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SPATIAL DISTRIBUTION PATTERN AND SEQUENTIAL SAMPLING PLAN FOR *PARATYLENCHUS DIANTHUS*

by

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Paratylenchus dianthus (Jenkins *et* Taylor) is an important pest of carnation in the province of Naples. Fumigation prior to planting prevents damage until the second growing year when nematode populations may build up to damaging levels. Because treatment during cultivation is expensive in relation to the value of the crop, it would be useful to have a method of estimating the distribution and densities of nematode populations as a basis for decision on the economic advantages of treatment.

The spatial distribution of *P. dianthus* was determined in the investigation described here and a sequential sampling procedure established from the data.

MATERIALS AND METHODS

1. *Spatial distribution*

A study was undertaken in the province of Naples, from April 1982 to May 1984, in a plastic unheated greenhouse of 600 m² area. Old carnation plants were rooted out on 28 April 1982. The soil was fumigated on 2 May with DD + Vapam at the rate of 800 and 1,000 l/ha respectively. One month later carnation cv. Astor rooted cuttings were planted. Soil samples were taken once every two weeks from

9 May 1982 to the end of the investigation in 1984. On each occasion 100 samples were collected to a depth of 20 cm according to a stratified-random design (Southwood, 1978). This was done by dividing the total area into 6 plots of 100 m² each and each plot into subplots of 1 m²; a sample was taken from each 1 m² area within a randomly selected plot.

The soil samples were kept in a refrigerator at 5°C until the nematodes were extracted from a 10 ml aliquot by modified Baermann's method.

To analyze the spatial distribution of the nematodes the regression method of mean crowding on the mean proposed by Iwao (1968) was used. The mean crowding (Lloyd, 1967) was estimated for each sample by means of the equation: $m^* = m + (\sigma^2/m - 1)$, where m^* = mean crowding; σ^2 = variance; m = mean density. This method assumes that mean crowding is linearly related to mean density, i.e., $m^* = \alpha + \beta m$, where α and β are constants for a given species. The intercept α , referred to by Iwao as « index of basic contagion » indicates whether the basic units of a population are individuals or groups; when $\alpha = 0$ the basic units are individuals, but when $\alpha > 0$ the basic units are groups. The slope β , referred to as « density contagiousness coefficient » describes the distribution of the basic units of the population within their habitat; when $\beta = 1$ the distribution is random and as β increases the aggregation increases.

Changes of spatial distribution were followed utilizing Lloyd's « patchiness index » by calculating m^*/m for each sample. When the « patchiness index » is greater than 1, less than 1 or equal to 1, the population is respectively aggregated, uniform or randomly distributed.

2. *Sequential sampling procedures*

A sampling plan is defined as sequential when the number of samples is not determined and single units are examined one after the other until a statistically valid result is reached.

Traditional sequential sampling procedures are based upon pre-determined fit of sample data to the theoretical distribution models e.g., binomial, Poisson, negative binomial (Onsager, 1976; Southwood, 1978). However, as the spatial distribution often changes with time, and since in the negative binomial the « k » value can vary systematically according to the mean density of the population (Bliss and Owen,

1958), it follows that the applicability of traditional procedures is rather restricted. The realization of a sequential sampling plan without preliminary knowledge of theoretical distribution models is made possible using Iwao's method (Iwao, 1975) and fixing an appropriate economic threshold. As far as we know, there are no specific studies on this but a number of personal observations seem to indicate that a mean density of 250 nematodes per 10 ml soil must not be exceeded.

By this method the intercept and the slope of the regression line obtained, together with the economic threshold were used to calculate the upper and lower limits of the sequential sampling plan. The upper (T') and lower (T'') limits were calculated by the equations:

$$T' = q m_0 + t \sqrt{q [(\alpha + 1) m_0 + (\beta - 1) m_0^2]} \quad (1)$$

$$T'' = q m_0 - t \sqrt{q [(\alpha + 1) m_0 + (\beta - 1) m_0^2]} \quad (2)$$

T' and T'' are respectively the upper and lower limits for a given value of q , m_0 is the economic threshold, α and β are respectively intercept and slope of the regression line of m^* on m , t the tabulated value of Student's « t » for ∞ degrees of freedom and for a preassigned probability level; q is the independent variable and represents the number of analyzed samples; the equations (1) and (2) were calculated for successive values of q (1).

