Effect of Interaction of Inoculum Dose, Cultivar, and Geographic Location on the Development of Foliar Symptoms of Bacterial Ring Rot of Potato

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ABSTRACT

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Proportional hazards models were used to estimate hazard and survival functions for both the onset and maximum incidence of foliar symptoms of bacterial ring rot in three potato cultivars (Norchip, Norland, and Russet Burbank) grown at seven locations across the United States (Colorado, Maine, New York, North Dakota, Oregon, Washington, and Wisconsin) over the period 1988–1990. The models predicted a minimum of 50 and 82 days after planting for survival probabilities of $P < 0.95$ and $P < 0.05$ for symptom onset and maximum disease incidence, respectively. The time frame predicted for these events was affected by cultivar, location, and cultivar by location interaction. There was no proportional increase in the relative hazards for symptom onset and maximum disease incidence due to increased inoculum dose. An increased probability of survival, however, was associated with an increased incidence of right-censoring of the data in plants given the lower inoculum doses. The relationship between cultivar maturity and the relative hazard for maximum disease incidence was not consistent, an observation attributable to the fact that Russet Burbank displayed stunting (dwarf rosette) as an early season symptom of bacterial ring rot. Symptom onset and maximum disease incidence were influenced most by location and location by cultivar interaction, with certain locations displaying a trend toward an increased hazard for these events. Differences in the relative hazard for onset and maximum disease incidence among locations, in some cases, were associated with higher rates of right-censoring of the data. However, environmental conditions also appeared to influence the time when onset and maximum disease incidence occurred, because significant differences in the hazards for these events were observed in locations in which no right-censored observations were recorded.

Additional keywords: *Clavibacter michiganensis* subsp. *sepedonicus*, survival analysis.

Bacterial ring rot, caused by the gram-positive bacterium *Clavibacter michiganensis* subsp. *sepedonicus*, is a disease of particular concern to the seed potato industry. Symptoms of this disease appear as wilting, necrosis, and, in some cultivars, stunting of foliage (8). Because *C. m. sepedonicus* is readily transmitted through tuber-handling processes, such as seed cutting and planting, traditional control strategies for this disease have concentrated on the elimination of the causal organism from seed stocks. Central to the control of this disease is the imposition of a zero-tolerance limit for bacterial ring rot on seed potatoes eligible for certification. The zero-tolerance limit is currently enforced through a series of pre- and postharvest inspections of seed potato lots and depends on the ability of inspectors to detect symptomatic plants and tubers. The efficacy of this control strategy, however, has been constrained by a general lack of knowledge concerning the factors that influence symptom expression.

Previous investigations have shown that variables such as inoculum dose (2,16,23,25), cultivar (2,3,7,15,22,25), and environment (1,13,14,17–20) are important in determining the magnitude of bacterial ring rot symptom expression. Because the incidence of symptom expression in infected seed lots is often low (7), inspections of seed potatoes for bacterial ring rot ideally should be conducted when the probability of finding the disease is highest. However, except for a recent investigation of the influence of both inoculum dose and cultivar on foliar symptom onset (2), little attention has been given to this aspect of the epidemiology of bacterial ring rot.

One possible approach to the problem of optimal timing of inspections is to develop models that predict two critical events in the development of foliar symptoms of bacterial ring rot: symptom onset and maximum disease incidence. Recently, we have shown that the magnitude of symptom expression is attributable largely to the interactions of inoculum dose, cultivar, and geographic location (25). Based on these results and others (2), we believe that these variables also affect the time of foliar symptom development. This study describes the use of the proportional hazards model (6) to estimate hazard and survival functions for both the onset and maximum incidence of foliar symptoms of bacterial ring rot. Our objective was to determine the effect of inoculum dose, cultivar, and geographic location on these events and to provide estimates of the probability of symptom onset and maximum incidence occurring at given times during the growing season.

MATERIALS AND METHODS

Experimental design. Melon balls of potato cultivars Russet Burbank, Norchip, and Norland were inoculated with 0, 10^2, 10^6, or 10^9 colony forming units (cfu) of *C. m. sepedonicus* strain SS43. The 12 inoculum dose-cultivar combinations were planted as a randomized complete block design consisting of plots of 25 plants replicated three times (1988) or plots of 15 plants replicated four times (1989 and 1990) in diverse potato growing regions of the United States (Colorado, Maine, New York, North Dakota, Oregon, Washington, and Wisconsin) during the years 1988–1990. Disease incidence (i.e., the proportion of plants in each plot with bacterial ring rot symptoms) was recorded weekly on the basis

of interveinal chlorosis and wilting of the leaves and/or stems, and, in the case of Russet Burbank, stunting and rosetting of the foliage. Additional details of the methods used in this study have been described previously (25).

