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THE EFFECT OF PLOT SIZE ON SOME *PRATYLENCHUS PENETRANS (*PHYTONEMATODE) POPULATION PARAMETERS

AMINAYANABA ONARI ASIMIEA
Department of Crop and soil Science, Faculty of Agriculture,
University of PortHarcourt P. M. B.5323, Port Harcourt
E-mail: mina_2000laurel@yahoo.Com. Tel.234-80-33107774

ABSTRACT
Pratylenchus penetrans counts obtained from a rose field, sampled sequentially by decreasing the plot sizes were computed to obtain the respective sample means, variance and k-value of the negative binomial distribution. Plots 21 m x 80 m, 3.6 m x 3.6 m and 0.6 m x 0.6 m were sampled for the nematode. It is reported that the k-value is influenced by plot size ($P < 0.05$) and depends on the sample mean.

Keywords: core number, negative binomial distribution, k-value, plot size, *Pratylenchus penetrans*.

INTRODUCTION
The distribution of plant parasitic nematodes within an ecosystem is determined by such factors, as the length of time the population has been present in the field. Variation in factors that influence habitat suitability, like soil texture, moisture and drainage pattern are important. In addition to these, selection pressure of the host plant, life history (including feeding strategies) of the nematode and cultural practices of the farmer (e.g. extent of tillage) are also influential. These factors act in concert to determine the degree of aggregation in the spatial population pattern of nematode (Ferris *et al.,* 1990).

Various attempts have been made to improve sampling plans and data processing - methods, as to obtain a more representative mean of the nematode population in the field. In addition to this, clustered spatial patterns of various nematodes have been illustrated in different type of maps (Barker and Campbell, 1981 Seinhorst 1982). Aggregated populations have been studied and described by various techniques among which the negative binomial model is earlier and well discussed (Anscombe, 1950; Gates and Ethridge, 1970; Elliott, 1977; Southwood, 1978; Goodell and Ferris, 1980). The negative binomial probability distribution, which often describes aggregated population, is characterized by the parameters of the population mean and the exponent k. These parameters provide the basis for estimating the number of samples required to attain a given level of sampling precision. The k-parameter is related to the degree of aggregation and is considered as an index of dispersion (Barker *et al.,* 1985).

Despite these potentials, some studies have reported shortcomings associated with the use of the negative binomial distribution (Taylor *et al.,* 1979; Noel and Campbell, 1985). The k value is dependent on the population mean and due to poor information on cluster sizes, nematode population estimates vary widely with sampling procedure. This introduces the element of sampling error, a parameter that is not predetermined accurately prior to sampling. From the forgoing, it is clear that the k-value is useful for evaluating frequency distribution but offers little information on the precise nature and scale of aggregation (Taylor *et al.,* 1979; Taylor, 1984; Noe and Campbell, 1985). So far available reports on nematode cluster sizes are those on marine nematodes by Delmotte (1975), Findlay (1981, 1982b) and Hogue (1982). This investigation is aimed at evaluating the importance of plot size in the use of negative binomial distribution for the determination of the cluster size of *Pratylenchus penetrans* an endoparasitic nematode with free moving stages. Reports on *Pratylenchus penetrans* and other free moving nematodes are scarce.

MATERIALS AND METHODS

Samples were collected from a rose field located in Wetteren, about 15 kilometers from the city of Ghent in Belgium. The plot of land has been used for the cultivation of tree nursery of *Robinia pseudoaeaeia* and *Aeer plaranoides* (Rose plant), in the past 5 years without the application of nematicide since its inception. Tilling was done by ploughing and harrowing at a depth of about 30 cm with conventional machines. The plot of land measuring 1680 m^2 (21 m x 80 m) was divided into grids of 3.65m x 8m and a total of 50 single core samples were collected at points of intersection of the grid lines. The samples were collected with an auger that is 20 cm in length and 2.5 cm in diameter. They were immediately put into plastic bags, labeled and taken to the laboratory for storage at 50^oC for subsequent extraction and counting.

