Analysis of Spatial Patterns of Virus-Diseased Tobacco Plants

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ABSTRACT

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Epidemics caused by tobacco etch virus (TEV) and tobacco vein mottling virus (TVMV) were monitored in six experimental fields of tobacco in Kentucky from 1983 to 1985. Aggregation of virus-diseased plants was determined by dividing fields into contiguous quadrats and using point pattern (e.g., variance-to-mean ratio and Lloyd's patchiness [m^*/m]) and spatial autocorrelation analyses. Spatial distribution of diseased plants was neither solely clustered nor random, but changed with time during the epidemics. All indices of aggregation indicated a random pattern at the beginning of the epidemics if the first disease assessment was early enough. Patchiness increased to a maximum ($m^*/m > 2$) and then declined throughout the remainder of the epidemics. In many fields, patchiness also indicated randomness ($\sim$ 1) by the last assessment time. First-order autocorrelations (r_w, r_a) increased throughout most epidemics, eventually indicating clustering (> 0.23). Autocorrelations often exceeded 0.5 at their maxima. When mean disease density approached 100% incidence, autocorrelations declined at the end of the epidemics. Spatial correlograms suggested a first-order autoregressive process. Iwao's regression of mean crowding (sensu Lloyd) on mean virus disease density indicated a random pattern of clusters (slope $\cong$ 1). The sequence of diseased plants per row also was found to change with time based on separate ordinary runs analyses. Percentage of tobacco rows with a clustered pattern increased during most of the epidemics from a low initial level ($< 10\%$); the percentage declined as disease incidence approached its maximum. In agreement with spatial autocorrelation analysis, maximum percentage of rows with a clustered pattern was reached before the last assessment date if disease density was high.

Additional key words: dispersion, *Nicotiana tabacum*, potyviruses, quantitative epidemiology.

Taylor (21) stated that "spatial distribution is one of the most characteristic ecological properties of species." It is now broadly accepted by epidemiologists that the spatial component of plant disease epidemics is as important as the temporal component (2,10). The importance of describing the spatial distribution of virus-diseased plants has been recognized for over 40 yr (1,25). Since 1946, most investigators have quantified virus patterns by assessing the sequence of diseased and healthy plants in each row using a form of doublet or runs analysis (10–13,25). These analyses,

although very easy to perform, fail to account for the two-dimensional aspect of aggregation within a field and also may produce ambiguous results when some rows exhibit a random sequence and others a clustered sequence.

Gray et al (6,7) developed and successfully used a method to quantify two-dimensional patterns of virus-diseased plants based on distances between infected plants. Other more general techniques also are available if a field is divided into quadrats and the numbers of virus-diseased plants per quadrat are counted (2,3,10,24). The first of these methods is point pattern analysis, which entails determining the mean, variance, and sometimes the frequency distribution of diseased plants or other counted variables. Used frequently for assessing the aggregation of fungal pathogens and soilborne inoculum (2), the approach has also been used with plant viruses (11,13). An alternative method is spatial autocorrelation analysis, which entails correlating disease level in each quadrat with disease in the proximal quadrats. Several autocorrelation test statistics can be calculated to measure aggregation. This approach was only recently introduced to plant pathology (16,19) and had not been used to analyze the pattern of virus-diseased plants before the present study. The advantages and disadvantages of assessing aggregation using point patterns and autocorrelation have been discussed (3,10,16).

Epidemics caused by tobacco etch virus (TEV) and tobacco vein mottling virus (TVMV) are common whenever burley tobacco (*Nicotiana tabacum* L.) is grown (5,18). Both of these potyviruses are transmitted nonpersistently by several aphid species, only some of which colonize tobacco. It was shown that disease incidence due to both viruses increased logistically in susceptible tobacco cultivars grown in Kentucky (14) and in some years reached 100%. The objectives of this study were to characterize the aggregation of virus-diseased tobacco plants and determine how aggregation changes with time during virus disease epidemics in Kentucky over 3 yr.

MATERIALS AND METHODS

Field data collection. A detailed description of the field data collection was given in Madden et al (14), and only a summary is presented here. Six field sites were selected on the University of Kentucky "South Farm" near Lexington. Three pairs of plots were established; one field of each pair was treated with insecticides (disulfoton and acephate 75% EC) to suppress colonization by aphids. The suffix -I was used to label the insecticide-treated field; -N was used for the fields not treated. The greatest distance between any two plots was about 825 m and the least about 230 m. Tobacco cultivar Burley 21 was used in 1983 and cultivar Kentucky 14 in 1984 and 1985. The first two field pairs (A and B) had 22 rows of 150 plants each, whereas the C field pairs had 50 rows of 60 plants each. Distance between rows was 102 cm and plant spacing was 46 cm, except in 1985 when it was 41 cm, due to an error in calibration of the transplanter. Dates of transplanting were 8–13 June 1983; 7–8 June 1984; and 23–28 May 1985. Maps of each field were constructed to mark the location of each plant and that of missing plants.

The A fields were monitored for virus-infected plants approximately three times a week and the others at least once a week. Plants were marked when symptoms first appeared, and the infecting virus was recorded. For the first several weeks of each season, the accuracy of identification was checked by assay of random samples with antisera specific to TEV and TVMV. For the balance of the season only visual assessment was used. Symptoms only could be observed for the first infection if plants were infected by both viruses. For simplicity and to avoid the confounding of double infection, most of the analyses and descriptions of virus-disease patterns are presented for the combined-virus data.

Data analyses. The spatial pattern of virus-diseased plants was assessed using point pattern and spatial autocorrelation methods. The A and B fields were divided into 15 × 5 quadrats, each consisting of four rows and 10 plants per row. The two outer rows were not used. The C fields were divided into 10 × 5 quadrats, each containing 10 rows with six plants per row. Before choosing this partitioning of the fields, a modified Greig-Smith procedure (8) was used to determine the quadrat size at which maximum variance occurred. This quadrat size represents the approximate size of the clusters of diseased plants. Maximum variance occurred at the above listed quadrat sizes in $> 80\%$ of the fields and times (L. V. Madden, *unpublished*). Therefore, all reported results are for fixed quadrat sizes. Because of a large number of missing plants, the B fields in 1983 were not analyzed.

The mean (m) and variance (v) of virus-diseased plants per quadrat were calculated for each assessment time. With these statistics, the following indices of aggregation were calculated: variance-to-mean ratio ($VTM = v/m$); Lloyd's mean crowding ($m^* = m + [VTM - 1]$); and Lloyd's patchiness (m^*/m) (24). For the number of quadrats in this study, m^*/m is equivalent to Morisita's index of aggregation (22). Two regressions were performed for each field: (*i*) $\ln(v)$ on $\ln(m)$; and (*ii*) m^* on m. For both regressions, the dependent and independent variable came from the separate assessment times. Regression *i* corresponded to Taylor's (20) power relation; if the log-transformed relation is linear, then the slope is an index of aggregation. Regression *ii*, attributable to Iwao (9), can also be used to quantify aggregation; if linear, the slope can indicate the pattern of clusters.

