

# Dispersion and Distribution of *Pratylenchus scribneri* and *Hoplolaimus galeatus* in Soybean Fields<sup>1</sup>

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**Abstract:** Examination of dispersional characteristics of *Pratylenchus scribneri* and *Hoplolaimus galeatus* indicated that there were patches within soybean fields in which both survival and reproduction were enhanced in spite of apparent homogeneity of soil type and topography. Treatment with carbofuran reduced the patchiness (or increased the dispersion) for *H. galeatus* while it had the opposite effect for *P. scribneri*. *P. scribneri* was less highly dispersed in conventional tillage plots than in the zero tillage plots. Populations from quadrats contained entirely within the patches could be described by the normal distribution (in the case of *P. scribneri*) or by the Poisson distribution (in the case of *H. galeatus*), while populations from quadrats contained entirely outside the patches could be described by the Poisson distribution for both nematodes. None of the distributions tested (Poisson, normal, negative binomial, Neyman's) gave an adequate fit when populations from both inside and outside the patches were considered together. In all instances,  $\log_{10}$  and  $\ln$  transformations reduced the goodness of fit of the data to all of the distributions tested. Even with logarithmic transformations, parametric statistics were not appropriate for analysis of data in most instances. **Key words:** nematode frequency, population ecology, index of dispersion, plot size selection, data transformation.

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The nematode pests of soybeans, *Pratylenchus scribneri* Steiner and *Hoplolaimus galeatus* (Cobb) Filipjev and Shuurmans Stekhoven, are widespread in Indiana and invariably exhibit a patchy occurrence. Other workers (1) have observed that several plant parasitic nematode species tend to occur in distributions characterized by patches of high nematode concentration surrounded by, and contained within, areas of low or moderate concentration. Goodell and Ferris (4) discussed the necessity for describing these distributions in order to determine whether the assumptions which underlie parametric statistical tests are met. Proctor and Marks (10) described methods of determining normalizing transformations for nematode count data. It would be desirable to use an analytical method which does not assume any particular type of frequency distribution to quantify patchiness in order that spatial distribution could be perceived and studied more efficiently.

## MATERIALS AND METHODS

Sites used in the tillage and chemical pesticide studies were located within a

single field in southern Indiana which contained a moderately to well drained silt loam. That area of the field used for the tillage study was planted to the soybean cultivar 'Bonus.' The area used in the chemical pesticide study was planted to the cultivar 'Cutler 71.' Carbofuran 10G was applied in a 7-in band at the rate of 33.6 g a.i./100 m of row in treated plots. For each study, plots were arranged in a randomized complete block design with six replications. One replicate was dropped from the tillage study analysis because of excessive weediness, thus reducing that study to five replications.

Sites used for determining the effect of plot (quadrat) size on estimates of dispersion and distribution of *Pratylenchus scribneri* and *Hoplolaimus galeatus* were located in a second field with a moderately to well drained silt loam and were planted to the soybean cultivar 'Wells II.' Each of these quadrat size studies consisted of nested large, intermediate, and small quadrats. A straight line running diagonally across the study area was sampled every 20 m with a 25-mm-d soil probe to ascertain approximate patch size and location. Quadrat size study areas were established so that each area would completely contain a patch of high nematode concentration and an area of low nematode concentration. Each of eight large quadrats was divided into quarters to form the intermediate size quadrats. Eight adjoining intermediate quadrats were divided into quarters to form 32 small quad-

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rats. The sizes of the large, intermediate, and small quadrats were  $50 \times 50$  m,  $25 \times 25$  m, and  $12.5 \times 12.5$  m, respectively, for the areas with *P. scribneri*, and  $100 \times 100$  m,  $50 \times 50$  m, and  $25 \times 25$  m, respectively, for the areas with *H. galeatus*.

Each soil sample consisted of twenty  $2.5 \times 20$ -cm cores taken such that ten equally spaced cores were obtained along each diagonal of an X pattern within a plot. During the growing season, samples were taken within 7.5 cm of the plants in order that the full effect of the chemical treatments could be obtained. Nematodes were extracted from a 500-cm<sup>3</sup> aliquot portion of each sample by a decanting and sieving technique, followed by suspension of the residues in Baermann funnels. The nematodes thus extracted were killed by gentle heat and fixed in 5% formalin. Two aliquot portions, each representing 1/100th of the total sample, were counted and added together. The total number of nematodes counted was thus taken to represent nematodes per 10 cc of soil.

