

## Single and Combined Effects of the Lesion Nematode and *Colletotrichum graminicola* on Growth and Anthracnose Leaf Blight of Corn

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### ABSTRACT

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Corn plants were grown to maturity in the greenhouse in the presence or absence of the lesion nematode (*Pratylenchus hexincisus*) (initial soil populations of 89.6 and 0.6 nematodes per 50 cc, respectively), and leaves were inoculated with the fungus at 14, 35, or 55 days after planting. The effects of the nematode, the fungus and the nematode  $\times$  fungus interaction were assessed by measuring disease development, leaf senescence, and various aspects of plant growth. Anthracnose leaf blight severity increased significantly in plants that were also infected with the nematode. Plants grown either in the presence of the nematode or inoculated with the fungus

at all times also senesced faster than the controls, and the nematode and fungus together caused an even greater rate of individual leaf senescence. Dry root weight, but not shoot weight, was significantly reduced by nematodes. Plants inoculated with the fungus at the seedling stage had significantly lower root and shoot dry weights. The fungus, but not the nematode, significantly reduced extended leaf height and stem circumference. The results demonstrate that nematode infection in corn can cause earlier appearance and increase the severity of anthracnose symptoms.

*Additional key words:* pathogen interactions, susceptibility, *Zea mays*.

Anthracnose leaf blight, caused by the fungus *Colletotrichum graminicola* (Ces.) Wils, has become an economic threat to corn (*Zea mays* L.) production throughout the corn belt and eastern corn-growing regions of the United States (8,16). Normally, the disease occurs at the seedling stage and then again around the time of anthesis. When it occurs at intermediate stages of plant growth the disease is severe and typically appears in irregular patterns within a field. The infrequency of disease at intermediate stages of plant growth and the irregular pattern of disease development in the field suggest associations with other stress factors such as nematode infection.

Although lesion nematodes (*Pratylenchus* spp.) are important pathogens of corn (15,18), little has been reported on their influence on disease interactions in corn (19,20,23). The purpose of this investigation was to determine whether nematode infection influences anthracnose severity or alters the normal pattern of leaf blight development.

### MATERIALS AND METHODS

**Nematode.** *Pratylenchus hexincisus* Taylor and Jenkins was cultured on corn roots planted in field soil (Raub silt loam) in which experimental corn was to be grown. Containers for treatments lacking nematodes were kept fallow and dry for 9 mo prior to planting, and the Baermann funnel technique (1) was used to assay the soil in each container for nematode population and species determination. Sample suspensions (1 ml) from Baermann funnels were placed in a Peters eelworm slide, and nematodes were identified and counted with a compound microscope. Containers in which the nematode had been cultured averaged 89.3 nematodes (*P. hexincisus*) per 50 cc of soil. Although some nematodes survived the fallow and desiccation treatment, the level was sufficiently low (0.6 per 50 cc) that we regard this treatment as lacking nematodes at the time the experiment was initiated. This

method of reducing nematode populations was chosen over steam or chemical sterilization to avoid drastic changes in soil microflora which might mask the role of the nematode.

**Plant material.** Three seeds of the corn hybrid Mo17<sub>HI</sub>  $\times$  B73<sub>HI</sub>, which is susceptible to *C. graminicola* (25), were planted in each of 96, 56-L containers. Upon emergence, the seedlings were thinned to one per container.

Prior to seed planting, the soil in each container was assayed for fertilizer requirements and adjusted to a rate equivalent to 224.5 kg N, 112.3 kg P, and 112.3 kg K/ha. An additional application of fertilizer equivalent to 84.2 kg N, 42.1 kg P, 42.1 kg K/ha was applied topically to each container 30 days after seeding. Supplemental lighting (230  $\mu\text{E}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$  photosynthetically active radiation at a distance of 50 cm from plants) was supplied from fluorescent tubes. Plants were watered uniformly to avoid water deficit.

**Fungus.** *C. graminicola* was cultured on oatmeal agar under fluorescent light (60  $\mu\text{E}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ ). Spores were harvested from 10-day-old cultures, and suspensions were adjusted to  $5 \times 10^5$  spores per milliliter. Tween-20 (50  $\mu\text{l}$ /100 ml) was added to suspensions as a wetting agent.

**Experimental design and inoculation treatments.** There were eight treatment groups. Plants were grouped into nematode and no-nematode treatments which were further divided into four groups according to inoculation with the fungus. The four inoculation treatments included an uninoculated control and groups inoculated with the fungus at 14, 35, and 55 days after planting. Plants in the various treatment groups were arranged in a completely randomized design in the greenhouse to minimize potential differences in heating, cooling, and light exposure. At the times of inoculation, 4, 7, and 13 leaves, respectively, had emerged from the whorl.

Plants were inoculated with spore suspensions from an atomizer pressurized at 0.5 atmospheres, and each leaf on each plant was inoculated. After inoculation, leaves were sprayed with a fine mist of water to ensure that they were covered with water droplets, and the plants were then enclosed in moisture chambers which surrounded the entire plant and growth container. Moisture chambers were constructed from plastic bags and wire fencing. The experiments were carried out from October through January and repeated twice. Results presented are from the second experiment.