Based on the nematode counts obtained from each sampling point in the first sampling stated above, three categories of sites were chosen to represent points of low, medium and high populations (0-100, 101-200 and >200 *P. penetrans*/100g soil and above respectively). Plots (3.60 m x 3.60 m) were obtained using the points of intersection as center. These plots were then subdivided by perpendicular grid lines, 0.6 m apart and 36 samples were collected at point of intersections of these grid lines. Furthermore, using these last points as center, plots of 0.6 m x 0.6 m were mapped out and divided by perpendicular grid lines that are 0.1 m apart. Finally, at the point of intersection of these lines, 36 samples were collected. This procedure was repeated for each of the three replicates of each category of the bigger plots (3.6 m x 3.6 m) labeled A, B and C respectively in fig.2 and each of the two replicates of each category of the smaller plots (0.6 m x 0.6 m) labeled D, E and F respectively in fig. 3. A total of 590 samples were taken for nematode extraction and counting. All the samples taken were single core samples and were treated as such.

Nematodes were extracted from the soil samples using the centrifugal floatation methods introduced by Caveness and Jensen (1955) and modified by Coolen and D'Herde (1972). The nematodes were then washed off the sieve and suspended in 100 ml water. Two replicates of 10 ml each were counted in a De Grisse counting dish and the average count multiplied by 10 gave the nematode count per 100 gm soil. This procedure was repeated for all the 590 samples. The mean (x) and variance (S^2) were calculated using the statistical package of Lotus 1-2-3. These parameters were calculated for each set and subset of samples and the formula for the k-value, $k = (X)^2/(S^2X)$ which is an index of aggregation based on the negative binomial distribution was calculated. Soil particle size was analyzed, using a COULTER^R LS analyzer. The particle sizes were determined by percentage volume of various particle size ranges occurring in a sub sample pulled from all the samples collected.

RESULTS

A contour map of the macrodistribution of *P. penetrans* in the 21 m x 80 m plot is shown in Fig. 1. The population mean (X) is 61.36, variance (S^2) is 4956.15 and the k-value of the negative binomial distribution is 0.77. This result suggests a highly clustered distribution. The frequency distribution of *P. penetrans* in the 3.6 m x 3.6 m plots is shown in Fig. 2. In this figure A, B and C represent frequency distributions of samples taken in 3 replicates of plots of high, medium and low densities respectively. There are no remarkable differences between A, B and C categories of plot in terms of frequency classes. Similarly, in Fig. 3 frequency distributions of *P. penetrans* in the 0.6 m x 0.6 m plots are presented. In the high-density plots D, densities are concentrated in higher density class ranges while in the medium density plots E, the frequencies are spread over a wide range. In the lower density plots (F) the spread is minimal and occurs within lower density classes.

Table 1. Statistics of the frequency distributions of nematode counts as influenced by plot size and nematode density.

Plot size		x	S^2	k-value
3.6 m x 3.6 m	A	319.61	20391.96	5.09
		278.89	13104.32	6.06
	B	125.69	7557.15	2.13
		271.94	26226.77	2.85
		381.61	21861.96	4.71
	C	225.83	17934.30	2.88
		133.89	6323.77	2.89
		209.44	21044.00	2.11
0.6 m x 0.6 m	D	386.67	6811. 11	23.27
		336.39	9184.18	12.79
	E	411.11	22715.43	7.58
		277.50	17007.64	4.60
	F	136.94	9554.55	1.19
		76.94	9632.33	3.31

The results obtained for the negative binomial k-values in all of the sampled plots are presented in Table 1. As stated earlier, the k-value describes the tendencies of a population to be clustered. The results indicate that in the 3.6 m x 3.6 m plots high k- values are obtained with higher means. A similar trend is also noticeable in the 0.6 m x 0.6m plots. However, the result show higher k-values in the smaller plots with high nematode densities than in plots with corresponding densities range in the set of 3.6 m x 3.6 m plots.

