Effect of Interaction of Inoculum Dose, Cultivar, and Geographic Location on the Magnitude of Bacterial Ring Rot Symptom Expression in Potato

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ABSTRACT

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The effect of the interaction of inoculum dose of *Clavibacter michiganensis* subsp. *sepedonicus*, potato cultivar, and geographic location on the magnitude of bacterial ring rot symptom expression was investigated by planting an inoculum dose × cultivar factorial experiment in several potato-growing regions of the United States. Inoculum dose was positively correlated with the expression of both foliar and external tuber symptoms of bacterial ring rot and negatively correlated with total yield and vine length. The magnitude of the response to inoculum dose was modulated primarily by its interaction with cultivar and location. Three-way interaction between inoculum dose, cultivar, and location was generally not significant. The relative effect of cultivar on the inoculum dose response for foliar symptom expression, vine length, and yield remained constant (Russet Burbank > Norchip ≥ Norland), irrespective of geographic location, whereas its effect on tuber symptom expression was variable. The relative effect of location on dose response was more variable than that of cultivar but followed the general pattern: Washington ≥ Maine, New York, Oregon > Colorado, Wisconsin ≥ North Dakota. Variability in the effect of location on dose response may indicate that environmental conditions specific to each location influence the expression of bacterial ring rot symptoms. Variation in the effect of location on the different disease response variables was also observed within a single growing season. This, combined with the poor correlation observed among these variables, indicated that environmental conditions may affect these responses differentially and that they are likely to be of little value as predictors of one another.

Additional keyword: general linear model

Bacterial ring rot of potato (*Solanum tuberosum* L.), caused by the gram-positive bacterium *Clavibacter michiganensis* subsp. *sepedonicus,* is a major threat to the North American seed potato industry. Bacterial ring rot is controlled primarily through enforcement of a zero tolerance limit for the disease in seed potato lots (5), a practice currently achieved through a series of pre- and postharvest inspections. Since enforcement of the zero tolerance limit is dependent upon visual detection of symptomatic plants and/or tubers, the efficacy of this control method is influenced directly by factors controlling symptom expression. Thus, a more thorough understanding of the parameters that influence bacterial ring rot symptoms could contribute to an improved efficiency in the detection of this disease and a more effective application of the zero tolerance limit.

Both foliar and tuber symptoms of bacterial ring rot can be expressed by afflicted plants. Foliar symptoms of bacterial ring rot are manifested typically as interveinal chlorosis followed by wilting and eventual necrosis of the affected tissues. Stunting and rosetting have also been described as foliar symptoms in some cultivars (1,6). Tubers displaying symptoms of bacterial ring rot are characterized by a breakdown of the vascular ring of the tuber. In severe cases, the destruction of the vascular ring can extend to and cause cracking of the tuber periderm, with a concomitant loss in tuber yield. Expression of these symptoms is affected by a number of factors including *C. m. sepedonicus* strain (1), inoculum concentration (1,11,20), host cultivar (1,3,5,6,10,19), and environment (2,7–9,13,14,16,18). Typically, these studies considered only the effect of a single variable (e.g., inoculum dose or cultivar) on a single disease response (e.g., incidence of tuber symptoms). As a result, there is little or no information on how these factors interact and how this interaction affects the magnitude of bacterial ring rot symptom expression. Furthermore, only a single disease variable usually is examined, so the effect of the interaction of factors such as inoculum dose, cultivar, and environment on the simultaneous expression of several disease variables is unknown.

The objective of this study was to determine the nature of the interaction among inoculum dose, cultivar, and geographic location and to assess the effect of such interaction on several disease response variables (incidence of foliar and tuber symptoms, stunting, and yield loss). In the process of making this determination, quantitative comparisons of the relative reaction of three potato cultivars (Norchip, Norland, and Russet Burbank) grown in several locations across the United States were made.

MATERIALS AND METHODS

Inoculum preparation. *C. m. sepedonicus* strain SS43 (1) was maintained in lyophilized form in a 7% peptone and 7% sucrose solution and was cultured on nutrient broth-yeast extract agar (NBY) (21) at ambient temperature (approximately 23 C). Bacteria were renewed from lyophilized cultures after a maximum of five NBY transfers. After approximately 5 days of growth on NBY, several *C. m. sepedonicus* colonies were transferred via an inoculating loop to a 100-ml flask containing 50 ml of filter-sterilized (0.2 μm) LM medium (22) and agitated on a rotary shaker for approximately 72 h at 150 rpm and 24 C. Five milliliters of this solution was then transferred to a 1-L flask containing 500 ml of LM and grown on a rotary shaker for 5–7 days at 24 C and

150 rpm. Cells were then harvested by centrifugation for 20 min at 15,000 *g* at 4 C, resuspended in 0.05 M phosphate buffer (PB) at pH 7.2, and adjusted to cell densities of 10^4, 10^8, or 10^{11} colony-forming units (CFU)/ml in PB based on $A_{600} = 0.1$ being equivalent to 1.0×10^8 CFU/ml.

Seed preparation and inoculation. Certified seed potatoes (cvs. Norchip, Norland, and Russet Burbank), obtained from a single source, were removed from cold storage a minimum of 72 h before cutting. Melon scoops were used to prepare uniform seed pieces (approximately 5–10 g), each having a single sprout. Seed pieces were then placed in loosely closed plastic bags for 48 h to allow suberization prior to inoculation. After suberization, seed pieces were wounded by inserting a pipette tip at a 45-degree angle on either side of the sprout such that the wounds met directly beneath the sprout, and 10 μl of PB (control plants) or bacteria suspended in PB were then injected into the wound so that the seed pieces each received an inoculum dose of 0, 10^2, 10^6, or 10^9 CFU of *C. m. sepedonicus*. After inoculation, seed pieces were delivered via overnight express to cooperators and stored at 4 C until planted.