The whole root system of each of five plants was taken on each sampling date. One plant was removed approximately half-way along each leg of the X pattern described above, and the fifth plant was taken near the intersection of the plot diagonals; i.e., the center of the X. All root systems from each plot were processed by incubating in aerated water for 2 weeks. The nematodes were removed and counted and the water replaced at the end of the first week. Nematodes were removed and counted and the water discarded at the end of the second week. The two counts were added together and divided by five to standardize to nematodes per plant.

Dispersion of *P. scribneri* and *H. galeatus* was measured using the Morisita's  $I_{\delta}$  index of dispersion (6,7,8) as follows:

$$I_{\delta} = \frac{n \sum_{i=1}^n X_i(X_i - 1)}{T(T - 1)}$$

Where  $T = \sum_{i=1}^n X_i$

and  $X_i$  is the number of individuals in the  $i$ th sample.

This index is related to Lloyd's patchi-

ness,  $\frac{*}{m}$ , named  $C$  by Pielou (9), by the following equation:

$$I_{\delta} = \frac{N}{N-1} C$$

In cases where the frequency distribution of sample counts can be described by a negative binomial distribution,  $I_{\delta}$  is related to the parameter  $k$  by the following equation:

$$I_{\delta} = \frac{1}{1-k}$$

In cases where the Poisson distribution describes the count data (8):

$$I_{\delta} = 1.0$$

Frequency distributions were tested for goodness of fit using the Kolmogorov-Smirnov test from the NPAR nonparametric statistical analysis package version 40.09 from the computer institute for Social Science Research at Michigan State University, East Lansing, Michigan. Differences in population level were tested using the median test described in Siegel (11).

## RESULTS

### *Pratylenchus scribneri*

Fluctuations in the levels of  $I_{\delta(s)}$  (based on soil samples) and  $I_{\delta(r)}$  (based on root samples) over the growing season are depicted in Figure 1 and the median values for the soil (number per 10 cc) and root (number per root system) counts in Figure 2. Counts were expressed as  $\log_{10}$  values for ease of representation. Prior to planting,  $I_{\delta(s)}$  rose slowly with time (Fig. 1), whereas the soil nematode counts decreased (Fig. 2). After planting, neither soil nematode counts nor  $I_{\delta(s)}$  appeared to follow any particular pattern in the untreated plots, although  $I_{\delta(s)}$  fluctuations tended to be in directions opposite those of the soil counts.  $I_{\delta(r)}$  in the untreated plots was generally (but not consistently) higher than the corresponding  $I_{\delta(s)}$  and followed a more easily discernable seasonal pattern characterized by a slight rise through the early growing season and a decline in the late season through plant senescence. The root counts in the untreated plots had a pattern similar to that

