

Spatial pattern of the Río Cuarto corn disease vector, *Delphacodes kuscheli* Fennah (Hom., Delphacidae), in oat fields in Argentina and design of sampling plans

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Abstract: The spatial pattern of the Río Cuarto Corn Disease vector, *Delphacodes kuscheli* (Hom., Delphacidae), was analysed in oat fields within the endemic area of the disease, during the growing seasons 1993 and 1994. The spatial pattern was analysed by fitting the probabilistic models Poisson and negative binomial and estimation of single-date and overall aggregation indices. The population of the different stage classes, sex, and wing forms showed a significant trend to aggregation as the negative binomial model fitted the observed frequency distributions in more than 78% of the cases (sampling dates) while the Poisson model fitted well in only 28% of cases or less. Single-date aggregation index, C_A , ranged from 0.3 to 0.84. Overall (whole season) aggregation index, C_A^* , estimated through the Bliss and Owen's regression method, ranged from 0.18 (female adults) to 1.08 (nymphs I–II), indicating a moderate degree of aggregation compared with other planthopper species. There were no significant relationships between aggregation and population density. The minimum number of sampling units and critical lines for sequential sampling plans were calculated based on the estimation of C_A^* for the precision levels (D) 0.1, 0.2 and 0.3. Even low degrees of aggregation, like that of adults, demand much more sampling effort than randomly distributed populations, particularly at high densities. General implications and limitations of the proposed sampling plans for monitoring the vector population abundance are discussed.

1 Introduction

The Mal de Río Cuarto del Maíz (MRC), (Río Cuarto Corn Disease), has become the most important disease of corn fields over a large area in Central Argentina and particularly in the south-west of Córdoba Province. Economic losses caused by MRC were estimated around 100 million U.S. dollars during the 1983–93 period and a devastating epidemic occurred during the 1996/97 growing season (MARCH et al., 1997). MRC is caused by the Mal de Río Cuarto Virus (MRCV, Fiji-virus) (CONCI and MARZACHI, 1993) which is transmitted by the planthopper *Delphacodes kuscheli* Fennah (Hom., Delphacidae) (REMES LENICOV et al., 1985). The epidemiology of MRC closely resembles Maize Rough Dwarf Disease in countries around the Mediterranean region (CONTI, 1984). *Delphacodes kuscheli* does not breed on corn (VIRLA and REMES LENICOV, 1991), but on several wild gramineaceous plants and winter cereals, mainly oats (*Avena sativa* L.) (TESÓN et al., 1986; REMES LENICOV et al., 1991; VIRLA and REMES LENICOV, 1991; BOITO et al., 1993; ORNAGHI et al., 1993). In the endemic area, in south-west Córdoba, a high proportion of fields are cultivated with oats, which makes this crop the most important overwintering host and the main source from which the vector migrates to corn fields in spring (TRUM-

PER, 1996). In addition, oat has been identified as one of the natural hosts of the MRCV (GIMENEZ PECCI et al., 1993).

One of the tactics that growers use for the management of the MRC is early sowing, to avoid overlapping of the susceptible phenological stage of the maize plants (3 weeks after plant emergence) with the vector population peak. An empirical predictive model, based on climatic variables and abundance of *D. kuscheli* in oats fields, has been developed to help growers make a decision about cultivar selection and sowing date for a particular area within the edemic region (MARCH et al., 1997). With a similar objective but a different approach GRILLI and GORLA (1997) presented evidence that a vegetation index derived from meteorological satellite imagery could be used to monitor the abundance within endemic and nonendemic areas. Despite the growing relevance of MRC as a significant constraint to corn production in Argentina, no previous attempt has been made to develop plans for monitoring the vector populations through field sampling. The development of monitoring programmes to estimate the vector abundance can contribute to increase the efficacy of such management tactics. A first step in the elaboration of sampling plans for *D. kuscheli* in oat fields is the analysis of its spatial pattern.

Delphacodes kuscheli has a seasonal life cycle with most of the generations occurring during late spring and early summer. In November and December, one generation is completed in approximately 1 month (VIRLA and REMES LENICOV, 1991). The immature stage is represented by five instars. Both male and female adults show wing polymorphism, i.e. macropterous (long-winged) and brachypterous (short-winged) forms. Wing form determination in many planthopper species occurs during the nymphal stage, and would be influenced by the stress produced by high levels of aggregation in overcrowded populations (DENNO and RODERICK, 1990; KUSMAYADI et al., 1990).