Experimental design. A factorial treatment design of inoculum dose and cultivar (4 × 3) was used. Treatments were assigned to plots according to a randomized complete block design. Each treatment was replicated three times (25 plants/plot) in 1988 or four times (15 plants/plot) in 1989 and 1990 in diverse potato-growing regions of the United States (Table 1). Stand (number of plants) and foliar disease incidence (i.e., the proportion of plants displaying bacterial ring rot symptoms) were recorded weekly for each plot. Plants were scored as symptomatic on the basis of interveinal chlorosis and wilting of leaves and, in the case of Russet Burbank, stunting and rosetting of foliage. Vine length of at least three randomly selected plants per plot was measured approximately 80 days after planting. At harvest, at least 20 randomly selected tubers from each plot were scored for external symptoms of bacterial ring rot, and the total yield of each plot was recorded. Yield data were analyzed as the mean yield per plant (total yield/stand) for each plot to correct for differences in final stand among plots.

TABLE 1. Location of experiments by year of study

Location	Year 1988	 1989	 1990
San Luis Valley, Colorado	+	+	+
Presque Isle, Maine	+	+	+
Ithaca, New York	−	−	+
Fargo, North Dakota	+	+	+
Corvallis, Oregon	+	+	+
Prosser, Washington	+	+	+
Arlington, Wisconsin	+	−	−

Statistical analysis. General linear models were fit for vine length, incidence of external tuber symptoms, total yield, and relative area under disease incidence curves (AUDIC) (17) with PROC GLM in SAS (SAS Insitute, Cary, NC); data for Russet Burbank grown in Washington (1989 and 1990) or Wisconsin (1988) were used as the baseline response against which the other cultivars and locations were compared. Data were transformed to correct nonlinearity and unequal variances as appropriate, and the relative effects of both individual cultivars and locations on the magnitude of bacterial ring rot symptom expression were determined by comparison of their overall contributions to the inoculum dose response (i.e., the slope) for each disease response variable. Only differences in slope were considered, because the intercept of each model represents the baseline response for each cultivar and location against which the disease response was measured. Models were initially reduced by removal of insignificant ($P > 0.05$) interaction terms. In the case of significant interaction, model reduction was carried out within the term, i.e., by the elimination of insignificant coefficients ($P > 0.05$) and/or the combination of coefficients that were not significantly different from one another on the basis of their standard errors. Loss of predictive value of the model due to reduction of slope terms was assessed at each step by the *F* test for reduction in model sums of squares (15), with a significance level of $P > 0.05$ used as the criterion for acceptance of the reduced models. *P* values given for model terms and coefficients are based on type III sums of squares.

RESULTS

Although typical symptoms of bacterial ring rot were observed for each cultivar in all locations, the magnitude of response to inoculation with *C. m. sepedonicus* varied among cultivars and locations. Analysis of the various disease response variables by the general linear model procedure explained a minimum of 53.2% of the observed variability in the data (Table 2). Based on inspection of the partitioned model sums of squares (Table 3), most of the variability explained by the models was attributable to differences in the response of the control plants grown at the various locations. Over the 3 yr of this study, an average of 17–62, 83–99, 46–78, and 55–89% of the model sums of squares were attributed to the intercept terms (i.e., cultivar, location, and cultivar by location terms) of the models representing AUDIC, vine length, tuber symptoms, and yield, respectively. The remaining variability explained by the models was attributable to the slope terms and is discussed below.

TABLE 2. Analysis of variance table for general linear models describing four bacterial ring rot disease variables for the years 1988–1990[a]

Variable[b]	Year	R^2	Model df	Error df	Model sum of squares	Error sum of squares	Mean square error
AUDIC	1988[c]	0.720	22	193	6.46	2.51	0.01
	1989[c]	0.723	15	176	10.43	4.00	0.02
	1990[c]	0.848	19	219	13.66	2.44	0.01
Vine length	1988	0.821	13	129	147,779.84	3,219.84	24.96
	1989	0.665	14	177	10,235.68	5,165.88	29.19
	1990	0.662	19	267	16,295.12	8,308.21	31.12
Tuber symptoms	1988[d]	0.676	16	127	124.38	59.57	0.47
	1989[d]	0.785	16	175	252.77	69.22	0.40
	1990[d]	0.532	16	222	177.20	155.69	0.70
Yield	1988[c]	0.842	18	161	5.48	1.03	0.01
	1989	0.826	17	174	55.32	11.68	0.07
	1990	0.697	18	220	83.16	36.11	0.16

[a] ANOVA results given are for reduced general linear models and were obtained from SAS output. Results were obtained with inoculum dose levels that had been transformed by the natural logarithm.

[b] AUDIC = relative area under disease incidence curves; tuber symptoms = proportion of a minimum of 20 randomly selected tubers showing external symptoms of bacterial ring rot; yield = total yield/final stand.

[c] Square root transformation of dependent variable.

[d] Logit transformation of dependent variable.

TABLE 3. Partitioned model sums of squares for general linear models describing four bacterial ring rot disease variables for the years 1988-1990[a]

			Slope terms			
				Interaction terms		
Variable[b]	Year	Intercept[c]	Dose	Location	Cultivar	Location × cultivar
AUDIC	1988[d]	2.5	0.3	0.2	0.5	0.6
	1989[d]	1.0	4.1	0.7	0.03	0.1
	1990[d]	2.8	1.4	3.0	0.0	0.4
Vine length	1988	12,757.8	0.0	246.0	0.0	0.0
	1989	9,741.6	1,085.2	311.1	531.4	0.0
	1990	15,011.6	1,028.9	0.0	500.5	0.0
Tuber symptoms	1988[e]	89.1	24.0	7.8	2.0	7.4
	1989[e]	123.9	17.6	8.5	5.3	4.1
	1990[e]	80.0	87.5	4.5	0.0	0.0
Yield	1988[d]	4.0	0.0	0.4	0.04	0.0
	1989	43.1	18.3	15.9	0.3	1.2
	1990	58.2	18.1	11.4	1.6	0.0

[a] Type III model sums of squares obtained for reduced general linear models from SAS output. Results were obtained with inoculum dose levels that had been transformed by the natural logarithm. Values are rounded to one decimal place.