DISCUSSION

Factors that influence the horizontal distribution of plant parasitic nematodes have been outlined in literature (Barker and Nusbaum, 1971). These factors act in concert to determine the carrying capacity of the microenvironment of nematodes. In a sandy soil like the experimental site being considered, with all other factors constant, nematode densities within a given plot may determine horizontal distribution and cluster size. The skewness of the observed frequency distribution of *P. penetrans* in all the sets of samples in this study is variable. However, patterns observed as shown in Figures 2 and 3 depend on population density and plot size. Although they were all found to fit the negative binomial distribution, these figures suggest that at higher densities the distribution of *P. penetrans* tends towards normal within a given plot size. Consequently, the plot size within which nematode distribution is being considered is important. The smaller the plot size the less clustered the distribution, given a relatively high density, nevertheless at lower density nematode maintain their tendency to cluster even in the smaller plots. There is a positive correlation of the k-value on the population mean ($P < 0.05$) in the smaller plots. In the larger plots however the relationship is weak. The graph of the regression line is shown in fig.4, A and B for the 3.6 m x 3.6 m and 0.6 x 0.6 m lots respectively.

The concept of plot size has been considered indirectly in attempts to improve sampling accuracy and precision by increasing number of cores per sample in a given area (Boag *et al.,* 1987, Francl, 1986). However, bulking core samples and sub-sampling for density estimates negates the idea of accuracy and precision. Fitting a negative binomial distribution to frequency distribution of nematode sample counts is reported extensively. In this context, the k-parameter is used as an index of aggregation and small k-values indicate a clumped distribution (Bliss and Fisher, 1953). Although k-values were not related to physical cluster sizes in these and other studies, the results obtained in the present study (Table 1) indicates a relationship not only between population means and K values but more importantly, plot size and k-values. In an attempt to define cluster size in the context of horizontal distribution, there IS the need to define the physical limits and nature of spread of nematodes at various densities. This is because the spread of nematodes seems to be density dependent among other factors.

CONCLUSION

This study shows that the cluster size of *Pratylenchus penetrans* populations in terms of horizontal distribution is irregular and depends, on the plot size. The k-value of the negative binomial distribution, which is an index of cluster or aggregation, is significantly influenced by experimental plot size and depends on the population mean of the sample. Within close frequency class range the k-value tend to be higher in smaller plots. Finally, it is suggested that the k-value of the negative binomial may be more useful in the estimation of nematode population if sampling is designed to consider plot size. This will require further research on smaller plots.

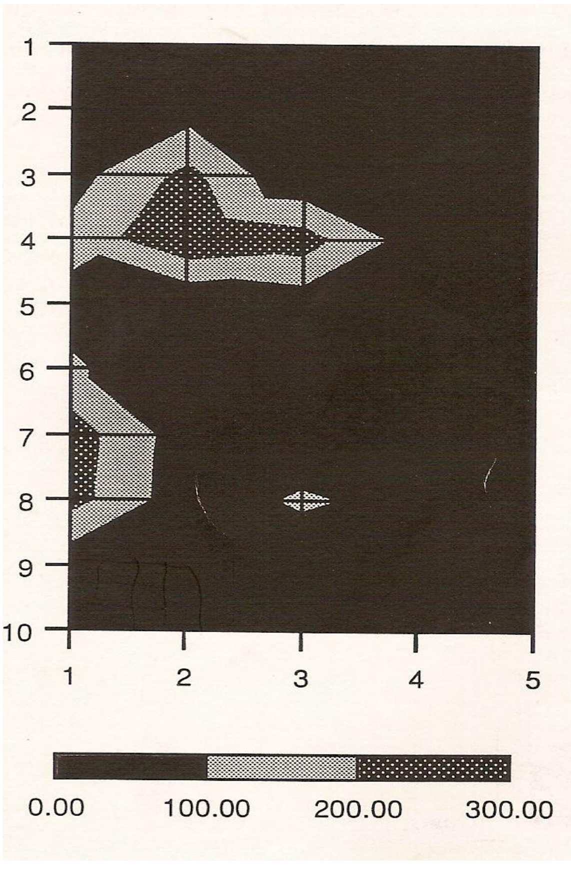

Fig.1. Contour map of the macrodistribution of *Pratylenchus penetrans* on a 21 m x 80 m plot