Statistical analysis. Data that describe time to a particular event, e.g., symptom onset or maximum disease incidence, are generically referred to as failure time data (syn.= survival data, life data). As a rule, techniques such as linear regression are inappropriate for the analysis of these data because complete knowledge of the failure times for an event may not be available due to censoring, i.e., the failure times for some subjects are not observed but are only known to have occurred before (left-censored), after (right-censored), or during some time interval within the study period (interval-censored). Furthermore, failure times often do not follow a normal distribution (12).

In this study, the data obtained for symptom onset and maximum disease incidence were interval-censored, included some right-censored observations, and were of unknown distribution. As a result, proportional hazards models (6), one of a number of models that fall under the general category of survival analysis and allow for the analysis of censored data, were used to obtain estimates of the hazard and survival functions of symptom onset and maximum disease incidence. The proportional hazards model has been discussed in detail by Kalbfleisch and Prentice (12), and its application to the analysis of epidemiological data has been reviewed by Breslow (4,5).

A hazard function generated by the proportional hazards model describes the instantaneous hazard for an event (i.e., the instantaneous rate of failure at time $T = t + \delta t$, given that the event has not occurred at time $T = t$). Unlike the more traditional models used in survival analysis, the proportional hazards model does not depend on a particular distribution of the failure times being modeled but instead assumes an arbitrary, unspecified baseline hazard function:

$$H(t|x) = h_0(t)\, g(x).$$

The terms $h_0(t)$ and $g(x)$ may involve additional parameters; thus the hazard function may be written in the form:

$$H(t|x) = h_0(t) \exp(x\beta),$$

in which x represents a vector of covariates and β represents a vector of regression coefficients. This situation is somewhat analogous to that of multiple regression. However, the effects of these covariates on the hazard function are multiplicative rather than additive, as is the case in linear regression, and the hazard for an event is assumed to be modified proportionally by the addition of covariates to the models (i.e., the ratio of two hazard functions $H(t|x_1)/H(t|x_2)$ with regression vectors x_1 and x_2 is constant over time). If the assumption of proportionality is met, then there is some ordering of the hazard functions, e.g., $H(t|x_1) > H(t|x_2)$. Thus, the function can be used to assess the relative effect of some factor on the hazard for a particular event. Written in terms of a survival function, the hazard function:

$$H(t|x) = h(t) \exp(x\beta)$$

takes the form:

$$S(t|x) = S_0(t)^{\exp(x\beta)}$$

and gives the probability of future survival (i.e., an event not occurring given that it was not observed at $T = t$). This function can be used for prognosticative purposes.

The software used to estimate the hazard and survival functions for symptom onset and maximum disease incidence is based on a discrete model described by Kalbfleisch and Prentice (12) and was designed for use with continuous data that may include right-censored observations. For the purposes of our analysis, symptom onset was assumed to have occurred on the day that foliar symptoms of bacterial ring rot were first observed in each plot, even though symptoms were assessed weekly. In the case of maximum disease incidence, the problem of interval-censoring of the data was circumvented by obtaining estimates of the time to maximum disease incidence for each plot by the use of the SAS procedure NONLIN (SAS Institute, Cary, NC) to fit exponential quadratic models to disease incidence curves via the parameterization:

$$Y = a \exp[b(X - c)^2],$$

in which Y is $\mathrm{Sin}^{-1}\sqrt{\%\text{ disease incidence}}$, X is the number of days after planting, and c is an estimate of maximum disease incidence.

Models describing the hazard and survival functions for both onset and maximum symptom expression were obtained by the SAS procedure PHGLM (11). Proportionality of the covariates was assessed by the $Z{:}PH$ statistic given by the SAS output, with a value of $Z{:}PH \leq 1.5$ used as the cutoff value for the acceptance of covariates in the models. Final proportional hazards models were obtained by dropping nonsignificant terms ($P > 0.05$) and combining coefficients that were not significantly different from one another based on inspection of their standard errors. Loss of predictive value due to reduction of the model was assessed by the likelihood ratio test (12), with $P > 0.05$ used as the criterion for acceptance of the reduced model.