The study of *D. kuscheli* spatial pattern can provide relevant information to improve the understanding of both the vector ecology and the epidemiology of MRC (TRUMPER, 1996). Spatial pattern may change along with population growth. The aggregation index of *Nilaparvata lugens*, another plant virus vector delphacid, showed some variation in short periods of time as the population increased (KUSMAYADI et al., 1990). The spatial pattern of a vector population in a field could determine the probability of virus transmissions. The greater the probability of finding more than one insect in the same habitat patch (an oat plant) the more likely are healthy insects to acquire the virus. Mathematical models describing the epidemiology of MRC in oat fields could take into account the spatial dynamics by explicitly considering the spatial pattern of the vector in the model equations (TRUMPER, 1996).

The objective of this work was to study the spatial pattern of *Delphacodes kuscheli* in oat fields and its relation to population density. Fixed and variable intensity sampling plans for estimating population abundance are proposed, based on aggregation parameters.

2 Materials and methods

2.1 Experimental fields and sampling methods

This study was developed during two growing seasons (1993 and 1994) on 5-ha oat fields used for cattle grazing, located at El Espinillo, Córdoba Province (Argentina), within the endemic area of MRC. Samplings were carried out from approximately the third week after plant emergence (October), and interrupted when significant crop decay was observed (December).

Eight and six samplings were taken during the first and second season, respectively. Forty-two samples were taken on each sampling date. Each sample unit consisted of the material collected along 3 m of an oat row with a gas-powered suction machine.

The insects collected were sorted into species, sex, wing form and development stage using a stereomicroscope and the abundance of each category was recorded. Nymphs were sorted to classes I–II, III, IV and V. No insecticide was used throughout the study and no practice other than grazing cattle, was carried out.

2.2 Data analysis

The information recorded from each sampling date of both seasons and corresponding to each category identified, was analysed fitting two mathematical models to the observed frequency distribution. The Poisson model assumes purely random or independent disposition of individuals (KUNO,

1991). The Poisson distribution is characterized by the equality of mean to variance. To determine whether the distribution of each category departs from randomness, the count of *D. kuscheli* per sampling unit was examined in terms of the variance–mean ratio. In order to test the fitness of the observed distribution to the random (Poisson) distribution, the statistic $T = (n - 1)s^2/m$ was used, which is known to follow approximately the chi-square distribution with $n - 1$ degrees of freedom.

The negative binomial model is known to be most widely applicable for describing the patchy spatial distribution of insects (SOUTHWOOD, 1978; KUSMAYADI et al., 1990). The fitness of the observed distributions to the negative binomial model was tested using a chi-square test (RABINOVICH, 1978).

2.3 Aggregation index

A further step in the analysis of the spatial pattern of populations whose observed frequency distributions show departure from randomness is quantifying the degree of aggregation. A number of indices have been used for this purpose, but most of them are known to be mathematically similar (KUNO, 1977). The index adopted in this paper is the C_A proposed by KUNO (1991) which is the reciprocal of the k parameter of the negative binomial model. Unbiased estimates of C_A for each category were calculated using equation (1):

$$C_A = \frac{s^2 - m}{m^2 - \frac{s^2}{n}} \quad (1)$$

where S^2 , m and n are variance, the mean and number of samples, respectively. The greater C_A , the more aggregated the population.

To interpret the general feature of *D. kuscheli* distribution throughout the period of the population buildup, the overall distribution pattern was analysed with the BLISS and OWEN'S (1958) method (KUSMAYADI et al., 1990). A single parameter, 'common C_A ' (C_A^* , from hereafter) can be obtained as the slope of the linear regression of y' on x' passing through the origin, where y' is $s^2 - m$ and x' is $m^2 - s^2/n$. This slope parameter represents an aggregation index that characterizes the general spatial pattern of the population (ALLSOP and BULL, 1989).

2.4 Sampling plans

The minimum number of sampling units and critical lines for sequential sampling plans were calculated based on the estimation of C_A^* for the precision levels (D) 0.1, 0.2 and 0.3. The relative precision $D = 0.1$ might be required for research purposes, such as the estimation of population parameters from stage frequency data (MANLY, 1989), whereas $D = 0.2$ or $D = 0.3$ are often suggested for use in pest management (SOUTHWOOD, 1978).

