Influence of Rainfall, Drip Irrigation, and Inoculum Density on the Development of Phytophthora Root and Crown Rot Epidemics and Yield in Bell Pepper

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ABSTRACT

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Peppers (*Capsicum annuum*) were grown in three fields in which main plots were irrigated either on a more frequent or less frequent basis. Soil in subplots was left uninfested or was infested 5–6 wk after transplanting with one of three inoculum densities of *Phytophthora capsici*. In the Clayton field in 1988, where rainfall was low (16 cm), disease onset occurred 26 days after infestation in plots that were drip irrigated more frequently, whereas disease onset occurred 39 days after infestation in plots that were drip irrigated less frequently. Final disease incidence, the rate of disease increase, and mean area under the disease progress curve (AUDPC) values were significantly greater in infested plots irrigated on the more frequent than the less frequent schedule. More rainfall occurred at the Clinton and Clayton fields in 1988 and 1989, respectively (31 and 22 cm), than at the Clayton field in 1988. Disease in plants in the higher rainfall fields progressed rapidly at all inoculum densities after a single rainfall >2.0 cm, and onset was earlier (5–7 days after infestation) than at the low rainfall field. Final disease incidence was independent of the density of inoculum applied at all three locations. However, inoculum density and rainfall had significant linear effects on the mean AUDPC values and yield. Mean AUDPC from the three field locations were 24.4, 68.2, and 60.9%-days per day and mean yields were 10.4, 2.3, and 2.1 kg/plot in fields receiving 16, 22, and 31 cm of total rainfall, respectively. Irrigation also increased mean AUDPC values significantly when means were calculated over all locations. Mean AUDPC values were 57.5%-days per day in plots irrigated more frequently and 44.8%-days per day in plots irrigated less frequently. Irrigation increased disease progress to the greatest extent in the low rainfall field in Clayton in 1988. The cumulative centimeters of rainfall between infestation of plots and the final disease assessment was correlated negatively with yield ($r = -0.51$) and correlated positively with AUDPC ($r = 0.47$). There was a negative correlation between AUDPC and yield ($r = -0.84$), and a positive correlation between the time of disease onset and yield ($r = 0.73$). Plant-to-plant spread of *P. capsici* resulted in severe disease in uninfested plots in high rainfall fields, whereas less plant-to-plant spread occurred in the low rainfall field. Both rainfall and irrigation had larger effects on the time of onset and final incidence of disease than the range of inoculum densities evaluated. Disease incidence became very high when either a heavy (>2.0 cm) rainfall or more frequent drip irrigations occurred. Final disease incidence was independent of inoculum densities, indicating that Phytophthora root and crown rot is a truly polycyclic disease on pepper in North Carolina.

Phytophthora root and crown rot of bell pepper (*Capsicum annuum* L.), caused by *Phytophthora capsici* Leonian, is a severe disease on peppers, tomatoes, and cucurbits in many areas of the United States and worldwide. Although considerable research has been conducted on chemical control strategies and breeding for resistance to the disease (15,19), only a few studies have been conducted on the epidemiology of the disease in the field (3,25). Better management of Phytophthora root rots requires more specific knowledge of factors that influence epidemic development in the field.

Diseases caused by *Phytophthora* spp. are potentially polycyclic, with many cycles of inoculum production and infection occurring during a single season (4). Limited research has been done to evaluate the role of initial inoculum of *Phytophthora* spp. in disease development in the field (8,11,13,14). Low densities of inoculum may be sufficient to cause high levels of disease development for some species of *Phytophthora*. Inoculum densities of 0.5 colony-forming units per gram of soil (cfu/g) of *P. parasitica* var. *nicotianae* resulted in high levels of disease in a susceptible tobacco cultivar (11). The density of initial inoculum of *P. p. nicotianae* was related directly to disease incidence in a resistant but not a susceptible tobacco cultivar (8). Extreme aggregation of initial inoculum delayed disease development in the susceptible tobacco cultivar. Plant-to-plant spread of *P. p. nicotianae* was related to the level of host resistance (28). Analysis of data from epidemics caused by *P. p. nicotianae* indicated that the logistic model was the most frequently appropriate fit to disease progress data (5). Disease incidence and severity caused by *P. parasitica* in fields of furrow-irrigated processing tomato increased significantly with increased densities of inoculum applied to 6-wk-old plants of a susceptible cultivar (14). The influence of inoculum density on subsequent epidemic development caused by *P. capsici* in pepper fields has not been evaluated.

Changes in soil matric potential may significantly alter inoculum density-disease relationships and disease progress by affecting pathogen behavior directly (6,13). Saturated soil conditions associated with rainfall or irrigation can increase the severity of Phytophthora root rots by stimulating release of zoospores from sporangia and thus, increase the effective amount of inoculum present in soils and available for infection of plant roots increases, (2,3,24). Dispersal of inoculum also occurs during conditions of soil saturation (6). In addition, *P. capsici* produces pedicellate sporangia on infected tissue, which can be dispersed with wind-driven rain (3,22).

Growers in North Carolina routinely use drip irrigation to supplement natural rainfall for pepper production. The incidence of Phytophthora root and crown rot has increased in recent years in some irrigated fields (22). In the research reported here, experiments were conducted at three field locations to evaluate the interactive effects of rainfall and soil infestation with different inoculum densities of *P. capsici* on the development of Phytophthora root and crown rot epidemics and yield of bell pepper

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under more frequent and less frequent drip-irrigation episodes. A preliminary report of a portion of this work has been published (21).

MATERIALS AND METHODS

Inoculum production. *P. capsici* was isolated frequently from pepper and cucurbit plants in North Carolina between 1987 and 1989 (22). One isolate (Sc 2A, Type A1) pathogenic to pepper was used to infest soil in field plots in this study. Cultures were maintained on cornmeal agar (Difco Laboratories, Detroit, MI) slants or V8 agar in petri dishes (800 ml of water, 200 ml of V8 juice, 2 g of $CaCO_3$, 18 g of agar). Inoculum was prepared by culturing the fungus at 25 C for 6–7 wk in 500 cm^3 of vermiculite and 250 ml of V8 broth contained in 1-L mason jars. Inoculum consisted of hyphae and sporangia of the fungus.

