 0.20 to zero. At an $m$ value of 2.31 (ET) (Figure 6). The $O C$ curve showed that the actual error rate in classifying infestation as being $x_{1}$ when, in fact, it is above $x_{2}(\beta$ error) was 0.50.


Figure 5: The stop lines of SPRT for classifying the mean densities of $P$. ulmi relative to an economic threshold ( $E T$ ).


Figure 6: Operating characteristic (OC) curve for the sequential probability ratio test sampling plan ( $\alpha=$

$$
\left.\beta=0.2, x_{1}=2.26, x_{2}=2.36, k_{\mathrm{c}}=1.58\right)
$$



Figure 7: Average sample number ( $A S N$ ) curve for the sequential probability ratio test sampling plan ( $\alpha$

$$
\left.=\beta=0.2 ; x_{1}=2.26 ; x_{2}=2.36 ; k_{c}=1.58\right) .
$$

The $A S N$ curve showed that for classifying the range densities of 0.22-9.29, about 8-149 sample units are needed with more samples for classifying the mean density at the ET of 2.31mites per leaf. Each of the 24 independent data sets used to evaluate the performance of the sequential sampling plan was based on 100 sample units. Therefore, with 100 sample units, mean densities of $\leq 2.14$ or $\geq 4.12$ can be correctly
classified on average concerning the fixed ET. However, for mean densities of Panonychus ulmi between 2.14 and 4.12, we can expect that more than 100 sample units will be required (Figure 7). The uncertainty in classifying the mean densities of P. ulmi increases near the threshold. Therefore, the $A S N$ is near the $E T$. More sample units needed to be examined to cross either the upper or lower stop line when $P$. ulmi density was between $x_{1}$ and

[^2]$x_{2}$ than when it was less than $x_{1}$ or than $x_{2}$. Uncertainty of classifying mite density at the $E T$ results in a large number of samples being required to reach a decision (Binns, Nyrop, Werf, \& Werf, 2000). A pest mite control measure should be applied when the mean density or infestation level is at or above the economic threshold. The use of these sequential sampling plans in integrated pest management programs for pests will result in the application of pest control measures only when needed (Morris, 1960; Binns and Nyrop, 1992; Binns, 1994).

Several methods of calculating $k_{c}$ have been discussed (Pedigo and Buntin, 1994) and several estimates of a common $k$ have been described (Beall, 1942; Kleczkowski, 1949; Anscombe, 1949a; 1949b; 1950; Bliss \& Fisher, 1953), but a simple method based on the use of linear regression to calculate $k_{c}$ is often used (Bliss and Owen, 1958)

The stability of the parameter $k$ is not always present. The parameter $k$ tends to increase with the increase of the mean, and its biological interpretation is not obvious (Pedigo and Buntin, 1994; Robinson and Smyth, 2008). Ruesink suggests the use of Taylor's power law when the average density of sampled populations varies by more than one order of magnitude (Ruesink, 1980). If there is a $k$, the Wald procedure can be used to develop a sequential sampling plan.

Using Bliss \& Owen's (1958) model, the $k_{c}$ value was 1.58 . Generally, $k_{c}$ of the negative binomial distribution closes to 2 . The high $k_{c}$ corresponds to the Poisson distribution. Wald's sequential sampling is evaluated by examining the operating characteristic (OC) and average sample number (ASN) functions (Nyrop et al., 1999). The OC curve describes the probability of making a no intervention decision (crossing the lower decision boundary) as a function of the mean pest density. A $k$ value of 1.58 , The $O C$ curve is flatter and increases the classification accuracy of the plan (the smaller type I and type II error rates) (Nyrop et al., 1994). The $A S N$ curve indicates the expected number of samples required to decide on as a
function of the mean pest density. Generally, for Wald's sequential probability ratio test, a flatter $O C$ function means less robustness and precision of the sampling plans. A flatter $A S N$ function means that fewer samples are required to classify pest population density.

### 3.5 Iwao sequential sampling plan

The linear regression equation between mean crowding and mean density of $P$. ulmi is $x^{*}=$ $1.10 m+0.9\left(r^{2}=0.99, d l=22, P<0.05\right)$ (Figure 8). The value of the contagion index corresponds to the intercept ( 0.90 ), and the value of the distribution coefficient corresponds to the slope of the regression (1.10).

At an error level of $5 \%$, and $D=0.50$, the equations of the non-parallel stop lines of the sequential sampling plan developed according to Iwao procedure are: $y=2.31 x+1.71(4.89 x)^{1 / 2}$ and $y=2.31 x-1.71(4.89 x)^{1 / 2}$ for the upper and lower limits, respectively (Figure 9). The same rules apply concerning the decision to process (value above the upper limit) or not (value below the lower boundary) or to continue sampling (the middle zone between the upper and lower limits).