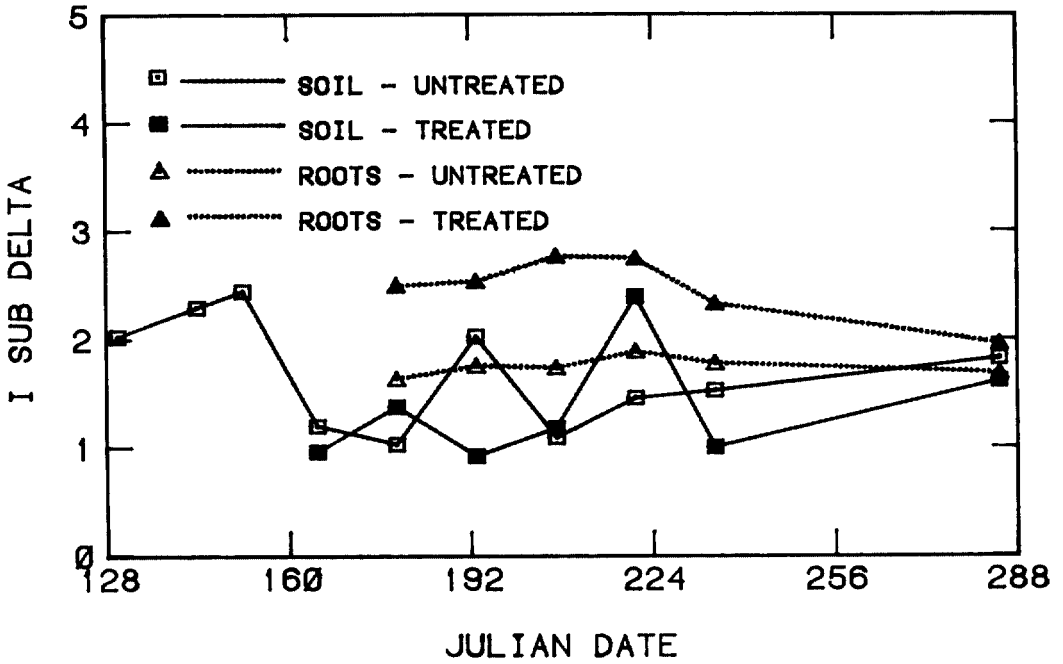


Fig. 1.  $I_6$  for *Pratylenchus scribneri* in untreated and carbofuran treated plots. Julian dates (JD) on X-axis with JD 130 = 10 May, planting date = JD 152 = 1 June, and JD 285 = 12 October. Each point represents six observations.

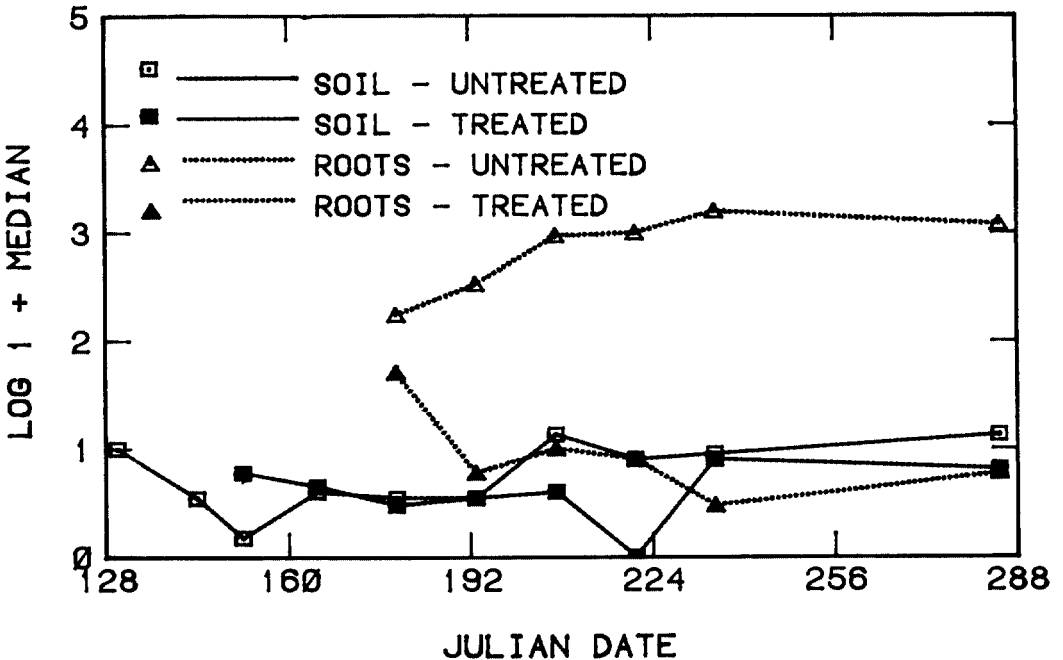


Fig. 2.  $\log_{10}$  of 1 + median number of *P. scribneri* per root system and per 10 cc soil in untreated and carbofuran treated plots. Julian dates (JD) on X-axis with JD 130 = 10 May, planting date = JD 152 = 1 June, and JD 285 = 12 October. Each point represents six observations.

of  $I_{\delta(r)}$ , but a slight decline did not occur until senescence.

**Effect of chemical treatment:** The trend for  $I_{\delta(r)}$  in the chemically treated plots paralleled that of the untreated plots, but the absolute values were greater (Fig. 1). The trend for root counts in the treated plots, however, was opposite that of the untreated plots for most of the growing season. Of interest is the rise in root counts for the treated plots on the last sampling date (Fig. 2).