It was also possible to establish the maximum number of samples to analyze when $m = m_0$. In this situation the sampling could continue for a long time because the cumulative number of nematodes counted would be included between T' and T'' for a long sequence of analysis. Assigning in advance the confidence interval d_0 , the estimation of the maximum number of samples to be taken before stopping the sampling was calculated with the equation:

$$q_{\max} = \frac{t^2}{d_0^2} [(\alpha + 1) m_0 + (\beta - 1) m_0^2] \quad (3)$$

(1) Please note that m , m^* , σ^2 , α and β are parameters related to the population of a given species. But, because they are calculated with sampling data the obtained values are only estimates and being such they should be indicated in this way: \bar{x} , \bar{x}^* , s^2 a and b . For simplicity and clarity reasons we preferred to use for them the first symbols.

RESULTS

1. Spatial distribution

The calculated regression equation of mean crowding on the mean for *P. dianthus* (Fig. 1) was:

$$m^* = 35.85 + 1.38 m \quad (r = 0.99)$$

On the basis of the estimated values of α and β we concluded that the basic units of nematode population were groups of individuals because α was significantly greater than 0, and that their distribution was aggregated because β was significantly greater than 1 ($P = 0.05$).

Lloyd's « patchiness index », calculated for every sampling occasion and plotted against the mean density of population (Fig. 2) indicated that at low population levels the spatial distribution was aggregated and when the population increased the aggregation decreased, even though it never resulted in a random spatial distribution.

2. Sequential sampling procedures

The graph of the sequential sampling plan for *P. dianthus* is shown in Fig. 3. A decision with a 95% confidence level can be derived as follows:

- a) from a number of samples picked at random between 1 and 79⁽²⁾, if $T_o > T'$ ⁽³⁾ ($m > m_o$)⁽⁴⁾: *Treatment necessary*;
- b) from a number of samples picked at random between 1 and 79, if $T_o < T''$ ($m < m_o$): *Treatment not necessary*;
- c) from 79 samples picked at random, if $T' > T_o > T''$ (m is close to m_o): *Sampling to be repeated at a later date or decision to be taken on the basis of developing potentialities*.

As an alternative to plotting the sequential sampling plan the tabulated data of T' and T'' for every value of q (Tab. I) can be utilized. The comparison of T_o with T' and T'' can be more immediate and precise.

(2) For $m_o = 250 \pm 40$, q_{max} resulted to be equal to 79.

(3) T_o = cumulative number of counted nematodes after q samples.

(4) m = mean density of population present in the field .

