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Comparative Analyses of Lettuce Drop Epidemics Caused by *Sclerotinia minor* and *S. sclerotiorum*

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ABSTRACT

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Temporal and spatial patterns of lettuce (*Lactuca sativa*) drop caused by *Sclerotinia minor* and *S. sclerotiorum* were determined in lettuce fields in the Salinas, Santa Maria, and San Joaquin Valleys in California during 1995 to 1998. Of the 25 commercial fields assessed, 14 had predominantly *S. minor*, 9 had predominantly *S. sclerotiorum*, and 2 had varying levels of both species. *Sclerotinia* infections were classified based on symptoms: those caused directly by eruptive germination of sclerotia (type I) and those caused by the airborne ascospores (type II). The precise location of diseased and healthy plants was mapped and lettuce drop progress was determined at different crop growth stages. Spatial patterns of disease incidence were analyzed using 1-by-1-, 2-by-2-, 3-by-3-, and 4-by-4-m quadrat sizes. Regardless of the analytical method employed, disease incidence with type I infection showed an aggregated pattern in a majority of the fields evaluated and random patterns in fields where incidence was low. In all fields with type I infection, disease progress followed the monomolecular model, typical of soilborne diseases. For fields with aggregated distribution, spatial dependence was observed up to 10 m and was either isotropic or random in direction, suggesting the potential influence of tillage operations on inoculum distribution and disease incidence. Lettuce drop incidence in fields with type II infection was erratic in time and peaked within a very short time. However, disease incidence showed an aggregated pattern in all fields evaluated. Spatial dependence of quadrats generally was detected in two adjacent directions, suggesting a directional gradient perhaps caused by wind direction during ascospore dissemination. Increasing quadrat sizes usually increased the degree of aggregation of lettuce drop, but not the distribution pattern itself. These results demonstrate that the source of inoculum and the type of infections they cause are most likely to determine spatial patterns of lettuce drop in the field.

Lettuce drop occurs worldwide and causes significant yield losses on lettuce (*Lactuca sativa* L.). In California, where nearly 60% of U.S. lettuce production occurs, losses of up to 60% have been recorded in individual fields (33). The causal agents of lettuce drop are two closely related species, *Sclerotinia minor* Jagger and *S. sclerotiorum* (Lib.) de Bary (1,33). Although general symptoms caused by the two species on lettuce are similar, close examination of diseased plants reveals inherent differences. These differences are the result of different infection modes employed by the two species for infection. In nature, *S. minor* infects lettuce exclusively by mycelium from eruptively germinating sclerotia that are located near the tap root system or the plant crown (3,9,17,26,34,35). Thus, it is typically a soilborne pathogen, although infections attributed to ascospores from the carpogenic germination of sclerotia occasionally have been reported in New Zealand or in laboratory conditions (12,14,19). Our extensive surveys, however, have not revealed ascospore production or infection from ascospores by *S. minor* in commercial fields.

In contrast, *S. sclerotiorum* infects lettuce primarily by ascospores (4,28,30) and infections from mycelial germination of sclerotia occur in low frequencies (1). Ascospore infections cause water-soaked areas on lettuce crowns, mycelial mat formation, and sclerotial development with time (4,5,20). Infection from direct germination of *S. sclerotiorum* sclerotia is similar to that of *S. minor*, but is easily differentiated by the larger size of *S. sclerotiorum* sclerotia on infected plants.

S. minor and *S. sclerotiorum* have different modes of infection; therefore, lettuce drop caused by these two species is expected to follow different spatial patterns (25,27,35). Because *S. minor* is typically a soilborne pathogen, lettuce drop caused by this species is expected to reflect the distribution of sclerotia in the soil profile due of the high correlation between inoculum density and disease incidence (7,9,13). The pattern of lettuce drop incidence caused by *S. sclerotiorum* depends on not only sclerotia but also the conditions such as soil temperature and moisture that affect the production of apothecia, and wind speed and direction that affect ascospore discharge and deposition (1,28,32). Comparing the spatial pattern and progress of lettuce drop caused by the two *Sclerotinia* spp. is necessary to not only describe attributes of epidemics caused by different types of inocula but also potentially aid in the development of sampling and novel control strategies (7,23).

A spatial pattern analysis procedure of lettuce drop incidence caused by *S. sclerotiorum* is lacking, and spatial pattern analyses of lettuce drop caused by *S. minor* are limited to the ordinary runs analysis procedure (9,25). The current study, therefore, compares the spatial patterns of lettuce drop caused by the two species of *Sclerotinia* for comparative analyses of the epidemics caused by different sources of inoculum.

Spatial pattern analysis has been used to describe the relationship between inoculum density and disease incidence (6,7,23), inoculum distribution pattern (9,20,36) and movement affected by edaphic and other environmental factors (21,31), tillage and other practices (34,35), and distribution of diseases as influenced by vectors (24), as well as to develop accurate sampling methods (15). One of the cardinal assumptions in the literature is that the different mechanisms of disease spread result in different spatial patterns of disease (7,23). A number of techniques have been employed for spatial pattern analyses, such as mapping (9,21,25,36), frequency distributions (6,9,15,16,22,24), two-dimensional distance class analysis (27), spatial autocorrelation analyses (23), and geostatistics (8,18,32). Geostatistics offers several advantages over the techniques described above (8,18). It takes into consideration both the random and systematic characteristics of spatially distributed variables, and requires flexible assumptions of stationarity compared with spatial autocorrelation techniques (8,18). In addition, geostatistics can be applied to detect spatial dependence either in specific directions or collectively in all directions using semivariograms, which quantify spatial dependence by measuring the variation between samples, separated by a vector, the "lag distance".

The differential geographic distribution of the two *Sclerotinia* spp. causing lettuce drop in California offers an ideal opportunity to make comparative spatial pattern

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analyses of lettuce drop caused by the two species. *S. minor* is the dominant species in coastal California and *S. sclerotiorum* is prevalent in San Joaquin Valley; whereas, in Santa Maria Valley, both species occur in different fields, with *S. sclerotiorum* occurring at a higher frequency than in Salinas Valley.

The hypothesis tested was that different sources of inoculum of *Sclerotinia* spp. will result in different spatial patterns of the disease. Specific objectives of this work were to compare (i) the spatial patterns of lettuce drop incidence caused by *S. minor* and *S. sclerotiorum* and (ii) the sclerotial versus ascosporic infections in time and space. Preliminary results of this study have been published (11).