[b] AUDIC = relative area under disease incidence curves; tuber symptoms = proportion of a minimum of 20 randomly selected tubers showing external symptoms of bacterial ring rot; yield = total yield/final stand.

[c] Includes sums of squares attributed to cultivar and location main effects and location × cultivar interaction.

[d] Square root transformation of dependent variable.

[e] Logit transformation of dependent variable.

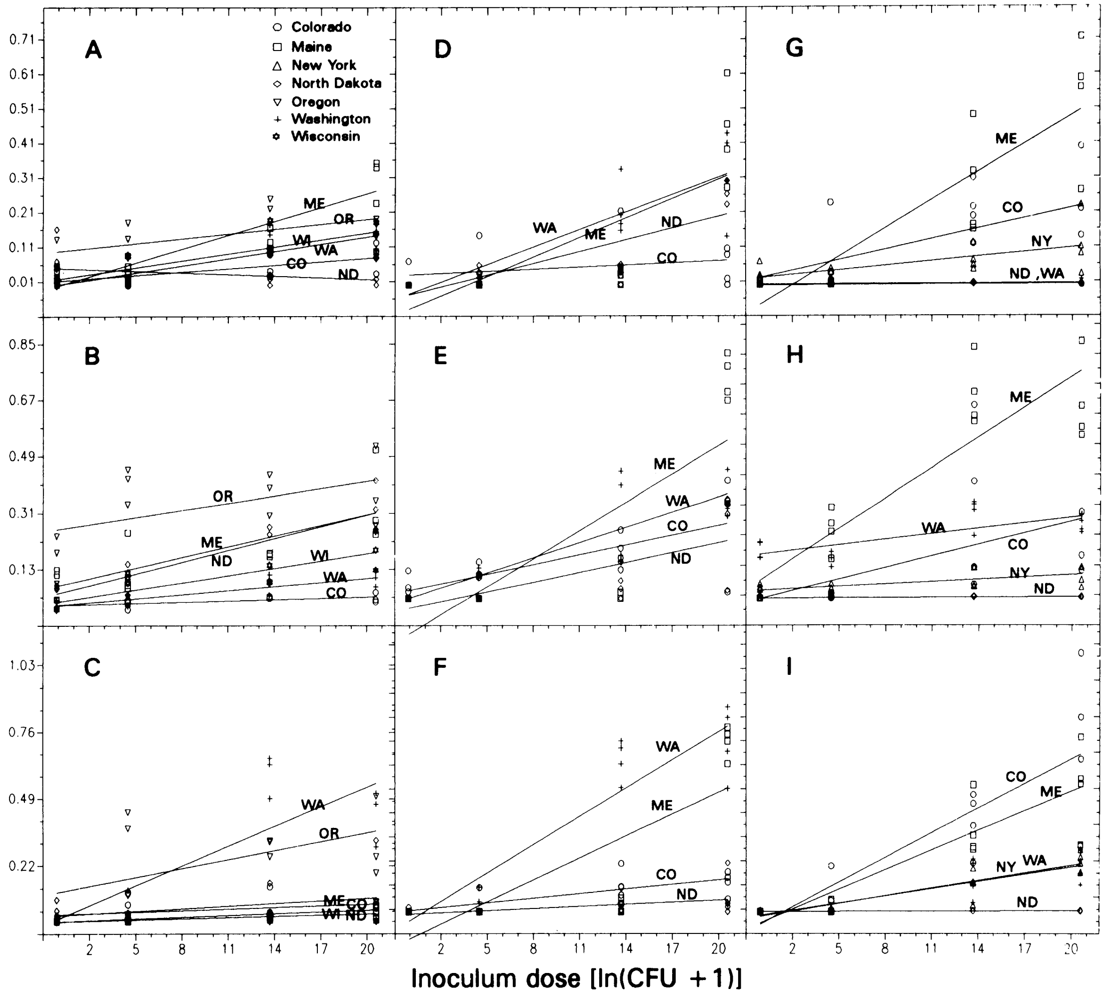


Fig. 1. Effect of inoculum dose of *Clavibacter michiganensis* subsp. *sepedonicus* on relative area under bacterial ring rot disease incidence curves (AUDIC) of three potato cultivars grown in several locations across the United States. **A-C,** Respective responses for cultivars Norchip, Norland, and Russet Burbank in 1988, **D-F,** in 1989, and **G-I,** in 1990. Regression lines, included to indicate trends, were generated by the graphics software and do not necessarily describe intercepts and slopes estimated by the general linear models described in Tables 2 and 3 for transformed data. The regression line for each location is indicated by the state abbreviation.

Effect of inoculum dose. The overall magnitude of response for each of the disease variables was correlated with inoculum dose and accounted for 6 to 68% of the variation not explained by differences in the model intercepts (Table 3). Both foliar disease incidence, as judged by AUDIC (Fig. 1), and the incidence of external tuber symptoms (Fig. 2) were positively correlated with increased inoculum dose in all 3 yr ($P = 0.0001$ for each model in each year), whereas vine length (Fig. 3) and yield (Fig. 4) were negatively correlated with increased inoculum dose in 1989 and 1990 ($P = 0.0041$ and 0.0001 for vine length in 1989 and 1990, respectively, and $P = 0.0001$ and 0.0001 for yield in both years). No significant effect of inoculum dose on vine length and yield was observed in 1988 ($P = 0.0646$ and 0.1032, respectively).

Interaction of cultivar and inoculum dose. Inspection of plots of disease response variables versus increasing inoculum dose (Figs. 1-4) indicated potentially significant inoculum dose-by-cultivar interaction. Although such interaction was not significant for AUDIC in either 1989 or 1990 ($P = 0.2366$ and 0.8227, respectively), vine length in 1988 ($P = 0.1123$), or external tuber symptoms in 1990 ($P = 0.5015$), it was a significant factor in the remaining two-thirds of the models and accounted for 0.4-12.4% of the variability explained by these models (Table 3). When significant interaction existed, the trend was for an overall decrease in the inoculum dose response of Norchip and Norland relative to that of Russet Burbank (Fig. 5). For example, the overall inoculum dose responses of Norland and Norchip for AUDIC in 1988 were not significantly different from one another but were significantly decreased relative to that of Russet Burbank ($P = 0.0001$).