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**Disease development and leaf senescence.** Disease development was measured at 7-day intervals after inoculation by a visual estimate of percent lesion coverage on each leaf. To avoid bias, plants were coded by one experimenter and independently rated by two others. Because the same two individuals did the rating throughout the experiment, variance in visual ratings should be minimized.

Leaves were rated for senescence seven times at weekly intervals beginning 35 days after planting. Senescence was considered to have begun when the leaf tip exhibited a characteristic yellow-orange color. Ratings were assigned quantitative values on a scale from 0 through 2 in which 0 represented no senescence and 2 represented complete (100%) discoloration of the leaf, regardless of whether the tissue was also desiccated.

**Extended leaf height and stalk circumference.** Extended leaf height of each plant was measured as the distance from the soil line to the tip of the highest leaf at weekly intervals beginning 15 days after planting. Stem circumference at the middle of the first internode above the soil surface was also measured at weekly intervals after planting.

**Dry root and shoot weights and final nematode counts.** At 85 days after seeding, the plants were separated into root and shoot tissues. Soil was gently washed from roots, and the root and shoot tissues from each plant were separately bagged and placed in an air-flow grain dryer at 38–40 C for 14 days before being weighed. Before the roots were dried, 10 g samples of root tissue were assayed for the presence of lesion nematodes by placing them in a mist chamber for 1 wk. Nematodes that emerged were counted, the root tissues were oven-dried (100 C for 24 hr), and the number of nematodes per gram of dry root tissue was determined.

**Statistical analyses.** Data were analyzed by two-, three-, and four-way analyses of variance (ANOVA), and differences in means were evaluated by using the Student-Newman-Keuls sequential range test. Some data were recorded at weekly intervals and these sequential observations were treated in the ANOVA as a split plot in time. Because some leaves were lost during the course of the experiment, data were analyzed to account for unequal sample number. Analyses were as follows: extended leaf height and stem circumference, a  $4 \times 2 \times 7$  (inoculation group  $\times$  nematode  $\times$  observation date) factorial ANOVA; root and shoot weights, a  $4 \times 2$  (inoculation group  $\times$  nematode) ANOVA; percentage disease, a  $2 \times 4 \times 3$  (nematode  $\times$  leaf number  $\times$  assessment date) ANOVA for the 14-day inoculation group, a  $2 \times 7 \times 6$  ANOVA for the 35-day inoculation group, and a  $2 \times 9 \times 3$  ANOVA for the 55-day inoculation group (nematode presence or absence was a between factor and leaf number and assessment date were within factors);

TABLE 1. Split-plot (in time) analysis of variance of extended leaf height and stem circumference of corn plants as affected by *Pratylenchus hexincisus* and *Colletotrichum graminicola*

Source of variation <sup>y</sup>	Extended leaf height		Stem circumference	
	Degrees of freedom	Mean square <sup>z</sup>	Degrees of freedom	Mean square <sup>z</sup>
Nematode	1	237.47	1	242.26
Inoculation group	3	8,670.50***	3	4,044.98***
Nematode $\times$ inoculation group	3	59.87	3	49.23
Error a	84	318.26	84	73.38
Observation date	6	86,949.68***	6	4,236.75***
Observation date $\times$ nematode	6	56.38**	6	11.16*
Observation date $\times$ inoculation group	18	441.99***	18	23.34***
Observation date $\times$ nematode $\times$ inoculation group	18	22.13	18	4.02
Error b	504	18.24	504	4.45

<sup>y</sup>Nematode = presence or absence of the nematode; inoculation group = uninoculated plants and plants inoculated with the fungus at 14, 35, or 55 days after planting; and observation date = times of data collection.

<sup>z</sup>Asterisks \*, \*\*, and \*\*\* indicate significance at  $P < 0.05$ , 0.01, and 0.001, respectively.

leaf senescence, a  $4 \times 2 \times 7 \times 10$  (inoculation group  $\times$  nematode  $\times$  assessment date  $\times$  leaf number) ANOVA with inoculation group and nematode as between factors and dates and leaf number as within factors; senescence by inoculation group, a  $2 \times 7 \times 10$  (nematode  $\times$  assessment date  $\times$  leaf number) ANOVA for the uninoculated control and the 14-, 35-, and 55-day inoculation groups (nematode presence or absence was a between factor and assessment dates and leaf number were within factors).

## RESULTS

**Extended leaf height and stem circumference.** For extended leaf height data, the main effect of fungal inoculation was significant. But neither the main effect of the nematode nor the interaction of nematode with fungal inoculation group was significant (Table 1). As expected, there were significant differences based on when the data were collected (observation date).

Inoculation with *C. graminicola* at 14 days after planting significantly reduced extended leaf height, whereas inoculation at 35 days after planting had no effect on extended leaf height (Fig. 1). A Student-Newman-Keuls test ( $P = 0.05$ ) conducted on data for each observation date showed that, beginning on the second observation date, plants in the first inoculation group (14 days) were significantly shorter than plants in other inoculation groups.