**Effect of tillage treatment:** The effect of tillage treatment on  $I_{\delta(s)}$  and  $I_{\delta(r)}$  is depicted in Figure 3 and the effect on soil and root counts in Figure 4. The seasonal patterns within these plots were similar to those exhibited by the untreated plots discussed above with little difference between the conventional tillage plots and the zero tillage plots in  $I_{\delta(s)}$  (Fig. 3) or soil counts (Fig. 4).  $I_{\delta(r)}$  followed parallel patterns in the two treatments with lower absolute values for the zero tillage plots (Fig. 3). The root counts in the two treatments were roughly parallel, although the counts in the zero tillage plots were lower between 26 July (Julian date 207) and 23 August

(Julian date 235) ( $P < .10$ ) (Fig. 4). The difference was not statistically significant for 12 October (Julian date 285).

**Effect of quadrat size:** The effect of plot size selection as utilized in this study was to alter the relationship between the size of the area sampled and the patch size, thus providing plots with a wide range of population densities. The  $I_{\delta}$  values for the quadrat size study are presented in Table 1. The greatest  $I_{\delta}$  value occurs for the intermediate size quadrats and the smallest in the small quadrats. The late season sampling gave similar results, with the intermediate size quadrats retaining the highest value for  $I_{\delta}$

Table 1. Effect of quadrat size on  $I_{\delta}$  values for *P. scribneri*.

Quadrat size	Early $I_{\delta(s)}$	Late $I_{\delta(r)}$	No. of quadrats
Large (50 × 50 m)	1.45	1.26	8
Intermediate (25 × 25 m)	2.69	1.96	32
Small (12.5 × 12.5 m)	.96	1.20	32
Inside patch	1.14	1.13	32
Outside patch			

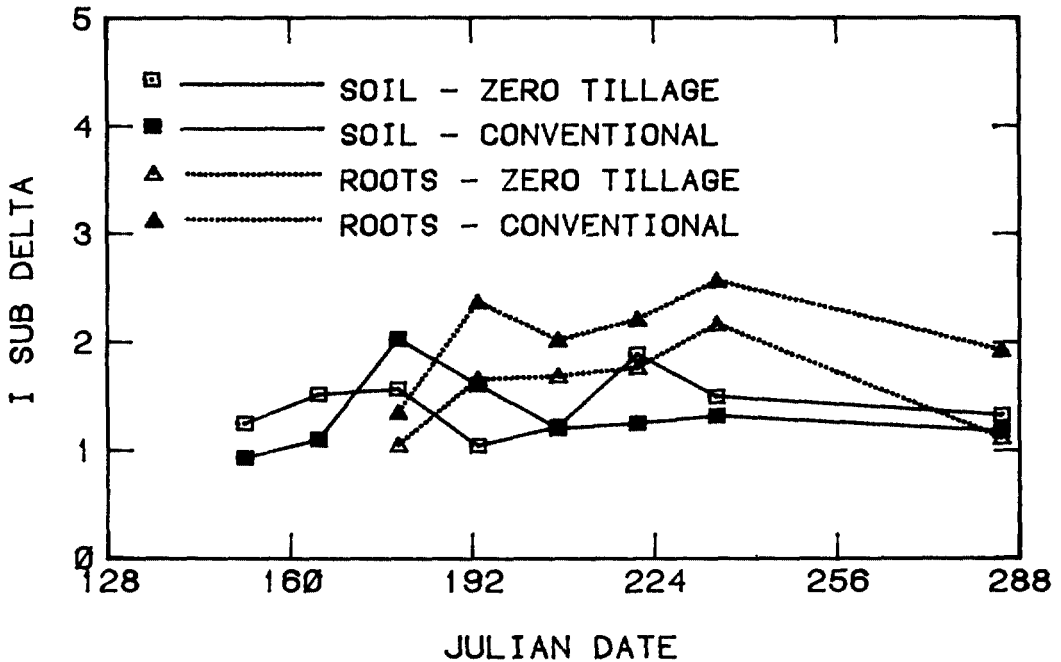


Fig. 3.  $I_{\delta}$  for *P. scribneri* in conventional and zero tillage plots. Julian dates (JD) on X-axis with JD 152 = 1 June, planting date = JD 174 = 23 June, JD 207 = 26 July, JD 235 = 23 August, and JD 285 = 12 October. Each point represents five observations.