Field experiments. Seeds of susceptible pepper cultivar Keystone Resistant Giant were planted in flats containing a 1:1 (v/v) mix of Norfolk sandy loam soil and Metro Mix 220 (W. R. Grace and Co., Cambridge, MA) in the greenhouse. Plants were thinned after emergence and fertilized with a modified Hoagland's solution (9).

Experiments were conducted in 1988 and 1989 on a Johns sandy loam soil at the Central Crops Research Station in Clayton, NC, and in 1988 on an Orangeburg loamy sand soil at the Horticultural Crops Research Station in Clinton, NC. Eight-week-old pepper seedlings were transplanted into raised single-row beds of soil fumigated previously with methyl bromide-chloropicrin (392 kg/ha, Great Lakes Chemical Company, West Lafayette, ID). Experimental units were 12.2 m long and beds were 1.1 m wide. Treatments were arranged in a split-plot design, with irrigation as main plots and densities of inoculum as subplots. All treatments were replicated four times. Irrigations were applied to main plots with a drip system. A single, drip-irrigation line (Typhoon 20, Netafim Irrigation Inc., Valley Stream, NY) with emmitters spaced 40 cm apart was buried 10 cm below the soil surface and approximately 10 cm from one side of the plant row in each plot. Plots were either irrigated less frequently (only after soil infestation with the pathogen or to avoid water stress) or more frequently, three times per week for a 4-h duration. Plots were not irrigated on days when rainfall occurred. Water was applied at a rate of 1.9 L/min per 30.5 m of row at a pressure of 68.9 kPa. All plots in the Clayton field area were drip irrigated uniformly for 4 h either 1 or 3 days after infestation with the pathogen in 1988 and 1989. Rainfall occurred within 2 days after infestation of the plots in the Clinton field in 1988.

Soil in subplots was either left uninfested or infested 43, 35, or 31 days after transplanting (Clayton 1988, Clinton 1988, and Clayton 1989, respectively) with different inoculum densities of *P. capsici* grown on V8 vermiculite media (24). Because yield data were to be collected, soil in plots was infested after transplanted plants were established to avoid loss of plant stands. Inoculum density in this research was defined as the number of colony-forming units per gram of dry soil. Inoculum of *P. capsici* in V8 vermiculite medium was applied to subplots at the highest level (1.0× level) at a rate of 208 cm^3 per meter of row approximately 20 cm from either side of the plant row in each plot. The inoculum was covered with soil to a depth of approximately 10 cm. Care was taken to avoid damage to the roots at the time of infestation of soil with the pathogen. Inoculum was diluted 10-fold and 100-fold with uninfested V8 vermiculite medium and incorporated into soil at the same rate to give 0.1× and 0.01× levels of inoculum. Soil in control plots was not infested and did not contain detectable levels of the pathogen at the first soil sampling date. Plots were separated with soil berms and borders, 6.1 m long.

Data collection. The incidence and severity of disease on shoots were evaluated visually during the growing season. Disease was evaluated at more frequent intervals during the phases of rapid increase as shown (Figs. 1–3). Disease severity on shoots was evaluated on a scale of 0–4, in which 0 = no symptoms; 1 = wilting of plant, without a stem lesion; 2 = wilting and stem lesion without girdling; 3 = girdled by stem lesion; 4 = dead plant. Disease incidence data are reported in the figures, because trends in disease progress were similar when disease severity and