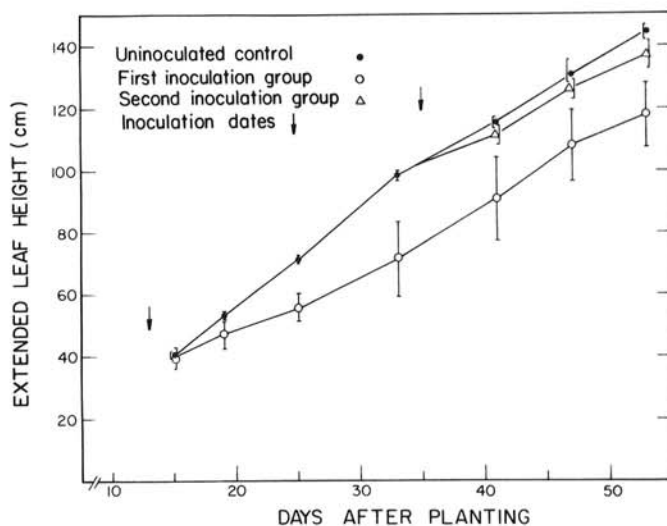


Fig. 1. Extended leaf height of corn as influenced by inoculation with *Colletotrichum graminicola* at different plant ages. First and second inoculation groups represent inoculations at 14 and 35 days after planting. Values are means and the vertical bars represent the 95% confidence limits for no fewer than 12 samples.

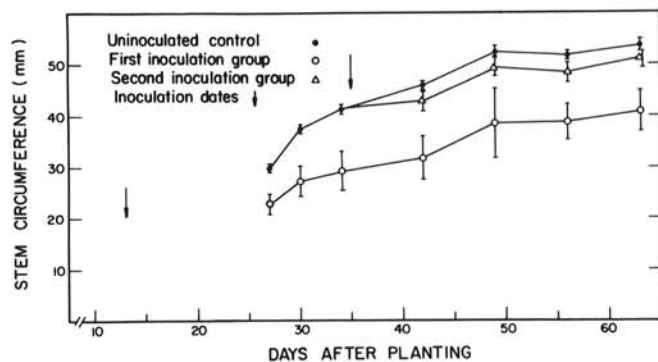


Fig. 2. Stem circumference of corn as influenced by inoculation with *Colletotrichum graminicola* at different plant ages. First and second inoculation groups represent inoculations at 14 and 35 days after planting. Values are means and the vertical bars represent 95% confidence limits for no fewer than 12 samples.

TABLE 2. Factorial analysis of variance of root and shoot dry weights of corn plants as affected by *Pratylenchus hexincisus* and *Colletotrichum graminicola*

Source of variation <sup>y</sup>	Dry root weight		Dry shoot weight	
	Degrees of freedom	Mean square <sup>z</sup>	Degrees of freedom	Mean square <sup>z</sup>
Nematode	1	83.67*	1	1,215.14
Inoculation group	3	246.47***	3	9,449.77***
Nematode × inoculation group	3	7.14	3	502.56
Error	84	21.00	84	839.27

<sup>y</sup>Nematode = presence or absence of the nematode, and inoculation group = uninoculated plants and plants inoculated with the fungus at 14, 35, 55 days after planting.

<sup>z</sup>Asterisks \* and \*\*\* indicate significance at  $P < 0.05$  and  $0.001$ , respectively.

TABLE 3. Dry root and shoot weights of corn plants infected with *Colletotrichum graminicola* and *Pratylenchus hexincisus*<sup>y</sup>

Treatment	Dry weight (g)	
	Roots	Shoots
Nematodes		
Absent	10.86 a	75.03 a
Present	8.58 b	66.33 a
Fungal inoculation group		
Uninoculated	11.66 a	89.79 a
Inoculated at 14 days	4.16 b	35.12 b
Inoculated at 35 days	9.97 a	79.86 a
Inoculated at 55 days	13.09 a	77.94 a

<sup>y</sup>Values are means and when followed by the same letter they do not differ significantly ( $P = 0.05$ ) according to a Student-Newman-Keuls' sequential range test. Means represent no fewer than 45 samples for nematode treatments and no fewer than 11 for fungal inoculation group treatments.