TABLE 1. Final proportional hazard models obtained for onset and maximum incidence of foliar symptoms of bacterial ring rot of potato

Year	Model	R value[a]	Hazard function[b,c]
1988	Onset	0.463	$h_{ij}(t) = h_0(t)_{\mathrm{ME,WI,RB,NRC}}\ \exp[1.24_{\mathrm{NRL}} + 3.97_{\mathrm{CO,OR}} + (-1.71)_{\mathrm{ND}} + 7.56_{\mathrm{WA}} + (-1.74)_{\mathrm{CO,ORNRL}} + (-7.13)_{\mathrm{WA,NRL}} + 4.21_{\mathrm{ME,NRC}} + (-1.94)_{\mathrm{OR,NRC}} + (-4.97)_{\mathrm{WA,NRC}}]$
	Maximum	0.532	$h_{ij}(t) = h_0(t)_{\mathrm{CO,RB}}\ \exp[2.54_{\mathrm{NRL,NRC}} + (-3.01)_{\mathrm{ME,OR}} + (-8.28)_{\mathrm{ND}} + 3.4_{\mathrm{WA}} + (-5.34)_{\mathrm{WI}} + (-2.51)_{\mathrm{OR,NRL}} + (-9.08)_{\mathrm{WA,NRL}} + (-2.64)_{\mathrm{ND,NRC}} + (-3.09)_{\mathrm{OR,NRC}} + (-6.14)_{\mathrm{WA,NRC}}]$
1989	Onset	0.242	$h_{ij}(t) = h_0(t)_{\mathrm{ME,ND,RB,NRL,NRC}}\ \exp[1.0_{\mathrm{CO}} + 2.31_{\mathrm{WA}} + (-1.06)_{\mathrm{WA,NRL}}]$
	Maximum	0.435	$h_{ij}(t) = h_0(t)_{\mathrm{CO,RB,NRL,NRC}}\ \exp\,[-3.05)_{\mathrm{ME,ND}} + 1.65_{\mathrm{WA}} + (-2.13)_{\mathrm{WA,NRL}}]$
1990	Onset	0.437	$h_{ij}(t) = h_0(t)_{\mathrm{CO,WA,RB}}\ \exp[(-3.73)_{\mathrm{NRL}} + 2.35_{\mathrm{ME}} + (-4.19)_{\mathrm{ND}} + 4.51_{\mathrm{ND,NRL}}]$
	Maximum	0.399	$h_{ij}(t) = h_0(t)_{\mathrm{CO,ME,RB,NRL}}\ \exp[(-0.05)_{\mathrm{NRC}} + (-1.63)_{\mathrm{ND}} + 3.03_{\mathrm{NY}} + 1.07_{\mathrm{ME,NRL}} + 2.01_{\mathrm{ND,NRL}} + 0.87_{\mathrm{NY,NRC}}]$

[a] $R^2 = (\text{model chi-square} - 2p)/[-2L(0)]$, in which p is the number of variables in the model, and $L(0)$ is the model log likelihood with all regression coefficients set to 0. This statistic is analogous to the multiple correlation coefficient when correction is made for the number of parameters estimated.

[b] $h(t)$ is the hazard function, which specifies the instantaneous rate of failure for symptom onset or maximum disease incidence at time $T = t + \delta t$ given that the event has not occurred at time $T = t$, and $h_0(t)$ is the baseline hazard function. Written in terms of a survival function, the hazard function $h_{ij}(t) = h_j(t) \exp(X_{ij}\beta)$, takes the form $s_{ij}(t) = s_j(t)^{\exp(X_{ij}\beta)}$ and gives the probability of future survival (i.e., symptom onset or maximum disease incidence not occurring).

[c] Subscripts are used to indicate cultivar(s), location(s), and/or cultivar by location interaction(s) represented by individual coefficients. The cultivars Russet Burbank, Norland, and Norchip are represented by the abbreviations RB, NRL, and NRC, respectively. Locations are represented by state postal abbreviations.

TABLE 2. Percentage of blocks not developing detectable foliar symptoms of bacterial ring rot (right-censored observations) in potato cultivars (Russet Burbank, Norland, and Norchip) given initial inoculum doses of 0, 10^2, 10^6, or 10^9 colony forming units of *Clavibacter michiganensis* subsp. *sepedonicus* and grown in seven locations in the United States[a]