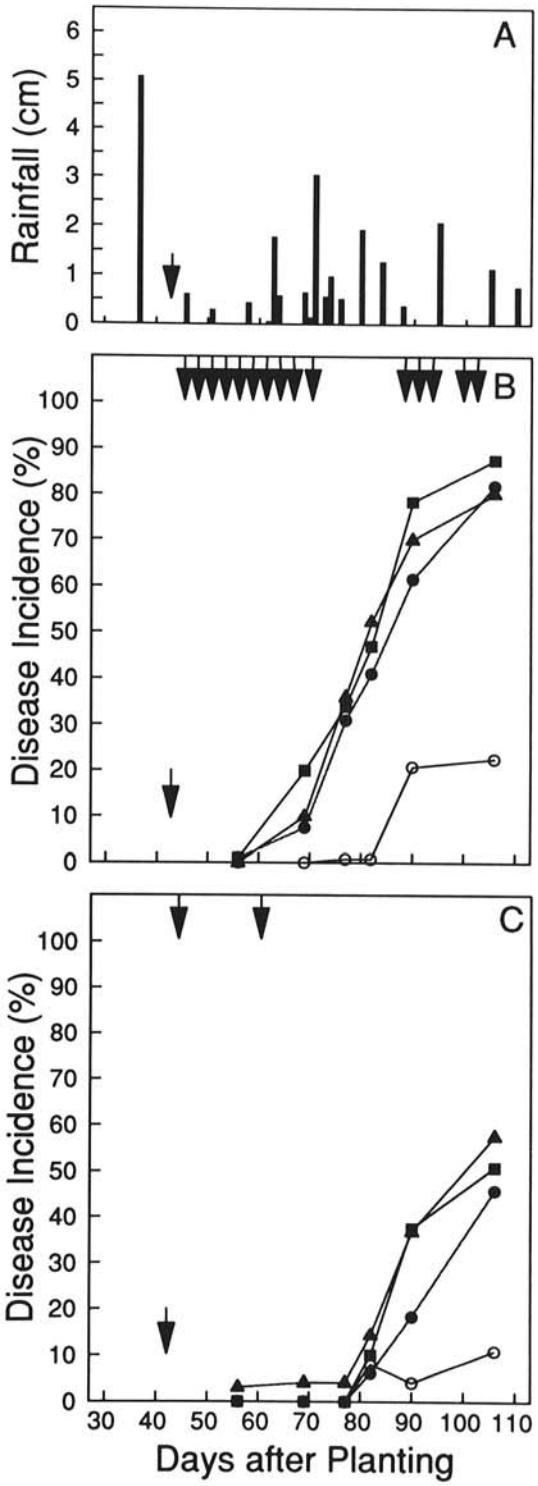

Fig. 1. Total rainfall distribution and progression of Phytophthora root rot in bell peppers in plots infested with *Phytophthora capsici* at the Clayton field area in 1988. **A,** Total rainfall distribution; **B,** disease incidence over time in plots irrigated more frequently and **C,** less frequently in plots infested at four inoculum densities applied 43 days after transplanting, including uninfested (○), 0.01× (●), 0.1× (△), or 1.0× (■). Actual inoculum densities were 0, 3.9, 5.9, or 9.4 cfu/g of dry soil, respectively. For comparisons of final disease incidence within or between an irrigation level, $LSD_{0.05} = 30$ and 28, respectively. Arrows at the top of the figure indicate the time of application of drip irrigations to the plots. Arrow at the bottom of the figure indicates the time of infestation of the plots with *P. capsici*.

disease incidence data were analyzed separately. Roots were surface-disinfested and plated on a selective medium (11) to reisolate the pathogen and confirm that infections were caused by *P. capsici*. Yield was measured in multiple harvests at each location at the end of the season. Fresh weights of fruits in various size categories and cull fruits were measured. The total harvestable yield is reported from three harvests at the Clayton field in 1988, and two harvests from the other locations.

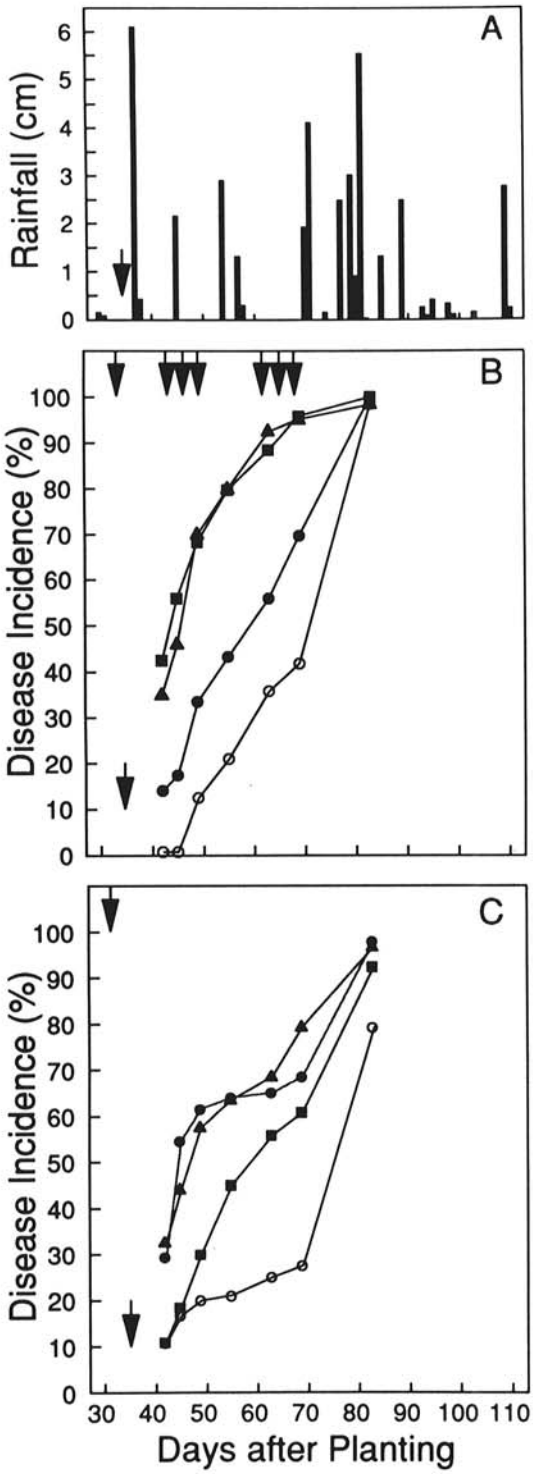

Fig. 2. Total rainfall distribution and progression of Phytophthora root rot in bell peppers in plots infested with *Phytophthora capsici* at the Clinton field area in 1988. **A,** Total rainfall distribution; **B,** disease incidence over time in plots irrigated more frequently and **C,** less frequently at four inoculum densities applied 35 days after transplanting, including uninfested (○), 0.01× (●), 0.1× (△), or 1.0× (■). Actual inoculum densities were 0, 1.9, 7.9 and 37.7 cfu/g of dry soil, respectively. For comparisons of final disease incidence within or between an irrigation level, $LSD_{0.05} = 16$. Arrows at the top of the figure indicate the time of application of drip irrigations to the plots. Arrow at the bottom of the figure indicates the time of infestation of the plots with *P. capsici*.

