SAMPLING EFFICIENCY OF DIFFERENT SAMPLE UNITS AND PROCEDURES TO SAMPLE SPITTLEBUG EGGS[1]

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ABSTRACT - A sampling study of spittlebug eggs in pastures of *Brachiaria decumbens* Stapf was conducted by using samples of 25, 50 and 75 cm^2 of the pasture. The sampling efficiency of three sample units was compared by calculating the number of samples necessary to obtain a certain level of precision (proportion of standard error to mean expressed as a percentage). The sampling efficiency of 50 cm^2 sample unit was poor in comparison to 25 and 75 cm^2. To obtain the same level of precision, e.g., 20%, the total pasture area to be examined was slightly smaller for 75 cm^2 unit than for 25 cm^2 at lower egg density (1/25 cm^2); however, such advantage of the 75 cm^2 disappeared at egg density of 4/25 cm^2. Based on this finding and the fact that fewer samples would be taken in the case of 75 cm^2 than the 25 cm^2 unit, the use of former was suggested. The distribution pattern of number of eggs per 25 or 50 cm^2 of pasture fitted the negative binomial series.

Index terms: *Zulia entreriana*, *Deois flavopicta*, Cercopidae, *Brachiaria decumbens*, pasture.

A EFICIÊNCIA DE AMOSTRAGEM DE DIFERENTES UNIDADES E PROCEDIMENTOS PARA AMOSTRAR OVOS DAS CIGARRINHAS-DAS-PASTAGENS

RESUMO - Foi conduzido um estudo sobre amostragem de ovos de cigarrinhas em pastagens de *Brachiaria decumbens* Stapf, usando-se as amostras de 25, 50 e 75 cm^2 da pastagem. A eficiência de amostragem das três unidades foi comparado calculando-se o número necessário de amostras para um certo nível de precisão (a proporção do erro-padrão, a média, em percentagem). A eficiência de amostragem da unidade de 50 cm^2 foi baixa em comparação à de 25 ou 75 cm^2. Para se obter um mesmo nível de precisão, por exemplo, de 20% a área total de pasto a ser examinado foi um pouco menor para a unidade de 75 cm^2 do que para a de 25 cm^2 quando a densidade de ovos era baixa (1/25 cm^2), porém essa vantagem desapareceu quando a densidade foi de 4 ovos/25 cm^2. Com base nisto, e o fato de que um menor número de amostras seria necessário no caso da unidade de 75 cm^2, do que com a de 25 cm^2, o uso da anterior foi sugerido. O número de ovos das cigarrinhas por 25 e 50 cm^2 de área de pastagem mostrou uma distribuição do tipo binomial negativa.

Termos para indexação: *Zulia entreriana*, *Deois flavopicta*, Cercopidae, *Brachiaria decumbens*, pastagem.

INTRODUCTION

Knowledge about spittlebug egg densities in various parts of a pasture would be useful when considering control methods such of fire in selective areas (Martin 1983) and pasture management tactics where predictions about severity of the forthcoming infestation are necessary (Nilakhe 1983). However, the examination of soil and plant samples for eggs is a tedious process. The basic objectives of their work were to estimate egg densities with the minimum possible effort, and with an acceptable level of precision or relative variation (proportion of standard error to mean expressed as a percentage). Using these 2 criteria, they compared the sampling units of different sizes and reported a sampling unit of 75 cm^2 to be superior over those of 150, 225 and 625 cm^2. The next step then was to determine whether reducing the sample size further would improve the sampling efficiency.

Herein, the sampling efficiency of a 75 cm^2 unit to that of 50 and 25 cm^2 is compared. Information is also included on the distribution pattern of spittlebug eggs in *Brachiaria decum-*

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bens Stapf pastures, number of samples necessary for various levels of precision, and choice of sample sites in the sampling area.