The overall effect of cultivar on dose response for vine length and yield was similar to that observed for AUDIC. In 1989 and 1990, there was no significant difference in the inoculum dose response of Norland and Norchip on vine length, whereas the inoculum dose response of these cultivars was significantly less than that of Russet Burbank ($P = 0.0013$ for 1989 and 0.0001 for 1990). Similarly, there was no significant difference in the overall inoculum dose response for total yield of Norland and Norchip, whereas this response was significantly less for these cultivars than that of Russet Burbank ($P = 0.015$, 0.0406, and 0.0021 for 1988-1990, respectively).

In contrast, the effect of cultivar on the dose response for the incidence of external tuber symptoms was not consistent. In 1988 and 1989, the responses of Russet Burbank and Norchip to inoculum dose were not significantly different from one another but were significantly lower than Norland in 1988 ($P = 0.0409$) and significantly higher than Norland in 1989 ($P = 0.0003$).

Interaction of location and inoculum dose. In addition to the cultivar effects described above, examination of Figures 1-4 indicated an effect of location on the dose response. Interaction between inoculum dose and location was found to be significant in all of the models except that describing vine length in 1990 ($P = 0.2194$). The percentage of variation explained by the models that was accounted for by this interaction ranged from 1.9 to 39.5%. The effect of inoculum dose-by-location interaction was

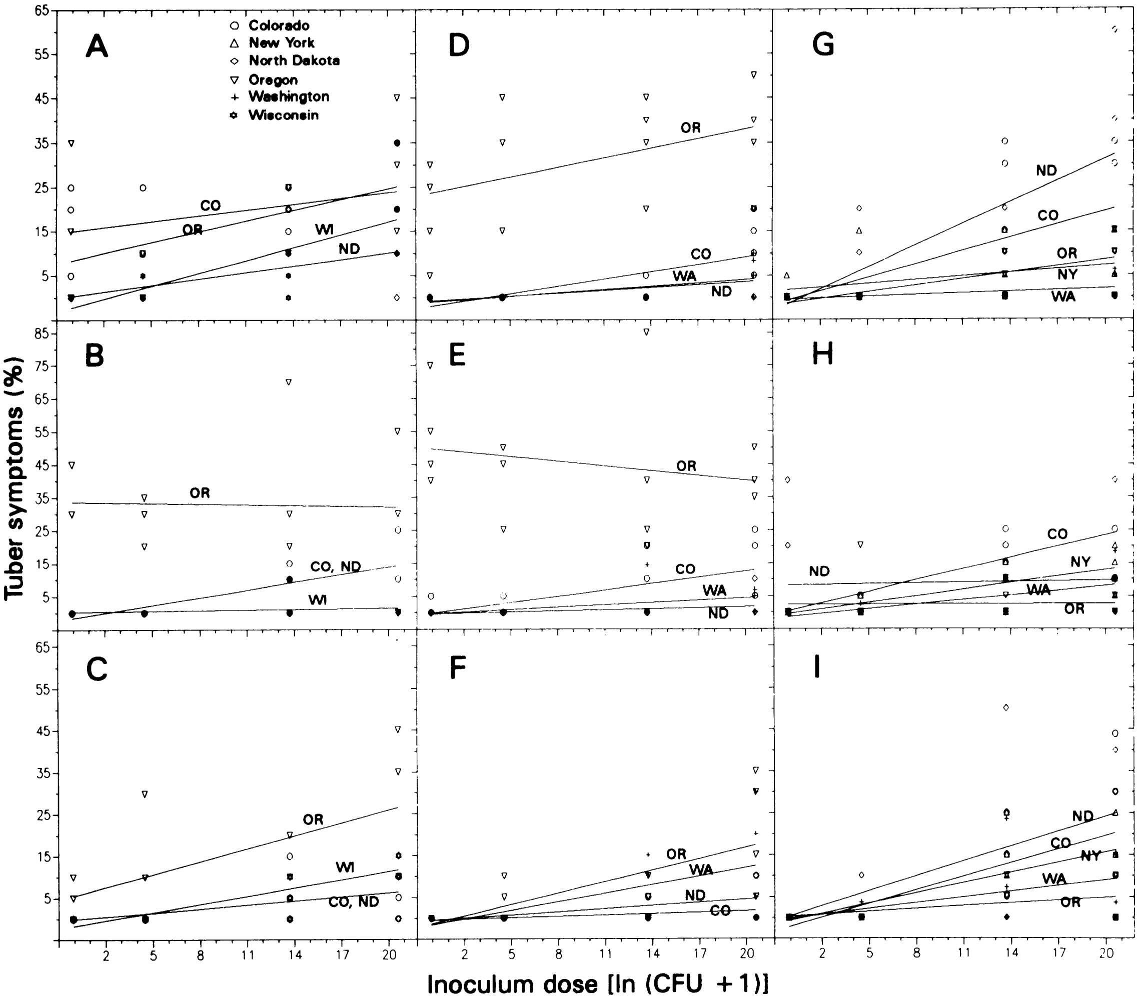


Fig. 2. Effect of inoculum dose on the incidence of external tuber symptoms of bacterial ring rot in potato cultivars inoculated with *Clavibacter michiganensis* subsp. *sepedonicus* and grown in several locations across the United States. **A-C**, Respective responses for cultivars Norchip, Norland, and Russet Burbank in 1988, **D-F**, in 1989, and **G-I**, in 1990. Regression lines, included to indicate trends, were generated by the graphics software and do not necessarily describe intercepts and slopes estimated by the general linear models described in Tables 2 and 3 for transformed data. The regression line for each location is indicated by the state abbreviation.

not, however, as consistent as that observed for the inoculum dose-by-cultivar interaction (Fig. 5), with variation in location effect occurring across disease variables as well as year of study. For example, reduction of the model describing AUDIC in 1988 resulted in a common coefficient representing Colorado, North Dakota, and Oregon, which significantly ($P = 0.0004$) reduced the inoculum dose response relative to that for Maine, Washington, and Wisconsin; the results obtained for the incidence of external tuber symptoms in the same year indicated that the response to inoculum dose was significantly increased in Colorado ($P = 0.0009$) and decreased in North Dakota ($P = 0.0266$) relative to Oregon and Wisconsin. Similarly, the results for AUDIC in 1988 differ somewhat from those obtained for the same disease variable in 1990, where the inoculum dose response was significantly increased in Colorado ($P = 0.0001$) and Maine ($P = 0.0001$) relative to that of North Dakota, New York, and Washington.