MATERIALS AND METHODS

Field sites. The data for spatial analysis of disease incidence were collected during 1995 to 1998 in 25 commercial lettuce fields in coastal and central California. There were 9 fields in Huron in the San Joaquin Valley, CA, (HUR01 to 05, HUR11 to 14), 12 in the Salinas Valley (FST01 to 03, GZL01 and 02, HIT01 to 05, SMV01, and SPK01), and 4 in the Santa Maria Valley (STM01 and 02, and LPC01 and 02) (Table 1). The criteria for field selection included high drop incidence and similarity in bed configuration (1 m between bed centers), tillage operations, distance between plants, irrigation, lettuce types, and ignored cropping history of the fields. All fields were planted with crisphead lettuce cv. Salinas in two rows per bed (1 m between bed centers) with 25- to 30-cm spacing between plants. Sprinkler irrigation after direct seeding was used in all fields to facilitate germination and emergence of lettuce seedlings before thinning. After thinning, sprinkler, furrow, or subsurface drip irrigation was used, depending on the choice of the growers. In each field, three plots were established arbitrarily for data collection, with plot areas in the range 16 to 32 by 30 to 32 m (Table 1).

Lettuce drop caused by the two species was differentiated based on the location and size of sclerotia on infected plants. Each field plot was mapped only once, 1 week prior to harvest when the drop incidence was the highest (9,13). In each field plot, the precise location of diseased and healthy plants, along with a notation on whether the plant was infected by ascospores or direct germination of sclerotia, was recorded. On infected plants, *S. minor* sclerotia are spherical, approximately 2 mm in diameter, and are localized around the crown near the soil surface. *S. sclerotiorum* sclerotia are spherical to flat, and vary from 2 mm to 2 cm. Sclerotia are distributed on the surface of lettuce heads when infections are initiated by airborne ascospores. Isolations from a random sample were made to confirm the species causing the disease. For convenience, the terms type I and type II infections were used to describe infections caused by sclerotia and ascospores, respectively.

Because the relative position of each plant was recorded, the plot areas were divided into grids of contiguous quadrats in a computer program. Three sizes of quadrats were arranged for each plot: 1 by 1, 2 by 2, and 3 by 3 m, depending on the analysis. Disease incidence in each quadrat was calculated by dividing the number of diseased plants by the total number of plants in each quadrat and expressing them as percentages. Using a custom computer program (DMAP, *unpublished*), maps of lettuce drop incidence distribution in the fields were generated.

Sclerotial population estimates. Soil samples were collected from Huron fields in the spring of 1997, a month after lettuce harvest following incorporation of the residue, and from Salinas experimental fields in 1996 and 1997 before planting (11). The preceding crop in these plots was lettuce and the soil samples were collected 6 weeks after the previous crop was incorporated after harvest. Twenty soil samples of 100 cm^3 each were collected arbitrarily to a depth of 7 cm (10) from each of the six fields in Huron during the surveys. Twelve soil samples of 100 cm^3 each were collected from three plots at planting at Hartnell and Spence sites (11). Sclerotia were recovered using the wet sieving method (2), counted, and their viability tested on water agar (32).

Temporal disease progress. To study the temporal progress of lettuce drop caused by the two species, additional data were collected from commercial fields at

Table 1. Commercial lettuce fields in California from which data for spatial pattern analyses of lettuce drop caused by *Sclerotinia minor* or *S. sclerotiorum* were collected

Field[w]	Pathogen	Type[x]	Location[y]	Plot size (m)	Date[z]
FST01	*S. minor*	I	Salinas	32 by 32	21 April 1995
FST02	*S. minor*	I	Salinas	32 by 32	21 April 1995
FST03	*S. minor*	I	Salinas	30 by 26	21 April 1995
GZL01	*S. minor*	I	Gonzales, Salinas	32 by 22	31 May 1996
GZL02	*S. minor*	I	Gonzales, Salinas	32 by 32	31 May 1996
HIT01	*S. minor*	I	Salinas	32 by 30	10 May 1996
HIT02	*S. minor*	I	Salinas	32 by 30	10 May 1996
HIT03	*S. minor*	I	Salinas	30 by 26	25 April 1995
HIT04	*S. minor*	I	Salinas	22 by 20	25 April 1995
HIT05	*S. minor*	I	Salinas	32 by 30	25 April 1995
HUR01	*S. sclerotiorum*	II	Huron	32 by 30	14 March 1996
HUR02	*S. sclerotiorum*	II	Huron	32 by 30	13 March 1996
HUR03	*S. sclerotiorum*	II	Huron	32 by 30	13 March 1996
HUR04	*S. sclerotiorum*	II	Huron	32 by 30	14 March 1996
HUR05	*S. sclerotiorum*	I	Huron	32 by 30	22 March 1996
HUR11	*S. sclerotiorum*	II	Huron	32 by 32	2 May 1998
HUR12	*S. sclerotiorum*	II	Huron	32 by 16	25 April 1998
HUR13	*S. sclerotiorum*	II	Huron	32 by 32	2 May 1998
HUR14	*S. sclerotiorum*	II	Huron	32 by 32	25 April 1998
LPC01	*S. minor* + *S. sclerotiorum*	I	Lompoc, SMV	34 by 32	14 July 1995
LPC02	*S. minor* + *S. sclerotiorum*	I	Lompoc, SMV	20 by 32	14 July 1995
SMV01	*S. minor*	I	Somavia, Salinas	32 by 32	25 August 1996
SPK01	*S. minor*	I	Salinas	32 by 20	11 July 1996
STM01	*S. minor*	I	Santa Maria, SMV	32 by 32	7 June 1995
STM02	*S. minor*	I	Santa Maria, SMV	32 by 32	7 June 1995

[w] Field designation.
[x] Two types of infection caused by *Sclerotinia* spp. were differentiated based on symptoms: type I = the infection caused by mycelium from eruptively germinated sclerotia and type II = the infection caused by ascospores.
[y] Nearest town. SMV = Santa Maria Valley.
[z] Date disease incidence was recorded, all 1 week before harvest.

Huron for *S. sclerotiorum* (HUR01-5 and HUR11-14; Table 1) and from experimental plots at Hartnell and Spence in Salinas for *S. minor* (11). Disease progress was monitored weekly in three replicated plots of 8 by 12 m at Spence and four replicated plots of 6 by 10 m at Hartnell. At the Huron fields, lettuce drop progress data were collected during the winter (February to March each year) from five 32-by-32-m areas arbitrarily established at the center and four corners of the fields. The total number of plants and the number infected were counted in each of these areas and averaged to obtain disease incidence for that field. Mean incidence and the corresponding standard errors of the mean were computed and incidence was plotted against the dates of data collection.