2.5 Sample size and the required precision level

The standard procedure for estimating the population density is to take an appropriate number of samples from the field to estimate the mean density and then assign a level of precision to the estimation in terms of the error relative to the mean ($D = \sqrt{\text{variance}/m}$).

Most insect populations show a clumped pattern (SOUTHWOOD, 1978). Among several two-parameter mathematical models proposed to describe aggregated spatial patterns, the negative binomial model is the most widely used because it is generally a good descriptor of field data (KUNO, 1991). In

such cases, the parametric variance is given by $\sigma^2 = m + m^2/k = m + C_A m^2$ and the variance estimate of the mean population density is given by $S^2 = (1/n)\sigma^2 = (1/n)(m + C_A m^2)$. The required precision level is then $D = \sqrt{(1/n)(1/m + C_A)}$. From this equation, the minimum sample size for securing a fixed precision level D is given by:

$$n = \frac{1}{D^2} \left(\frac{1}{m} + C_A \right) \tag{2}$$

The precision of population abundance estimation is greatly affected by the mean density. Consequently, a preliminary estimate of m should be obtained to calculate the appropriate sampling size. This procedure can be costly in terms of time and required labour. Sequential sampling is an alternative approach in which the sample size n is not a predetermined constant but a variable to be estimated during the sampling process. In sequential sampling, data collection is interrupted when the cumulative total number of individuals so far observed, Tn , intersects a critical line which has been previously calculated by the equation:

$$Tn = \frac{1}{D^2 - \frac{C_A}{n}} \tag{3}$$

(KUNO, 1969, 1977).

3 Results and discussion

3.1 Spatial pattern of *D. kuscheli* population

During the 1993 season, *D. kuscheli* population showed an exponential growth until the end of November. Afterwards, the population density decreased abruptly (fig. 1a). During the 1994 season, the population density increased in an early phase, although the population growth pattern was irregular (fig. 1b). Population abundance during the first season was much higher than in the second season.

Pooling the data from both growing seasons, the negative binomial model fitted the observed frequency distributions in more than 78% of the cases (sampling dates) while the Poisson model fitted well in only 28% of cases or less (table 1). These results clearly show that *D. kuscheli* populations in oat fields have an aggregated spatial pattern. This trend is most evident in nymphs. Frequency distributions of the first four immature stadia fitted the Poisson model in none of the cases while they fitted the negative binomial in 86 to 93% of the cases (table 1), showing a very clear trend to aggregation. It should be noted that a significant fit to the Poisson model does not necessarily mean that the population is actually randomly distributed. Indeed, at low population densities the distribution of an organism cannot depart markedly from the Poisson expectation for statistical limitations even though the organism may have some tendency to aggregation (TAYLOR, 1984; KUSMAYADI et al., 1990).

Temporal changes in spatial distribution pattern of *D. kuscheli* were analysed in terms of the C_A index. During the 1993 season, C_A lay within the 0.3–0.58 range, except on November 6, when $C_A = 1.15$ (fig. 1a). During the 1994 season, C_A was more variable and lay within the 0.32–0.84 interval (fig. 1b). The correlation analysis of insect density and the C_A index showed no significant relationship.

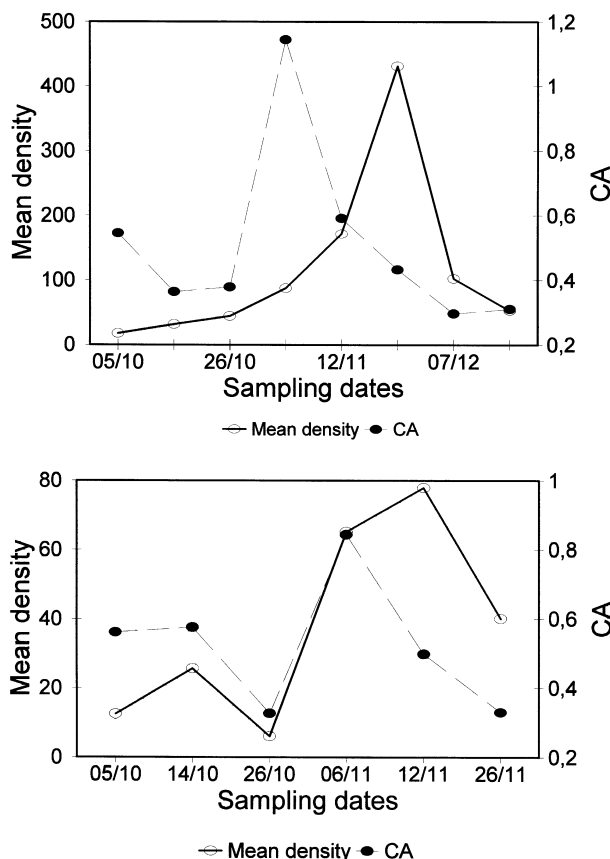


Fig. 1. Temporal fluctuation of insect density and aggregation index C_A ($=1/k$) during growing seasons (a) 1993 and (b) 1994