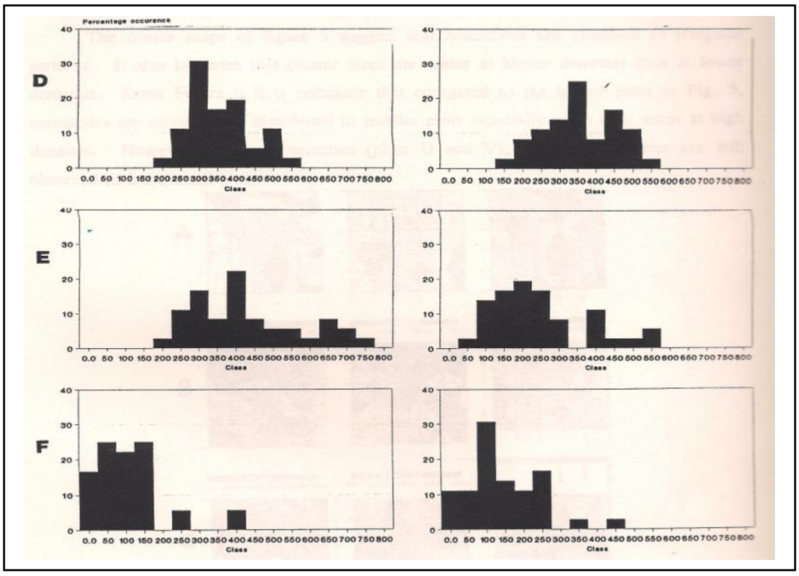

Fig. 3. Frequency distribution of nematodes in 0.6 m x 0.6 m plots. D, E and F are plots of high, medium and low population densities respectively.

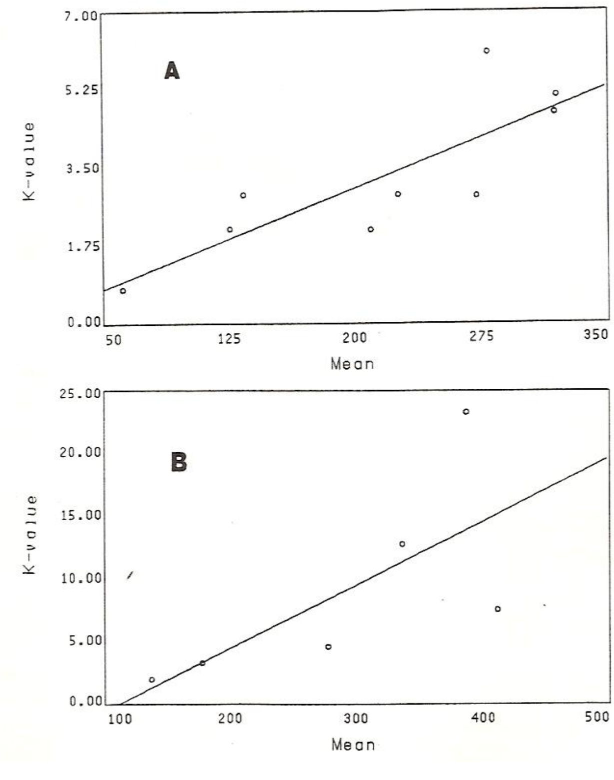

Fig. 4. Regression lines of k-value on sample mean for 3.6 m x 3.6 m (A) and 0.6 m x 0.6 m (B) plot sizes

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