Frequency distribution analysis on disease incidence. Unless mentioned specifically, all analyses were performed using data from 2-by-2-m quadrats. Representative maps of distribution of lettuce drop incidence caused by *S. minor* and *S. sclerotiorum* were generated using the DMAP computer program (*unpublished*). Frequency distribution analyses were performed to detect patterns using the computer program for fitting a β-binomial distribution (BBD), which is appropriate for binary data such as disease incidence (22). The program calculates moment estimates of parameters p (probability of diseased plants) and θ (index of aggregation), maximum likelihood estimates of the parameters and standard errors of the estimates, and goodness of fit for β-binomial distribution. The two variance ratio tests, the Z statistic in $C(\alpha)$ test, and D (the index of dispersion) also were calculated. Spatial patterns were considered to be aggregated if $D > 1$ and randomly distributed when $D = 1$. Similarly, the parameter $\theta > 0$ in the β-binomial distribution suggests an aggregated distribution, and $\theta < 0$ indicates a random pattern. The null hypothesis of $\theta > 1$ was tested using a t test. These statistics were used together to describe the spatial distribution and homogeneity of lettuce drop incidence caused by both species of *Sclerotinia* in these fields.

Geostatistical analysis of disease incidence. Data points used for analysis were the disease incidence from each quadrat in the plot. Geostatistical software, VARIOWIN (29), was used to detect the spatial dependence or correlation among sample sites in this study. Semivariograms were calculated for four directions, 0, 45, 90, and 135º, where 0º represents the direction along the rows and 90º represents the direction across rows and those between. Semivariograms for each direction were plotted against the lag distances.

Effect of quadrat sizes on disease distribution. To examine the effect of quadrat sizes on spatial distribution of lettuce drop incidence, the data were rearranged into different sizes of quadrats. Incidence data were calculated for quadrat sizes of 1 by 1, 2 by 2, 3 by 3, and 4 by 4 m for each field. To compare the effect of quadrat size on the observed spatial pattern, point analysis was performed using BBD and autocorrelation analyses were performed using VARIOWIN. The index of dispersion (D), range, and sill-nugget of semivariograms from these analyses were subjected to analysis of variance to determine the effects of quadrat sizes, and means were compared using a least significant difference test. Only data from fields or directions in which spatial dependence was detected were used for this purpose.

RESULTS

Population of sclerotia. The population of *S. minor* sclerotia was 2.6, 13.4, and 11.9 per 100 cm^3 of soil in 1996, 1997, and 1998, respectively. There was a high correlation between the number of sclerotia of *S. minor* in soil and the incidence of drop ($R^2 = 0.77$, $P < 0.0001$). The density of *S. sclerotiorum* in the soil of sampled fields averaged 0.06 sclerotia per 100 cm^3 of soil at Huron (*data not shown*). Although the density of sclerotia in soil was very low, lettuce drop incidence was as high as 14% (Fig. 1).

Temporal disease progress. The incidence of lettuce drop caused by *S. minor* varied from 0.7 to 73.1% and that caused by *S. sclerotiorum* from <1 to 14.1% during the 3-year study (Fig. 1). In all fields infected by *S. minor*, sclerotia were observed only at the crown and root portions of the plants, suggesting the predominance of direct infections by soilborne sclerotia of this species. In contrast, in plots with higher incidence of lettuce drop caused by *S. sclerotiorum*, all infected plants had a large number of sclerotia on the exposed

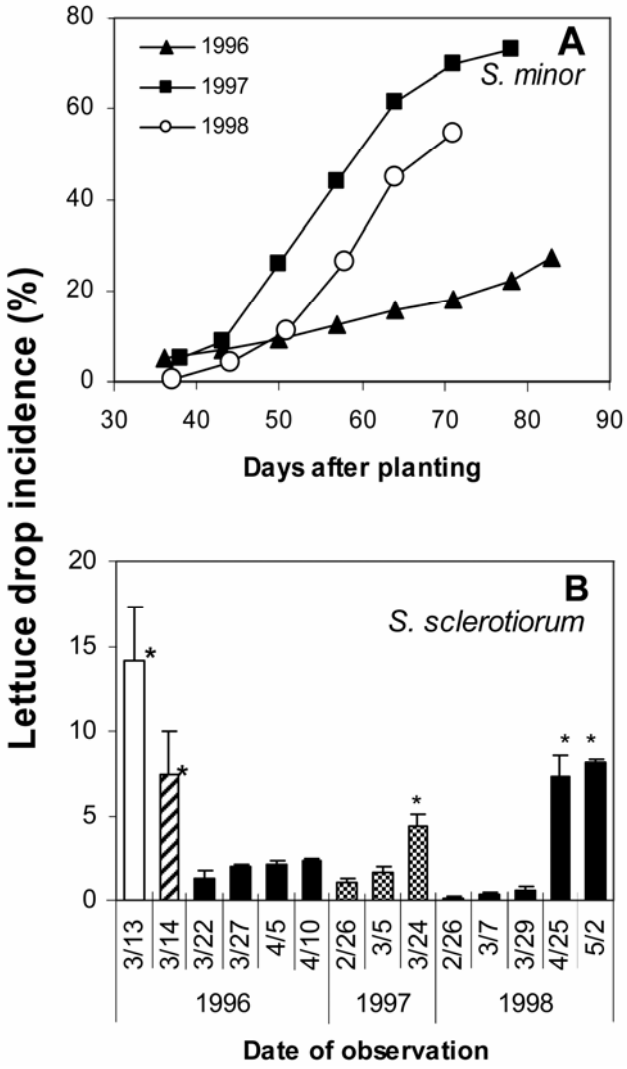

Fig. 1. A, Lettuce drop incidence (%) caused by *Sclerotinia minor* at the Spence site, Salinas, in 1996, and at the Hartnell site, Salinas, during 1997 and 1998 and **B,** lettuce drop incidence from *S. sclerotiorum* infections at Huron, CA during 1996 to 1998. The disease in Salinas was evaluated weekly in the same field each year and in Huron at 5- to 27-day intervals in different fields. Data for 1996 were derived from three different fields and in 1997 and 1998 from the same field multiple times. Bars followed by asterisks indicate infection ascosporic infection by *S. sclerotiorum*. Dates of planting at Huron were 10 January (two fields) and 1 March (one field) in 1996, and one field each on 20 January 1997 and 15 February 1998.

surface of lettuce heads, suggesting infection by airborne ascospores. However, in plots with less than 1% incidence of lettuce drop, sclerotia of *S. sclerotiorum* were observed at the base of plants, showing infection by sclerotia germination.