Although the variability in the effect of location on inoculum dose response of the various disease variables was greater than that observed for dose-by-cultivar interaction, there was a general trend towards a lower inoculum dose response in locations such as North Dakota, whereas locations such as Washington tended to have a higher overall response. Based on inspection of the groupings resulting from reduction of the inoculum dose-by-location interaction terms of the models (Fig. 5), the overall pattern of the relative effect on dose response of the individual locations, in order of decreasing effect, appeared to be Maine, Washington > Oregon, Colorado, New York ≥ Wisconsin, North Dakota.

Interaction of inoculum dose, cultivar, and location. The inoculum dose response for each of the disease variables appeared to be affected primarily by interaction with cultivar and location. However, in approximately 8% (14/168) of the total cultivar-location combinations included in the models, the dose response was further modified by significant three-way interaction of inoculum dose, cultivar, and location (Fig. 5). With the exception of AUDIC in 1988, such interaction accounted for less than 6% of the variability explained by each of the models (Table 3). In all cases, the effect of this interaction was to reduce the inoculum dose response of the affected cultivar-location combination. The frequency of significant three-way interactions appeared to be related to the degree of subjectivity associated with the assessment of each disease variable, as was the proportion of the variability accounted for by this interaction (Table 3). There was no significant inoculum dose-by-cultivar-by-location interaction in the models describing vine length in any of the 3 yr of this study ($P = 0.5576$, 0.3391, and 0.514, respectively, for 1988, 1989, and 1990 full models). Inoculum dose-by-cultivar-by-location interaction was not significant in the models describing yield in 1988 or 1990 ($P = 0.2677$ and 0.1304, respectively) and had a significant effect on yield in 1989 only (for the cultivars Norland and Norchip in Colorado, $P = 0.0037$ and 0.0021, respectively). In contrast, the inoculum dose response for the incidence of external tuber symptoms was affected by significant three-way interaction for Norland in Colorado ($P = 0.0001$) in 1988 and in Oregon ($P = 0.0015$) in 1989; the majority (71%) of the significant three-way interactions occurred in the models for AUDIC,

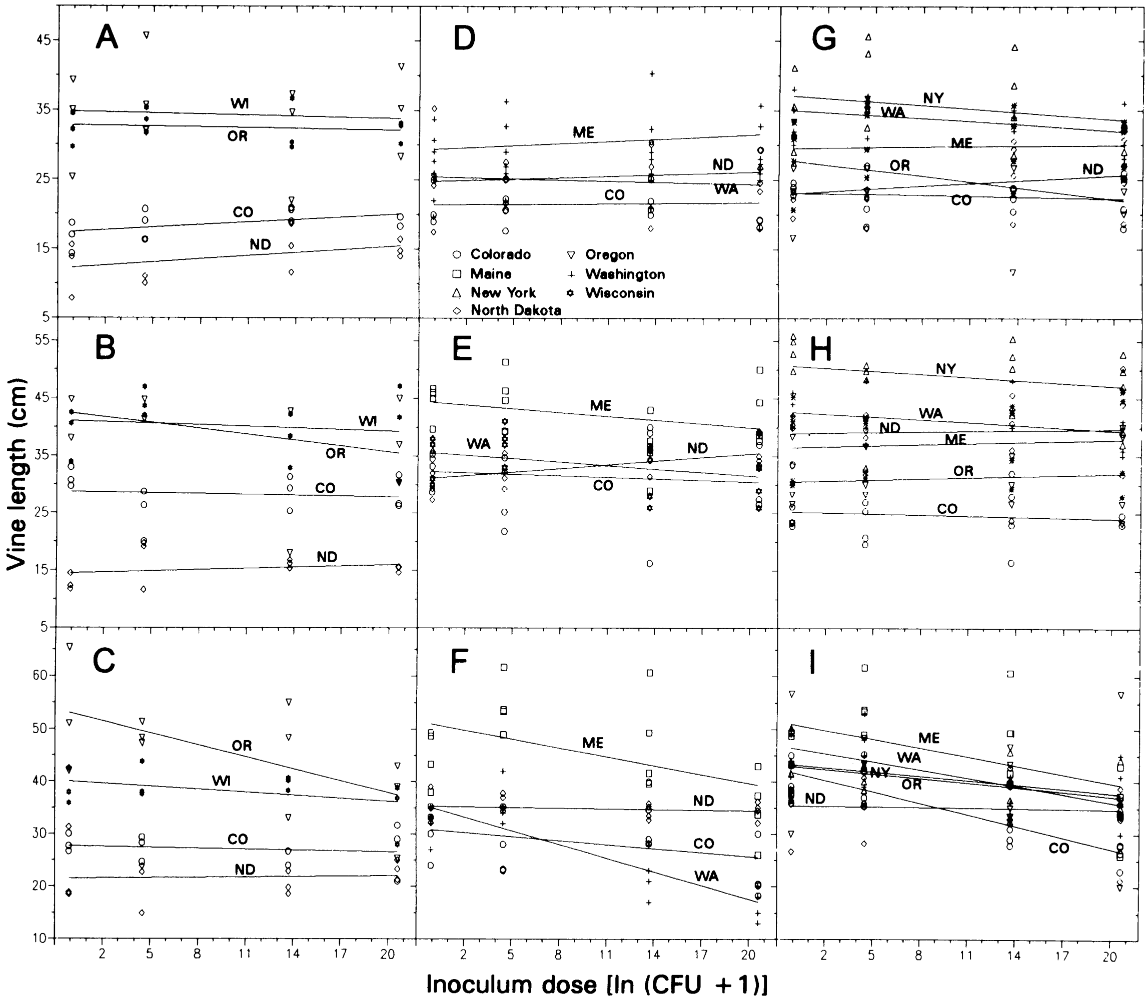


Fig. 3. Effect of inoculum dose on vine length of three potato cultivars inoculated with *Clavibacter michiganensis* subsp. *sepedonicus* and grown in several locations across the United States. **A–C,** Respective responses for cultivars Norchip, Norland, and Russet Burbank in 1988, **D–F,** in 1989, and **G–I,** in 1990. Regression lines, included to indicate trends, were generated by the graphics software and do not necessarily describe intercepts and slopes estimated by the general linear models described in Tables 2 and 3 for transformed data. The regression line for each location is indicated by the state abbreviation.