Autocorrelations within (r_w) and across (r_a) "rows" were determined, as well as the combined "rook's" case autocorrelations (r), at each assessment time using the methods and program of Modjeska and Rawlings (15). Autocorrelations were determined for adjacent quadrats (i.e., first-order lag), quadrats separated by one quadrat (second-order lag), and so on, to calculate spatial correlograms. "Row" corresponds to the longer dimension of the field. For first-order autocorrelations, r_w is calculated by relating each $[i, j]$ quadrat with the $[i - 1, j]$ and $[i + 1, j]$ quadrat; r_a is calculated by relating $[i, j]$ with $[i, j - 1]$ and $[i, j + 1]$.

The average quadrat location of virus-diseased plants, i.e., "center of mass," was determined for each time. For this calculation, each quadrat was represented by its ordered position from an arbitrary corner of the field. The A and B fields had quadrats ranging from [1, 1] to [15, 5]; the quadrats of the C fields ranged from [1, 1] to [10, 5]. The frequency of diseased plants per quadrat was then used to calculate the average quadrat location, its variance, and coefficient of determination. The data can be considered a sample of quadrat locations with the frequency of each location given by the number of diseased plants at the location (quadrat). If virus diseased plants were mainly located in one particular area, the average location would be substantially different from the field center ([8, 3] for A and B fields; [5.5, 3] for C) or the location variance would be small relative to the average.

Patterns of diseased plants within rows were assessed with ordinary runs analysis (12,13). The number of runs of diseased and healthy plants was determined for each row separately, and significant departure from a random sequence was appraised with a standard-normal test (11,12).

RESULTS

Spatial patterns. Disease incidence varied considerably among years and locations, being highest in 1984 and lowest in 1983 (Table 1). Effect of insecticide treatment on mean disease density also varied with year and location (cf. 14). In general, the average location ("center-of-mass") of diseased plants was within one quadrat distance of the field center. Results only are given for the final assessment and the time at which density was 50% of its observed maximum (Table 1). Mean location was substantially different from field center only for A-N in 1983 and B-N in 1984. However, coefficients of variation (CV) for mean location were greater than 40% for all fields and times when disease incidence was greater than zero. In fact, three-quarters of the CVs were greater than 50%. Thus, there was little evidence that occurrence of virus diseased plants was associated with particular areas of the fields.

Spatial patterns varied over time in all fields (Figs. 1–3). Depending on the assessment time, either randomness or clustering was indicated. Despite the variability, a general process can be described with only minor exceptions. At the earliest times

of the epidemics, patchiness and spatial autocorrelations indicated randomness (e.g., A-I and A-N in all years). With fewer assessments and a higher mean disease density at the first observation, it was possible that we missed this early characteristic in some cases (e.g., B-N in 1984 and 1985 and C-I in 1984).

Patchiness increased to a maximum and then declined for the remainder of the epidemics. This maximum usually occurred fairly early during the epidemics, whether disease level was high (A-I and A-N in 1984 and 1985) or low (A-I and A-N in 1983). Maximum patchiness, therefore, was not strictly dependent on mean disease density. In most fields, patchiness declined to a value near the expected value for a random point pattern ($m^*/m = 1$). In 1983, final patchiness was greater than one ($P < 0.05$), but mean disease density was very low (Table 1).

Concurrent with the change in patchiness, first-order autocorrelations (r_w and r_a) increased during the epidemics. Eventually, values significantly different from zero ($P < 0.05$) were obtained (e.g., A-N and A-I in 1985, C-I in 1984, and C-N in 1983). Unlike patchiness, r_w and r_a often increased throughout the epidemics with no clear maximum (e.g., A-I and C-N in 1983; A-N, B-I, and C-I in 1985). When mean disease density was very high and approached 100% of the plants in a quadrat, autocorrelation coefficients did reach a maximum and then declined. This was especially true in 1984. The maximum autocorrelation coefficients, however, declined *after* the time of maximum patchiness (Fig. 2).

Autocorrelations never were less than −0.2 and were greater than 0.8 in only one field (B-N in 1984). In most fields, r_w and r_a were similar. However, there were a few fields in which the two coefficients substantially differed over time (e.g., C-I in 1983 and C-N in 1984). When there was a substantial difference in the magnitude of the two coefficients, r_w was larger (e.g., B-I and C-I in 1984). In these fields, there was a greater similarity in virus-disease level within than across quadrat rows.

Spatial correlograms exhibited a decline in autocorrelation with lag order at most times. The autocorrelations for the first-, second-, and third-order lags (rook's case) corresponding to the median disease levels are given in Table 1. There was only a slight increase from the first- to second-order lag for two fields. The correlograms were representative of a first-order autoregressive process that can conveniently be summarized with the first-order coefficients (3).

Insecticide treatment had no apparent or consistent effect on aggregation. Any effect was indirect, through the influence of insecticide on mean density and rate of increase, which then influenced aggregation (cf. 14).

The relationship between $\ln(v)$ and $\ln(m)$ was not linear for most fields. When mean disease density was very high, as in 1984, $\ln(v)$ usually reached a maximum and then decreased with increasing ln(m) (e.g., Fig. 4). One field (C-N in 1984) displayed no significant relationship between $\ln(v)$ and $\ln(m)$ (Table 2). There was, however, a linear relationship between m^* and m for all fields (Table 2), even when m approached a maximum (Fig. 4). All coefficients of determination were greater than 0.9. The slope of this regression represents the patchiness of the clusters under certain theoretical conditions (9). The many estimated slopes near 1 suggest that, although diseased plants often were aggregated, clusters of diseased plants were random. Fields in 1983 all had slopes greater than 1, indicating aggregation of the clusters.

Percentage of tobacco rows with a clustered sequence of virus-diseased plants based on ordinary runs analysis varied with time (Fig. 5). In 1983, there was a general increase throughout the epidemics. During 1984 and 1985, in which disease densities were high, there was an increase to a maximum and then a decrease. The decrease was greater in 1984 than 1985. The maximum percentages were always reached later than the maximum patchiness (cf. Figs. 1–3). If autocorrelations reached a maximum, the time was about the same or later than the time of maximum percentage of clustered rows.

As with the combined virus-disease data, there was considerable variation in the patterns of TEV- and TVMV-diseased plants (data not shown). In general, the sequence described for the change in pattern over time with combined data also was found for the individual viruses. There was, however, no reduction in r_w and r_a at the end of the 1984 epidemics with the individual viruses. This likely was because neither virus approached 100% disease incidence.