		Russet Burbank				Norland				Norchip			
Year	Location	0	10^2	10^6	10^9	0	10^2	10^6	10^9	0	10^2	10^6	10^9
1988	Colorado	100	0	0	0	0	33	0	0	0	33	0	0
	Maine	100	67	0	0	0	0	0	0	0	0	0	0
	N. Dakota	0	67	33	67	0	0	0	0	0	100	33	33
	Oregon	67	0	0	0	0	0	0	0	0	0	0	0
	Washington	67	67	0	0	33	0	0	0	33	33	0	0
	Wisconsin	100	67	33	33	100	0	0	0	100	0	0	0
1989	Colorado	75	75	0	0	25	25	0	0	75	50	50	25
	Maine	100	100	50	0	100	100	75	0	100	100	75	0
	N. Dakota	100	100	50	25	100	100	25	0	100	50	25	0
	Washington	100	50	0	0	100	25	0	0	100	25	0	0
1990	Colorado	100	50	0	0	100	75	0	0	100	25	0	25
	Maine	100	75	0	0	75	0	0	0	75	75	0	0
	N. Dakota	100	25	0	0	100	0	0	0	100	0	0	0
	New York	75	50	0	0	0	0	0	0	0	0	0	0
	Washington	100	100	0	0	0	0	0	0	100	100	100	75

[a] Percentage of three blocks of 25 plants (1988) or four blocks of 15 plants (1989 and 1990) per plot.

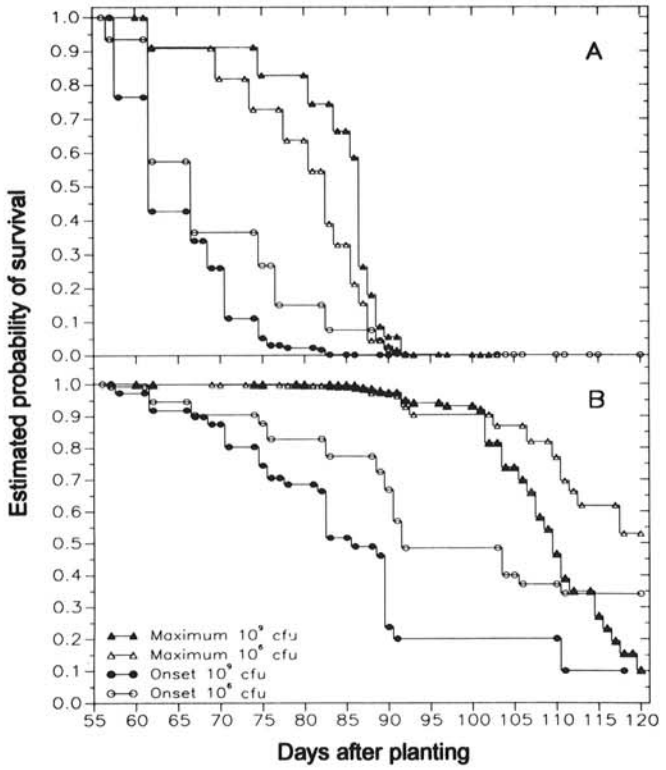

Fig. 1. Kaplan-Meier estimates of survival curves for onset and maximum incidence of foliar symptoms of bacterial ring rot in potato cultivar Russet Burbank given an initial inoculum dose of 10^6 or 10^9 colony forming units of *Clavibacter michiganensis* subsp. *sepedonicus* and grown during 1989 in **A,** Washington and **B,** Maine.

RESULTS

Typical symptoms of bacterial ring rot (e.g., chlorosis and wilting of leaves and stunting) were observed in all cultivars at all locations. Disease onset and time to maximum incidence of these symptoms were variable and ranged from 34 to 122 and 50 to 122 days after planting, respectively. Analysis of the data by proportional hazards models indicated that this variability could be attributed to the effects of cultivar, location, and cultivar-location interaction. Final proportional hazards models for symptom onset and maximum disease incidence (Table 1) are discussed.

Effect of inoculum dose on onset and maximum incidence of foliar symptoms. Initial attempts at modeling symptom onset and maximum disease incidence included models that incorporated terms describing the levels of inoculum dose. In these models, the hazard rates for the various inoculum dose-location-cultivar combinations were not proportionate over the levels of inoculum dose, with the estimated hazards of the higher doses tending to increase relative to those of the lower doses as the growing season progressed ($Z{:}PH > 0.48$ for inoculum-dose terms in all models). To meet the assumption of proportionality of the covariates required by the proportional hazards model, the data were stratified over the levels of inoculum dose. Only the 10^6 and 10^9 cfu doses were included in the models due to the high degree of right-censored observations present in the data at the lower inoculum levels (68.5, 42.6, 12.4, and 4.9% right-censored observations for 0, 10^2, 10^6, and 10^9 cfu, respectively [Table 2]). As a result, only qualitative statements can be made as to the effect of dose on onset and maximum disease incidence. Based on inspection of survival curves generated from the models shown in Table 1, there did not appear to be any significant effect of inoculum dose on either symptom onset or maximum disease incidence (Fig. 1). When apparent differences existed, however, the trend was toward an increased probability of survival at the lower inoculum doses, i.e., these plants tended to have an increased probability of surviving the entire season without developing symptoms. This relationship was most pronounced for symptom onset and maximum disease incidence during 1989 and was associated with a higher incidence of right-censored observations during that year (Table 2).