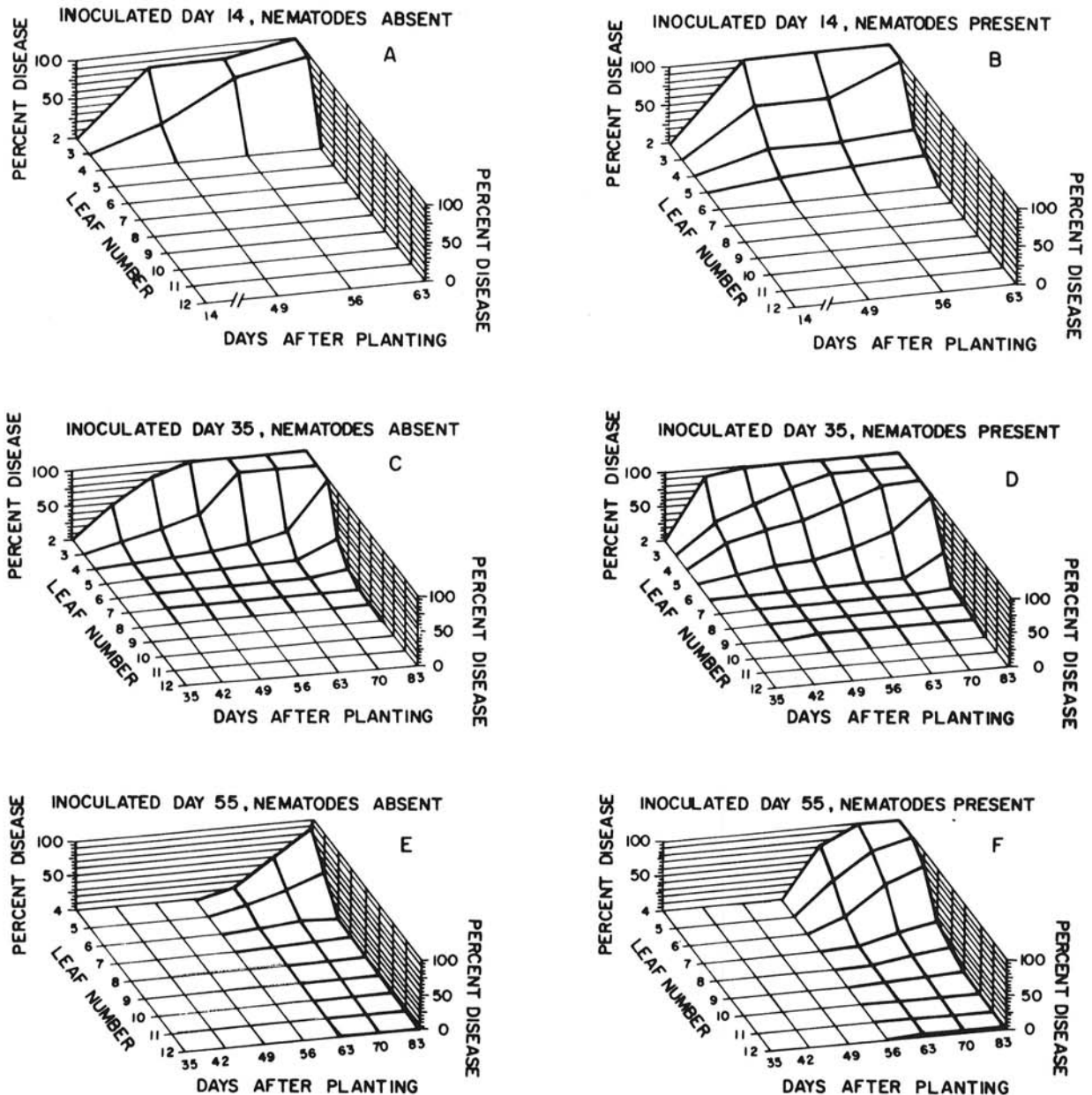


Fig. 3. Influence of infection by the nematode *Pratylenchus hexincisus* on foliar leaf blight caused in corn by the fungus *Colletotrichum graminicola*. A and B, leaves inoculated with the fungus 14 days after planting and with nematodes either A, absent or B, present. C and D, leaves inoculated with the fungus at 35 days after planting and with nematodes either C, absent or D, present. E and F, leaves inoculated with the fungus at 55 days after planting and with nematodes either E, absent or F, present.

Leaf heights for plants in the second and third inoculation groups did not differ from uninoculated controls at any time. Data in Fig. 1 represent the mean extended leaf heights for inoculation groups regardless of nematode presence since nematodes did not affect plant height for any inoculation group (Table 1).

For stem circumference there were significant main effects of fungal inoculation and observation date and significant interactions of observation date with nematode, and observation date with inoculation group (Table 1). No other main effects or interactions were significant.

As with extended leaf height, inoculation with *C. graminicola* significantly reduced stem circumference of plants inoculated at 14 days after planting (Fig. 2). Inoculation at 35 days after planting also significantly reduced stem circumference, and the reduction was evident as early as 7 days after inoculation (Fig. 2). Circumferences in the 55-day group did not differ from those of the uninoculated controls at any time. Data in Fig. 2 represent the mean circumference for inoculation groups regardless of nematodes since nematodes did not affect circumference for any inoculation group (Table 1).

**Dry root and shoot weights.** Analysis of dry root weight data showed significant main effects for nematode and inoculation group, but the interaction between these factors was not significant (Table 2). Analysis of shoot weight data showed that inoculation was the only significant main effect and that neither the presence of the nematode nor the nematode  $\times$  inoculation group interaction affected shoot weight (Table 2).

Dry root weights of plants grown in the presence of nematodes were significantly lower than plants grown in the absence of the