Example. Maps of field A-I in 1984 are presented in Figure 6 to display some of the typical patterns and change in pattern over

TABLE 1. Final number of virus-diseased plants per quadrat (density), mean location ("center-of-mass") of diseased plants (in units of quadrats) at the final assessment time and when mean density was approximately 50% of the final disease density, and first-(r_1), second-(r_2), and third-(r_3) order spatial autocorrelations at the time of approximate median virus disease density for tobacco fields grown in Kentucky over 3 yr

	Final disease		Median disease			
				Autocorrelations[d]		
Field[a]	Density[b]	Center-of-mass[c]	Center-of-mass[c]	r_1	r_2	r_3
1985						
A-I	25.2	7.8, 3.0	7.5, 3.2	0.43	0.12	−0.14
A-N	26.8	8.5, 3.1	8.6, 3.0	0.36	0.14	−0.06
B-I	11.6	8.7, 2.8	8.9, 2.8	0.20	0.04	0.03
B-N	26.5	8.3, 2.8	8.9, 2.7	0.42	0.30	0.17
C-I	16.4	5.5, 2.7	5.0, 2.7	0.25	0.05	−0.01
C-N	50.8	5.3, 2.9	4.9, 2.9	0.58	0.56	0.43
1984						
A-I	34.6	8.1, 3.0	8.1, 2.8	0.30	0.20	0.13
A-N	35.7	8.0, 3.1	8.0, 2.9	0.30	0.02	−0.06
B-I	29.0	8.0, 3.0	7.9, 3.0	0.29	−0.04	−0.05
B-N	32.4	8.1, 2.9	10.1, 2.9	0.87	0.80	0.69
C-I	44.6	5.4, 3.0	4.8, 2.7	0.38	0.45	0.40
C-N	53.4	5.5, 3.0	5.2, 3.3	0.47	0.27	0.25
1983						
A-I	2.0	7.4, 3.4	6.9, 3.3	0.06	0.12	−0.07
A-N	4.5	5.9, 2.7	5.1, 2.7	0.57	0.33	0.33
C-I	7.6	4.9, 2.6	4.7, 2.7	0.29	0.17	0.13
C-N	14.5	4.9, 2.5	4.8, 2.6	0.38	0.30	0.34

[a] Fields labeled with "I" were treated with insecticides and those with "N" were not.
[b] Theoretically, maximum number of virus-diseased plants per quadrat is 40 for the A and B fields and 60 for the C fields. Because of missing plants, actual maximums are less.
[c] Exact center is [8, 3] for the A and B fields and [5.5, 3] for the C fields. All coefficients of variation were greater than 40%.
[d] Rook's case autocorrelation coefficients; values greater than 0.23 are significantly different from 0 ($P = 0.05$).

time. Spatial statistics for the maps are given in Table 3. Numbers of virus-diseased plants per quadrat are grouped for presentation, not for the analyses.

There were few diseased plants on the first date shown (180), and these were randomly located. Only three of the quadrats had plants with virus symptoms. All aggregation indices indicated randomness. Four days later (184), 17 quadrats had diseased plants. Some newly "occupied" quadrats were near the original quadrats, but others were well separated. By this time, clustering was indicated based on point patterns (VTM or m^*/m).

One week later (191), mean density increased fivefold, and more than 70% of the quadrats had one or more diseased plants. By this date, r_w and r_a were significantly greater than 0 ($P < 0.05$), and VTM (or m^*/m) also indicated clustering. Two weeks later (205), all quadrats were occupied and one quadrat had more than 20 diseased plants. Quadrats with relatively high levels of disease were never surrounded by quadrats with the lowest levels of disease. Clusters (foci) could be be visualized at this time. Clustering was indicated by all indices.

By 10 August (223), VTM and m^*/m indicated random point patterns. However, spatial autocorrelations still were significantly greater than 0. Three days later (226), at relatively high mean density, all indices of aggregation indicated randomness. The grouping of plants for presentation purposes (Fig. 6) only gives the impression of clustering at this final time.

DISCUSSION

The pattern of virus-diseased tobacco plants was neither solely clustered nor random. Aggregation depended on time and mean disease density, the latter being partially determined by the growing season. Combined results for point patterns and autocorrelation analysis can be used to hypothesize the spatial process of tobacco virus epidemics (3, page 92). Early in an epidemic at low mean density, all aggregation indices indicated randomness. According to Cliff and Ord (3), this suggests a true (simple) Poisson process. The early random distribution also implied no major (nearby) local source for the viruses. Rather,