Effect of cultivar on symptom onset and maximum disease incidence. The overall effect of individual cultivars on symptom onset and maximum disease incidence was determined through the elimination or combination of coefficients within the cultivar term of the models (Table 1). Although no significant effect of cultivar on the hazard for symptom onset or maximum disease incidence was observed in 1989 ($P = 0.412$ and 0.417, respectively), cultivar was a significant determinant in the hazard for these events in 1988 and 1990. However, its effect was not consistent over years and between onset and maximum disease incidence. In 1988, for example, Russet Burbank and Norchip did not differ significantly in the hazard for symptom onset ($P = 0.149$), but both cultivars had significantly lower hazard for this event than did Norland ($P = 0.0006$). Conversely, in 1990 the hazard for symptom onset was significantly higher in Russet Burbank than in Norland ($P = 0.0$). Similar variability in cultivar effects on the hazard for maximum disease incidence also was observed. For example, the relative estimated hazard rate for maximum disease incidence in 1988 was significantly lower for Russet Burbank, relative to Norland or Norchip, which were not significantly different from one another ($P = 0.666$). In 1990, the overall hazard for maximum disease incidence was significantly lower for Nor-

chip ($P = 0.0496$) relative to Russet Burbank and Norland, which were not significantly different from one another ($P = 0.56$).

Effect of location on symptom onset and maximum disease incidence. The overall effect of individual locations on symptom onset and maximum disease incidence was determined by inspecting the full proportional hazards models and subsequently eliminating or combining coefficients within the location terms. Based on reduction of the full models, there was a significant effect of location on symptom onset and maximum disease incidence (Table 1), with certain locations displaying a trend toward a lower hazard for symptom onset and maximum disease incidence (Table 3). This trend remained relatively consistent over the three years of this study and between symptom onset and maximum disease incidence. For example, the ranking of locations, in order of increasing effect on the hazard of symptom onset, was North Dakota < Maine, Wisconsin < Colorado, Oregon < Washington in 1988; Maine, North Dakota < Colorado < Washington in 1989; and North Dakota < Colorado,Washington < New York in 1990 (based on reduction of the full models $P = 0.920$, 0.412, and 0.109 for 1988, 1989, and 1990, respectively). Similarly, for onset and maximum disease incidence in 1988, the rankings were North Dakota < Maine, Wisconsin < Colorado, Oregon < Washington; North Dakota < Wisconsin < Maine,Oregon < Colorado < Washington ($P = 0.115$ and 0.920 for onset and maximum disease incidence, respectively). The reduction in the hazard for symptom onset and maximum disease incidence was associated with a high number of right-censored observations in some locations (e.g., North Dakota in 1988 and 1989) but not the majority of locations (Table 2).

Effect of location by cultivar interaction on symptom onset and maximum disease incidence. After eliminating insignificant interaction terms from the models ($P > 0.05$), approximately 21% of the location by cultivar combinations included in our analysis were affected by significant location by cultivar interaction (Table 3). The effect of such interaction was not entirely consistent, but the trend was for the hazard of symptom onset and maximum disease incidence to be reduced by such interaction (76% of the total significant location by cultivar interactions) relative to that which would have been predicted if the interaction term had not been included in the model.

Prediction of foliar symptom development. Because the proportional hazards models can be used to obtain estimates of the survival probabilities for symptom onset and maximum disease incidence, they allow for the prediction of these events and the scheduling of inspections of potato seed lots. Table 4 shows the number of days after planting required at each location for survival probabilities of $P < 0.95$ and $P < 0.05$ to be realized for symptom onset and maximum disease incidence, respectively. These data indicated that symptom onset may occur as early as 50 days after planting for Russet Burbank in Oregon or as late as 72 days after planting for Norchip in North Dakota. Similarly, the models indicated that the number of days after planting where maximum

TABLE 3. Relative estimated hazard rates for both onset and maximum expression of foliar symptoms of bacterial ring rot in three potato cultivars grown in seven locations in the United States[a]