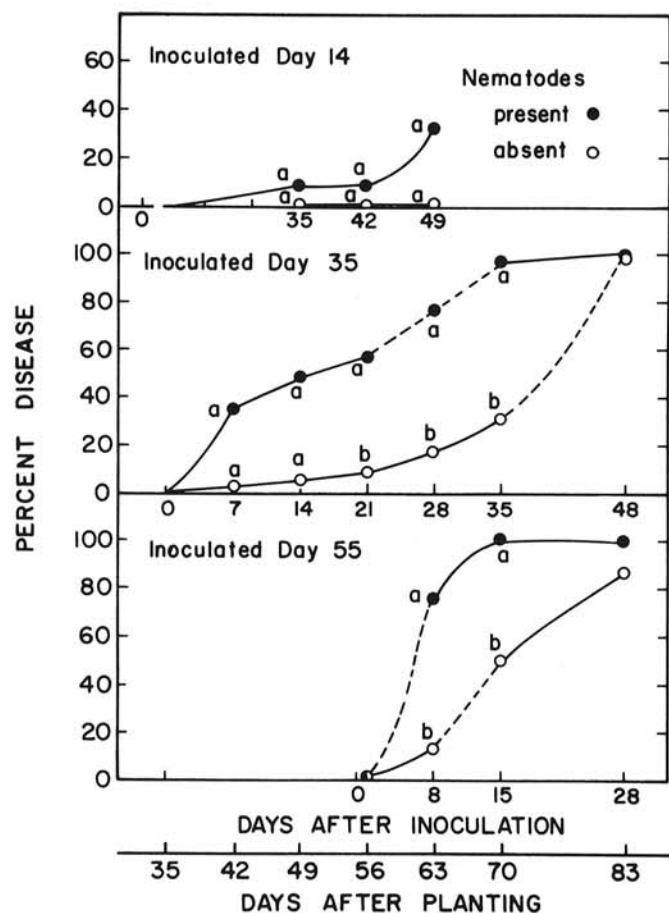


Fig. 4. Disease development on leaf 4 for corn plants grown in the presence or absence of the nematode *Pratylenchus hexincisus*, and inoculated with the fungus *Colletotrichum graminicola* at 14, 35, or 55 days after planting. Letters indicate significant differences ( $\alpha = 0.05$ ) in nematode treatment groups. Dashed lines indicate a significant change ( $\alpha = 0.05$ ) in the amount of disease between two successive observations in the same treatment group.

TABLE 4. Split-plot analysis of variance of percent lesion coverage and senescence in corn leaves as affected by *Pratylenchus hexincisus* and *Colletotrichum graminicola*

Source of variation <sup>y</sup>	Lesion coverage (%)		Senescence	
	Degrees of freedom	Mean square <sup>z</sup>	Degrees of freedom	Mean square <sup>z</sup>
<b>First fungal inoculation group (inoculated at 14 days)</b>				
Nematode	1	1,722.66	1	0.004
Error a	6	1,763.89	6	0.69
Assessment date	2	3,603.18**	6	2.90***
Date $\times$ nematode	2	131.30	6	0.03
Error b	12	292.92	36	0.06
Leaf number	3	46,911.82***	9	43.04***
Leaf $\times$ nematode	3	220.16	9	0.05
Error c	18	582.59	54	0.19
Date $\times$ leaf	6	1,736.93***	54	0.57***
Date $\times$ leaf $\times$ nematode	6	325.47	54	0.03
Error d	36	242.73	324	0.06
<b>Second fungal inoculation group (inoculated at 35 days)</b>				
Nematode	1	53,120.16***	1	9.55***
Error a	19	1,354.75	20	0.29
Assessment date	5	29,018.55***	6	26.96***
Date $\times$ nematode	5	81.29	6	0.10
Error b	95	176.58	120	0.09
Leaf number	6	167,772.11***	9	93.19***
Leaf $\times$ nematode	6	7,067.92***	9	0.73***
Error c	114	411.02	180	0.12
Date $\times$ leaf	30	4,440.91***	54	3.28***
Date $\times$ leaf $\times$ nematode	30	1,659.43***	54	0.38***
Error d	570	140.76	1,080	0.07
<b>Third fungal inoculation group (inoculated at 55 days)</b>				
Nematode	1	45,674.34***	1	4.47*
Error a	20	1,281.26	20	0.34
Assessment date	2	16,824.72***	6	37.58***
Date $\times$ nematode	2	860.61*	6	0.93***
Error b	40	166.64	120	0.09
Leaf number	8	45,173.29***	9	84.39***
Leaf $\times$ nematode	8	6,892.31***	9	0.54***
Error c	160	349.61	180	0.08
Date $\times$ leaf	16	2,479.13***	54	4.07***
Date $\times$ leaf $\times$ nematode	16	1,121.31***	54	0.26***
Error d	320	134.50	1,080	0.05
<b>Uninoculated</b>				
Nematode			1	9.09***
Error a			39	0.23
Assessment date			6	53.56***
Date $\times$ nematode			6	0.24*
Error b			234	0.09
Leaf number			9	164.17***
Leaf $\times$ nematode			9	0.53***
Error c			351	0.71
Date $\times$ leaf			54	6.97***
Date $\times$ leaf $\times$ nematode			54	0.49***
Error d			2,106	0.05

<sup>y</sup>Nematode = presence or absence of the nematode; assessment date = number of times data were collected; and leaf number = number of leaves considered on a plant.

<sup>z</sup>Asterisks \*, \*\*, and \*\*\* indicate significance at  $P < 0.05$ , 0.01, and 0.001, respectively.