The incidence of lettuce drop caused by *S. minor* was low in early growth stages and increased linearly or curvilinearly as the lettuce matured (Fig. 1A). For fields with *S. sclerotiorum* in Huron, where fields were monitored at 5- to 27-day intervals,

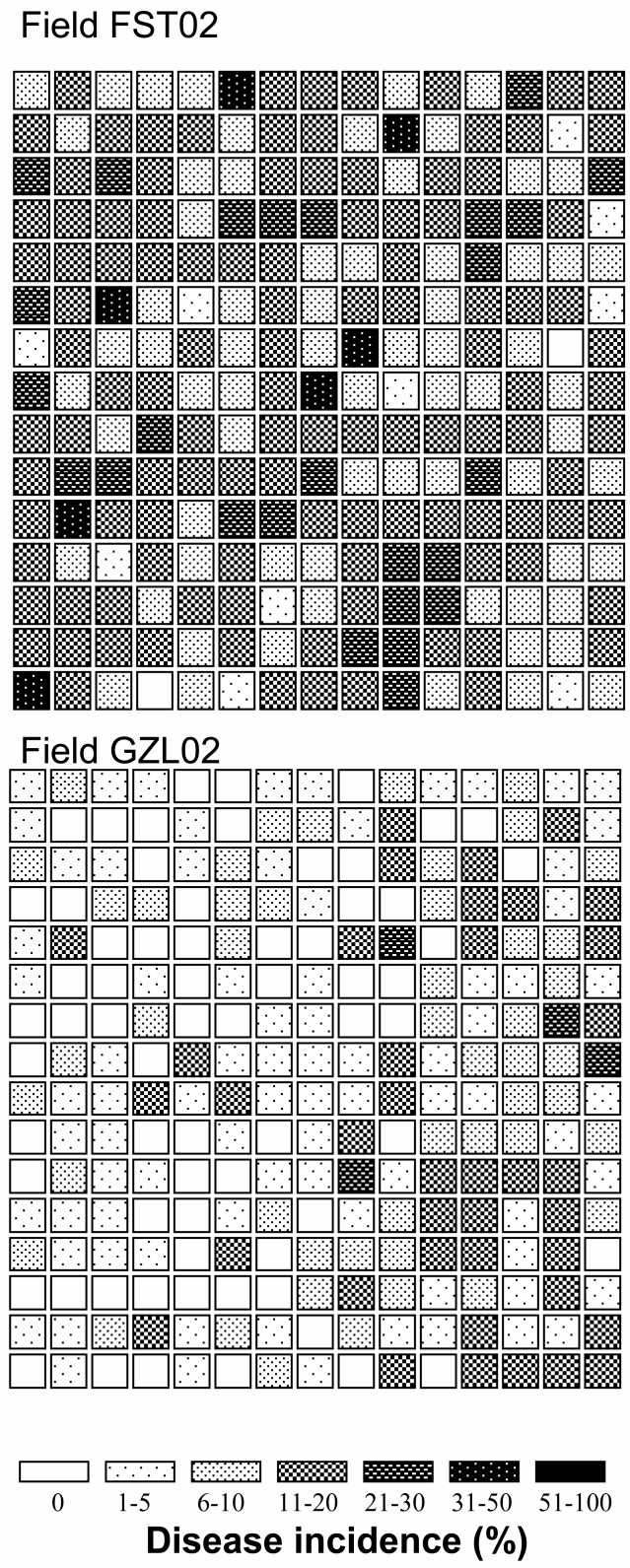

Fig. 2. Distribution of lettuce drop incidence (%) caused by *Sclerotinia minor* in two commercial lettuce fields, representing type I infection, in California. Each small square represents incidence in a 2-by-2-m quadrat, with about 24 plants each. The different patterns represent incidence classes shown in the legend. Disease incidence was rounded to nearest range class.

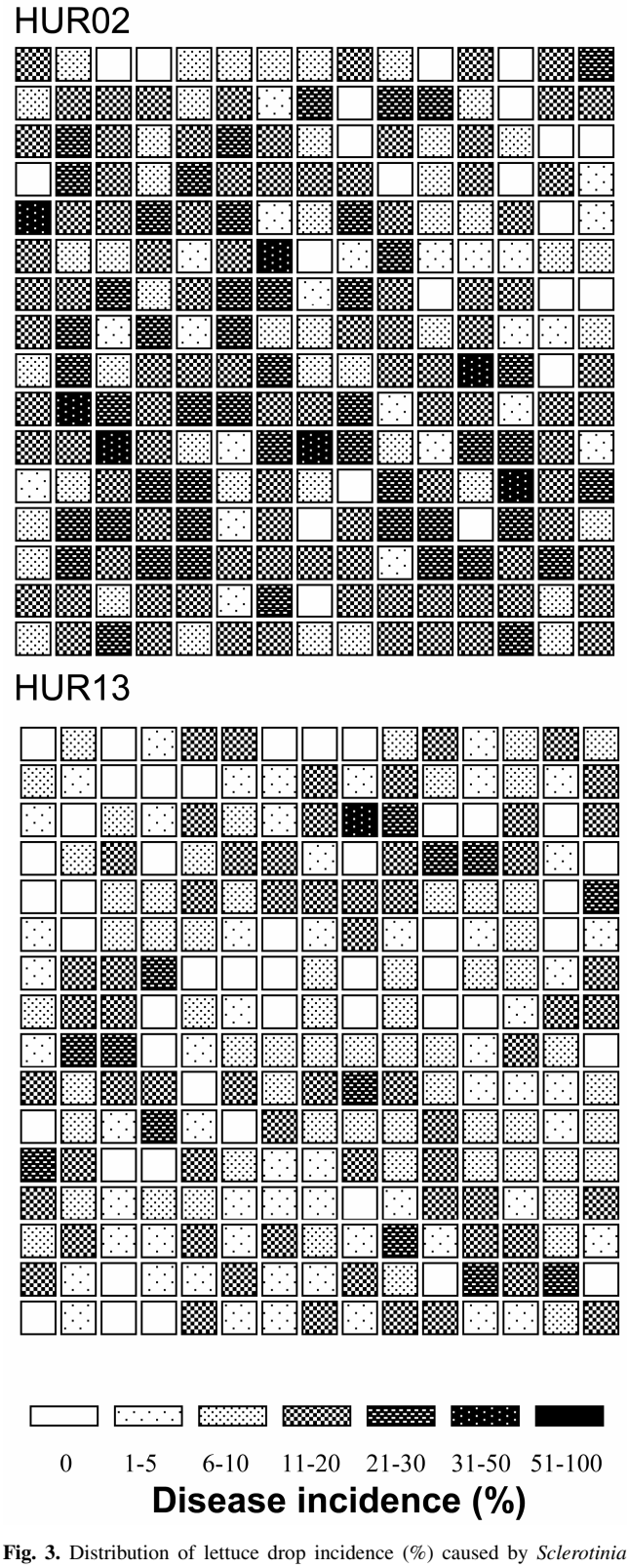

Fig. 3. Distribution of lettuce drop incidence (%) caused by *Sclerotinia sclerotiorum* in two commercial lettuce fields, representing type II infection, in California. **A,** Data from field HUR02 and **B,** data from field HUR13. Each small square represents incidence in a 2-by-2-m quadrat, with about 24 plants each. The different patterns represent incidence classes shown in the legend. Disease incidence was rounded to nearest range class.

the incidence of lettuce drop caused by direct germination of sclerotia of *S. sclerotiorum* was very low and no infection by ascospores occurred at an early stage (Fig. 1B). The final disease of each field on 13 and 14 March 1996, 24 March 1997, and 2 May1998 showed ascosporic infection and had higher disease incidence, except in one field on 10 April 1996 (Fig. 1B).