MATERIALS AND METHODS

Egg samples from 12 pastures were obtained during June-September (before beginning of egg hatch) of 1985 near Campo Grande and in the region of Dourados. The majority of spittlebug adults in these areas were species *Zulia entreriana* (Berg.) and *Deois flavopicta* Stal; therefore it was assumed that eggs sampled belonged to these two species. In each pasture, an area of 100 m x 100 m was designated as the field, and was divided into four blocks and each block was further divided using a 2 x 2 grid in four equal plots, thus giving a total of 16 plots per field. In each plot, three 50 cm^2 (8 cm diameter) and three 25 cm^2 (5.6 cm diameter) areas of the pastures were selected at random for sampling. A sample consisted of the above-ground portions of grass plants clipped at about 8 cm height and soil inside the sample area to a depth of 2.5 cm. After clipping the grass plants in the sampling area, the sharper end of the metal sampler (15 cm length) was driven into the ground by beating on one end with a heavy wooden block. Prior to clipping the plants, plant cover was estimated using a scale of 0 to 10, where, 0 = no grass plants up to 10 = entire area covered with plants. In the laboratory the sample was washed and dried (Nilakhe et al. 1984a), passed through a blower to remove the plant debris (Nilakhe et al. 1984c) and then examined for spittlebug eggs by spreading on black paper. Additionally, 20 egg samples of 75 cm^2 (9.8 cm diameter) were obtained from each of the two 1 ha pastures in November 85.

To test the distribution pattern of spittlebug eggs for conformity to the Poisson distribution, the index of dispersion I_D was calculated (Southwood 1978). The negative binomial parameter (k) was also calculated as given by Southwood 1978. To study the importance of variation between blocks, and plots within blocks, the egg counts from each field were subjected to an analysis of variance. The variation between rows and between columns was also evaluated in this manner. Because the spatial pattern of spittlebug eggs within a field conformed to the negative binomial distribution, the variance was stabilized by transforming the data as Log (x + 1), where "x" was the observed count. The association between the plant cover in the sample area and the number of eggs was examined by calculating correlation coefficients.

RESULTS

The number of spittlebug eggs obtained in the 25 and 50 cm^2 samples are given in Tables 1 and 2, respectively. The numbers show that there was a good representation of different densities in the study. Values of relative variation (RV) were higher in the 25 cm^2 samples as compared to the 50 cm^2. For example, in the 50 cm^2 unit the RV exceeded 25% in three of the twelve cases whereas in the 25 cm^2 unit it exceeded in six instances. This indicated that with equal sample numbers, the 50 cm^2 unit showed better precision than the 25 cm^2.

The index of aggregation (k) was always less than 1, indicating that the egg counts tended to clump and conform to the negative binomial distribution. None of the I_D values conformed to the Poisson distribution. A contagious distribution of eggs was also supported by the variance mean ratios, since in all the cases the variance exceeded the corresponding mean.

The mean egg counts in two fields sampled during rainy season were 3.1 ± SE of 0.66 in field 1, and 4.7 ± 1.10 in field 2. The relative variation value for the field 1 was 21%, and it was 24% for field 2. The k values were 1.73 and 1.14 for the field 1 and 2, respectively. These egg counts also conformed to the negative binomial distribution pattern.

The number of samples required for precision levels of 10%, 15% and 20% are given in Tables 1 and 2. These estimates are shown graphically in Figs. 1 and 2. The graphs show that the number of samples required for a certain level of precision was inversally proportional especially at the lower egg densities. The number of samples remained about the same for egg densities of four or greater, in the 25 cm^2 unit, and eight or greater in the 50 cm^2 unit.

To compare further the sampling efficiency of the 25 cm and 50 cm units, the number of samples required for RV of 20% at egg densities of 1 and 4/25 cm^2, and 2 and 8/50 cm^2 was obtained from the Fig. 1 and 2, and are presented in Table 3. The number of samples for the 75 cm^2 unit was obtained from Nilakhe et al. (1985). The total pasture area to be examined with the 50 cm^2 unit tended to be larger than that for the 25 or 75 cm^2. At an egg density of 1/25 cm^2 (3 for 75 cm^2 unit), the pasture area to be examined was slightly smaller when 75 cm^2 unit was used

($3225\ cm^2$), as compared to the $25\ cm^2$ unit ($4100\ cm^2$). However, the advantage of the $75\ cm^2$ disappeared when the egg density was increased to $4/25\ cm^2$. Nevertheless, based on these findings and on the fact that far fewer samples would be necessary using the $75\ cm^2$ unit than the $25\ cm^2$, the former is preferred.