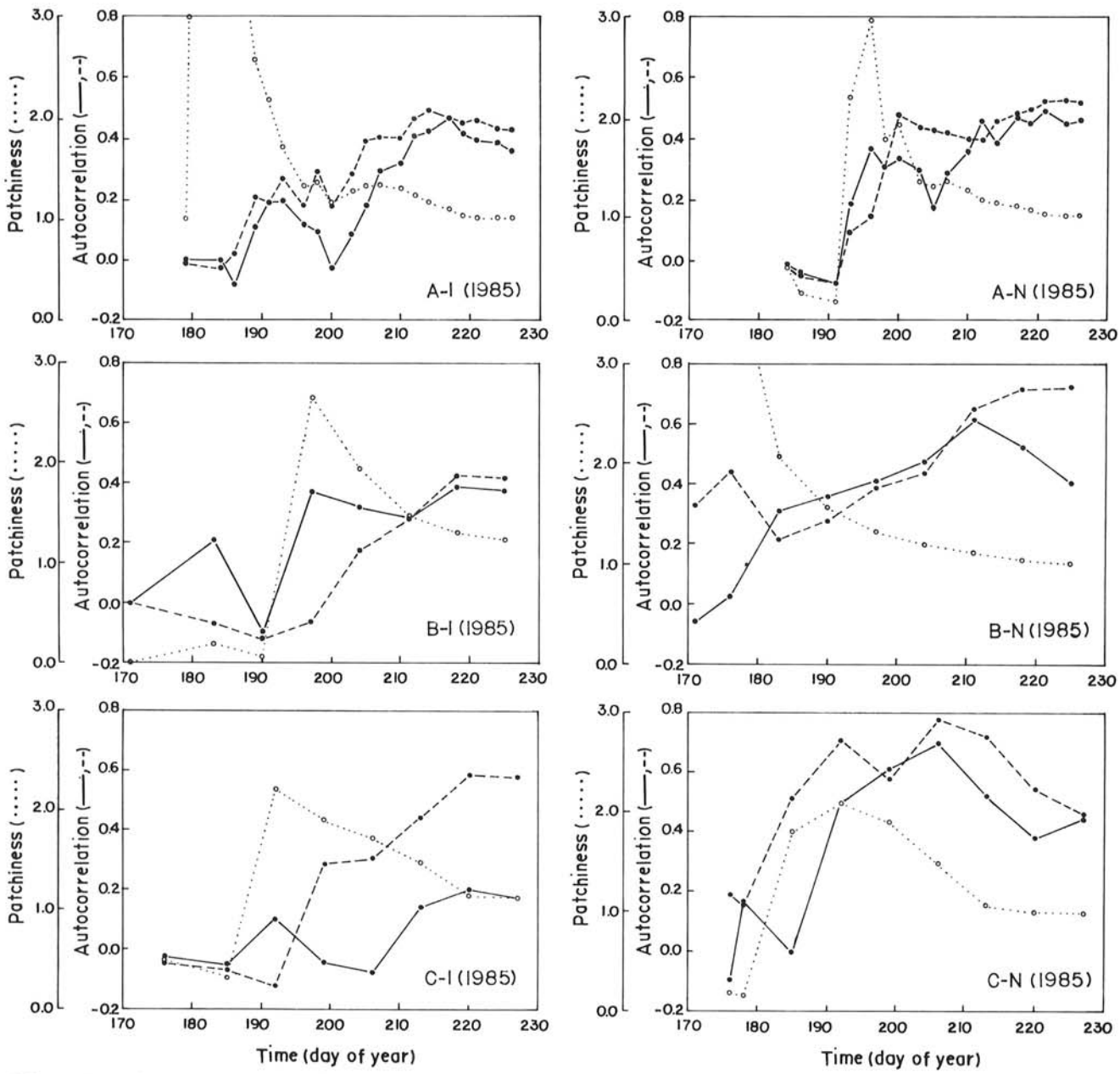

Fig. 1. First-order spatial autocorrelation within (r_w; dashed line) and across (r_a; solid line) rows, and patchiness (m^*/m; dotted line) for each assessment date during epidemics of tobacco viruses in Kentucky in 1985. Fields labeled -I were treated with insecticides and those labled -N were not. To use a consistent scale, some patchiness values are off the scale (A-I and B-N).

multiple sources at many distances were likely.

After this random beginning, point patterns (e.g., m^*/m) indicated clustering that could be due to a generalized- or compound-Poisson process as represented by the negative binomial or Neyman A distribution. However, spatial autocorrelations indicated randomness at this stage of the epidemics (e.g., see A-I for 1985 in Fig. 1). Such a combination indicated that clusters were small and seldom overlapped quadrat boundaries (3). If one assumed that small clusters were attributable to true contagion (= generalized Poission), then our data suggested spread of the tobacco viruses in the immediate vicinity of the originally infected plants.

A third stage in the pattern of virus-diseased plants was usually discernible. Both patchiness (or VTM) and the spatial autocorrelation coefficients indicated clustering. This combination suggested that clusters were relatively large and covered areas (much) greater than quadrat sizes. One could assume that large clusters are due to apparent contagion (= compound Poisson process) in which the expected number of diseased plants per quadrat (μ) is not constant, but varies slowly over space (μ_{ij}). Then we would conclude that, at this stage of the epidemics, aggregation was attributed to apparent contagion. However, the spatially varying μ_{ij} would not necessarily be due to soil or environmental conditions. Extensive spread of the virus diseases from the earlier diseased plants, and from clusters of diseased plants, would lead to relatively large, overlapping, clusters. Expected number of diseased plants could then vary with distance from the edge or center of a cluster.

A fourth stage in the changing pattern of virus-diseased plants occurred when point patterns were random, but spatial autocorrelation was significant and positive. Cliff and Ord (3) labeled this combination as questionable apparent contagion but gave no explanation. Disease values in proximal quadrats were similar, and the variance was close to the mean. Fields were being "saturated" with diseased plants in many cases, but the trend toward lower point pattern aggregation was also seen for the low density year of 1983. Disease progress curves for 1983 showed that virus-disease incidence approached a low maximum at a rate similar to the other years in which the maximum was much higher, rather than approaching a higher maximum at a slow rate (14).

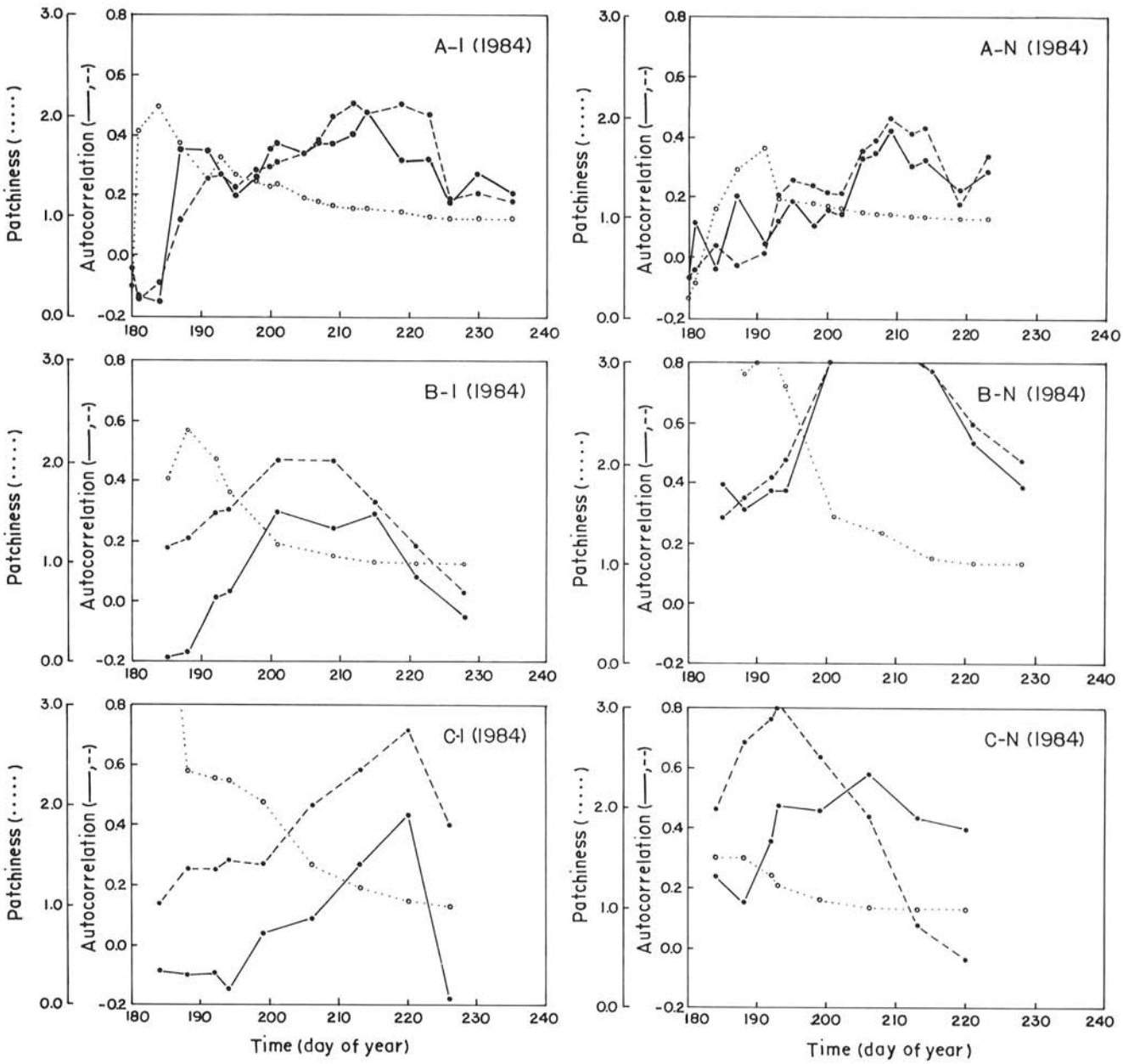

Fig. 2. First-order spatial autocorrelation within (r_w; dashed line) and across (r_a; solid line) rows, and patchiness (m^*/m; dotted line) for each assessment date during epidemics of tobacco viruses in Kentucky in 1984. Fields labeled -I were treated with insecticides and those labeled -N were not. To use a consistent scale, some patchiness (B-N and C-I) and autocorrelation (B-N) values are off the scale.