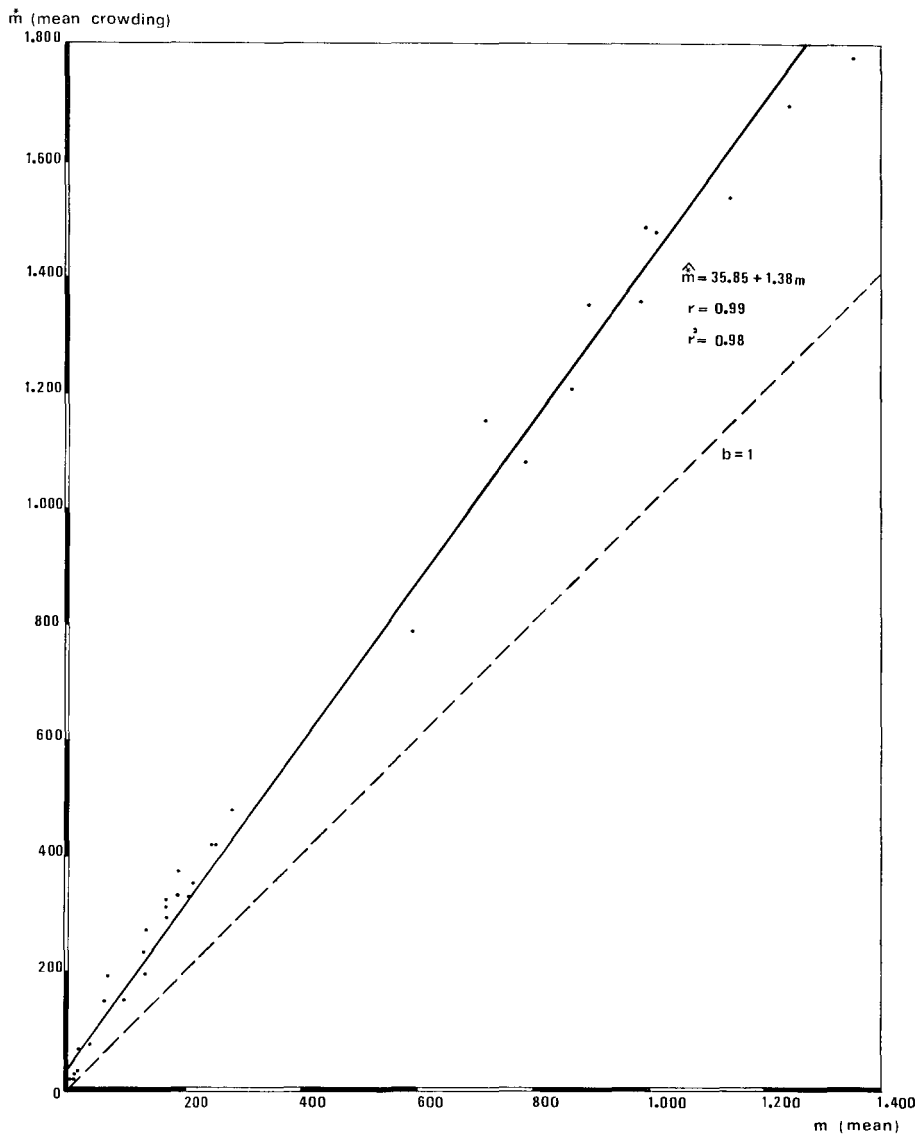


Fig. 1 - Relationship of mean crowding (m^*) to mean (m) number of nematodes/10 ml of soil. The dashed line is relative to a theoretical population where the basic units are individuals randomly distributed.

LLOYD'S "patchiness index" (\bar{m} / m)