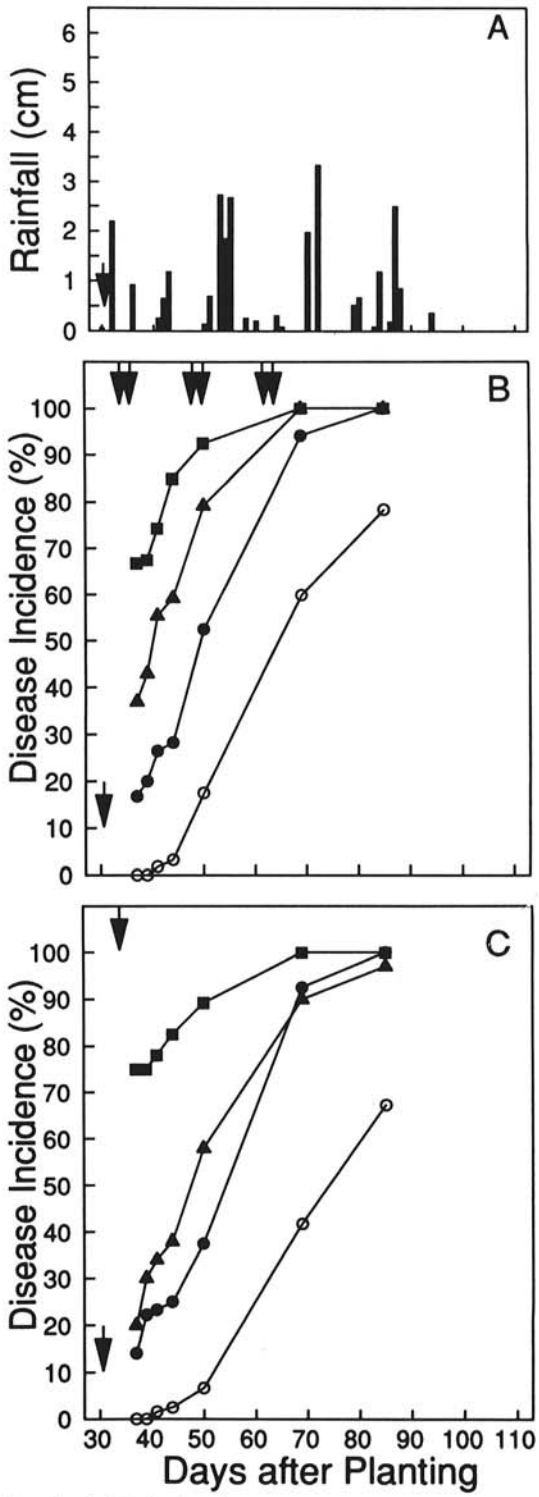

Fig. 3. Total rainfall distribution and progression of Phytophthora root rot in bell peppers in plots infested with *Phytophthora capsici* at the Clayton field area in 1989. **A,** Total rainfall distribution; **B,** disease incidence over time in plots irrigated more frequently and **C,** less frequently at four inoculum densities applied 31 days after transplanting, including uninfested (○), 0.01× (●), 0.1× (△), or 1.0× (■). Actual inoculum densities were 0, 5.3, 17.7, and 40.7 cfu/g of dry soil, respectively. For comparisons of final disease incidence within or between an irrigation level, $LSD_{0.05} = 31$ and 32, respectively. Arrows at the top of the figure indicate the time of application of drip irrigations to the plots. Arrow at the bottom of the figure indicates the time of infestation of the plots with *P. capsici*.

Soil temperature and moisture were monitored with thermistors and soil moisture blocks attached to a CR21 micrologger (Campbell Scientific, Inc, Logan, UT). The sensors were buried 20 cm deep in an uninfested plot of each irrigation treatment. Rainfall was recorded with tipping-bucket rain gauges located near the plots on each research station.

Soil from three replicates of each treatment was sampled immediately after infestation to estimate population densities of *P. capsici*. Fifteen cores (1.9 cm diameter × 20 cm deep) were sampled from each plot approximately 20 cm from both sides of the plant row where the inoculum was placed initially. The top 5 cm of soil from each core was discarded, and cores were combined into a composite sample for each side of the plant row. Three soil samples were removed and three separate dilutions were made from each soil sample with 0.25% water agar. Soil from plots infested with the highest level (1.0×) of inoculum was diluted 1:10 (w/v), whereas soil from uninfested plots and plots infested with the medium (0.1×) level of inoculum was diluted 1:5. Soil from plots infested with the lowest (0.01×) level of inoculum was diluted 1:2. From each dilution, a 1-ml sample was spread onto Masago's medium (12) amended with hymexazol at 20 μg/ml (99.5% a.i.) in each of five petri dishes. Dishes were incubated at 25 C in the dark for 48–72 h, rinsed with water, and colonies of *P. capsici* were counted. Moisture content of soil samples was measured gravimetrically, and data were transformed to colony-forming units per gram of dry soil. Data are presented as means for each treatment.

Statistical analysis. Data were tested for normality and homogeneity of variance before analysis of variance with the Statistical Analysis System (SAS Institute, Cary, NC). Separate analyses of variance were conducted for each location. Multivariate analyses of variance were conducted on disease incidence data to account for serial correlations between the dependent variable with the repeated measures option of the SAS General Linear Models procedure. Rates of disease increase were calculated from data transformed for a logistic model. The appropriateness of the model was tested by observations of scatter plots of the residual error terms (4). Mean areas under the disease progress curve (AUDPC) were calculated with the formula:

$$\Sigma_1^{n-1}[(y_i + y_{i+1})/2](t_{i+1} - t_i)$$

in which y_i = disease incidence at the ith observation, t_i = time at the ith observation, and n = total number of assessment times (4,26). Values of mean AUDPC were standardized to compare epidemics of different duration and were expressed in units of percent-days per day. Means from significant treatment effects were separated with the appropriate LSDs. Data from the three locations were combined, and a split-plot analysis was performed on the standardized AUDPCs and yield data with location as the main plot, irrigation as the subplot, and inoculum density as the sub-subplot. To test whether the cumulative amount of rainfall could explain the different levels of disease observed at the three locations, significant location sums of squares were partitioned into linear and lack-of-fit components for rainfall. Sums of squares also were partitioned for significant inoculum main effects, and linear, quadratic, and lack-of-fit components were calculated.