Table 1. Percentage of cases during growing seasons 1993 and 1994 in which the observed frequency distributions of the number of insects per sample fitted the negative binomial and Poisson probabilistic models

Insect category	Fit percentage	
	Negative binomial	Poisson
Males	100	28
Females	100	21
Macropteros	78	21
Brachipteros	78	43
Adults	93	21
V Nymphs	100	21
IV Nymphs	86	0
III Nymphs	93	0
I–II Nymphs	93	0
Total	79	0

The percentage of macropterous adults in the 1993 season increased gradually from 40% on the first sampling date up to almost 100% at the end of the season, while it was always higher than 75% during the 1994 season. However, no relevant differences in C_A values from both seasons were found. Neither does population density explain the difference, since the highest percentages of long-winged adults occurred during the least crowded season.

Table 2. Estimates of the common aggregation index (C_A^*) through Bliss and Owen's method. C_A^* , common aggregation index; SE, standard error; R^2 , coefficient of determination; P, significance level of Bliss and Owen's regression coefficient

Category	1993			1994			1993-94		
	$C_A \pm SE$	R^2	P	$C_A \pm SE$	R^2	P	$C_A \pm SE$	R^2	P
Males	0.26 ± 0.014	0.98	<0.001	0.39 ± 0.13	0.70	0.037	0.26 ± 9E-3	0.98	<0.001
Females	0.18 ± 0.018	0.94	<0.001	0.34 ± 0.043	0.94	0.001	0.19 ± 0.01	0.95	<0.001
Macropterous	0.25 ± 0.017	0.97	<0.001	0.33 ± 0.05	0.90	0.003	0.25 ± 0.01	0.97	<0.001
Braquipterous	0.27 ± 0.1	0.53	0.04	0.54 ± 0.1	0.87	0.007	0.28 ± 0.06	0.67	0.003
Adults	0.23 ± 0.17	0.97	<0.001	0.28 ± 0.051	0.88	0.006	0.23 ± 0.01	0.97	<0.001
V Nymphs	0.47 ± 3E-3	0.999	<0.001	0.47 ± 0.028	0.98	<0.001	0.47 ± 2E-3	0.99	<0.001
IV Nymphs	0.41 ± 5E-3	0.999	<0.001	0.52 ± 8E-3	0.99	<0.001	0.42 ± 4E-3	0.99	<0.001
III Nymphs	0.52 ± 0.03	0.98	<0.001	0.48 ± 0.017	0.99	<0.001	0.52 ± 0.02	0.98	<0.001
I-II Nymphs	0.63 ± 0.06	0.95	<0.001	1.08 ± 0.025	0.99	<0.001	0.64 ± 0.04	0.94	<0.001
Total	0.43 ± 0.017	0.99	<0.001	0.61 ± 0.12	0.87	0.007	0.43 ± 0.01	0.99	<0.001

The values of the overall aggregation index, C_A^* , estimated by the Bliss and Owen's regression method (BLISS and OWEN, 1958) for each category of *D. kuscheli* identified and each growing season are given in table 2. The lowest and highest C_A^* values were 0.18 (females 1993) and 1.08 (nymphs I-II 1994), respectively. Among the different categories identified, the nymphs, particularly those of the I-II class, showed the highest degrees of aggregation. This could be explained by the lower ability and opportunity of young nymphs for dispersing from the plant where they were born. These relationships apply in both seasons, although C_A^* estimates from the first season were in general lower than those for the second season. One example of the regression lines obtained, corresponding to total *D. kuscheli* is represented in fig. 2, where the regression slope indicates the value of C_A^* .

The C_A^* estimates obtained in this work are lower than those obtained for the planthopper *Nilaparvata lugens* Stål, a rice virus vector in south-east Asia (KUSMAYADI et al., 1990). However, such differences could be a consequence of using different sampling methods.