Year	Variable	Cultivar	Location						
			CO	ME	ND	NY	OR	WA	WI
1988	Onset	R. Burbank	1.00	0.02	0.003	n.d.[b]	1.00	36.30	0.02
		Norchip	1.00	1.28	0.003	n.d.	0.14[c]	0.25[c]	0.02
		Norland	0.61[c]	0.07	0.012	n.d.	0.61[c]	0.10[c]	0.07
	Maximum	R. Burbank	1.00	0.05	0.0003	n.d.	0.05	29.95	0.01
		Norchip	12.73	0.62	0.0032[c]	n.d.	0.03[c]	0.82[c]	0.06
		Norland	12.73	0.62	0.0032	n.d.	0.05[c]	0.04[c]	0.06
1989	Onset	R. Burbank	1.00	0.36	0.36	n.d.	n.d.	3.58	n.d.
		Norchip	0.97	0.36	0.36	n.d.	n.d.	3.58	n.d.
		Norland	0.97	0.36	0.36	n.d.	n.d.	1.24[c]	n.d.
	Maximum	R. Burbank	1.00	0.05	0.05	n.d.	n.d.	5.19	n.d.
		Norchip	1.00	0.05	0.05	n.d.	n.d.	5.19	n.d.
		Norland	1.00	0.05	0.05	n.d.	n.d.	0.62[c]	n.d.
1990	Onset	R. Burbank	1.00	10.49	0.15	n.e.[d]	n.d.	1.0	n.d.
		Norchip	n.e.	n.e.	n.e.	n.e.	n.d.	n.e.	n.e.
		Norland	0.02	0.25	0.03[c]	n.e.	n.d.	0.02	n.d.
	Maximum	R. Burbank	1.00	1.00	0.20	20.67	n.d.	n.e.	n.d.
		Norchip	0.62	0.62	0.12	30.53[c]	n.d.	n.e.	n.d.
		Norland	1.00	2.91[c]	1.45[c]	20.67	n.d.	n.e.	n.d.

[a] Relative estimated hazard defined as $h(t)/h_0(t)$ in which $h(t) = \exp(\Sigma\beta_i X_i)$ and is the hazard function at time $T = t$ and $h_0(t)$ is the hazard function for Russet Burbank in Colorado. Locations are represented by postal abbreviations.
[b] Foliar symptom data were not provided by this location.
[c] Significant ($P \leq 0.05$) cultivar by location interaction.
[d] No estimate available due to violation of the proportionality assumption of the proportional hazards model.

TABLE 4. Estimated number of days after planting required for onset and maximum incidence of foliar symptoms of bacterial ring rot[a] for three potato cultivars grown in seven locations in the United States

Variable	Cultivar	Location[b]						
		CO	ME	ND	NY	OR	WA	WI
Onset ($P < 0.95$)	R. Burbank	53.0 ± 5.6	61.3 ± 9.0	68.0 ± 11.1	n.e.[c]	50.0 ± 0.0	50.7 ± 6.2	71.0 ± 0.0
	Norland	58.7 ± 4.4	59.3 ± 7.5	65.3 ± 9.3	n.e.	56.5 ± 7.8	61.7 ± 3.4	66.0 ± 1.4
	Norchip	55.0 ± 6.0	56.0 ± 6.9	72.0 ± 11.6	n.e.	65.0 ± 0.0	59.8 ± 2.6	71.0 ± 0.0
Maximum ($P < 0.05$)	R. Burbank	101.3 ± 6.6	108.3 ± 1.3	115.0 ± 2.0	89.0 ± 1.4	108.5 ± 2.1	84.8 ± 5.00	113.0 ± 0.0
	Norland	99.3 ± 9.6	98.0 ± 3.2	107.7 ± 4.7	89.0 ± 1.4	106.5 ± 0.7	108.0 ± 2.8	106.0 ± 0.0
	Norchip	100.5 ± 11.0	103.0 ± 10.0	116.3 ± 3.1	82.5 ± 3.5	111.0 ± 2.8	91.3 ± 3.0	106.0 ± 0.0

[a] Estimates of survival probabilities were obtained by the models described in Table 1. Values shown are the mean estimated days after planting ± standard deviation for survival probabilities for onset and maximum disease incidence to reach $P < 0.95$ and $P < 0.05$, respectively, obtained for the years 1988–1990 for plants inoculated with 10^6 or 10^9 colony forming units of *Clavibacter michiganensis* subsp. *sepedonicus*.
[b] Locations are represented by postal abbreviations.
[c] Estimates were not available to violation of proportionality assumption of the proportional hazards model.

disease incidence was likely to have occurred ranged from 83 to 115 days after planting for Norchip in New York and Russet Burbank in North Dakota, respectively.