RESULTS

Disease progress at the Clayton field in 1988. In 1988 at the Clayton field, 16 cm of cumulative rainfall occurred between the time of infestation of soil with *P. capsici* and final disease assessment 107 days after planting (DAP) (Fig. 1A). Only two rainfall events, each <1.0 cm, occurred between the time of infestation of the plots 43 DAP and the first disease assessment at 56 DAP. Disease onset did not appear to be associated with the rainfall events >1.0 cm that occurred 63 and 71 DAP (Fig. 1A), if a 3-day pathogen incubation period is assumed (22), because little disease occurred in the less frequently irrigated plots until 82 DAP (Fig. 1C). However, irrigation had a significant effect on the rate of disease increase over time (Wilks' lambda value for time × irrigation effect significant at $P = 0.0002$). Observed disease onset occurred 69 DAP in infested plots that were irrigated more frequently (Fig. 1B), whereas disease onset occurred 82 DAP in infested plots that were irrigated less frequently (Fig. 1C). Estimated rate parameters (r_L) for disease increase calculated with the logistic model were 0.13/unit per day in infested plots irrigated more frequently, whereas estimated r_L were less than 0.09/unit per day in infested plots irrigated less frequently. Final disease incidence at all inoculum densities was lower ($P < 0.001$) in plots that were irrigated less frequently (Fig. 1C) than in plots that were irrigated more frequently (Fig. 1B). In addition, mean AUDPC values also were significantly lower in infested plots irrigated on the less frequent than the more frequent schedule of this field location (irrigation effect was significant at $P = 0.05$) (Table 1).

Inoculum densities estimated by dilution plating at the time of infestation were 0, 3.9, 5.9, and 9.4 cfu/g of dry soil in uninfested plots and plots infested at the 0.01×, 0.1×, and 1.0× levels of inoculum, respectively. Inoculum density had significant linear and quadratic effects on mean AUDPC values at $P = 0.0007$ and 0.0001, respectively. Mean AUDPC values were lower in

TABLE 1. Influence of irrigation schedule and inoculum density of *Phytophthora capsici* applied either 43, 35, or 31 days after transplanting at three field locations on the development of Phytophthora root and crown rot in bell pepper as indicated by the area under the disease progress curve (AUDPC)

	AUDPC (percent-days per day)[a]					
	Clayton field 1988[b]		Clinton field 1988[c]		Clayton field 1989[d]	
Inoculum level[e]	More frequent irrigation	Less frequent irrigation	More frequent irrigation	Less frequent irrigation	More frequent irrigation	Less frequent irrigation
None	8.81	2.76	38.30	32.35	39.87	28.50
0.01×	39.08	12.47	57.12	68.72	69.91	69.43
0.1×	43.62	21.72	83.14	70.34	84.76	71.14
1.0×	47.90	18.52	83.81	53.45	93.25	92.82

[a] AUDPC was calculated by methods described previously (4).

[b] For comparisons of means within and between irrigation levels at the Clayton field in 1988, $LSD_{0.05}$ = 12.9 and 18.9, respectively. Inoculum density had significant linear and quadratic effects on AUDPC values at $P = 0.0007$ and 0.0001, respectively. The irrigation effect was significant at $P = 0.05$.

[c] For comparisons of means within and between irrigation levels at the Clinton field in 1988, $LSD_{0.05}$ = 31.1 and 33.5, respectively. Inoculum density had significant linear and quadratic effects on AUDPC values at $P = 0.0228$ and 0.0099, respectively.

[d] For comparisons of means within and between irrigation levels at the Clayton field in 1989, $LSD_{0.05}$ = 19.4 and 22.8, respectively. Inoculum density had significant linear and quadratic effects on AUDPC values at $P = 0.0004$ and 0.0001, respectively.

[e] Inoculum of *P. capsici* was applied undiluted to subplots at the 1.0× level at a rate of 208 cm^3 per meter of row. Inoculum was diluted 10-fold and 100-fold in uninfested V8 vermiculite and incorporated at the same rate to give 0.1× and 0.01× levels of inoculum. Actual inoculum density ranges were 0, 3.9–5.3, 5.8–17.6, and 9.4–40.7 cfu/g of dry soil, at the 0, 0.01×, 0.1×, and 1.0× levels of inoculum, respectively, for all locations at the time of infestation.

uninfested than infested plots (Fig. 1B and Table 1). Significant differences in the rate of disease increase, final disease incidence (Fig. 1B,C), or mean AUDPC values (Table 1) were not observed among plots infested with different densities of inoculum and irrigated at the same frequency. However, the rate of disease increase over time and the final incidence of disease was lower in uninfested than infested plots (Wilks' lambda value significant for the time × inoculum interaction at $P = 0.0009$). Relatively low levels of disease were apparent in initially uninfested plots at the end of the season in the Clayton field in 1988 (Fig. 1B,C).

Disease progress at the Clinton field in 1988. A greater amount of cumulative rainfall occurred in the Clinton field than in the Clayton field in 1988 (Figs. 1A and 2A), that is, 31 cm of rainfall occurred between infestation of the plots and final disease assessment date 83 DAP (Fig. 2A). The greatest single rainfall event in the Clinton field occurred 37 DAP (6.53 cm) and only 2 days after the plots initially were infested (Fig. 2A). Observed disease onset occurred in all infested plots within 5 days after this rainfall, and disease increased with time (Wilks' lambda value for time significant at $P = 0.0001$) (Fig. 2B,C). Irrigation did not have a significant effect on the time of onset or the final incidence of disease. However, the mean rate of increase in disease incidence between 45 and 49 DAP was significantly greater ($P < 0.05$) in plots irrigated more frequently (0.25/unit per day) than in plots irrigated less frequently (0.16/unit per day).

Inoculum densities at infestation were 0, 1.9, 7.9, and 37.7 cfu/g of dry soil in uninfested plots and plots infested at the 0.01×, 0.1×, and 1.0× levels of inoculum. Inoculum densities had significant linear and quadratic effects on the mean AUDPC values at $P = 0.0228$ and 0.0099, respectively. Mean AUDPC values were significantly less in uninfested plots than in infested plots irrigated on the more frequent schedule (Table 1). Inoculum densities did not affect the rate of disease increase over time (time × inoculum interaction was not significant). Disease incidence reached 100% in most plots at all inoculum densities by the end of the season. The duration of the epidemics was shorter and final disease incidence was higher in plots at the Clinton field in 1988 than in plots at the Clayton field in 1988 (Figs. 1 and 2).