The analyses of variance showed that the blocks, and the plots within blocks were significant in 5 of the 12 instances both for the 25 and $50\ cm^2$ units. The rows or columns were significant in 3 of the 24 instances. The correlation coeficient values (r) for degree of plant cover and egg counts were significant in 3 of 12 instances in the $50\ cm^2$ unit and in 2 of 12 instances for the $25\ cm^2$ unit.

DISCUSSION

For comparison purposes, egg densities of one and $4/25\ cm^2$ and 20% level of precision were chosen - one may choose different egg densities and different precision levels, e.g., 10% or 15%. An egg density of 1 represented the lowest of the densities observed in the study and at densities higher than 4, the number of samples needed remained about the same.

A high degree of aggregation of spittlebug eggs as indicated by low k values, suggested that within a pasture certain sites are highly attractive to spittlebug for oviposition. Overall, lack of association between the plant cover and the number of eggs in the sampling area indicated that factors other than plant cover could be important in the female's choice of oviposition sites.

Studies such as those of Nilakhe et al. 1984a, and the present study permit comparison of sampling efficiency of different sample units. Initially, we started with a large sample unit ($625\ cm^2$) and then the sample size was reduced; the smallest size studied was $25\ cm^2$. Thus among sample units of 625, 225, 150, 75, 50 and $25\ cm^2$, the $75\ cm^2$ was chosen.

TABLE 1. Mean number of spittlebug eggs in $25\ cm^2$ areas in pastures of *Brachiaria decumbens*, sampling variability, mathematical distribution characteristics of the counts and estimates of numbers of samples required for 3 levels of precision, Mato Grosso do Sul, 1985.

Field	$\bar{x} \pm SE$[1]	Variance (S^2)	CV[2]	RV[3]	Index of aggregation (k)[4]	Index of dispersion (I_D)[5]	No. of samples required for SE of a certain percentage of mean[6]		
							10	15	20
1	0.69 ± 0.24	2.66	236	35	0.24	181	557	248	139
2	0.75 ± 0.19	1.79	179	25	0.54	112	320	142	80
3	0.79 ± 0.40	7.62	349	51	0.09	453	1218	541	305
4	1.13 ± 0.26	3.31	161	23	0.59	138	269	115	65
5	1.23 ± 0.31	4.67	176	25	0.44	180	310	138	77
6	1.31 ± 0.25	3.10	134	19	0.96	111	180	80	45
7	1.73 ± 0.41	8.00	164	24	0.48	217	269	120	67
8	2.89 ± 0.51	12.25	121	18	0.89	199	146	65	37
9	3.95 ± 1.46	102.82	257	37	0.16	1223	660	294	165
10	4.06 ± 0.97	45.56	166	24	0.40	527	276	122	69
11	4.35 ± 1.17	65.45	186	27	0.31	707	346	154	86
12	9.89 ± 2.24	240.56	157	23	0.42	1143	246	110	62

[1] Mean based on 48 samples; SE = Standard Error
[2] $CV = (Std. dev./\bar{x}) \times 100$
[3] $RV = (SE/\bar{x}) \times 100$
[4] $k = \bar{x}^2/(S^2 - \bar{x})$
[5] $I_D = S^2\ (n - 1)/\bar{x}$
[6] No. of samples for SE of a certain probability. $(P) = (CV/P)^2$

TABLE 2. Mean number of spittlebug eggs in 50 cm^2 areas in pastures of *Brachiaria decumbens*, sampling variability, mathematical distribution characteristics of the counts and estimates of numbers of samples required for 3 levels of precision, Mato Grosso do Sul, 1985.