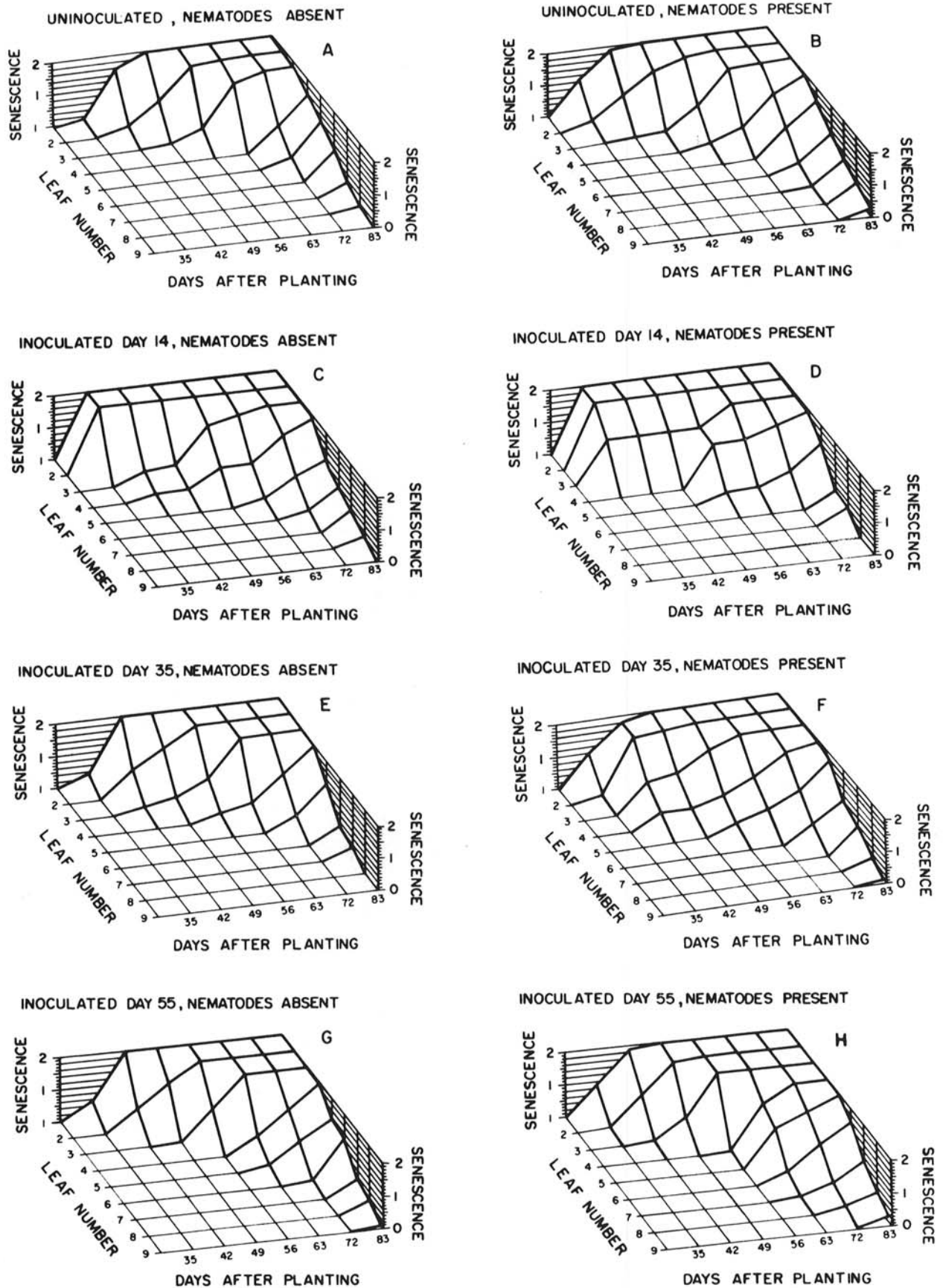


Fig. 5. Influence of root infections by the nematode *Pratylenchus hexincisus* and leaf infections by the fungus *Colletotrichum graminicola* on leaf senescence in corn. **A and B**, leaves not inoculated with the fungus and with nematodes either **A**, absent or **B**, present. **C and D**, leaves inoculated with the fungus at 14 days after planting and with nematodes either **C**, absent or **D**, present. **E and F**, leaves inoculated with the fungus at 35 days after planting and with nematodes either **E**, absent or **F**, present. **G and H**, leaves inoculated with the fungus at 55 days after planting and with nematodes either **G**, absent or **H**, present.

nematode (Table 3). However, nematodes had no significant effect on dry shoot weights (Table 3).

Inoculation with *C. graminicola* at 14 days after planting significantly reduced both dry root weight and dry shoot weight in comparison to other times of inoculation and to the uninoculated controls (Table 3).

**Influence of plant age at the time of inoculation with *C. graminicola* and presence of the nematode on anthracnose leaf blight development.** Three 3-way analyses of variance for percentage disease were performed, one for each inoculation group (Table 4). The separate analyses take into account that only leaves present at the time of inoculation could develop disease since secondary spread of anthracnose does not occur under greenhouse conditions.

Significant main effects of assessment date and leaf number were found for the first inoculation group (Table 4). The date  $\times$  leaf interaction was also significant, but no other main effect or interaction was significant, which indicates that neither the main effect of nematodes nor any interaction involving nematodes was significant for plants inoculated at 14 days after planting.

For the second inoculation group the main effects of nematode, assessment date, and leaf number were significant as were the interactions of leaf  $\times$  nematode, assessment date  $\times$  leaf, and date  $\times$  leaf  $\times$  nematode. The date  $\times$  nematode interaction was not significant,  $P > 0.05$ .