Analyses of spatial patterns. The incidence of lettuce drop caused by *S. minor* (Fig. 2) and *S. sclerotiorum* (Fig. 3) was aggregated in most fields. Frequency distribution analyses indicated that disease incidence was aggregated in 9 of 14 fields with type I infection by *S. minor*, in 8 of 9 fields with type II infection by *S. sclerotiorum*, and in 2 of 2 fields where type I infections by both *S. minor* and *S. sclerotiorum* were present. The remaining six fields, all with type I infection, exhibited a random distribution pattern (Table 2).

The $\theta > 0$ in 7 of 17 fields with type I infection caused by both *Sclerotinia* spp. (Table 2), suggesting that lettuce drop incidence was highly aggregated in these fields. In all eight fields with type II infection caused by *S. sclerotiorum*, $\theta > 0$, also indicating a highly aggregated pattern of disease incidence (Table 2). The χ^2 test revealed that, among the eight fields with type II infection by *S. sclerotiorum*, disease incidence distribution in seven fitted the BBD best. Results from all analyses consistently demonstrated that type II infection resulted in aggregated lettuce drop distribution (Table 2).

The distribution of lettuce drop with type I infection was best fitted by the BBD in 10 of 14 plots; in the remaining fields, lettuce drop was distributed randomly according to all indices (Table 2). Thus, the distribution of lettuce drop incidence in fields with airborne ascospores of *S. sclerotiorum* (type II infection) was aggregated and, in fields where lettuce drop was caused by the direct germination of *S. minor* or *S. sclerotiorum* sclerotia (type I infection), the distribution varied from aggregated to random. There was a positive correlation between the degree of aggregation (*D*) and incidence of lettuce drop (DI) (Fig. 4). The correlation coefficient between DI and θ was 0.74 ($P = 0.001$), and that between DI and *D* was 0.75 ($P = 0.001$) for 2-by-2-m quadrats used in all analyses.

Spatial autocorrelation. Anisotropic spatial dependence was detected by geostatistical methods for lettuce drop incidence caused by *S. minor* (type I infection) in three representative fields (Fig. 5; FST03, LPC-1, and HIT05). In plot FST03, semivariance increased up to 10 m in all four directions (Fig. 5). In plot LPC-1, semivariance increased slightly in one or two directions, but the increase was small (Fig. 5). In plot HIT05, semivariance increased linearly in three of four directions up to a distance of 18 m, though it remained consistent in the direction of 45º (Fig. 5). This suggested that there was spatial dependence of disease incidence between quadrats, possibly in all directions. In other words, the aggregation of disease incidence was isotropic.

In fields with type II infection by *S. sclerotiorum*, semivariance increased in the 90º direction up to 8 m or more (Fig. 5; HUR01, HUR03, and HUR13). Increases in the 135º direction occurred in two fields (Fig. 5; HUR01 and HUR03). Semivariance rarely changed in the direction of 0º or within the furrow. Thus, there was a spatial dependence of disease incidence between quadrats separated by a distance >8 m, and disease incidence was more aggregated across the rows than in the diagonal directions.

Quadrat size and spatial analysis. Frequency analyses showed that the index of dispersion (*D*) was greatly affected by the quadrat size when the disease was aggregated (Fig. 6; Table 3). In these fields, *D* values increased as quadrat size increased (Table 3; Fig. 6). In general, in fields with a random pattern of disease incidence, *D* values were not significantly different from 1 in all four quadrat sizes tested.

Quadrat size had little effect on the geostatistical analysis of disease incidence caused by either *S. minor* or *S. scle-*

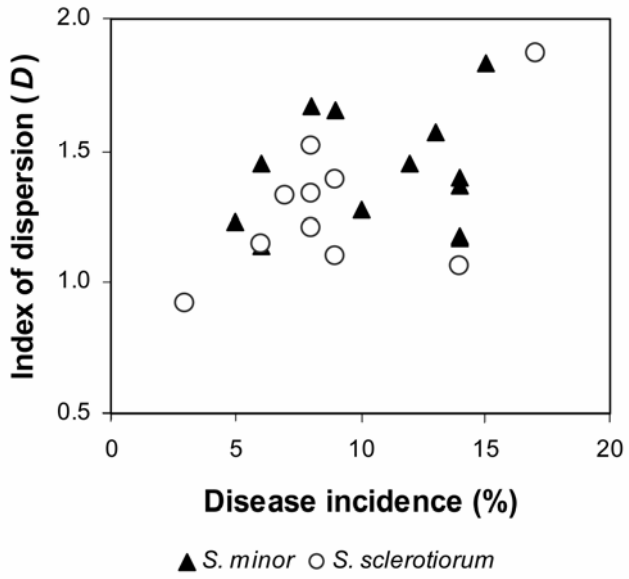

Fig. 4. Relationship between index of dispersion (*D*) and final lettuce drop incidence in 2-by-2-m quadrats in all fields examined. The data from *Sclerotinia minor* fit the polynomial model with a correlation coefficient of 0.57, and data from *S. sclerotiorum* fit the linear model with a correlation coefficient of 0.61.

Table 2. Summary statistics from the β-binomial distribution analyses of lettuce drop incidence caused by *Sclerotinia minor* and *S. sclerotiorum* in California commercial fields

				Goodness-of-fit[u]				
Species	**Type[v]**	**Plot[w]**	**DI (%)[x]**	**$\theta > 0$[y]**	**$P(Z) \leq \alpha$[z]**	**Bb > α**	**Bi > α**	**Fit Bb**
S. minor	I	14	12.1	7	9	13	9	10
S. sclerotiorum	II	8	9.8	8	8	7	0	7
S. sclerotiorum	I	1	3.1	0	0	0	1	0
S. minor + *S. sclerotiorum*	I	2	7.1	0	2	1	1	1