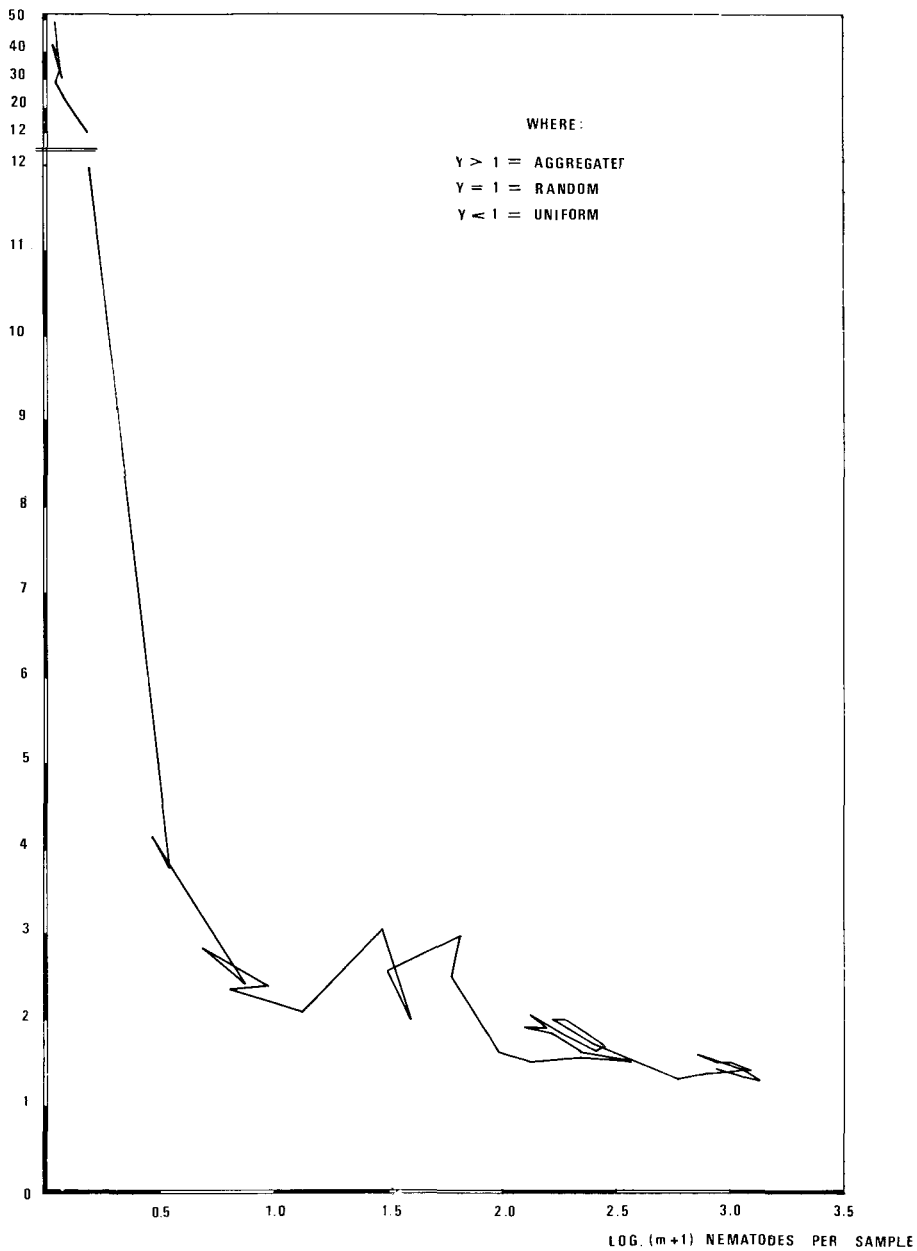


Fig. 2 - Analysis of temporal changes of *Paratylenchus dianthus* distribution by Lloyd's « patchiness index ». Each data point represents successive sampling dates.