DISCUSSION

Previous investigations focused on the effect of factors such as inoculum dose (2,16,23,25), cultivar (2,3,7,15,22,25), and environment (1,13,14,17–20) on bacterial ring rot symptom expression. Such information has been useful in explaining some of the phenomena associated with bacterial ring rot, e.g., symptomless infection. Unfortunately, the trend has been for investigators to consider the effect of these factors on the magnitude of symptom expression without regard to the time frame during which symptom development occurs. The time period during which symptoms develop is as important as the magnitude of symptom expression because, as others have noted (2), the difference between symptomless and symptomatic infection may be simply a function of when plants are scored for symptoms.

Inoculum dose is an important factor in determining whether plants become infected (16,21) and whether symptoms are expressed after infection (16,23). In a previous study, the magnitude of response to inoculation with *C. m. sepedonicus* for a number of bacterial ring rot disease variables was correlated with inoculum dose, and the response to inoculum dose was modulated by both cultivar and geographic location (25). In contrast to previous reports that indicated a correlation between inoculum dose and symptom onset (2,23), the results of this study indicate that no proportional relationship between the hazard for symptom onset or maximum disease incidence and inoculum dose exists. Instead, the effect of inoculum dose on both symptom onset and maximum disease incidence appears to be limited to whether foliar symptoms will be expressed. The difference between our results and those previously reported (2) may be explained in part by differences in assessment technique, as the earlier study defined symptom onset as 50% of plants with ≥20% of the foliage wilted or necrotic, whereas our assessments did not require that a prespecified level of symptom expression occur and were based on any recognizable symptom of bacterial ring rot. The latter criterion recognizes symptoms, such as stunting or rosetting of the apical foliage, that are cultivar-specific (2,10) and influenced by location (25) and also may account for our inability to demonstrate a previously reported relationship between cultivar maturity and symptom onset (2). The late-maturing cultivar Russet Burbank, for example, can develop a distinct rosette (10), a symptom that is readily recognizable and that enables detection several weeks earlier than the more generally characteristic foliar wilting and necrosis. This, combined with the differing propensities of these cultivars to exhibit symptoms of bacterial ring rot (25), may explain the variability in the effect of cultivar on foliar symptom development and also may explain the sensitivity of the hazard for symptom onset and maximum disease incidence to modification by the interaction of cultivar and location.

There was a consistent trend for certain locations to have a significantly lower risk for both symptom onset and maximum disease incidence (e.g., North Dakota) than others (e.g., Colorado and Washington). Furthermore, the trend remained consistent between symptom onset and maximum disease incidence. One of the more obvious explanations for the effect of location on these events is the influence of this variable on both the magnitude of symptom expression and the type of symptoms expressed. In a previous study, much of the variability in the magnitude of foliar symptom expression was attributed to environmental conditions at a particular location, and at certain locations, plants tended toward enhanced symptom expression, including an increased propensity to develop stunting symptoms (25). In this study, location clearly affected the hazard for symptom onset and maximum disease incidence by decreasing the occurrence of symptom expression (e.g., North Dakota). The higher incidence of stunting that occurred in some of these locations (25) probably influenced the hazard for these events, because this symptom appears earlier than wilting and necrosis of foliage. However, environment also may have a direct impact on these events because locations that did not differ with respect to the amount of stunting that occurred or the incidence of right-censored observations had different hazards for symptom onset and maximum disease incidence, and these hazards varied over seasons.

The influence of location is such that the current practice of scheduling inspections based solely on planting date (e.g., 60–70 and 90–100 days after planting for two inspections) seems inadequate. The survival probabilities predicted by the models indicate that, for many of the cultivar-location combinations in this study, symptom onset will have occurred by the time early inspections are conducted, a fact largely not realized in most locations. Furthermore, second inspections that traditionally key on bacterial ring rot detection may not occur as late as necessary to assure disease detection. Based on our results, an environmentally based model should be useful for predicting the onset or maximum incidence of foliar symptoms of bacterial ring rot. We are now evaluating environmental data collected at each location for relationships to both the magnitude and timing of symptom expression, with the goal of developing predictive models for bacterial ring rot symptom expression that can be used independently or as part of an integrated pest-management predictive program (9,24).

