

Structural Characterization of Bean Root Rot Epidemics

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ABSTRACT

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A 0.4-ha field of snapbeans (*Phaseolus vulgaris* 'Tendercrop') was divided into 100-contiguous quadrats. Twelve plants were removed from each quadrat at 10, 20, 30, 40, and 50 days after planting to assess disease severity and incidence of bean root rot induced primarily by *Rhizoctonia solani*, but also in part by *Fusarium solani* f. sp. *phaseoli*. A disease progress curve was developed to depict the epidemic occurring in each quadrat. The curve elements used to characterize these disease progress curves were: Weibull distribution function scale parameter; Weibull distribution function shape parameter; area under the disease progress curve; final disease severity; first-difference regression linear coefficient; first-difference regression quadratic coefficient; percent plants infected; and estimated time of disease onset.

Additional key word: comparative epidemiology.

The structure of the 100 disease progress curves was examined by conducting a multivariate Principal Axis Factor Analysis followed by a Varimax rotation. Four factors, accounting for 90% of the total variance, were selected to express the disease progress curve elements more simply and without confounding interactions among elements; i.e., to obtain greater parsimony. Factor 1 was an overall description of disease progression including shape, level, and rate parameters. Factor 2 showed a relationship between area under the disease progress curve and level of the epidemic. Factor 3, an indication of epidemic temporal location, identified the uniqueness of the estimated time of disease onset. Factor 4 identified a relationship between disease severity and disease incidence.

A disease progress curve (DPC) is a physical representation of an epidemic having certain definable characteristics. The analysis of these characteristics is an integral part of quantitative

epidemiology and can provide valuable information concerning the interrelationships among the components of epidemic systems.

Vanderplank (17) identified several important curve elements, including the time of disease onset and the rate of disease progression. Kranz (3-6), in a more complete characterization, specified 13 curve elements for a bilateral DPC. The relative

importance of individual elements in characterizing a DPC is determined by the degree of variability and intercorrelation among curve elements (5,6).

Numerous statistical techniques are available for the analysis of curve elements. One such is factor analysis which transforms an original set of variables, all of which are intercorrelated, into a set of fewer independent variables or factors. Curve elements chosen for inclusion in a factor analysis must accurately represent disease progression and must be reasonable, given the constraints of climatic conditions and crop type. Curve elements obtained for an annual, temperate-climate crop will be fewer than those obtained for a perennial, tropical crop with a complete bilateral DPC. Kranz (4,6) identified several biologically sound relationships by performing a Principal Axis Factor Analysis on the curve elements of 40 different host-pathogen combinations investigated for 2 yr.

A preliminary report on the structural characterization of bean root rot epidemics was presented (1). In the present study, factor analysis was used to identify interrelationships among eight DPC elements of naturally occurring bean root rot epidemics; interpretations of the resulting factors are presented.

MATERIALS AND METHODS

Cultural conditions and disease assessment. A 0.4-ha experimental field (soil type: Hagerstown loam) of beans (*Phaseolus vulgaris* L. "Tendercrop") was divided into 100 contiguous quadrats. Each 6 × 6-m quadrat had a row spacing of 0.92 m and a plant density of ~20 plants per meter of row. The field had not previously been planted to beans.

Twelve plants were arbitrarily selected and removed from each quadrat at 10, 20, 30, 40, and 50 days after planting. Plant specimens were brought to the laboratory, washed under running tap water for 8–10 min, and inspected to determine the severity of root rot induced by the complex of *Rhizoctonia solani* Kuehn and *Fusarium solani* (Mart.) Appel & Wr. emend. Snyder & Hans. f. sp. *phaseoli* (Burk.) Snyder & Hans. Disease severity was estimated as the proportion of the lower stem surface covered by fungal lesions. Disease incidence was taken as the proportion of diseased plants.

To verify the presence of *R. solani* and *F. solani* f. sp. *solani* in hypocotyl lesions, 50 plants with fungal lesions were selected from each 1,200-plant sample, and hypocotyl sections were surface-sterilized in a 5% sodium hypochlorite solution for 2–3 min. A small tissue sample was aseptically removed from the margin of at least one lesion of the hypocotyl section and placed on acidified 1.5% water agar in a 9-cm-diameter petri dish. Cultures were incubated for 7–10 days at room temperature and the fungi that were isolated were transferred to potato-dextrose agar for identification.

Identification of *R. solani* was based on hyphal characteristics (11). Isolates believed to be *F. solani* were grown under fluorescent light for 2 wk on potato-dextrose agar slants and then identified to species level on the basis of size and shape of macroconidia, presence of microconidia, and colony appearance (16). Isolates were identified to formae speciales by pathogenicity tests of selected isolates on bean tissue. For pathogenicity tests, a drop of an aqueous macroconidial suspension of an isolate was placed on an excised hypocotyl section of the bean cultivar Tendercrop and allowed to incubate in a covered petri dish lined with moist filter paper. The formation of a lesion and subsequent colonization of tissue beyond the limits of the water drop signified pathogenicity on the bean tissue.

Curve elements. A DPC was developed for each quadrat from the disease severity estimates. The disease proportion values were not transformed because any transformation implies certain underlying assumptions concerning the nature of disease progression.