Disease progress at the Clayton field in 1989. Approximately 22 cm of rainfall occurred during the season at the Clayton field in 1989, and a 2.2-cm rainfall occurred 1 day after infestation of the plots (Fig. 3A). Disease onset was observed within 5 days of this rainfall in all infested plots at the Clayton site in 1989. Initial levels of disease were highest in plots infested with the highest inoculum density, and they decreased with inoculum density. The rate of disease progress over time differed in plots

TABLE 2. Influence of irrigation schedule and inoculum density of *Phytophthora capsici* applied either 43, 35, or 31 days after transplanting at three field locations on total harvestable yield of bell pepper

	Yield of harvestable fruit (kg/plot)[a]					
	Clayton field 1988[b]		Clinton field 1988[c]		Clayton field 1989[d]	
Inoculum level[e]	More frequent irrigation	Less frequent irrigation	More frequent irrigation	Less frequent irrigation	More frequent irrigation	Less frequent irrigation
None	15.5	11.1	3.6	6.7	8.4	5.7
0.01×	8.9	11.6	1.9	1.7	0.9	0.8
0.1×	10.0	9.9	0.1	0.9	0.0	0.9
1.0×	8.8	7.0	0.2	3.4	0.0	0.0

[a] Yield data are shown as kilograms per 12.2-m length of single-row beds. Data are shown as total harvestable yields from three harvests in Clayton 1988 and two harvests for the other locations.
[b] For comparisons of means within and between irrigation levels at the Clayton field in 1988, $LSD_{0.05} = 5.8$ and 7.5, respectively. Inoculum density did not have a significant linear effect on yield.
[c] For comparisons of means within and between irrigation levels at the Clinton field, $LSD_{0.05} = 3.9$ and 3.9, respectively. Inoculum density had a significant linear effect on yield at $P = 0.0002$.
[d] For comparisons of means within and between irrigation levels at the Clayton field, $LSD_{0.05} = 4.9$ and 5.0, respectively. Inoculum density had a significant linear effect on yield at $P = 0.0003$.
[e] Inoculum of *P. capsici* was applied undiluted to subplots at the 1.0× level at a rate of 208 cm^3 per meter of row. Inoculum was diluted 10-fold and 100-fold in uninfested V8 vermiculite and incorporated at the same rate to give 0.1× and 0.01× levels of inoculum. Actual inoculum density ranges were 0, 3.9–5.3, 5.8–M.6 and 9.4–40.7 cfu/g of dry soil, at the 0, 0.01×, 0.1×, and 1.0× levels of inoculum, respectively, for all locations.

TABLE 3. The effect of rainfall, irrigation, and inoculum density of *Phytophthora capsici* applied either 43, 35, or 31 days after transplanting at three field locations on the area under the disease progress curve (AUDPC) and yield of pepper

		AUDPC[a]		Yield[b]	
Source	df	Mean square	$Pr > F$	Mean square	$Pr > F$
Location	2	17,626	0.0010	3,451	0.0011
Rain linear	1	26,748	0.0008	4,420	0.0015
Rain lack of fit	1	8,501	0.0200	2,481	0.0100
Block × location	9	1,091	...	218	...
Irrigation	1	3,842	0.0075	1	0.8830
Irrigation × location	2	422	0.3212	78	0.3315
Block × Irrigation × location	9	327	...	62	...
Inoculum	3	7,983	0.0001	687	0.0001
Inoculum linear	1	7,360	0.0001	865	0.0002
Inoculum quadratic	1	15,164	0.0001	1,097	0.0001
Inoculum lack of fit	1	1,424	0.0224	169	0.0862
Inoculum × location	6	452	0.1259	45	0.5594
Inoculum × irrigation	3	252	0.4087	28	0.6780
Inoculum × irrigation × location	6	320	0.2995	55	0.4385
Residual	54	257	...	55	...

[a] AUDPC was calculated by methods described previously (26).
[b] Yield data used in the analysis were total harvestable yields from three harvests in the Clayton field in 1988 and two harvests for the other locations.

treated with different inoculum densities (Wilks' lambda value significant for time × inoculum interaction at $P = .0019$) (Fig. 3B,C). However, final disease incidence reached high levels in all infested plots regardless of inoculum density, as was observed in the Clinton field in 1988 (Fig. 2B,C) and the more frequently irrigated plots in the Clayton field in 1988 (Fig 1B). Irrigation did not have a significant effect on disease onset or final disease incidence. However, the rate of disease increase between 41 and 44 DAP, respectively, in infested plots inoculated at the 1.0× and 0.1× levels of inoculum, was significantly greater in plots irrigated more frequently (0.30 and 0.19/unit per day) than in plots irrigated less frequently (0.11 and 0.06/unit per day).

Inoculum densities at infestation were 0, 5.3, 17.7, and 40.7 cfu/g of dry soil in uninfested plots and plots infested at the 0.01×, 0.1×, and 1.0× levels of inoculum. Inoculum densities had significant linear and quadratic effects on mean AUDPC values at $P = 0.0004$ and 0.0001, respectively (Table 1). Mean AUDPCs were significantly lower in uninfested than infested plots and increased with inoculum density.

Yield at each field location in 1988 and 1989. Disease reduced yield in plots inoculated with the highest inoculum density in the Clayton field in 1988 by 43% in plots that were irrigated more frequently and 37% in plots that were irrigated less frequently (Table 2). Yield was significantly lower in all infested than in initially uninfested plots on the more frequent irrigation schedule, whereas significant reductions in yield were not apparent between uninfested and infested plots irrigated less frequently in the Clayton field in 1988 (Table 2). In addition, inoculum density did not have a significant linear effect on yield in this location. Disease incidence at 69, 77, 82, 90, and 107 DAP was negatively correlated with total harvestable yield ($r = -0.45, -0.62, -0.73, -0.72, -0.72$, and -0.61, respectively).