A minimum number of 4 samples is necessary to cross the negative zone of the lower limit, whereas a maximum number of 57 sample units are needed to determine whether the population level is equal to the economic threshold at a confidence interval predetermined.


Figure 8: Mean-variance relationship using Iwao's model


Figure 9: Acceptance curves of a sequential sampling plan according to the Iwao procedure for classifying the mean densities of $P$. ulmi relative to an economic threshold (ET).

[^3]Table 3: Sequential decision table, giving the cumulative number of $P$. ulmi required for a decision from 4 to 20 sample units with an error level of 0.05 .

| Sample number | lower threshold | upper threshold |
| :---: | :---: | :---: |
| 4 | 0 | 16 |
| 5 | 3 | 20 |
| 6 | 5 | 23 |
| 7 | 7 | 26 |
| 8 | 8 | 29 |
| 9 | 10 | 31 |
| 10 | 12 | 34 |
| 11 | 13 | 37 |
| 12 | 15 | 40 |
| 13 | 17 | 43 |
| 14 | 19 | 46 |
| 15 | 21 | 48 |
| 16 | 22 | 51 |
| 17 | 24 | 54 |
| 18 | 26 | 57 |
| 19 | 28 | 59 |
| 20 | 30 | 62 |
|  |  |  |

The failure to establish a $k$ value from the sampling data precluded the use of traditional sequential sampling methods. In Figure 8, the regression of mean crowding ( $x^{*}$ ) on mean density ( $m$ ) accounts for $99 \%$ of the variation in the distribution of $P$. ulmi. The mathematical relationship between the mean density $m$ and the mean crowding ( $x^{*}$ ) describes some characteristics of the spatial distribution inherent in a species in a given habitat. Iwao (1968) has shown that this relationship is often linear and described by a simple regression line. Two parameters describe the type of spatial distribution of the mite. These are the contagion index $\alpha$ and the slope $\beta$, appointed distribution coefficient. The first of these parameters characterize the basic unit of the population, while the second describes the distribution of these basic units in space. The validity of the regression was checked using the correlation coefficient ( $r^{2}$ ) (Montgomery et al., 2012).

After calculating the upper and lower decision limits by using the formulas supplied by (Iwao,
1975), equations were fit to the data by the least-squares method. The resulting graphs (Figure 9) remind one of the typical sequential sampling graphs, except that the lines are not parallel. For pest management use, scouts prefer a table such as Table 3.

The Wald's and Iwao's sequential sampling plans are different in terms of the minimum number of samples required to leave the negative zone and the economic threshold used for decision making. The developed Wald's (1945) sequential plan is more restrictive since a minimum of 19 samples (Figure 5) is necessary to detect the presence of harmless populations of the mite compared to 4 samples for the sequential plan developed with the Iwao's procedure. The thresholds of the Iwao Plan seem more permissive as to the cumulative number of $P$. ulmi per sample unit required before deciding to intervene or not.

## IV. CONCLUSIONS

The sequential sampling plans presented here are powerful tools for monitoring and managing the
mite pest populations. Sampling needs an efficient procedure to estimate the damage related to pest mites and phytophagous insects and succeed integrated management programs in orchards. Sequential sampling protocols based on three hypothesis and calculated probabilities could be a quick method for making control decisions in biological control against mite populations by phytoseiidae and requires additional research on various aspects of predator-prey relationships. Population dynamics studies are also needed to transfer the action threshold from 2.31 mobile forms per leaf to the number of eggs, ie, how many eggs per sample unit must be observed to produce later an infestation of 2.31 mite pest per leaf?

It is possible to envisage the integration of a simultaneous sampling of different insect pests to make the sampling profitable, thus allowing the application of sequential plans for plant protection. The sequential sampling plans may be easier to apply over large areas, where the time and cost may be reduced by adequate sampling. In this context, current practices in arboriculture will need to be revised and focused more on sampling. The aggregative behavior of pest mites in small areas to be sampled seems to be the cause of the difficulty of applying a sequential design that minimizes the sampling effect.

Although sampling to classify the dynamic and distribution of the pest mites population, is generally expensive and involves the time and money, sampling remains essential for monitoring population dynamics and decision-making. However, sampling costs could be offset by an informed decision based on established concepts and developed sampling plans. The choice of an effective method is, therefore, crucial when the objective is to reduce sampling effort and associated costs.

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[^0]:    Development and Validation of Sampling Plans for the European Red Mite Panonychus Ulmi (Acari: Tetranychidae)

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