[u] The χ^2 goodness-of-fit test was used to test the frequency distribution of β-binomial and binomial distribution models. Bb > α, number of fields that cannot reject β-binomial distribution at a significance level of 0.05. Bi > α, number of plots that cannot reject binomial distribution at a significance level of 0.05. Fit Bb is the number of plots that fit β-binomial with a higher *P* value than the binomial distribution.
[v] Infection types: I = direct infection by eruptively germinated sclerotia and II = infection caused by ascospores.
[w] Plot = total number of fields observed.
[x] DI = averaged disease incidence estimate.
[y] Number of fields with $\theta > 0$; $\theta > 0$ when the calculated ratio of θ over its error is larger than the tabulated *t* value at the level 0.05; otherwise $\theta < 0$.
[z] Number of fields with $P(Z) \leq \alpha$, where *Z* = standard normal statistic of the $C(\alpha)$ test, and significance level (α) = 0.05.

rotiorum (Table 3). Regardless of the spatial pattern of lettuce drop, the semivariance increased or decreased similarly in all directions in the four quadrat sizes tested, except in one direction in the 3–by-3-m quadrat size (Fig. 7). For example, semivariance linearly increased as distance increased in the direction of 90 and 135º with all four quadrat sizes tested (Fig. 7). The increases in semivariance (sill – nugget) were not significantly different for the four quadrat sizes (Table 3). The ranges, however, were significantly smaller for quadrat size 1 by 1 m compared with the other quadrat sizes.

DISCUSSION

The distribution of lettuce drop incidence caused by eruptive, mycelial germination of *S. minor* sclerotia and mycelial germination of *S. sclerotiorum* sclerotia (type I infection) showed an aggregated pattern in a majority of the fields surveyed. This mode of infection also generated random patterns in a few fields with low levels of lettuce drop incidence. Lettuce drop caused by airborne ascospores of *S. sclerotiorum* (type II infection) also showed aggregated patterns consistently in all fields, contrary to our initial hypothesis that airborne inoculum would lead to random distribution of the disease. Both types of infections exhibited spatial dependence

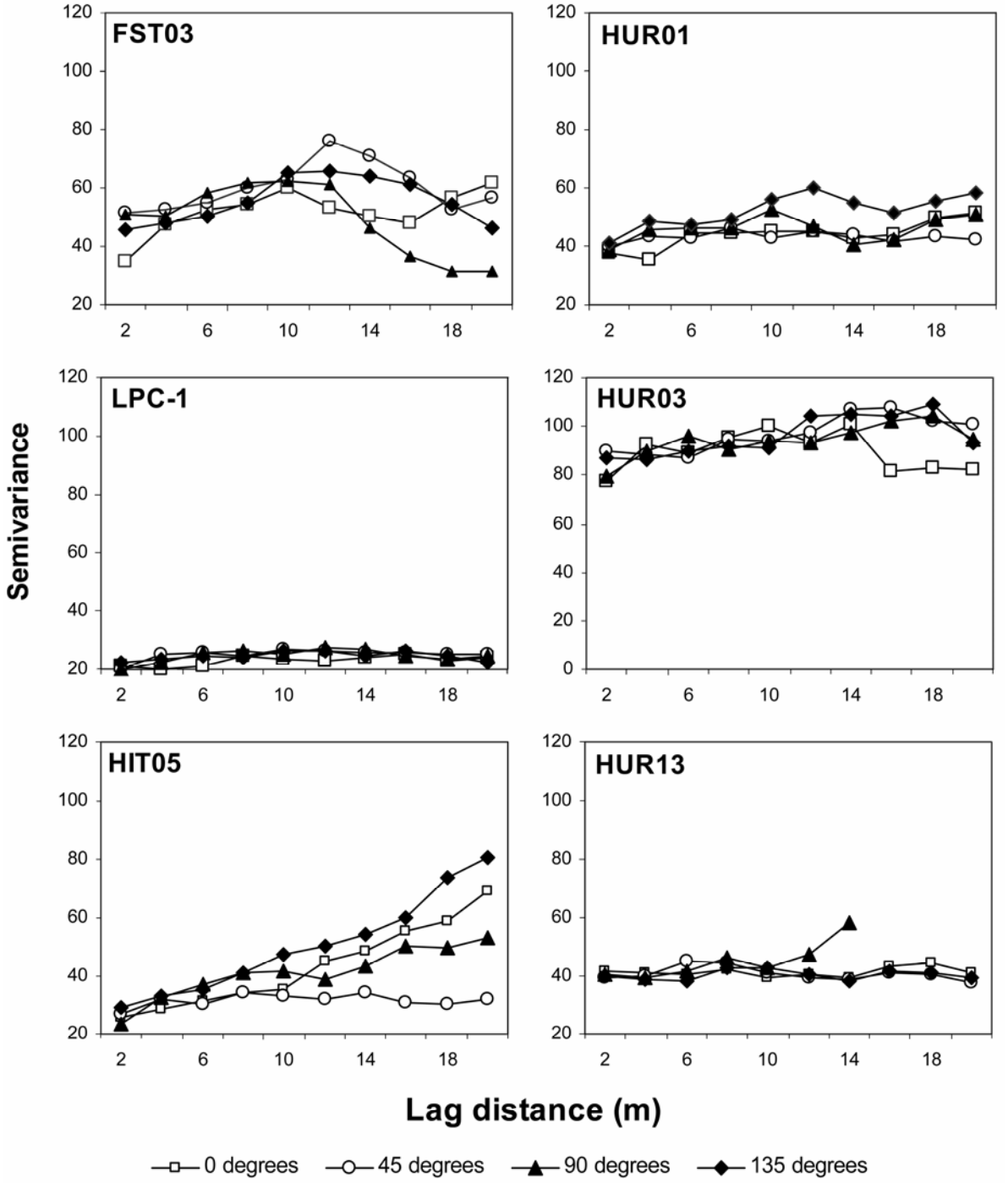

Fig. 5. Semivariograms of lettuce drop incidence caused by *Sclerotinia minor* and *S. sclerotiorum* in three representative fields for each infection type or species. The acronyms on the top left corner of each panel represent the fields from which the data were collected. FST03, LPC-1, and HIT05 had type I infection by *S. minor* or also by *S. sclerotiorum*; HUR01, HUR03, and HUR13 had type II infection by *S. sclerotiorum*. Semivariance was calculated from lettuce drop incidence in 2-by-2-m quadrats in the direction of plant rows (0º) or at 45, 90, and 135º relative to plant rows. Disease incidence distribution in all the six fields displayed here was aggregated as suggested by the β-binomial distribution.

of various degrees. The two types of primary inoculum, however, resulted in disease progress curves with contrasting dynamics.