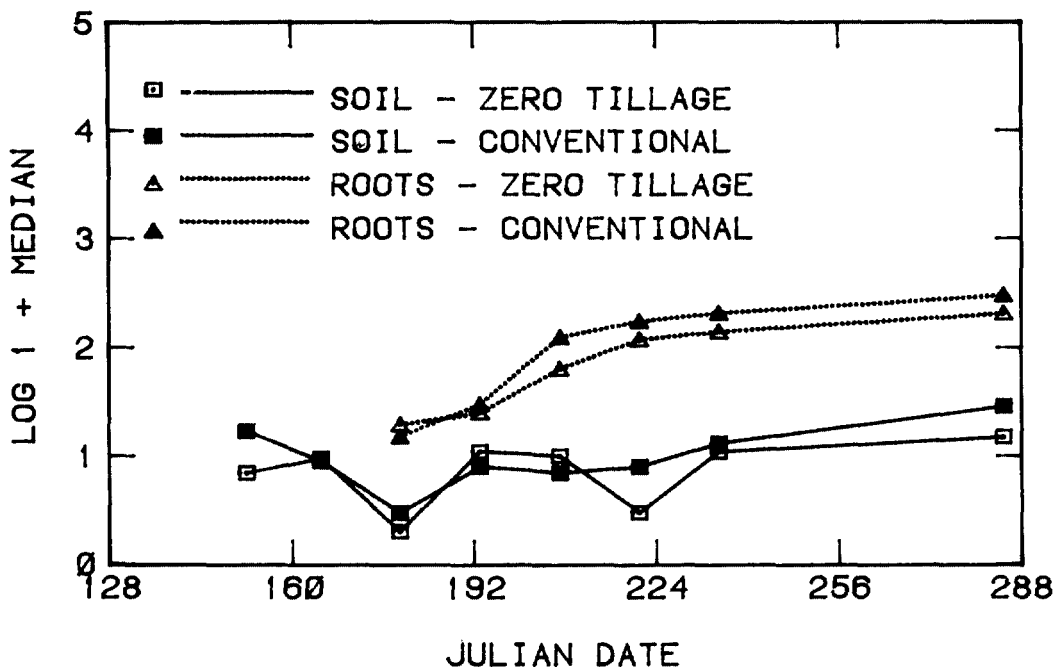


Fig. 4.  $\log_{10}$  of 1 + median number of *P. scribneri* per root system and per 10 cc soil in conventional and zero tillage plots. Julian dates (JD) on X-axis with JD 152 = 1 June, planting date = JD 174 = 23 June, JD 207 = 26 July, JD 235 = 23 August, and JD 285 = 12 October. Each point represents five observations.

and the small quadrats retaining the lowest values.

When counts of nematodes from the small quadrats which occurred outside the patches were tested against a Poisson distribution, they gave D values of 0.1496 and 0.1125 for left and right skew and a two-sided probability of fit of 0.91. The counts from the small quadrats inside the patches gave D values of 0.1859 and 0.1122 for left and right skew when compared to a normal distribution, resulting in a two-sided probability of fit of 0.72. In both instances, the  $\log_{10}$  and  $\ln$  transformation of the nematode counts greatly reduced the goodness of fit. Attempts to fit negative binomial distributions failed for both the intermediate size and small plots, even though highly efficient estimators of  $k$  were found using the maximum likelihood method of Bliss (3). A better ( $P = 0.50$ ) but still insufficient fit was obtained when a Neyman's distribution with Beall and Rescia's (2) parameter  $n$  set equal to 1 was tested against the intermediate size plots. No attempt was made to fit a frequency distribution to the largest quadrats since there were only eight of them

and it was felt that this was an inadequate sample to obtain a good fit.

#### *Hoplolaimus galeatus*

The seasonal levels of  $I_{\delta(s)}$  and  $I_{\delta(r)}$  for the carbofuran treated and untreated plots are depicted in Figure 5 and the soil counts in Figure 6.  $I_{\delta(s)}$  increased in value prior to planting (Fig. 5). In the untreated plots, a large increase in  $I_{\delta(s)}$  (Fig. 5) and a decrease in soil count (Fig. 6) occurred when the soybeans germinated. (A few *H. galeatus* were detected on this sampling date even though the median population level was 0.) This high degree of patchiness moderated as the growing season progressed.

*Effect of chemical treatment:*  $I_{\delta(r)}$  could not be calculated for the treated plots on the first sampling date on which root samples were taken because no *H. galeatus* were detected in the roots.  $I_{\delta(r)}$  values and root populations were lower in the treated plots than in the untreated plots for most of the growing season (Figs. 5, 6).