Yields were greatly reduced in the Clinton field area in 1988 and the Clayton field in 1989 at all inoculum densities when compared with uninfested controls, and the irrigation effect on yield was not significant (Table 2). However, inoculum density had a significant linear effect on yield at $P = 0.0002$ and 0.0003, at the Clinton and Clayton fields, respectively, in 1988 and 1989. Disease incidence at 42, 45, 49, 55, 63, and 69 DAP in the Clinton field in 1988 was negatively correlated with total harvestable yield ($r = -0.44, -0.48, -0.63, -0.72, -0.77$, and -0.83, respectively).

Analysis over all locations. There were significant differences among locations for AUDPCs and yield (Table 3). Rainfall had a significant linear effect on mean AUDPC values and yield. Mean AUDPCs were 24.4, 68.2, and 60.9%-days per day, and mean yields were 10.4, 2.3, and 2.1 kg/plot at the three locations receiving 16, 22, and 31 cm of total rainfall, respectively. However, the rainfall lack-of-fit effect also was significant, indicating that other factors contributed to the variation in AUDPC and yield among field locations in addition to rainfall. The AUDPCs among all locations also were affected by irrigation and inoculum densities at infestation (Table 3). Mean AUDPCs were 57.5%-days per day in plots irrigated more frequently and 44.8%-days per day in plots irrigated less frequently. Inoculum density also had significant linear and quadratic effects on AUDPC and yield (Tables 3 and 4). However, the inoculum lack-of-fit effect also was significant for AUDPC, indicating that a quadratic function did not completely describe the effect of inoculum density on AUDPC. None of the two-way or three-way interactions between rainfall, irrigation, and inoculum density were significant (Table 3).

Disease incidence at each assessment date at all inoculum densities was positively correlated with the total cumulative centimeters of rainfall preceding disease assessment ($r = 0.50$, 0.57, and 0.55 for the three field locations, respectively). The cumulative centimeters of rainfall between infestation of the plots and the final disease assessment date were negatively correlated with total harvestable yield ($r = -0.51$) and the time of disease onset ($r = -0.67$), and positively correlated with AUDPC ($r = 0.47$) when all the data from the three locations were analyzed. There was a significant negative correlation between AUDPC and yield ($r = -0.84$) and a positive correlation between yield and the time of disease onset ($r = 0.73$).

TABLE 4. Effect of inoculum density of *Phytophthora capsici* applied either 43, 35, or 31 days after transplanting at three field locations on the overall mean areas under the disease progress curve (AUDPC) and overall yield of pepper.

Inoculum level[a]	AUDPC[b] (percent-days/day)	Yield[c] (kg/plot)
0	25.1	8.51
0.01×	52.0	4.31
0.1×	62.5	3.64
1.0×	64.9	3.24

[a] Inoculum of *P. capsici* was applied undiluted to subplots at the 1.0× level at a rate of 208 cm^3 per meter of row. Inoculum was diluted 10-fold and 100-fold in uninfested V8 vermiculite and incorporated at the same rate to give 0.1× and 0.01× levels of inoculum. Actual inoculum density ranges were 0, 3.9–5.3, 5.8–17.6 and 9.4–40.7 cfu/g of dry soil, at the 0, 0.01×, 0.1×, and 1.0× levels of inoculum, respectively, for all locations.

[b] The AUDPC was calculated by methods described previously (4). Inoculum density had a significant effect on AUDPC values at $P = 0.0001$.

[c] Yield data are shown as kilograms per 12.2-m length of single-row beds. Data are shown as overall mean total harvestable yields from three harvests in Clayton in 1988, and two harvests for the other locations. Inoculum density had a significant effect on yield at $P = 0.0001$.

DISCUSSION

The development of epidemics caused by *P. capsici* in bell pepper in the low rainfall field in Clayton in 1988 was significantly increased by frequent drip irrigations (Fig. 1B). Plants in plots that were drip irrigated less frequently had a lower incidence of disease than plants in plots that were drip irrigated more frequently, and disease onset was delayed (Fig. 1C). Similar delays in disease onset occurred in fields of processing tomato that were furrow irrigated on a less frequent basis (24). Phytophthora root rot on citrus also was less severe under furrow irrigations applied less frequently than under drip irrigations applied more frequently (7). Continuous measurements of soil moisture in the plots were not possible; however, tensiometer readings taken during some irrigation episodes indicated that soil matric potentials were between 0 and −2.5 kPa for at least 4 h during irrigation episodes and were conducive for zoospore release from sporangia and infection of pepper plant roots (J. B. Ristaino, *unpublished data*). Because sporangia of *P. capsici* release zoospores readily under saturated soil conditions, fewer cycles of secondary inoculum production and dispersal were possible in plots that were irrigated less frequently than in plots that were irrigated more frequently in this field (2,6).

The probability of pathogen-root contacts also may have increased with frequent drip irrigations due to increased root production in soil. Although root growth was not quantified in the study reported here, others (1,7) have demonstrated that root growth is increased and concentrated in wet areas around drip emitters in crops such as citrus and tomato. However, root growth and water extraction by roots may be reduced by Phytophthora root rots, and the effects of disease on water extraction are greatest when disease is increasing (23).

Rainfall also had a significant effect on disease development. A single rain event >2.0 cm was sufficient for disease onset to occur at the Clinton field in 1988 and the Clayton field in 1989 (Figs. 2 and 3). These rain events that occurred 2 days after infestation of the plots in Clinton in 1988 and 1 day after infestation of the plots in Clayton in 1989 caused prolonged periods of soil saturation that contributed to the rapid increase in disease incidence. Rainfalls >2.0 cm only occurred twice at the Clayton field in 1988 (Fig. 1), and final disease incidence was lower in less frequently irrigated plots in this field (Fig. 1C) than at the other two locations where either nine or five rainfall events >2.0 cm occurred, respectively (Figs. 2C and 3C). Rainfall has been associated with increases in disease caused by *Phytoph-*

thora spp. (3,10,20). Predictive models relating precipitation, temperature, and number of drought days to the development of black shank epidemics in tobacco have been developed (10). Temperature and surface wetness conditions associated with a period before a rain event, as well as rainfall amount, have been used to predict the occurrence of strawberry leather rot caused by *P. cactorum* (20).