In general, the incidence of lettuce drop caused by *S. minor* (or type I infection) occurred in two phases. First, immediately after thinning, a few plants showed lettuce drop symptoms and died. The more important second phase, however, occurred after lettuce began forming heads, with more plants exhibiting lettuce drop symptoms just prior to crop harvest. Factors such as soil moisture and temperature notably affect sclerotial germination, and optimal conditions for germination are ideal just prior to harvest (1,3,9,26). Because of the rapid expansion of the heads and the tap root during this phase, the frequency of irrigation was greatest during this period. In addition to providing optimal conditions for the germination of sclerotia during this phase, the probability of expanding tap roots coming in contact with the mycelium also was greatest (highest competence volume for sclerotia sensu Grogan; 10). This rapid increase in the incidence of lettuce drop resulted in disease progress curves typical of a monomolecular model. The level of disease incidence caused by *S. minor* in the fields examined showed a high degree of correlation with the density of sclerotia in the soil, which is another attribute of diseases associated with the monomolecular model (3,9).

In contrast, incidence of lettuce drop caused by type II infections by *S. sclerotiorum* was sudden and high. Factors such as soil moisture and temperature affect both the timing and production of apothecia and release of ascospores (1,5,14,28). Rainfall, wind speed and direction, and leaf wetness can potentially affect ascospore discharge, deposition, and germination on lettuce leaves, with the lettuce growth stage during these events influencing infection (1). Because of the ephemeral nature of apothecial production and ascospore discharge, lettuce drop incidence can increase quickly if the environmental conditions favorable for ascospore production, dispersal, and infection coincide (1,4). In the San Joaquin Valley, where only one crop of winter lettuce is grown each year, conditions that trigger production of apothecia and ascospore discharge generally are encountered during the rainy winter. The sudden incidence of high levels of lettuce drop in San Joaquin Valley fields, a lack of subsequent disease increase in the same fields, and the symptoms observed on infected plants all suggest that airborne ascospores were the cause of infection. Although infection directly by mycelial germination of sclerotia in soil and by airborne ascospores from carpogenic germination of sclerotia both possibly could occur in the same fields at any given time, the symptoms observed suggest that the proportion of plants infected by mycelial germination of sclerotia of *S. sclerotiorum* was very low. Furthermore, very few *S. sclerotiorum* sclerotia were detected in soil samples from these fields, and there was no correlation between the numbers of sclerotia in soil and the level of disease incidence (4,28,30). These observations strongly suggest that the vast majority of lettuce drop caused by *S. sclerotiorum* in the current study was due to infection by airborne ascospores.

The source of inoculum and the type of infection it causes likely determine the spatial patterns of lettuce drop. Lettuce drop from type I infection caused by both *S. minor* and *S. sclerotiorum* had similar spatial distributions. Lettuce drop from type II infection by ascospores, however, resulted exclusively in aggregated spatial patterns. These results were at variance with our initial hypothesis that infection by mycelial germination of soilborne inoculum is likely to result in aggregated patterns, and infection by airborne inoculum is likely to result in a random distribution of lettuce drop. Although lettuce drop incidence showed aggregated distributions from both types of infection, they likely resulted from different epidemiological processes. As a soilborne disease, the distribution of type I infection depends largely on the distribution of propagules in the soil. The propagule distribution in soil is affected by agricultural activities that precede or follow a crop (35). Activities such as soil disking, ripping, and other tillage operations will influence the sclerotial distribution and, consequently, the disease distribution. The pattern of disease incidence from type I infection also is predictable from the patterns of sclerotial distribution (9). Furthermore, the prevalence of moisture in the top 5-cm soil profile will determine whether the plant will be infected or not when the inoculum is

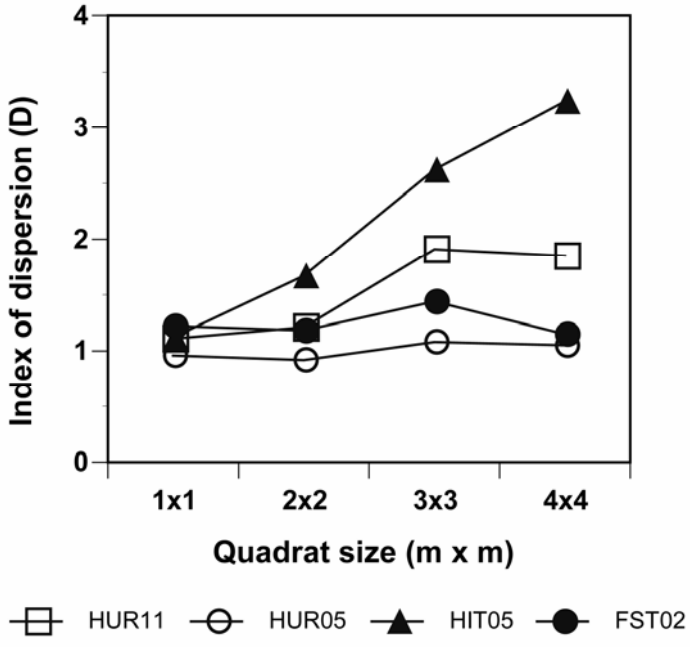

Fig. 6. Effects of quadrat sizes on the index of dispersion of lettuce drop incidence caused by *Sclerotinia minor* and *S. sclerotiorum* in selected commercial fields in California. Fields HUR05, HIT05, and FST02 had type I infection, whereas field HUR11 had type II infection. Disease incidence was aggregated in fields HUR11, HIT05, and FST02 and random in field HUR05.

Table 3. Effects of quadrat sizes on the spatial pattern of lettuce drop incidence[x]

		Geostatistics[y]	
Quadrat size (m)	**Mean of D^z**	**Mean of sill-nugget**	**Mean of range**
1 by 1	1.1756 a	13.753 a	6.850 a
2 by 2	1.4436 ab	12.350 a	11.700 b
3 by 3	1.6848 b	16.300 a	16.200 b
4 by 4	2.1752 c	13.900 a	15.100 b

[x] Means followed by the same letters are not significantly different ($P \leq 0.05$) according to the least significant difference test.

[y] In the semivariogram (plot of semivariance versus distance between samples), the range is the distance at which the semivariance reaches a plateau. The sill is the plateau that the semivariance reaches at the range. The nugget is the vertical jump from the value of 0 at the origin to the value of the variogram at extremely small separation distances.

[z] Index of dispersion (D), calculated by the ratio of observed variance over theoretical variance for a binomial distribution.

present (34). The distribution of type II infections, however, may depend partially on the distribution of the soilborne sclerotia but also on the environmental conditions that affect ascospore discharge and deposition. For both types of infection, the degree of aggregation was positively correlated with incidence of lettuce drop (i.e., higher incidence lead to higher degree of aggregation and vice versa; 9). Thus, spatial patterns in plots with low inoculum levels were not aggregated.