*Effect of quadrat size:* *H. galeatus* showed a pattern of  $I_{\delta}$  values similar to that for *P. scribneri* for the early sampling, with the

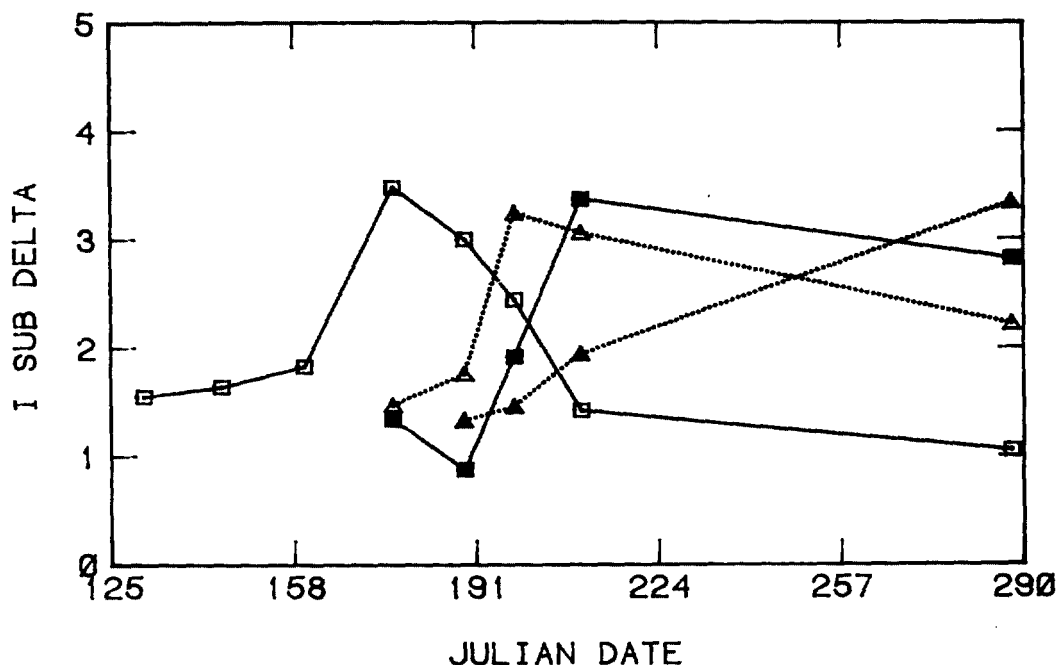


Fig. 5.  $I_8$  for *Hoplolaimus galeatus* in untreated and carbofuran treated plots. Julian dates (JD) on X-axis with JD 131 = 11 May, planting date = JD 160 = 9 June, and JD 288 = 15 October. Each point represents six observations. For legend, see Figure 1.