Field	$\bar{x} \pm SE$[1]	Variance (S^2)	CV[2]	RV[3]	Index of aggregation (k)[4]	Index of dispersion (I_D)[5]	No. of samples required for SE of a certain percentage of mean[6]		
							10	15	20
1	1.43 ± 0.33	5.34	162	23	0.52	176	262	117	66
7	1.69 ± 0.41	8.07	168	24	0.45	224	282	125	71
3	1.94 ± 0.54	14.06	193	28	0.31	341	372	166	93
6	2.98 ± 0.61	17.47	140	20	0.61	276	196	87	49
10	3.42 ± 1.18	66.91	239	35	0.18	920	571	254	143
2	3.46 ± 0.58	16.16	116	17	0.94	220	135	60	34
4	3.58 ± 0.77	28.40	149	22	0.52	373	222	99	56
9	5.93 ± 1.29	80.28	151	22	0.47	636	228	101	57
8	6.14 ± 1.13	61.78	128	18	0.68	473	164	73	41
5	6.89 ± 1.73	144.00	174	25	0.35	982	303	135	76
12	9.19 ± 1.78	151.78	134	19	0.59	776	180	80	45
11	12.88 ± 2.83	384.16	152	22	0.45	1402	231	103	58

[1] Mean based on 48 samples; SE = Standard Error
[2] $CV = (Std.\ dev./\bar{x}) \times 100$
[3] $RV = (SE/\bar{x}) \times 100$
[4] $k = \bar{x}^2/(S^2 - \bar{x})$
[5] $I_D = S^2 (n - 1)/\bar{x}$
[6] No. of samples for SE of a certain probability (P) = $(CV/P)^2$

Generally, smaller sampling units are reported to be more efficient than the larger units. For example, for sampling spittlebug nymphs, the 25 cm x 25 cm unit was more efficient than 50 cm x 50 cm or 100 cm x 100 cm (Nilakhe 1982). Working with soil insects, Fleming & Baker (1936), Burrage & Gyrisco (1954), and Guppy & Harcourt (1973) found the 0,09 m^2 unit to be more efficient than 0.36, or 0.81 m^2 units. Yates & Finney (1942) and Finney (1946) also found a greater sampling efficiency with smaller sampling units than with the larger ones.

However, other authors indicated that reducing the samples size beyond a certain minimum level may not be beneficial. For example, to sample wireworms a sampling unit of 0.09 m^2 was found to be more efficient than 0.0225 and 0,0056 m^2 (Jones 1937). In the case of spittlebug adults, in *B. decumbens* pastures, a sample of 10 sweeps of a sweep net was more efficient than 20 or 40 sweeps of a sweep net; however, reducing the sweep numbers from 10 to 5 did not show any additional benefits (Nilakhe et al. 1984b).

However, these numbers were suggested for egg densities of 2/75 cm^2 of pasture. Clearly at higher egg densities so many samples would not be necessary. For example, for moderately high egg density (12 per 75 cm^2, or 1600/m^2), 100 samples would probably be sufficient for the 10% level of precision. This level of precision would b necessary for studies where minute differences need detection. Some examples of studies needing 10% level of precision could be: egg counts before and after fire, change in egg densities during the dry season, life table, modelling etc. If prior informations of egg densities in the experimental area is available, then the sample numbers could be changed (reduced for higher egg densities) accordingly. The occurrence of a large number of spittlebug adults in the experimental area would also probably mean high egg densities in the field.

In the present study, the differences between blocks, and plots within blocks were important both for the 25 and 50 cm^2 units. However, that was not the case for the 75 cm^2 unit. Because the 75 cm^2 unit was chosen over the other two, the sampling procedure for this unit follows: Unless for some specific reasons, blocking within the sampling area of about one ha may not be necessary. Choose samples at random. Procedure

TABLE 3. Number of samples required for 20% level of precision for different spittlebug egg densities[1].

Sample size (cm^2)	Egg density/ sample	No. of samples required for SE of 20% of mean	Pasture area to be examined (cm^2)
25	1	164	4100
50	2	115	5750
75	3	43	3225
25	4	60	1500
50	8	46	2300
75	12	21	1575

[1] The no. of samples for the sample units of 25 and 50 cm^2 was obtained from Fig. 1 and 2, respectively. Data for the 75 cm^2 sample unit were from Nilakhe et al. (1985).