with the inoculum dose response of Norland and Norchip being reduced in Oregon ($P = 0.0004$) and Washington ($P = 0.0001$) in 1988, in North Dakota in 1989 ($P = 0.0252$), and in Maine and North Dakota ($P = 0.0001$) in 1990.

DISCUSSION

The manifestation of symptoms of bacterial ring rot is a complex consisting of several responses: chlorosis and wilting of the foliage, stunting, development of tuber symptoms, and reduction of yield. The expression of these symptoms is affected by several factors, including *C. m. sepedonicus* strain (1), inoculum dose (1,11,20), cultivar (1,3,4,10,19), interaction with other pathogens (12,14), and environmental factors (2,7–9,13,14,16,18). In these studies, the tendency has been for investigators to consider the effect of a single factor (e.g., inoculum dose or cultivar) on one or more of the responses to bacterial ring rot without regard to the effect that the interaction of these factors may have on symptom expression. Furthermore, the relationships among the various disease responses have been largely ignored, as have been the effects of factors such as inoculum dose and cultivar on their simultaneous expression. The approach used in this study was to consider the effects of two factors, cultivar and geographic location, on the inoculum dose response of several bacterial ring rot disease variables. This approach enabled us to examine the effect of *C. m. sepedonicus* over a range of inoculum doses and to take into account any background interference due to the presence of other diseases and the normal senescence of healthy plants (1). Our results confirmed the previously reported relationship between inoculum dose and the magnitude of expression of bacterial ring rot symptoms (1,11,20) and indicated that inoculum dose is an important factor in determining the incidence of foliar and tuber symptoms, stunting, and yield reduction. However, in this study, a significant amount (up to 78%) of the explainable variability in foliar and tuber symptom expression was accounted for by the intercept portions of the models. This interference is sufficient to explain most of the apparent variability in host reaction to *C. m. sepedonicus*, an observation that underscores the difficulty of diagnosing bacterial ring rot using symptom expression alone, and indicates that, under conditions that do not favor the expression of bacterial ring rot symptoms, this background level of interference may be sufficient to prevent detection of low levels of the disease.

The inoculum dose response observed for the each of the disease variables examined was further modulated by cultivar and location. This modulation was the result of the additive effects of cultivar and location on inoculum dose, rather than cultivar-location interaction. Relative cultivar expression of foliar symptoms of bacterial ring rot, stunting, and yield reduction was constant and not affected by variation in inoculum dose response due to location or growing season. Russet Burbank appears to be more sensitive to bacterial ring rot than either Norland or Norchip. However, this characterization is dependent upon the disease variable used to make the assessment.

Recently, incidence of tuber symptoms was used to gauge relative cultivar sensitivity to bacterial ring rot (4,10). Although the