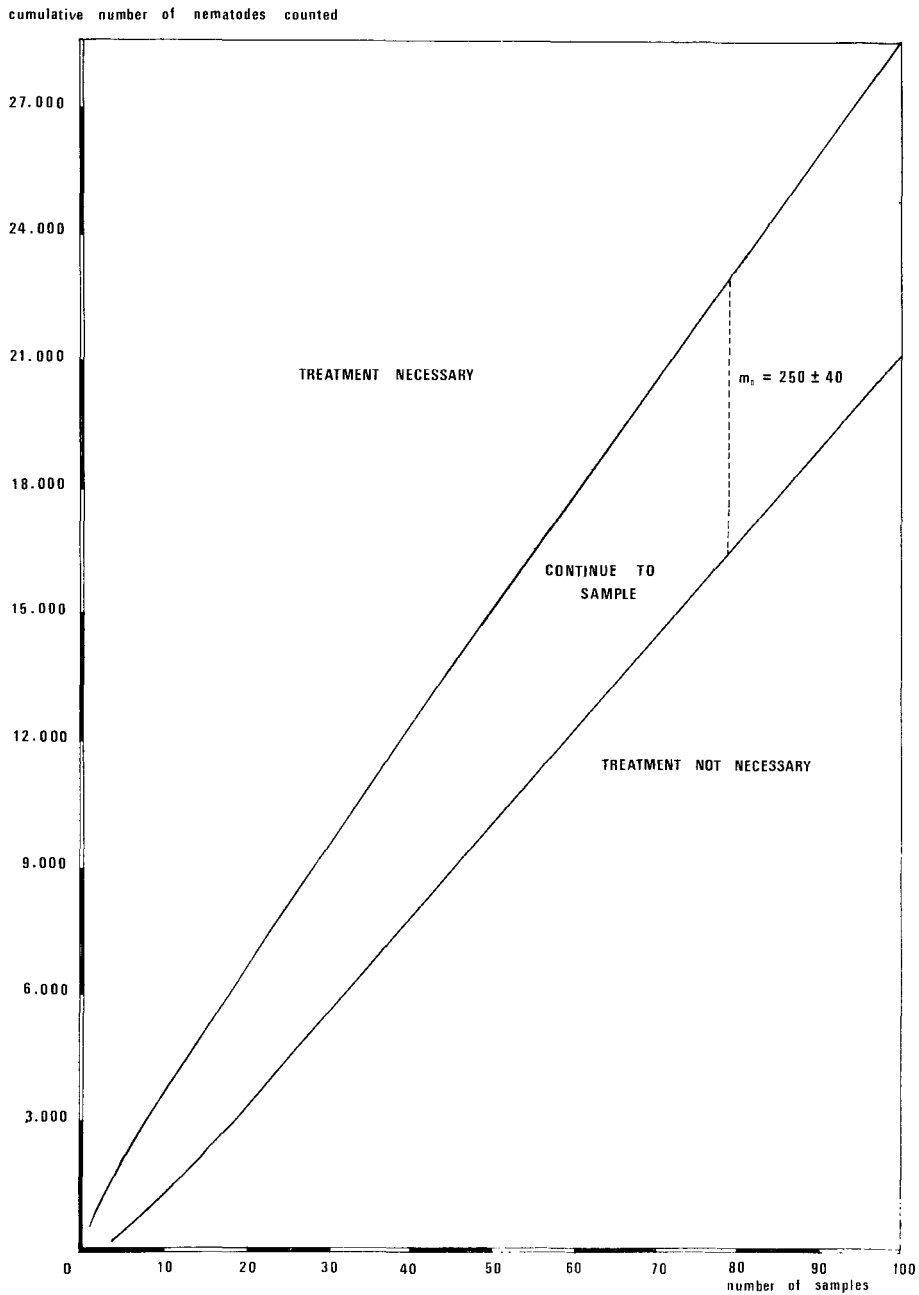


Fig. 3 - Graph of the sequential sampling plan for *P. dianthus*. The economic threshold of 250 nematodes/10 ml of soil and 95% confidence probability are used.

Tab. I - *Tabulated values of T' and T'', respectively upper and lower limits, calculated for successive values of q (number of samples collected). In T_o column the cumulative number of nematodes counted is to be reported. In this way the comparison of T_o with T' and T'' is more immediate and precise.*

| q | T' | T _o | T'' | q | T' | T _o | T'' |
|----|--------|----------------|-------|----|--------|----------------|--------|
| 1 | 607 | — | — | 41 | 12,531 | | 7,969 |
| 2 | 1,004 | | — | 42 | 12,808 | | 8,192 |
| 3 | 1,367 | | 133 | 43 | 13,086 | | 8,414 |
| 4 | 1,713 | | 287 | 44 | 13,363 | | 8,637 |
| 5 | 2,047 | | 453 | 45 | 13,639 | | 8,861 |
| 6 | 2,373 | | 627 | 46 | 13,916 | | 9,084 |
| 7 | 2,693 | | 807 | 47 | 14,191 | | 9,308 |
| 8 | 3,008 | | 992 | 48 | 14,467 | | 9,532 |
| 9 | 3,319 | | 1,181 | 49 | 14,743 | | 9,757 |
| 10 | 3,627 | | 1,373 | 50 | 15,019 | | 9,981 |
| 11 | 3,932 | | 1,568 | 51 | 15,294 | | 10,206 |
| 12 | 4,234 | | 1,766 | 52 | 15,569 | | 10,431 |
| 13 | 4,535 | | 1,965 | 53 | 15,843 | | 10,657 |
| 14 | 4,833 | | 2,167 | 54 | 16,117 | | 10,883 |
| 15 | 5,130 | | 2,370 | 55 | 16,392 | | 11,108 |
| 16 | 5,425 | | 2,575 | 56 | 16,666 | | 11,334 |
| 17 | 5,719 | | 2,781 | 57 | 16,939 | | 11,561 |
| 18 | 6,011 | | 2,989 | 58 | 17,213 | | 11,787 |
| 19 | 6,303 | | 3,197 | 59 | 17,486 | | 12,014 |
| 20 | 6,593 | | 3,407 | 60 | 17,759 | | 12,241 |
| 21 | 6,882 | | 3,618 | 61 | 18,032 | | 12,468 |
| 22 | 7,171 | | 3,829 | 62 | 18,305 | | 12,695 |
| 23 | 7,458 | | 4,042 | 63 | 18,577 | | 12,923 |
| 24 | 7,745 | | 4,255 | 64 | 18,850 | | 13,150 |
| 25 | 8,031 | | 4,469 | 65 | 19,122 | | 13,378 |
| 26 | 8,316 | | 4,684 | 66 | 19,394 | | 13,606 |
| 27 | 8,601 | | 4,899 | 67 | 19,666 | | 13,834 |
| 28 | 8,885 | | 5,115 | 68 | 19,937 | | 14,063 |
| 29 | 9,168 | | 5,332 | 69 | 20,209 | | 14,291 |
| 30 | 9,451 | | 5,549 | 70 | 20,480 | | 14,520 |
| 31 | 9,733 | | 5,767 | 71 | 20,751 | | 14,750 |
| 32 | 10,015 | | 5,985 | 72 | 21,022 | | 14,979 |
| 33 | 10,296 | | 6,204 | 73 | 21,293 | | 15,207 |
| 34 | 10,578 | | 6,423 | 74 | 21,564 | | 15,436 |
| 35 | 10,857 | | 6,643 | 75 | 21,835 | | 15,665 |
| 36 | 11,137 | | 6,863 | 76 | 22,105 | | 15,895 |
| 37 | 11,417 | | 7,083 | 77 | 22,375 | | 16,125 |
| 38 | 11,696 | | 7,304 | 78 | 22,646 | | 16,354 |
| 39 | 11,975 | | 7,525 | 79 | 22,916 | | 16,584 |
| 40 | 12,253 | | 7,747 | | | | |