There was a positive correlation between disease incidence and the cumulative amount of rainfall preceding the assessment date from the three field locations. In addition, the cumulative amount of rainfall was also negatively correlated with time of disease onset and yield and had a significant linear effect on the mean AUDPC values at the three locations. Others also have reported that the cumulative amount of rainfall is positively correlated with disease incidence and the rate of change in disease caused by *P. capsici* on pepper (3). The work reported here in fields with three different rainfall distributions confirms the work of others and also shows that rainfalls >2.0 cm (Figs. 2A and 3A) can have a larger effect on disease increase and final level of disease (Figs. 2C and 3C) than multiple rainfalls of lesser amounts (Fig. 1A,C). The amount of rain in a given rainfall can affect both the extent and duration of soil saturation and the amount of secondary spread of inoculum; less secondary spread of inoculum occurred in the low rainfall field in Clayton in 1988 (Fig. 1C) than at the other two locations (Figs. 2C and 3C). Thus, the cumulative amount of rainfall over time may be less important than the amount of rain in a single rainfall in initiating the development of epidemics.

P. capsici spread readily to uninfested plots. Populations of *P. capsici* were not detected in soil sampled at the first sampling date, and all plots were fumigated with methyl bromide before each experiment. Therefore, the presence of initial inoculum of *P. capsici* in the plots was minimized. The actual mechanism of dispersal was not determined in this study; however, wind-driven rain and surface runoff could have been factors in pathogen spread. Others also have demonstrated that *P. capsici* can spread with drainage water or rainfall from initial point sources of inoculum in the field (3,25). Plants in growers' fields that are affected by the disease in North Carolina often show patterns of disease associated with runoff of surface water. The isolate used in these field studies also produced caducous sporangia that were shed readily in water (22).

Phytophthora root and crown rot in pepper behaves as a polycyclic disease in the field. In polycyclic diseases, cycles of infection and inoculum production occur during the season. Differences in inoculum densities of *P. capsici* in soil infested 5–6 wk after transplanting did not affect final disease incidence in a susceptible pepper cultivar planted in infested plots at three field locations (Figs. 1–3). Inoculum densities of *P. capsici* reported in field soils in this study ranged from <1 to 40 cfu/g of soil and were within the range reported in naturally infested soils during the growing season (16). Initial inoculum densities of *P. capsici* present in growers' fields in North Carolina have not been measured. Tenfold dilutions of inoculum applied to the plots did not result in 10-fold differences in measured populations at the three field locations, and even the lowest densities (2–5 cfu/g) of inoculum evaluated were adequate for high levels of disease to develop, provided that soil matric potentials were conducive for release of zoospores and dispersal to plants. Inoculum densities reported here were estimates based on dilution plate assays, which are known to have limitations when populations of *Phytophthora* spp. are low (14).

Inoculum densities of <1 cfu/g have been reported for other *Phytophthora* spp. in field soils (11,13,14). Inoculum densities of *P. capsici* and other *Phytophthora* spp. are not static, but increase and decrease in soils over time (J. B. Ristaino, *unpublished data*; 11,13,14,24). Inoculum also was detected in uninfested plots later in the season. Multiple cycles of secondary inoculum production, infection, and plant-to-plant spread of the pathogen probably also contributed to the independence of final disease incidence and inoculum density. However, inoculum density had a significant linear effect on mean AUDPC values calculated among all locations (Table 4). Low densities of initial inoculum of *P. p. nicotianae* (0.04 cfu/g) also resulted in high levels of disease in tobacco when a single saturation period occurred (27). In contrast, the time of disease onset was delayed (8,17), and the rate and final level of disease were lower with low than with high inoculum densities of several other soilborne fungi (14,18). Low inoculum densities of *P. parasitica* applied 6 wk after transplanting caused low levels of disease in furrow-irrigated processing tomatoes that were not affected by rainfall (14). The inoculum density-disease relationships and rate parameters for disease increase in epidemics caused by *P. capsici* in pepper are more similar to epidemics caused by *P. p. nicotianae* in tobacco than to epidemics caused by *P. parasitica* in tomato.

The rate of disease progress and time of disease onset had a significant impact on yield of pepper grown at all locations (Table 2). Yields were significantly higher in plots irrigated less frequently than in plots irrigated more frequently at the Clayton field in 1988, because disease increase was less rapid in plots irrigated less frequently. Yields also were lower in infested plots at the higher rainfall fields than in infested plots at the lower rainfall field, because disease onset occurred earlier and progressed at a faster rate in the high rainfall fields. Plant-to-plant spread of *P. capsici* with heavy rainfall episodes had a major impact on final disease incidence and, ultimately, yield in pepper.

Overall, AUDPCs from the three field locations were significantly affected by rainfall, irrigation, and inoculum density at the time of soil infestation, and interactions were not significant. There were significant linear effects of rainfall and inoculum density on AUDPC and overall yields. However, irrigation also had a significant effect on AUDPCs. Irrigation had a greater effect on disease progress in the low rainfall than in the high rainfall fields. Both rainfall and irrigation had larger effects on the time of onset and final disease incidence than the range of inoculum densities evaluated in this work. Disease incidence became very high when either a heavy (>2.0 cm) rainfall or more frequent drip-irrigation events occurred. Plant-to-plant spread of *P. capsici* resulted in severe disease in uninfested plots in the high rainfall fields. Final disease incidence was independant of inoculum densities; thus, Phytophthora root and crown rot is a truly polycyclic disease on pepper in North Carolina fields (4).

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