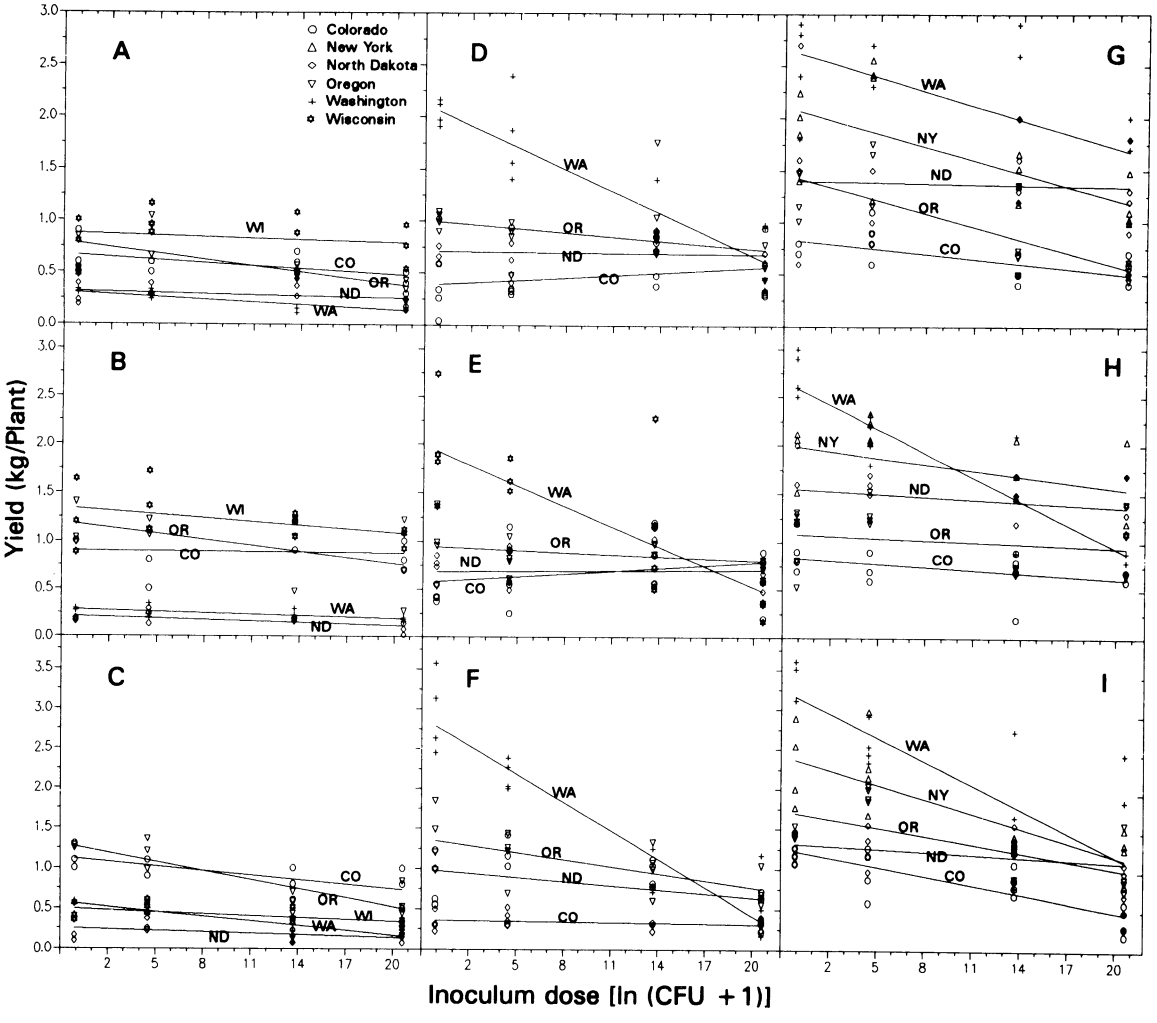
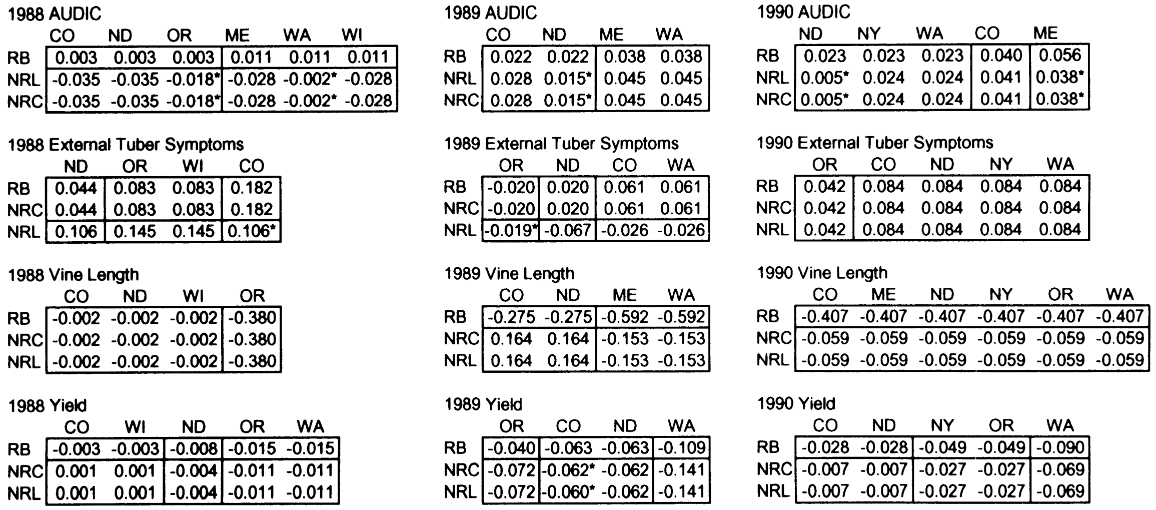


Fig. 4. Effect of inoculum dose on the yield of three potato cultivars inoculated with *Clavibacter michiganensis* subsp. *sepedonicus* and grown in several locations across the United States. **A–C**, Respective responses for cultivars Norchip, Norland, and Russet Burbank in 1988, **D–F**, in 1989, and **G–I**, in 1990. Regression lines, included to indicate trends, were generated by the graphics software and do not necessarily describe intercepts and slopes estimated by the general linear models described in Tables 2 and 3 for transformed data. The regression line for each location is indicated by the state abbreviation.

effect of cultivar on the expression of tuber symptoms should be understood, our results indicate that expression of tuber symptoms is highly variable between years and, therefore, a poor indicator of the relative sensitivity of cultivars to bacterial ring rot. On the other hand, foliar symptom expression remains constant among cultivars and can be expected to apply to all potato-growing regions. Furthermore, stunting in Russet Burbank did not appear to be limited to the western United States, as has been previously reported (6,10). The low correlation between the observed responses for the various disease variables examined in this study (Table 4) indicates that different factors may affect their expression and that none of the disease response variables can be used for accurate prediction of another. The practical implication of this observation is that separate models will be required for the prediction of the incidence of foliar and tuber symptoms of bacterial ring rot.

In the general linear models that were used to describe the disease variables, inoculum dose and the terms describing the interaction between inoculum dose, cultivar, and location encompass most of the sources of variability that affect the expression of bacterial ring rot symptoms. Location also modulates the inoculum dose-response relationship, but this modulation is not as consistent as the effect of cultivar. Variation observed in the modulation of the dose-response relationship by location may be explained by differences in the interpretation and assessment of bacterial ring rot symptoms by different cooperators. However, cooperator influence does not appear to be a significant factor, because one would expect a greater frequency of significant differences between locations than was observed if it were. Furthermore, the association of significant three-way interaction with the degree of subjectivity related to the assessment of the individual disease variables suggests that the models account for any cooperator influence by these interaction terms. Alternatively, environmental conditions specific to each location may account for the variation in the effect of location. Environmental conditions such as air temperature (2), soil temperature (9,18), soil fertility (8), day length (7), and total light energy (13) influence the degree to which bacterial ring rot symptoms are expressed. In this study, certain locations (e.g., North Dakota) displayed a trend towards reduced response to increased inoculum dose, while others (e.g., Washington) tended to have a higher overall response. This result supports the concept that environmental conditions directly affect symptom expression and that a change in these conditions, either within a locality or due to the transport of infected seed from one potato-growing region to another, will result in a change in the incidence of symptom expression. This finding provides one explanation for the sudden appearance of this disease in areas previously thought to be free of bacterial ring rot.