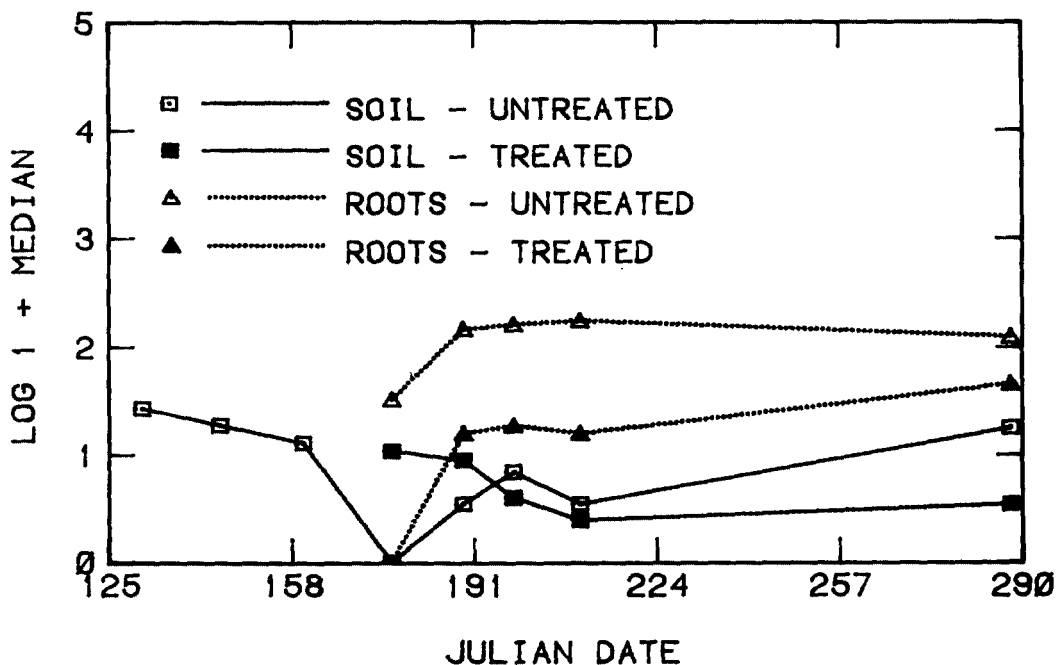


Fig. 6.  $\text{Log}_{10}$  of 1 + median number of *H. galeatus* per root system and per 10 cc soil in untreated and carbofuran treated plots. Julian dates (JD) on X-axis with JD 131 = 11 May, planting date = JD 160 = 9 June, and JD 288 = 15 October. Each point represents six observations.

intermediate size quadrats having the greatest  $I_{\delta}$  values (Table 2). In the later (early bloom) sampling, however, the  $I_{\delta}$  value for the large quadrats was greatest. The  $I_{\delta}$  values for the small quadrats remained small relative to those for the larger quadrats.

An excellent fit (D left = 0.0637, D right = 0.0925, two-sided probability 0.99) was obtained when the nematode counts in the small quadrats established inside a patch were compared to a Poisson distribution. The small quadrats outside the patches also produced a good fit (D right = 0.1151, D left = 0.0993, two-sided probability 0.97) when compared to a Poisson distribution. No fit could be obtained to the negative binomial distribution or to any of Neyman's distributions. As with *P. scribneri*, no attempt was made to fit any distribution to the nematode count data from the largest quadrats.

### DISCUSSION

$I_{\delta}$  was formulated to behave similarly for samples taken from any type of underlying frequency distribution.  $I_{\delta}$  increases in value as populations become less highly dispersed and more patchy or clumped. Values of  $I_{\delta}$  for a plot will not change as long as areas of high and low population levels within the plot retain the same relative numbers of organisms, even though the absolute numbers may change. By contrast, the parameter  $k$  decreases in value as populations become less highly dispersed and may be altered if the absolute densities change, unless the population can be described by a negative binomial distribution (9).

It must be emphasized that all count data in this study were means computed

Table 2. Effect of quadrat size on  $I_{\delta}$  values for *H. galeatus*.

Quadrat size	Early $I_{\delta(s)}$	Late $I_{\delta(r)}$	No. of quadrats
Large (100 × 100 m)	1.31	1.67	8
Intermediate (50 × 50 m)	1.85	1.53	32
Small (25 × 25 m)			
Inside patch	1.03	1.01	32
Outside patch	1.07	.94	32

from the entire sample and would thus result in lower values of  $I_{\delta}$  than if individual root systems or individual cores of soil were processed and counted separately. Furthermore, as the number of samples (number of root systems or soil cores) was increased, the values of  $I_{\delta}$  would decrease because the influence of individual subsamples on the entire sample mean would be decreased. Complete enumeration of the organisms within each plot would theoretically provide the lowest values of  $I_{\delta}$  (8).

The preplanting rise in  $I_{\delta(s)}$  for both *P. scribneri* (Fig. 1) and *H. galeatus* (Fig. 5), coupled with the reduction in the soil counts (Fig. 2 and Fig. 6, respectively), indicates a disparity in the mortality levels between the areas of high and low concentration. Assuming that the populations were not increasing prior to planting, it would appear that survival was enhanced inside the patches, producing a greater difference in the population levels between the areas of high concentration and the areas of low concentration and a greater value of  $I_{\delta(s)}$ . The rise in  $I_{\delta(r)}$  in check plots, concurrent with the rise in root counts during the early to mid growing season, indicates that reproduction was also enhanced inside the patches. It is likely that the disparity between the areas of high and low populations was increased not by depression of the low population areas but by elevation within the high population areas.

After the soybeans have germinated,  $I_{\delta(r)}$  is probably a more consistent indicator of dispersion than  $I_{\delta(s)}$  since the root counts appear to be more reflective of differences in the field population. When the individual plots were ranked according to soil and root counts and the rankings were compared among sampling dates, the plots tended to maintain their positions relative to each other when root rankings were used but not when soil rankings were used.