The distribution of sclerotia producing apothecia also can affect the distribution of lettuce drop caused by ascospore infections. Ascospore discharge from the source usually results in a gradient with the greatest number of ascospores being deposited near the source (5). Similarly, ascospore discharge and deposition from each sclerotium of *S. sclerotiorum* with apothecia also can result in a gradient that, in turn, can lead to aggregated patterns of lettuce drop incidence. Furthermore, the distribution of sclerotia can be affected by past cropping, cultivation, and disease history (*unpublished*). The number of sclerotia of *S. sclerotiorum* in fields evaluated in this study was very low, and the spatial pattern of type II infections may be more complicated when higher numbers of germinated sclerotia with apothecia are encountered within a field. Although no apothecia were observed at Huron during the course of this study, they were observed at the same location subsequently (B. M. Wu, *personal communication*). No other crops that are susceptible to *Sclerotinia* spp. were observed within at least 800 m of the fields evaluated in this study. Therefore, ascospores from the sclerotia within the fields studied were likely to have caused the lettuce drop that was observed. Although it is possible that ascospores also could have come from more remote fields (28), the results from geostatistical analyses suggest that the inoculum came from within the fields studied. Spatial dependence was observed only up to a maximum of 10 m in all fields, suggesting the local origin of the inoculum.

An anisotropic effect was detected for both types of infection in a few lags in the geostatistical analyses. However, the mechanisms that resulted in the spatial dependence of disease may be different for the two *Sclerotinia* spp. For *S. minor*, spatial dependence was limited to areas of less than 8 by 8 m, suggesting the importance of tillage operations. For *S. sclerotiorum*, however, the disease spread mostly in one or two directions, indicating possible influence of air currents prevalent at the time of ascospore dispersal. The semivariance changed significantly in 5 to 10 m in most aggregated distributions of type II infections; therefore, the ascospore dispersal appears to have had a small zone of influence.

The choice of quadrat size can have a significant effect on the spatial pattern analyses of plant diseases. Point analysis of the data in this study demonstrated that, although the quadrat size significantly affected the degree of aggregation of lettuce drop distribution in some plots, variation in quadrat size did not affect the spatial patterns. These results are consistent with those obtained by others in other pathosystems (24,25). The geostatistical analyses, however, were insensitive to the quadrat sizes chosen, with the exception of the 1-by-1-m quadrat.

In summary, results from this study demonstrate that the source of inoculum and the type of infection caused are most likely to determine spatial patterns of lettuce drop in the field. Lettuce drop from direct germination of *S. minor* and *S. sclerotiorum* sclerotia had similar spatial dis-

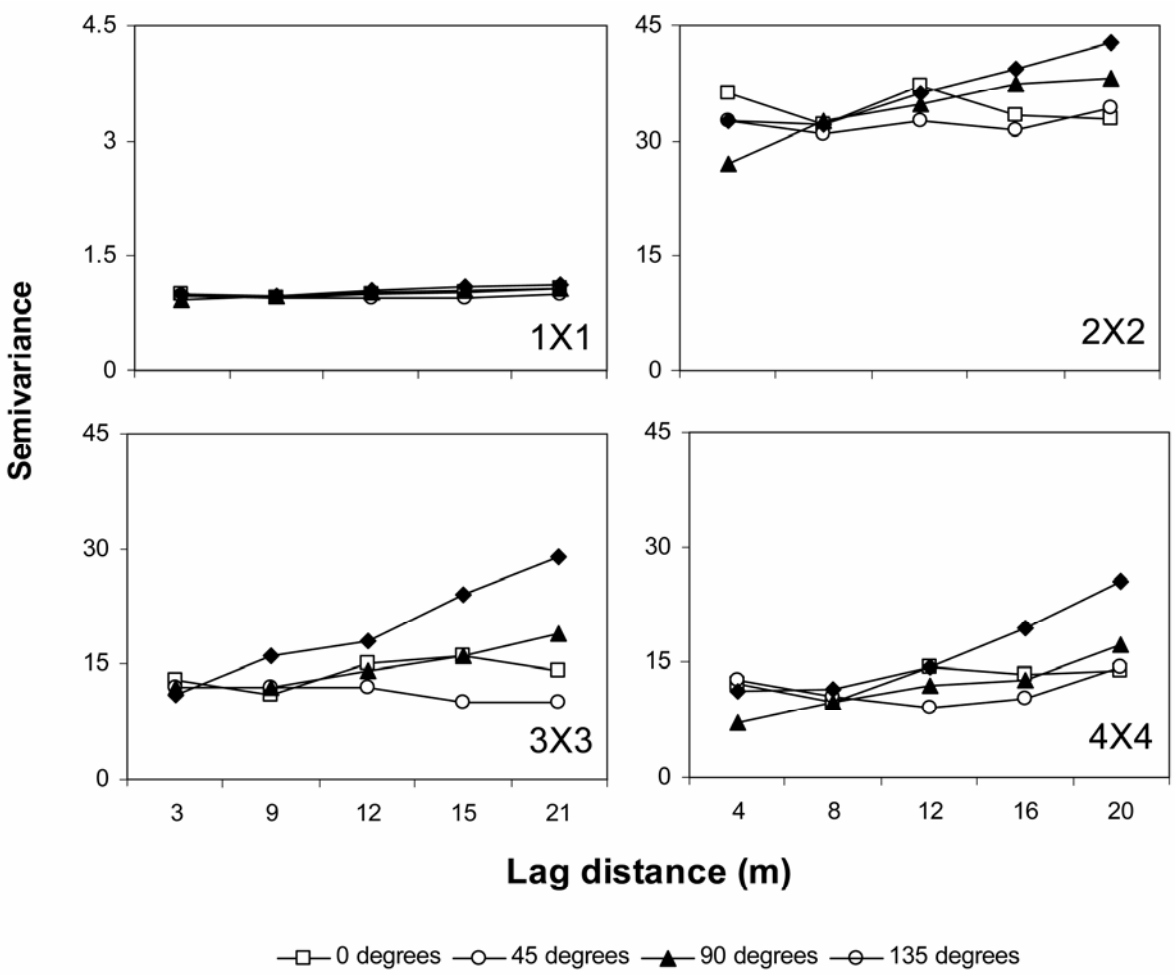

Fig. 7. Semivariograms of lettuce drop incidence caused by *Sclerotinia minor* for the four quadrat sizes in four directions. Data from field FST01 are shown as an example.

tributions. Lettuce drop from infections by *S. sclerotiorum* ascospores, however, resulted exclusively in aggregated spatial patterns. These results were at variance with our initial hypothesis that infection by mycelial germination of soilborne inoculum is likely to result in aggregated patterns and infection by airborne inoculum is likely to result in a random distribution of lettuce drop. Although lettuce drop incidence showed aggregated distributions from both types of infection, they likely resulted from different epidemiological processes. The pattern of disease incidence from direct germination of sclerotia also was predictable from the patterns of sclerotial distribution.

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