The effect of location on symptom expression, however, was inconsistent among the disease response variables within and between individual years of this study. The inability of a general indicator variable such as location to account consistently for the observed disease responses indicates that environmental conditions specific to both location and growing season influence the expression of bacterial ring rot symptoms. The inconsistency in the relative effect of location on the dose response of the various



1988 AUDIC

	CO	ND	OR	ME	WA	WI
RB	0.003	0.003	0.003	0.011	0.011	0.011
NRL	-0.035	-0.035	-0.018*	-0.028	-0.002*	-0.028
NRC	-0.035	-0.035	-0.018*	-0.028	-0.002*	-0.028

1989 AUDIC

	CO	ND	ME	WA
RB	0.022	0.022	0.038	0.038
NRL	0.028	0.015*	0.045	0.045
NRC	0.028	0.015*	0.045	0.045

1990 AUDIC

	ND	NY	WA	CO	ME
RB	0.023	0.023	0.023	0.040	0.056
NRL	0.005*	0.024	0.024	0.041	0.038*
NRC	0.005*	0.024	0.024	0.041	0.038*

1988 External Tuber Symptoms

	ND	OR	WI	CO
RB	0.044	0.083	0.083	0.182
NRC	0.044	0.083	0.083	0.182
NRL	0.106	0.145	0.145	0.106*

1989 External Tuber Symptoms

	OR	ND	CO	WA
RB	-0.020	0.020	0.061	0.061
NRC	-0.020	0.020	0.061	0.061
NRL	-0.019*	-0.067	-0.026	-0.026

1990 External Tuber Symptoms

	OR	CO	ND	NY	WA
RB	0.042	0.084	0.084	0.084	0.084
NRC	0.042	0.084	0.084	0.084	0.084
NRL	0.042	0.084	0.084	0.084	0.084

1988 Vine Length

	CO	ND	WI	OR
RB	-0.002	-0.002	-0.002	-0.380
NRC	-0.002	-0.002	-0.002	-0.380
NRL	-0.002	-0.002	-0.002	-0.380

1989 Vine Length

	CO	ND	ME	WA
RB	-0.275	-0.275	-0.592	-0.592
NRC	0.164	0.164	-0.153	-0.153
NRL	0.164	0.164	-0.153	-0.153

1990 Vine Length

	CO	ME	ND	NY	OR	WA
RB	-0.407	-0.407	-0.407	-0.407	-0.407	-0.407
NRC	-0.059	-0.059	-0.059	-0.059	-0.059	-0.059
NRL	-0.059	-0.059	-0.059	-0.059	-0.059	-0.059

1988 Yield

	CO	WI	ND	OR	WA
RB	-0.003	-0.003	-0.008	-0.015	-0.015
NRC	0.001	0.001	-0.004	-0.011	-0.011
NRL	0.001	0.001	-0.004	-0.011	-0.011

1989 Yield

	OR	CO	ND	WA
RB	-0.040	-0.063	-0.063	-0.109
NRC	-0.072	-0.062*	-0.062	-0.141
NRL	-0.072	-0.060*	-0.062	-0.141

1990 Yield

	CO	ND	NY	OR	WA
RB	-0.028	-0.028	-0.049	-0.049	-0.090
NRC	-0.007	-0.007	-0.027	-0.027	-0.069
NRL	-0.007	-0.007	-0.027	-0.027	-0.069

Fig. 5. Relative effect of location and cultivar on inoculum dose response of bacterial ring rot disease variables. Values shown indicate the slope for each combination of cultivar and location estimated with the general linear models described in Tables 2 and 3 for transformed data. Dividing lines within boxes indicate cultivar-location combinations that are not significantly different ($P > 0.05$) based on reduction of inoculum dose-by-cultivar and inoculum dose-by-location interaction terms of the full general linear models. For example, the overall response to inoculum dose of the relative area under disease incidence curves (AUDIC) in 1988 was greater for Russet Burbank (RB) than either Norchip (NRC) or Norland (NRL), which were not significantly different from one another. Similarly, the overall response to inoculum dose for AUDIC in 1988 in Colorado (CO), North Dakota (ND), and Oregon (OR) was not significantly different but was significantly less than that observed for Maine (ME), Washington (WA), and Wisconsin (WI). An asterisk indicates a significant ($P \leq 0.05$) inoculum dose-by-cultivar-by-location interaction.

TABLE 4. Spearman correlation of coefficients for bacterial ring rot disease response variables[a]

	AUDIC			Vine length			Tuber symptoms			Yield		
Variable[b]	ρ[c]	P[d]	n[e]	ρ	P	n	ρ	P	n	ρ	P	n
AUDIC	1.00	0.0001	647	−0.041	0.3223	574	0.411	0.0001	479	−0.207	0.0001	515
Vine length	−0.041	0.3223	574	1.00	0.0	622	−0.025	0.5716	526	0.508	0.0001	526
Tuber symptoms	0.411	0.0001	479	−0.025	0.5716	526	1.00	0.0	575	−0.092	0.0278	575
Yield	−0.207	0.0001	515	0.508	0.0001	526	−0.092	0.0278	575	1.00	0.00	611

[a] Correlation coefficients were calculated with data combined for all inoculum doses, cultivars, locations, and years of this study.

[b] AUDIC = relative area under disease incidence curves; tuber symptoms = proportion of a minimum of 20 randomly selected tubers showing external symptoms of bacterial ring rot; yield = total yield/final stand.

[c] Spearman correlation coefficient.

[d] P is the probability $>|R|$ under $H_0{:}\rho = 0$.

[e] The number of observations used to calculate the correlation coefficient.

disease variables is a further indication that environmental factors differentially influence their expression. If environmental conditions can be used to account for variation in disease response in a more consistent manner than is possible by a general indicator variable such as location, it may be possible to use such data to construct predictive models for bacterial ring rot. The construction of such models, particularly ones that predict the appearance of foliar symptoms of bacterial ring rot, would greatly enhance our ability to detect this disease in seed potato lots and thereby increase the efficacy of the zero tolerance limit as a control measure for bacterial ring rot.

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