CONSIDERATIONS AND CONCLUSIONS

Sequential sampling gives a harmonious synthesis between precision and practicality. In fact, with the parameters at our disposal (m , m^*), a comparison can be made between it and sampling at pre-determined size with the same level of accuracy. This comparison can be made by utilizing Ono's method (1967) which allows the pre-determination of the number of samples required to achieve a desired level of accuracy at different population densities. Especially at low population densities, the approximate number of samples required by the sequential sampling plan is shown to be less and this advantage is also associated with operational simplicity. These considerations are even more important in view of the widespread use that is made nowadays of economic thresholds in integrated control. Iwao's method proposed here has been utilized recently in applied entomology. Specific applications to aphids on strawberries (Trumble *et al.*, 1983), and to the pear Psylla (*Psylla pyricola* Foers.) (Burts and Brunner, 1981) are of interest.

In nematology this method could prove useful because it simplifies soil sampling. On the whole, we think that a wider use of this operative method could help rationalize the chemical control of nematodes of economic importance to crop production.

S U M M A R Y

The analysis of spatial distribution pattern and a sequential sampling plan for *Paratylenchus dianthus* (Jenkins *et Taylor*) are reported. The study was carried out from April 1982 to 1984 in a commercial crop of carnations grown in a greenhouse in the province of Naples. The analysis of spatial distribution was made by taking 100 soil samples, within a stratified-random design, every two weeks and using Iwao's regression of mean crowding (m) on the mean (m). Changes in spatial distribution were determined with Lloyd's index (patchiness index). It emerged that the basic components of population were groups of individuals with an aggregated distribution. The level of aggregation decreased as the mean density of population increased. Subsequently m^* on m regression was used, according to Iwao's method, to carry out a sequential sampling plan. In the conditions of this study, the highest number of samples to be taken to reach a decision with a 95% confidence level is 79 when the mean density of population present in the field (m) is close to the economic threshold of 250 nematodes/10 ml of soil. When the mean density is much greater or much lower than the economic threshold (treatment necessary or unnecessary) the decision is reached after a few samples.

L I T E R A T U R E C I T E D

- BLISS C. I. and DWEN A. R. G., 1958 - Negative binomial distribution with a common K. *Biometrika*, 45: 37-58.
- BURTS E. C. and BRUNNER J. F., 1981 - Dispersion statistics and sequential sampling plan for adult pear psylla. *J. Econ. Entomol.*, 74: 291-294.
- IWAO S., 1968 - A new regression method for analyzing the aggregation pattern of animal populations. *Res. Popul. Ecol.*, 10: 1-20.
- IWAO S., 1975 - A new method of sequential sampling to classify populations relative to a critical density. *Res. Popul. Ecol.*, 16: 281-288.
- LLOYD M., 1967 - Mean crowding. *J. Anim. Ecol.*, 36: 1-30.
- ONO Y., 1967 - Estimation of animal numbers. In: Christensen J. B., Gutierrez A. P., Cothran W. R. and Summers C. G., 1977. The within field spatial pattern of the larval Egyptian alfalfa weevil, *Hypera brunneipennis* (Coleoptera: Curculionidae): an application of parameter estimates in simulation. *Can. Ent.*, 109: 1599-1604.
- ONSAGER J. A., 1976. - The rationale of sequential sampling, with emphasis on its use in pest management. *USDA Tech. Bull.*, 1526, pp. 19.
- SOUTHWOOD T. R. E., 1978 - Ecological Methods with Particular Reference to the Study of Insect Populations. John Wiley & Sons, New York, pp. 524.
- TRUMBLE J. T., OATMAN E. R. and VOTH V., 1983 Thresholds and sampling for aphids in strawberries. *Calif. Agric.*, 37 (11-12): 20-21.

Accepted for publication on 2 April 1985.