Variability in diseased plants among quadrats thus decreased as incidence approached a maximum, even when the maximum was much less than the total number of plants per quadrat.

An additional stage was clearly seen only in 1984 when mean disease density was very close to the total number of plants per quadrat. For practical purposes, the analyses were assessing the patterns of the few remaining disease-free plants, which often were random in the fields.

Aggregation results were not due to the spread of the viruses from a single dominate source outside the fields. Such a source would produce a gradient of diseased plants within the fields and result in a virus "center-of-mass" very different from the field

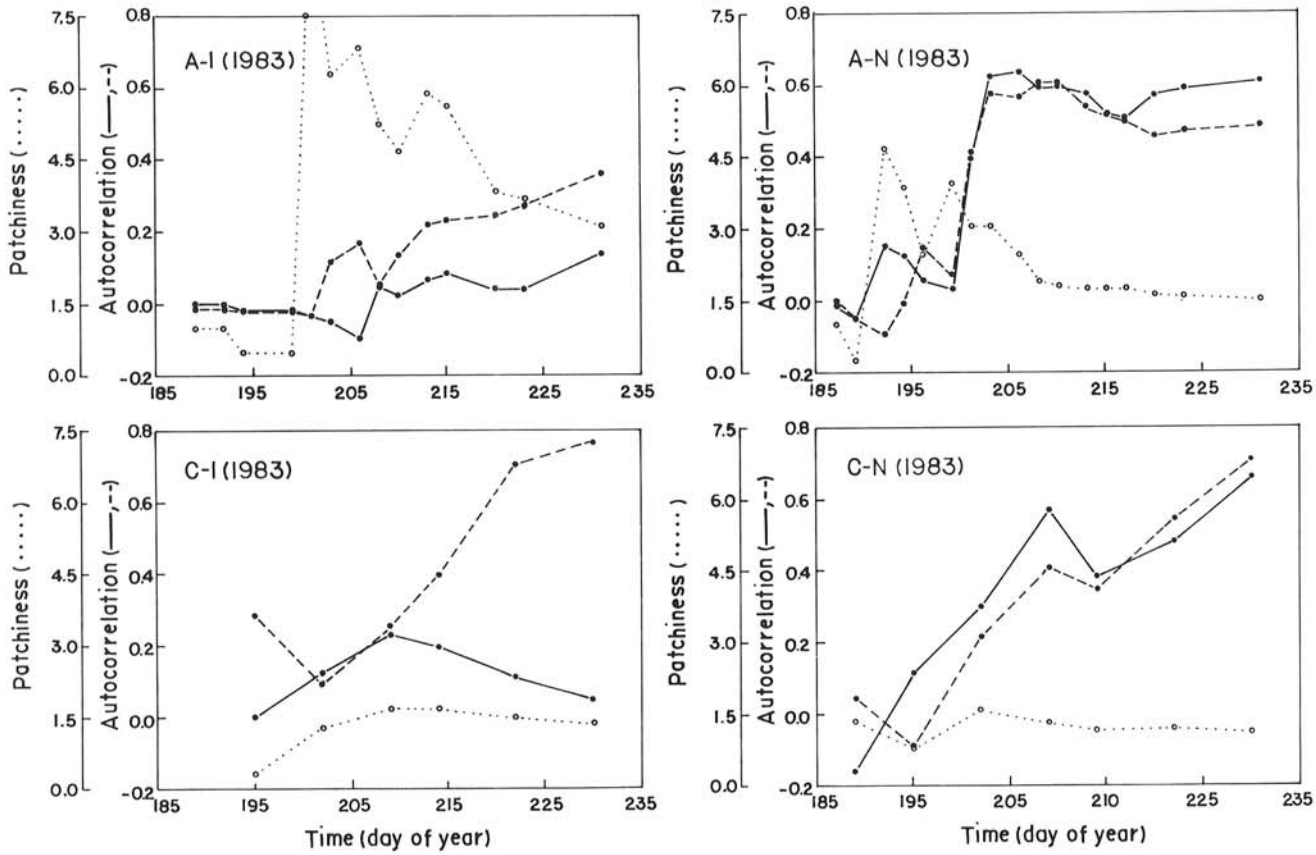

Fig. 3. First-order spatial autocorrelation within (r_w; dashed line) and across (r_a; solid line) rows, and patchiness (m^*/m; dotted line) for each assessment date during epidemics of tobacco viruses in Kentucky in 1983. Fields labeled -I were treated with insecticides and those labeled -N were not. To use a consistent scale, some patchiness values are off the scale (A-I). Note the scale change in patchiness compared with Figures 1 and 2.