3.2 Sampling plans

The necessary information (C_A^*) to calculate sampling plans for each *D. kuscheli* category identified is available

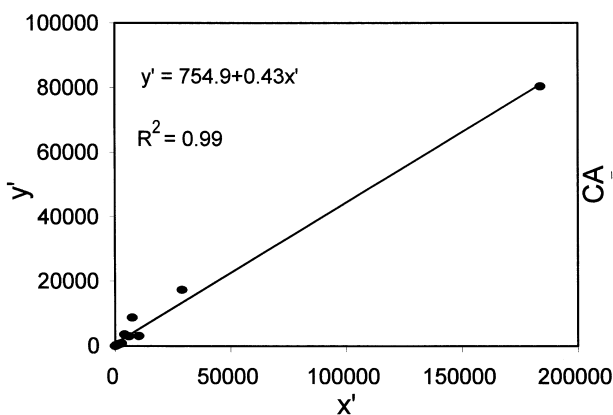


Fig. 2. Bliss and Owens's regression fitted with pooled data from growing seasons 1993 and 1994. x' : $m^2 - s^2/n$; y' : $s^2 - m$

in table 2. As an example, the sampling plans for total *D. kuscheli* are presented. The minimum number of sampling units necessary for estimating total *D. kuscheli* density with three precision levels is illustrated in fig. 3. The sampling effort demanded by a precision level $D = 0.1$ is much greater than that necessary should D be increased to 0.2. Indeed equation (2) shows that n is inversely proportional to the square of D . Consequently, in deciding on a monitoring plan, care must be taken not to adopt a unnecessarily low value of D (KUNO, 1977).

The lower the population density, the greater the difference between the sampling efforts demanded by two different precision levels. For $D = 0.1$, the minimum number of samples varies sensibly at low densities ($m = 1-20$). As density increases, the magnitude of change in minimum n decreases. If a mean population density of more than 20 individuals along the whole study period were expected, a constant sampling size of about 50 samples would warrant population estimates with a fairly constant precision level. On the contrary, if low population densities were expected, as in early

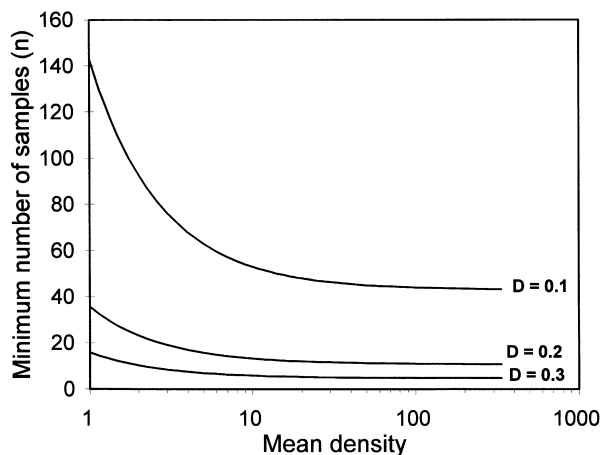


Fig. 3. Minimum number of sample units required to estimate total *Delphacodes kuscheli* density with fixed precision levels (D) and a common aggregation index $C_A^* = 0.43$

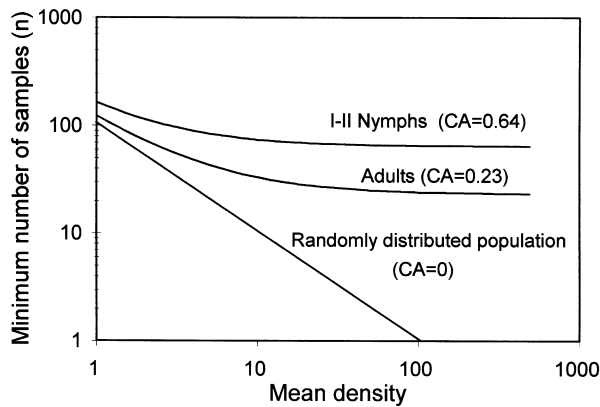


Fig. 4. Minimum number of sample units required to estimate *Delphacodes kuscheli* density with a fixed precision levels $D = 0.1$. Comparison between I-II nymphs ($C_A^* = 0.64$), adults ($C_A^* = 0.23$) and a hypothetical randomly distributed population ($C_A^* = 0$)