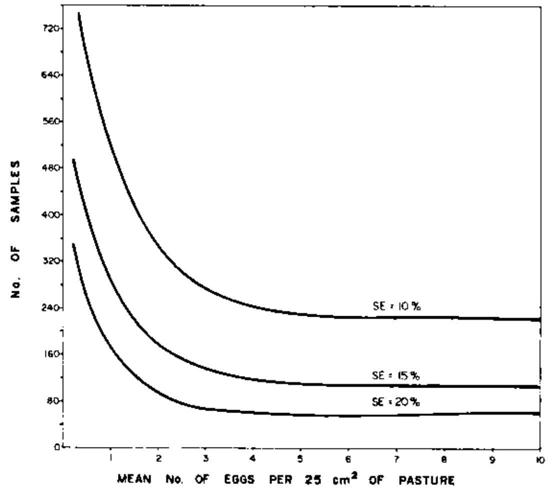

FIG. 1. Relationship between population density and number of samples necessary for three levels of precision (proportion of standard error to mean expressed as a percentage) for spittlebug eggs sampled with the 25 cm^2 sample unit in pastures of *Brachiaria decumbens*.

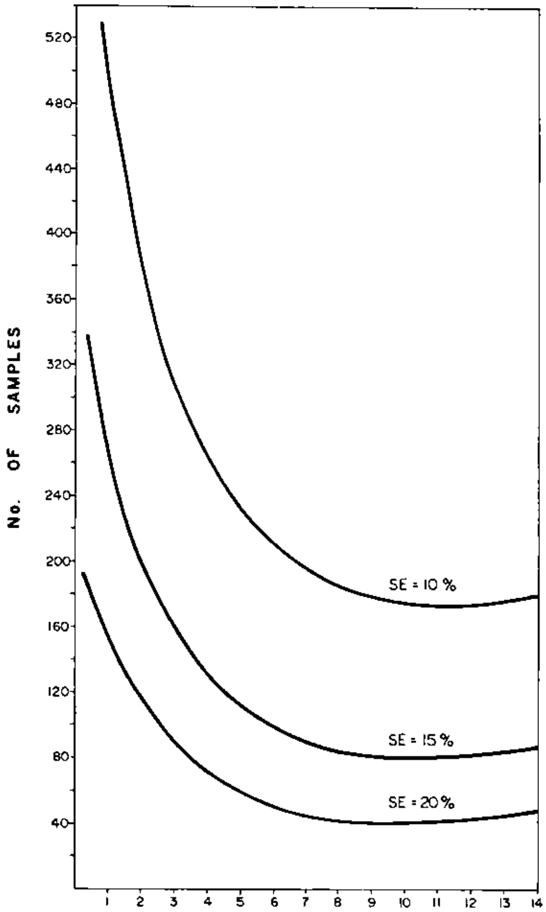

FIG. 2. Relationship between population density and number of samples necessary for three levels of precision (proportion of standard error to mean expressed as a percentage) for spittlebug eggs sampled with the 50 cm^2 sample unit in pastures of *Brachiaria decumbens*.

to sample in large pasture, e.g., 100 ha is not available. In such a pasture, blocking would probably be necessary. One may divide the pasture in 5 blocks and then at random obtain equal number of samples (for example 10) in each block.

Egg counts were taken in only two fields during the rainy season; however, the counts showed a similar degree of clumping as the counts made during the dry season. It is very likely that the number of samples needed for a certain level of precision would be equal for sampling done both in the dry and the rainy season. It is clear from these sampling studies that relatively high numbers are needed for an accurate determination of spittlebug egg densities. This would greatly increase the time spent in examination of soil for the eggs and thus development of faster methods of egg extraction is surely needed.

CONCLUSIONS

1. A high degree of aggregation of spittlebug eggs in a pasture indicated that certain sites are highly attractive to the females for oviposition.

2. Sampling efficiency of a series of sample units was compared and the use of the 75 cm^2 unit was suggested for estimating spittlebug egg densities in *Brachiaria decumbens* pastures.

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