For the third inoculation group, significant main effects of nematode, assessment date, and leaf number were found. The interactions of date  $\times$  nematode, leaf  $\times$  nematode, date  $\times$  leaf, and date  $\times$  leaf  $\times$  nematode were also significant.

The results demonstrated that when plants were inoculated with the fungus at 35 or 55 days, but not at 14 days, disease severity was greater when plants were also infected with nematodes (Figs. 3 and 4). Fig. 3A to F are three-dimensional plots of the means of percentage disease development for each leaf in each inoculation and nematode treatment group over the duration of the experiment. Graphs compare total disease development in the presence and absence of the nematode. Note that even for the 14-day inoculation group there was a trend toward greater disease severity in the presence of nematodes (Fig. 3A versus 3B) although the difference was not significant.

Student-Newman-Keuls' analyses of percent lesion coverage for each leaf in all six treatment groups were also carried out. Only the analyses for leaf 4 are presented here. Since data profiles for all leaves were similar, leaf 4 was chosen because it was present in each inoculation group. Fig. 4 presents the relationship between percent disease (lesion coverage) and the presence or absence of nematodes for leaf 4. For the 14-day inoculation group, the nematode had no significant effect on disease development. In the 35-day inoculation group a difference in disease severity due to the presence of the nematode was evident within 7 days after inoculation, but this did not become statistically significant until 21 days after inoculation. This difference is more striking in the 55-day group where at 8 days after inoculation disease severity was 75% with nematodes present compared to only 12% when nematodes were absent.

**Influence of *P. hexincisus* and time of inoculation with *C. graminicola* on leaf senescence.** A four-way analysis of variance on senescence data demonstrated statistical significance throughout the model. Therefore, as with disease development, separate analyses of variance for leaf senescence were performed on the data from each inoculation group (Table 4).

In the first inoculation group the main effects of assessment date and leaf number and the interaction of date  $\times$  leaf were significant. In each of the other inoculation groups all main effects were significant and all interactions (except date  $\times$  nematode for the second inoculation group) were also significant (Table 4).

As with disease development, the results for senescence are presented as three-dimensional graphs (Fig. 5A-5H). Uninoculated leaves tended to senesce more rapidly when plants were infected with nematodes (Fig. 5A versus 5B). Similarly, in the absence of nematodes, leaves inoculated with *C. graminicola* senesced more rapidly than uninoculated leaves (Fig. 5C, E, and G versus 5A). Leaves senesced even more rapidly when plants were infected with

both nematodes and the fungus than with the fungus alone (Fig. 5F versus 5E, 5H versus 5G).

Student Newman Keuls' analyses ( $P = 0.05$ ) were carried out for senescence of each leaf in all eight treatment groups. As for disease development, only the analyses for leaf 4 are presented (Fig. 6). Results show that except for the 14-day inoculation group, senescence was more advanced and began earlier when nematodes were present (Fig. 6).

**Final nematode counts.** Nematode populations were significantly higher in treatments in which nematode levels were substantial at the beginning of the experiment than in treatments where nematode levels were very low. There was no evidence of an effect of fungal inoculation on nematode populations in either nematode treatment group. The range of means for treatments with low initial nematode counts was 1,813 to 3,033 nematodes per gram of dry root tissue whereas the means for treatments with high initial counts ranged from 6,698 to 10,835 nematodes per gram of dry root tissue. The seemingly high populations obtained from roots of plants grown in soil with the initially low larval and adult nematode population probably resulted from eggs being present before planting. In any case, the final counts should only be considered to