$I_{\delta(r)}$  acted in one way for *H. galeatus* and in another for *P. scribneri* (as compared to checks) following treatment with carbafuran. The values for  $I_{\delta(r)}$  became higher for *P. scribneri*, owing to reduction of the nematodes to below detectable levels in some plots. Control was not so complete for *H. galeatus*, and nematodes did not fall below detectable levels within the roots

taken from any plot (after the first sampling date). Thus, even though fewer nematodes were present in the roots in both instances, *H. galeatus* became more highly dispersed whereas *P. scribneri* became less highly dispersed (or more clumped). The rise in the root counts of the treated plots at the last sampling date is due to the decrease in effectiveness of the pesticide and the late senescence for the carbofuran treated plots.

*P. scribneri* in the conventional tillage plots had higher values of  $I_{\delta(r)}$  than in the zero tillage plots (Fig. 3), probably as a result of greater plant vigor in the former. The root systems were larger and more robust in the conventional tillage plots, and thus the nematodes could achieve greater population densities within the patches, which resulted in a more dramatic expression of patchiness.

The  $I_{\delta}$  values for the large *P. scribneri* plots were smaller than those for the intermediate size plots because the plots were large relative to the patch size (Table 1). This resulted in a dilution effect wherein some of the soil cores comprising a sample were taken outside the patch and some inside the patch, thus reducing the variability among samples. The intermediate size quadrats were small relative to the patch size; some of them could be contained largely or entirely within a patch, whereas others could be entirely outside the patch thus increasing the variability among plots. The small quadrats had small values of  $I_{\delta}$  since they were established in two sets, either entirely inside or entirely outside the patch.

For *H. galeatus*, the  $I_{\delta}$  values for the different quadrat sizes did not retain their relative positions between the first and second sampling dates (Table 2). The small quadrats remained the most highly dispersed, but on the later date the intermediate size quadrats had an  $I_{\delta}$  value relatively smaller than that of the large quadrats. This may have resulted from an increase in patch size for *H. galeatus* during the growing season.

The distinct nature of the patches resulted in a bi-modality which made fitting of frequency distributions very difficult when all plots were considered together. Barker and Campbell (1) and Goodell and Ferris (4) noted that several nematode pop-

ulations could be described by the negative binomial distribution. However, this study shows that this type of distribution cannot be assumed for all nematode species in all field situations. As previously noted, a Neyman's distribution gave a better fit than the negative binomial distribution when all of the intermediate size *P. scribneri* plots were considered together. This was primarily due to the bi-modal tendency of the Neyman's distribution when  $n = 1$ , using Beall and Rescia's (2) modification of Neyman's distribution.

The good fit of the counts of *P. scribneri* in quadrat samples located inside the patches to the normal distribution indicates that parametric statistics may sometimes be used in population quantification, provided the fit is affirmed by some means such as the Kolmogorov-Smirnov test used here or the Chi-square test. However, the good fit of the aforementioned *P. scribneri* quadrat data to the normal distribution does not necessarily imply that parametric statistics might be useful in research plots established in such an area. As the size of the area sampled was altered, the form of the frequency distribution changed. Furthermore, the chemical treatments greatly increased the  $I_{\delta}$  value for *P. scribneri*, indicating that there was a shift toward a more contagious distribution within the carbofuran treated plots. Therefore, if parametric statistical tests were performed in this instance, the result might be comparison of a normally distributed population (the untreated plots) to a non-normally distributed population (the chemically treated plots). This also has implications concerning transformation of data. If the treatments had different distributions, a given transformation might make populations from one of the treatments behave like a normally distributed population, but not those from the other treatment.

Morisita (7) noted that sampling was more reliable when the  $I_{\delta}$  value could be minimized. Size selection and location of experimental plots could be quite important to efficient sampling. Greig-Smith (5) noted that mean squares of numbers of individuals per unit area could be altered by changes in plot size if those individuals had a patchy distribution. In applying this principle, consideration should be given to



timing of sampling, since changes in dispersion could alter interpretation of field events. Time of sampling probably should not be altered, since this might reduce representativeness, but a greater number of subsamples may be necessary (to increase reliability) at times when a large  $I_{\delta}$  value is anticipated.

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