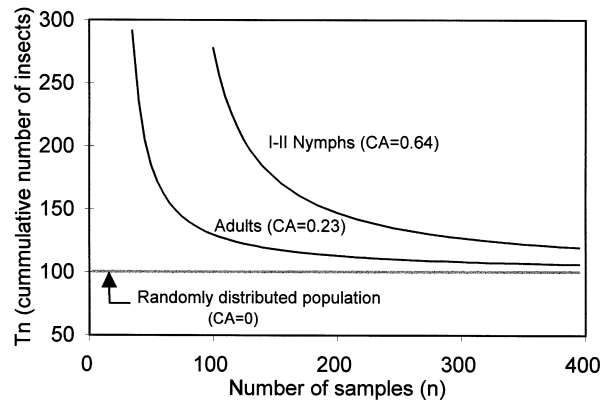


Fig. 6. Stop lines for sequential sampling to estimate density of *Delphacodes kuscheli* with a fixed precision level $D = 0.1$. Comparison between I-II nymphs ($C_A^* = 0.64$), adults ($C_A^* = 0.23$) and a hypothetical randomly distributed population ($C_A^* = 0$)

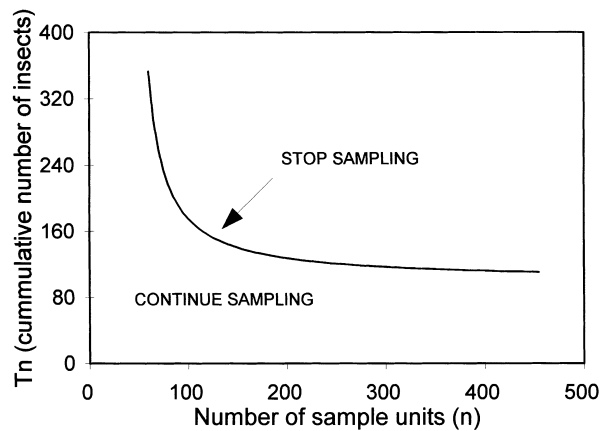


Fig. 5. Stop line for sequential sampling of total *Delphacodes kuscheli* to estimate density with a fixed precision level $D = 0.1$

to mid spring, preliminary estimates of the population mean would be necessary to determine the definitive sampling size required.

The minimum sampling size is also affected by the degree of aggregation of the population. Figure 4 compares the minimum sampling size required with a precision level $D = 0.1$, for I-II nymphs ($C_A^* = 0.64$), adults ($C_A^* = 0.23$) and a hypothetical randomly distributed population ($C_A^* = 0$). Even low degrees of aggregation, like that of adults, demand much more sampling effort than a random distribution, particularly at high densities. This observation illustrates the risk of calculating sampling plans under the assumption that the population has a random spatial pattern.

The boundary line for sequential sampling of total *D. kuscheli* was calculated using equation (3) (KUNO, 1969, 1977), based on the estimated parameter C_A^* ($=0.43$) of the Bliss and Owen regression (fig. 5). To execute this sampling plan, sample units are taken and planthoppers are counted. If the point (n_i, Tn_i) (where n_i the number of sample units taken and Tn_i is the cumulative planthopper counts on the n_i units), is below the line for the

required precision level (D), it is necessary to continue the sampling because the fixed, predetermined level of precision has not been achieved. On the contrary, when the point (n_i, Tn_i) is above the boundary line for the desired level of precision, the mean can be estimated as Tn_i/n_i and the required level of precision is obtained. Sequential sampling plans are also affected by the degree of aggregation of populations. Figure 6 compares the critical lines calculated for a precision level $D = 0.1$, corresponding to I-II nymphs ($C_A^* = 0.64$), adults ($C_A^* = 0.23$) and a hypothetical randomly distributed population ($C_A^* = 0$).

Monte Carlo simulation (NARANJO and FLINT, 1994) and resampling (NARANJO and HUTCHINSON, 1997) studies show that the precision level specified in a sequential sampling plan can differ from that actually achieved when the plan is executed. Consequently, sampling plans should be taken as a provisional guidance until they are validated through simulation or against independent field data. An additional limitation is the spatial scale with which this study was done. Definitive monitoring plans should be based on data gathered at different localities if they are to be adopted by farmers or agronomists. Despite these constraints, and considering the complete lack of alternative criteria for monitoring the MRC vector in Argentina, it is considered that the sampling plans proposed in this paper could be of much help, particularly for those who are using the predictive model based on *D. kuscheli* population density in oat fields.

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