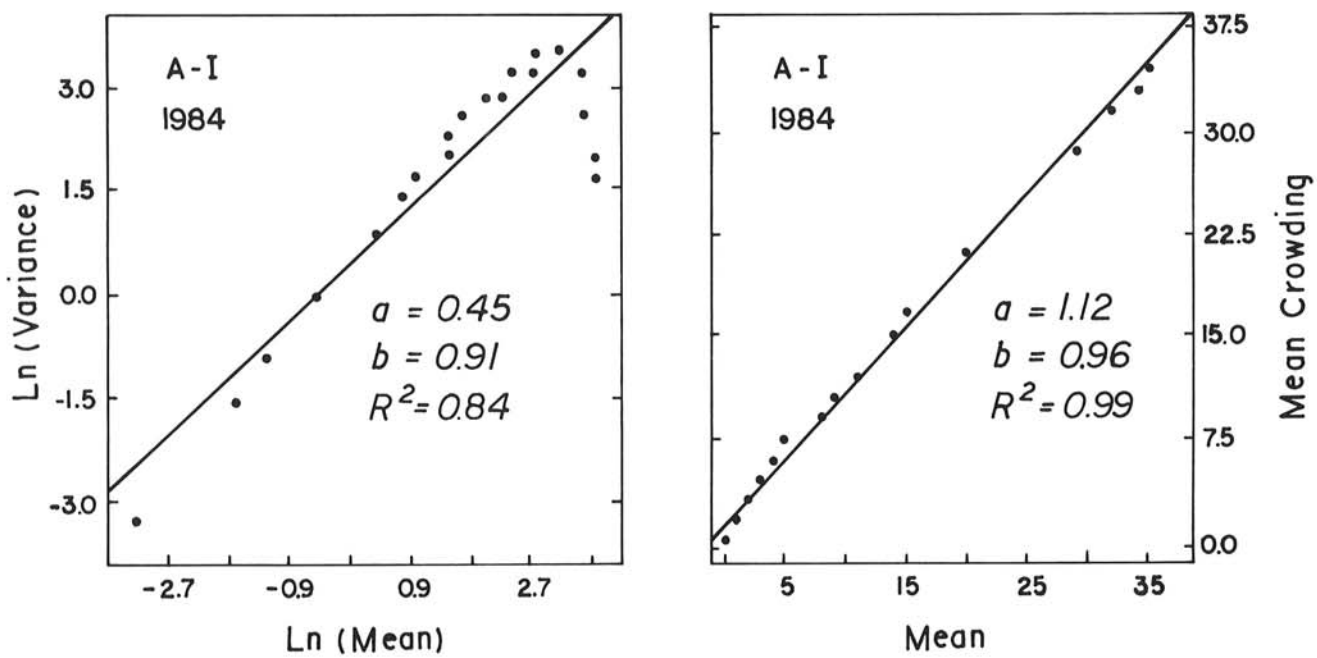

Fig. 4. Relationship between **A,** log of the variance (ln[v]) and log of the mean (ln[m]), and **B,** mean crowding ($m^* = m + [v/m - 1]$) and m over time for virus-diseased plants in a tobacco field. The regression equations $\ln(v) = a + b\ ln(m)$ and $m^* = a + bm$ were fit in **A** and **B**, respectively.

center. Likewise, there would have been significant negative high-order autocorrelation coefficients (3). The high coefficients of variation for mean location of diseased plants suggested that disease increase was attributable to infected tobacco plants within the fields and many other sources outside the fields.

One would expect the pattern of virus-diseased plants to be influenced by the movement and feeding behavior of aphid vectors. For instance, relatively immobile tobacco-colonizing aphids would be consistent with a generalized-Poisson process in which newly diseased plants arise in the immediate vicinity of the randomly located original diseased plants. Conversely, more mobile transitory aphids might be better represented by a compound-Poisson process in which diseased plants are in clusters generally larger than the quadrat size. We do not have data on the feeding and flight behavior of the aphid vectors in these tobacco fields. However, the insecticide treatments did eliminate virtually all aphid colonization in the monitored fields, but transitory aphids landed in both treated and nontreated fields (T. P. Pirone, L. V. Madden, and B. Raccah, *unpublished*). Suppression of aphid colonization had no discernible direct effect on the patterns of virus-diseased plants or how the patterns changed with time. Relating variation in aphid behavior to patterns of diseased plants would require more intensive field studies than reported here and may not be practically feasible.

It is well accepted that quadrat size influences the results of a spatial pattern analysis (3,4,17,24). By using a quadrat size that generally produced the largest variance with the Greig-Smith (8) procedure, we feel that we minimized the confounding effect of an arbitrary partitioning of the fields. Additionally, results based on quadrats were paralleled in many fields by the results from ordinary runs, which were based on the sequence of diseased and virus-free plants in the tobacco rows. As often observed for spatial autocorrelations and point patterns, the percentage of rows with a clustered pattern started low, rose to maximum, and then declined. This further documented the dynamic nature of aggregation of virus-diseased tobacco plants.

TABLE 2. Results from regressing log of variance (ln [v]) on log of mean (ln[m]) and mean crowding (m*) on m for tobacco virus disease epidemics in Kentucky over 3 yr

	$\ln(v) = a + b \ln(m)$			$m^* = a + b\,m$		
Field[y]	a	b	R^{2z}	a	b	R^{2z}
1985						
A-I	0.45	1.07	0.976	0.76	1.01	0.993
A-N	0.47	1.13	0.992	0.69	1.02	0.996
B-I	0.85	1.19	0.990	0.56	1.26	0.978
B-N	0.84	0.96	0.966	1.49	0.99	0.995
C-I	0.66	1.23	0.997	0.63	1.12	0.998
C-N	0.49	1.06	0.898	1.86	0.98	0.990
1984						
A-I	0.45	0.91	0.845	1.12	0.96	0.997
A-N	0.25	0.92	0.869	0.67	0.97	0.998
B-I	0.95	0.73	0.764	1.92	0.92	0.996
B-N	1.25	0.79	0.709	3.44	0.92	0.973
C-I	0.79	0.96	0.881	1.82	0.97	0.994
C-N	2.81	0.18	0.050	3.41	0.93	0.998
1983						
A-I	1.41	1.36	0.998	0.44	3.37	0.907
A-N	0.87	1.24	0.996	0.67	1.45	0.937
C-I	0.65	1.31	0.997	0.19	1.38	0.995
C-N	0.43	1.28	0.998	0.17	1.17	0.998

[y] Fields labeled with "I" were treated with insecticides and "N" were not.
[z] Coefficient of determination; because of different dependent variables, R^2 cannot be directly compared between the log-log and mean-crowding regressions.

TABLE 3. Aggregation statistics for field A-I in 1984 at six dates that are mapped in Figure 6

	Day of year[a]					
Statistic[b]	180	184	191	205	223	226
m	0.040	0.28	1.41	7.75	29.17	32.11
v	0.039	0.37	2.19	19.14	24.79	14.20
VTM	0.97	1.31*[c]	1.55**	2.47**	0.85	0.44
m^*/m	0.37	2.10**	1.39**	1.19**	0.99	0.98
r_a	−0.03	−0.15	0.35**	0.34**	0.32**	0.18
r_w	−0.04	−0.08	0.27*	0.35**	0.47**	0.19

[a] Field was planted 7 June 1984 (day of year 159).
[b] Mean (m), variance (v), variance-to-mean ratio (VTM), Lloyd's patchiness (m^*/m), and spatial autocorrelation within (r_w) and across (r_a) rows.
[c] Values followed by an * or ** indicate nonrandomness at $P = 0.05$ or 0.01, respectively.