LITERATURE CITED

1. Bishop, A. L., and Slack, S. A. 1982. Effect of temperature on development of ring rot of potato. (Abstr.) Phytopathology 72:1382.
2. Bishop, A. L., and Slack, S. A. 1987. Effect of cultivar, inoculum dose, and strain of *Clavibacter michiganense* subsp. *sepedonicum* on symptom development in potatoes. Phytopathology 77:1085-1089.
3. Bonde, R., and Covell, M. 1950. Effect of host variety and other factors on pathogenicity of potato ring-rot bacteria. Phytopathology 40:161-172.
4. Breslow, N. E. 1975. Analysis of survival data under the proportional hazards model. Int. Stat. Rev. 43:45-58.
5. Breslow, N. E. 1979. Statistical methods for censored survival data. Environ. Health Perspect. 32:181-192.
6. Cox, D. R. 1972. Regression models and life tables (with discussion). J. R. Stat. Soc. B 74:187-220.
7. De Boer, S. H., and McCann, M. 1990. Detection of *Corynebacterium sepedonicum* in potato cultivars with different propensities to express ring rot symptoms. Am. Potato J. 67:685-694.
8. De Boer, S. H., and Slack, S. A. 1984. Current status and prospects for detecting and controlling bacterial ring rot of potatoes in North America. Plant Dis. 68:841-844.
9. Ewing, E. E., Heym, W. D., Batutis, E. J., Snyder, R. G., Ben Khedher, M., Sandlan, K. P., and Turner, A. D. 1990. Modifications to the simulation model POTATO for use in New York. Agric. Syst. 33:173-192.
10. Guthrie, J. W. 1959. The early dwarf symptom of bacterial ring rot of potato in Idaho. Phytopathology 49:453-454.
11. Harrell, F. E. 1986. SAS SUGI Supplemental User's Guide. SAS Institute, Inc., Cary, NC.
12. Kalbfleisch, J. D., and Prentice, R. L. 1980. The Statistical Analysis of Failure Time Data. John Wiley & Sons, New York. 321 pp.
13. Kurowski, C. J., and Gudmestad, N. C. 1990. The effect of ethylene and abscisic acid on symptom expression of bacterial ring rot in eggplant and potato. Am. Potato J. 67:443-459.
14. Logsdon, C. E. 1967. Effect of soil temperature on potato ring rot. Am. Potato J. 44:281-286.
15. Manzer, F. E., and McKenzie, A. R. 1988. Cultivar response to bacterial ring rot infection in Maine. Am. Potato J. 65:333-339.
16. Nelson, G. A. 1982. *Corynebacterium sepedonicum* in potato: Effect of inoculum concentration on ring rot symptoms and latent infection. Can. J. Plant Pathol. 4:129-133.
17. Nelson, G. A., and Kozub, G. C. 1983. Effect of total light energy on symptoms and growth of ring rot-infected red pontiac potato plants. Am. Potato J. 60:461-468.
18. Nelson, G. A., and Kozub, G. C. 1987. Effect of temperature and latent viruses on atypical ring rot symptoms of Russet Burbank potatoes. Am. Potato J. 64:589-597.
19. Paquin, R., and Genereux, H. 1976. Effet du climat sur la fletrissure bacterienne de la pomme de terre et relation avec le contenu en sucres des tiges. Can. J. Plant Sci. 56:549-554.

20. Sherf, A. F. 1944. Infection experiments with potato ring rot and the effect of soil temperature on the disease. Am. Potato J. 21:27-29.
21. Sletten, A. 1980. Observations on systemic infection of potato plants grown from stem cuttings inoculated with *Corynebacterium sepedonicum*. Potato Res. 23:111-113.
22. Sletten, A. 1985. The effect of *Corynebacterium sepedonicum* on symptoms and yield of four potato cultivars. Potato Res. 28:27-33.
23. Starr, G. H. 1947. The effect of different concentrations of bacterial suspensions used in inoculations upon subsequent ring rot symptoms in the potato plant. Am. Potato J. 24:151-156.
24. Stevenson, W. R., Binning, L. K., Wyman, J. A., Curwen, D., Rice, G. J., Zadja, J. R., Smidl, J. E., Thorson, B. M., and Schmidt, R. W. 1989. PCM—The Integrated Systems Approach to Potato Crop Management. User's Guide for IBM and IBM-Compatible Personal Computers, Version 1.1. University of Wisconsin-Madison IPM Program.
25. Westra, A. A. G., and Slack, S. A. 1994. Effect of interaction of inoculum dose, cultivar, and geographic location on magnitude of bacterial ring rot symptom expression in potato. Phytopathology 84:228-235.