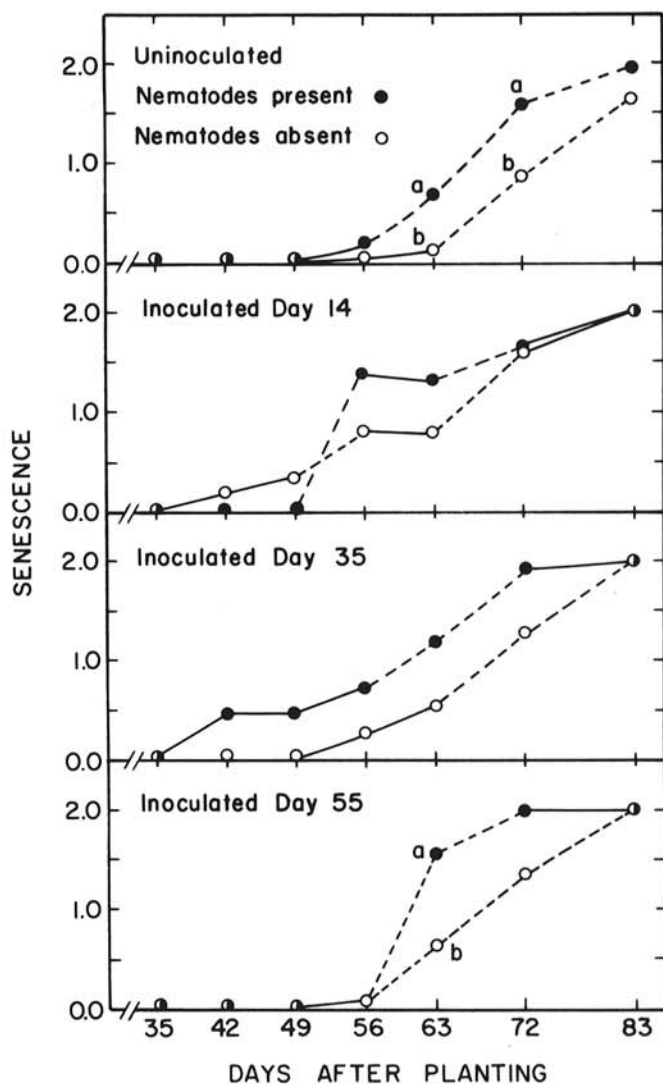


Fig. 6. Senescence of leaf 4 for corn plants grown in the presence or absence of the nematode *Pratylenchus hexincisus* and inoculated with the fungus *Colletotrichum graminicola* at 14, 35, or 55 days after planting. Letters indicate significant differences ( $\alpha = 0.05$ ) between nematode treatment groups. Dashed lines indicate a significant change ( $\alpha = 0.05$ ) in the amount of senescence between two successive observations in the same treatment group.

be rough estimates of the number of nematodes present while the root system was still alive.

## DISCUSSION

Results of this study demonstrate that plants infected with the nematode developed significantly more leaf blight than those without nematode infection (Figs. 3 and 4). The effect was especially evident in older leaves and was most pronounced in plants inoculated with the fungus at intermediate stages of growth when leaves are normally less susceptible to the fungus. One explanation for this increase in disease severity is that infection with the nematode hastens leaf senescence (Figs. 5 and 6), and this would be expected to favor anthracnose leaf blight (14). In the field, such early leaf senescence and the associated increase in susceptibility to anthracnose could significantly increase the potential for disease development and late-season epidemics (14).

As the corn leaf matures, anthracnose lesions normally fail to develop from new infections until around the time of anthesis (8,17). Even then, it is the lower and older leaves that first exhibit the disease. Thus, *C. graminicola*, like many other pathogens, exhibits latent disease development in mature plant tissue. Since there is no evidence for appressorial dormancy in *C. graminicola* (28) and because infection from an appressorium occurs without delay (22), the eventual development of anthracnose lesions in mature leaves appears to depend on changes in leaf physiology. Such changes have not been described but may be related to senescence (5).

There is ample precedence in the literature to suggest that senescence is associated with susceptibility to *C. graminicola* and other pathogens. For example, Leonard and Thompson (14) showed that anthracnose leaf blight was most severe in older, senescing corn leaves, and Katsanos and Pappelis (12) proposed that anthracnose stalk rot in sorghum was correlated with the onset of pith senescence. Rupe et al (24) reported that leaf senescence is important in gray leaf spot of corn, a disease that is similar to anthracnose. They found the onset and subsequent development of the disease to be influenced by the physiological age of the plant with initial symptoms occurring at anthesis on older leaves and eventually moving progressively up the plant. Payne and Waldron's (21) observation that gray leaf spot did not develop until late in the season even though inoculum was present supports the hypothesis that disease development is affected by leaf age (24).

Physical and chemical stresses also affect leaf senescence. For example, Schneider and Pendery (26) demonstrated that water stress applied in early stages of the growth of corn resulted in senescence of stalks and roots at an earlier-than-normal time. Importantly, water stressed plants were then predisposed to stalk rot. Hodges (7) attributed increased susceptibility of *Poa pratensis* to *Drechslera sorokiniana* to herbicide-induced senescence.

If senescence is a key factor in anthracnose leaf blight, then other stresses that hasten senescence should also favor earlier-than-normal blight development. Clearly, water stress both speeds senescence and augments stalk rot development (29). Our data demonstrate that *P. hexincisus* significantly reduced root mass (Table 3) and that this could also be expected to reduce the ability of the plant to take up water.

Reports of the interaction of nematodes and disease complexes in corn have dealt primarily with root-infecting fungi such as *Fusarium* spp. and the potential for greater disease development as a result of nematode infection (13,19,29). The importance of *Pratylenchus* spp. as pests of corn has only recently received emphasis (4,10,15,18,30). In most cases the primary role of this nematode in disease complexes is seen as a wounding agent (3), but effects on plant parts other than the root cannot be ruled out (6,11,27).

Although large numbers of nematodes are probably required to reduce yield, little is known about the involvement and potential importance of nematodes in pest interactions in corn (18). Our results demonstrated that nematode populations which alone had no obvious effect on shoot growth (extended leaf height, shoot weight, and stem circumference), nevertheless were associated with a significant increase in the severity of anthracnose leaf blight.

Thus, nematode infection caused plants to develop the leaf blight symptoms sooner than they normally would have if the nematode had not been present.

Norton (18) has pointed to the problem of identifying nematode levels that are damaging to the plant, and McSorley and Ferris (15) and Jaffee (9) suggest that it is the number of nematodes present early in plant growth which is critical since that is when the root system is actively growing and more susceptible to nematode damage. Our results demonstrate that nematode infection early in the growth of the plant should be recognized as a potential stress factor that can increase the severity of foliar and stalk rot diseases such as the anthracnose complex. It is also important to note that dense nematode populations occur at random in the field (2) and this may explain the random occurrence of anthracnose leaf blight during mid-season growth periods when leaves are normally less susceptible to the fungus.

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