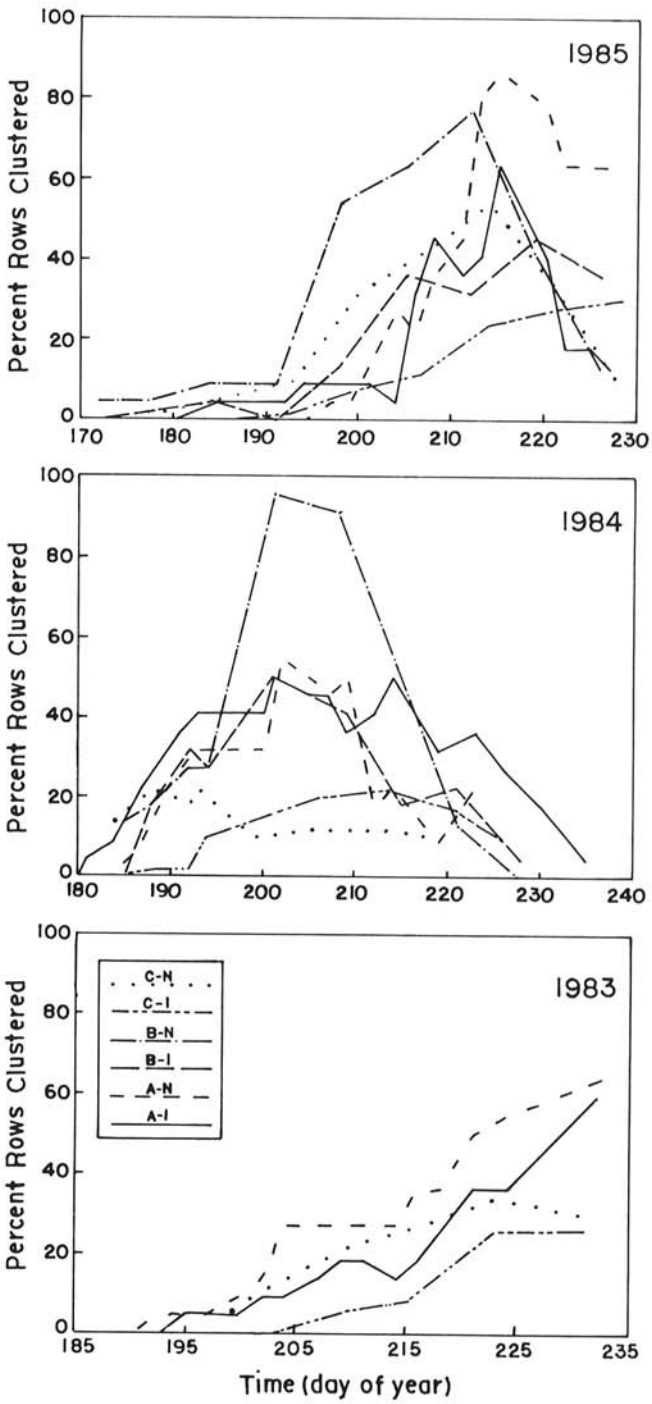

Fig. 5. Percentage of rows in which an ordinary runs analysis indicated clustering ($P < 0.05$) throughout the virus disease epidemics of tobacco over 3 yr. Significant clustering was determined with a standard normal test.

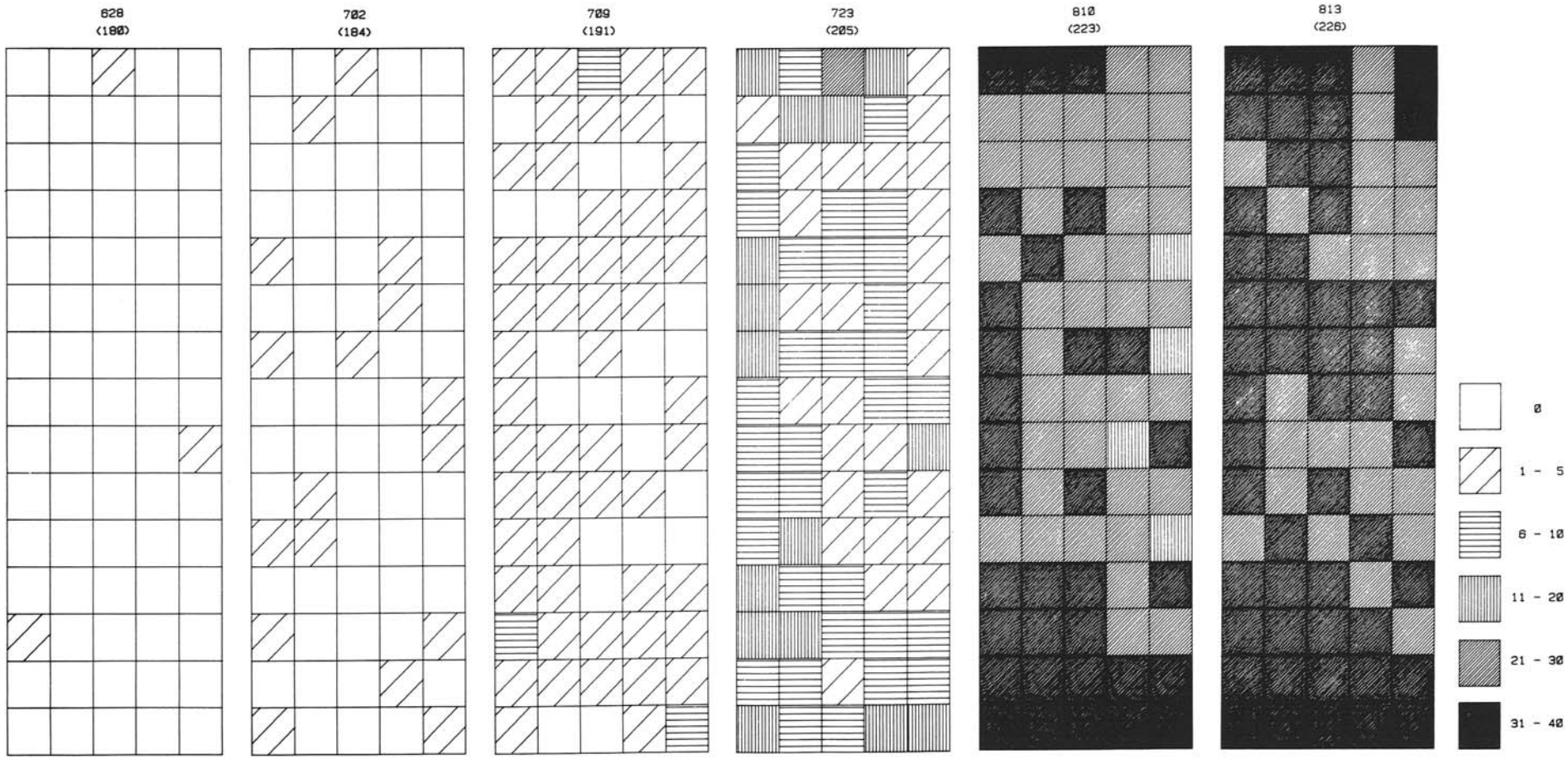

Fig. 6. Maps of field A-I in 1984 in which the field was divided into 75 quadrats of 40 plants each. Tobacco rows run along the long axis of the field. Numbers of diseased plants corresponding to each degree of shading also are shown. Virus-diseased plants are grouped only for presentation, not for analysis. Statistics associated with each date are given in Table 3.

In agreement with another potyvirus system (13), Taylor's power relation generally was not appropriate for describing aggregation in many tobacco fields. Unlike some other pathosystems (e.g., 23), mean density often was high and limited to a maximum level. Instead of increasing with m, v declined with m when m was near the maximum. Taylor's power relation was most appropriate when mean density was not near a maximum as in 1983. A further difficulty with our use of Taylor's relation was the use of data that were not independent. The individual variances and means were collected over time and, therefore, were correlated (11,14). As originally proposed, observations were assumed to be independent (20), although Taylor et al (22) later used some data collected over time. This serial correlation would affect the calculated standard deviations and significance tests (11). There also may be other effects, but serial correlation would not be responsible for the curvature seen in Figure 4.

The relation between m^* and m was nearly linear for all fields, even when mean density was very high, as in 1984. The linear regression model of $m^* = a + bm$ is equivalent to the quadratic equation $v=(a+1)m+(b-1)m^2$ (22). The process of an increasing v to a maximum and then a decline with increasing m can be represented with Iwao's regression equation if $b<1$. For tobacco virus epidemics, this property makes the crowding-mean regressions more useful than Taylor's power relation.

Under one theoretical formulation, the slope of Iwao's regression (b) equals the patchiness of the clusters, and the intercept (a) the mean crowding of "individuals" within the clusters (17). However, Taylor (21) raised serious questions regarding the underlying assumptions necessary to interpret the parameters in this specific manner. It is probably preferable to interpret the a and b parameters more generally as the "index of basic contagion" and "density contagiousness coefficient," respectively (9). For instance, b in a general sense "describes the manner in which individuals or groups of individuals [clusters] distribute themselves in their habitat with changing mean density" (9). The full implications of the results of Iwao's regressions for tobacco virus spatial patterns over time are not yet known.

The spatial pattern of virus-diseased plants likely is influenced by many interacting biotic (e.g., vectors) and abiotic (e.g., weather) factors. It is, therefore, not surprising that a wide range of patterns was observed in this study. Depending on the time of assessment, random or clustered patterns were observed. Using several statistical indices of aggregation, generally consistent and interpretable trends in patterns were found in the six tobacco fields over 3 yr despite this variability. These results indicate the importance of obtaining multiple assessments of mean density and aggregation for diseases caused by viruses such as TEV and TVMV.

LITERATURE CITED

1. Bald, J. G. 1937. Investigations on "spotted wilt" of tomatoes. III. Infection in field plots. Commonw. Aust., Council Sci. Ind. Res. Bull. 106. 32 pp.
2. Campbell, C. L., and Noe, J. P. 1985. The spatial analysis of soilborne pathogens and root diseases. Annu. Rev. Phytopathol. 23:129-148.
3. Cliff, A. D., and Ord, J. K. 1981. Spatial Processes: Models and Applications. Pion Ltd., London. 266 pp.
4. Gilligan, C. A. 1982. Statistical analysis of the spatial patterns of *Botrytis fabae* on *Vicia faba*: A methodological study. Trans. Br. Mycol. Soc. 79:193-200.
5. Gooding, G. V., and Rufty, R. C. 1987. Distribution, incidence, and strains of viruses in burley tobacco in North Carolina. Plant Dis. 71:38-40.
6. Gray, S. M., Moyer, J. W., and Bloomfield, P. 1986. Two-dimensional distance class model for quantitative description of virus-infected plant distribution lattices. Phytopathology 76:243-248.
7. Gray, S. M., Moyer, J. W., Kennedy, G. G., and Campbell, C. L. 1986. Virus-suppression and aphid resistance effects on spatial and temporal spread of watermelon mosaic virus 2. Phytopathology 76:1254-1259.
8. Greig-Smith, P. 1952. The use of random and contiguous quadrats in the study of the structure of plant communities. Ann. Bot. 16:293-316.
9. Iwao, S. 1968. A new regression method for analyzing the aggregation pattern of animal populations. Res. Popul. Ecol. 10:1-20.
10. Madden, L. V. 1988. Dynamic nature of within-field disease and pathogen distributions. In: The Spatial Component of Plant Disease Epidemics. M. J. Jeger, ed. Prentice-Hall, Inc., Englewood Cliffs, NJ. (In press.)
11. Madden, L. V., and Campbell, C. L. 1986. Descriptions of virus disease epidemics in time and space. Pages 273-293 in: Plant Virus Epidemics; Monitoring, Modelling, and Predicting Outbreaks. G. D. McLean, R. G. Garrett and W. G. Ruesink, eds. Academic Press, Sydney. 550 pp.
12. Madden, L. V., Louie, R., Abt, J. J., and Knoke, J. K. 1982. Evaluation of tests for randomness of infected plants. Phytopathology 72:195-198.
13. Madden, L. V., Louie, R., and Knoke, J. K. 1987. Temporal and spatial analysis of maize dwarf mosaic epidemics. Phytopathology 77:148-156.
14. Madden, L. V., Pirone, T. P., and Raccah, B. 1987. Temporal analysis of two viruses increasing in the same tobacco fields. Phytopathology 77:974-980.
15. Modjeska, J. S., and Rawlings, J. O. 1983. Spatial correlation analysis of uniformity data. Biometrics 39:373-384.
16. Nicot, P. C., Rouse, D. I., and Yandell, B. S. 1984. Comparison of statistical methods for studying spatial patterns of soilborne plant pathogens in the field. Phytopathology 74:1399-1402.
17. Pielou, E. C. 1977. Mathematical Ecology. John Wiley & Sons, New York. 385 pp.
18. Pirone, T. P., Gooding, G. V., and Smiley, J. H. 1973. Tobacco vein mottling virus on burley tobacco in Kentucky. Plant Dis. Rep. 57:841-844.
19. Shew, B. B., Beute, M. K., and Campbell, C. L. 1984. Spatial pattern of southern stem rot caused by *Sclerotium rolfsii* in six North Carolina peanut fields. Phytopathology 74:730-735.
20. Taylor, L. R. 1961. Aggregation, variance and the mean. Nature (London) 189:732-735.
21. Taylor, L. R. 1984. Assessing and interpreting the spatial distributions of insect populations. Annu. Rev. Entomol. 29:321-357.
22. Taylor, L. R., Woilwod, I. P., and Perry, J. N. 1978. The density dependence of spatial behaviour and the rarity of randomness. J. Anim. Ecol. 47:383-406.
23. Thal, W. M., and Campbell, C. L. 1986. Spatial pattern analysis of disease severity data for alfalfa leaf spot caused primarily by *Leptosphaerulina briosiana*. Phytopathology 76:190-194.
24. Upton, G., and Fingleton, B. 1985. Spatial Data Analysis by Example. John Wiley & Sons, Chichester, England. 410 pp.
25. Vanderplank, J. E. 1946. A method for estimating the number of random groups of adjacent diseased plants in a homogenous field. Trans. R. Soc. S. Afr. 3:269-278.