The progress curves were then characterized by eight associated variables. Two of the descriptive variables were obtained through regression analysis. The DPCs were described using a first-difference regression (FDR) model (14,15):

$$y_t - y_{t-1} = B_1(x_t - x_{t-1}) + B_2(x_t^2 - x_{t-1}^2) + (e_t - e_{t-1})$$

in which y = disease proportion, x = time of disease assessment, B_1 = linear regression coefficient, B_2 = quadratic regression coefficient, e = error term, and t = cardinal numeral of the disease assessment (ie, first, second, third ...).

The FDR model regresses the change in disease on the change in time which results in independent error terms and increasing variance over time (14,15). If the regression coefficient of the quadratic term was not significantly different from zero, the quadratic model was reduced to the linear first-difference regression model. Appropriateness of the linear or quadratic model was determined by analysis of residual plots from ordinary least-squares regression and significance of the regression coefficients (10).

Additional descriptive variables included: the area under the disease progress curve (ADPC) as estimated with a FORTRAN IV program utilizing the midpoint rule; the time of disease onset (X_0) as estimated by the midpoint of the interval between the last date when disease severity was zero and the first date when disease severity was greater than zero; final disease severity (Y_f , the disease proportion occurring in the sample taken 50 days after planting); and the percentage of plants infected (PPI) taken as the percentage of diseased plants in the sample 50 days after planting.

Epidemics also were characterized by fitting curves with the Weibull distribution function (WDF) (12,13,18):

$$y = 1 - \exp [-(x-a)/b]^c]$$

in which y = disease proportion, x = time of disease assessment, a = location parameter, b = scale parameter, and c = shape parameter.

The WDF is a model used extensively in reliability and life-testing studies (13,18); it effectively characterizes many disease progress curves (12,13).

Thus, the eight variables used to characterize each DPC were: WDF scale parameter, b ; WDF shape parameter, c ; area under the DPC, ADPC; final disease severity, Y_f ; FDR linear coefficient, B_1 ; FDR quadratic coefficient, B_2 ; percentage of plants infected, PPI; and estimated time of disease onset, X_0 .

Factor analysis. The structure of the 100 DPCs was determined by conducting a factor analysis on the eight variables. In the factor analysis model each of the eight variables is represented as a linear function of a small number of unobservable "common factors" and a single latent "unique factor." The factor model can be written as:

$$Z_i = a_{i1}f_1 + a_{i2}f_2 + \dots + a_{im}f_m + u_i$$

in which Z_i = i th variable (eg $Z_4 = Y_f$); f_j = j th common factor; a_{ij} = factor loading (parameter reflecting the importance of the j th factor in composition of the i th variable), and u_i = i th unique factor. The f 's account for correlations among the Z variables and the u 's account for the uniqueness or remaining variance of each variable (2,9). The parameter a_{ij} equals the correlation between Z_i and f_j .

A Principal Axis procedure followed by a Varimax rotation was used to estimate the factor loadings (2). All loadings greater than or equal to 0.35 were considered significant (5).

RESULTS

Isolation from root rot lesions. From the 250 bean hypocotyls selected and possessing fungal lesions, *R. solani* was isolated as the primary pathogen 230 times; *F. solani* f. sp. *phaseoli* was isolated alone five times and in conjunction with *R. solani* 10 times. Very few isolates of other fungi, which were considered to be secondary invaders, were obtained in the first two samples taken from each quadrat; however, as plant age increased, increasing numbers of isolates of secondary fungi, such as *F. oxysporum*, *F. roseum*, and *Alternaria* spp., were isolated in addition to the two primary pathogens. All 25 randomly selected isolates of *R. solani* were in anastomosis group 2.

Curve elements. The residual plots from ordinary least-squares regression signified that all 100 disease progress curves could be adequately described with a linear or quadratic FDR model. Values for the WDF scale parameter (b), which is inversely

proportional to the rate of disease increase (13), ranged from 35.42 to 81.54. The WDF shape parameter (c) (for which a value of 1.0 represents a "simple interest disease" and a value of 3.6 represents a "compound interest disease" [13]) ranged from 1.9 to 14.28. Means, standard deviations, and ranges for all factor analysis variables are listed in Table 1.

Factor analysis. The correlations among factor analysis variables are presented in Table 2. They indicate apparent relationships between curve elements without regard to confounding intercorrelations. The correlation matrix was used to calculate the factor loadings, ie, a_{ij} . The first four eigenvalues of the correlation matrix accounted for 90% of the total variance. Expansion to include a fifth factor only accounted for an additional 6% of the variance; therefore, we extracted four factors from the correlation matrix to express the variables of the data set more simply and without confounding interactions among curve elements, ie, to obtain greater parsimony. Results of the factor analysis are presented in Table 3.

The first factor, which accounted for 42.5% of the variance, showed a high positive intercorrelation among the WDF shape parameter (c), final disease severity (Y_f), and the FDR quadratic coefficient (B_2) coupled with a high negative intercorrelation of the WDF scale parameter (b) and the FDR linear coefficient (B_1). The WDF shape parameter indicates the nature of an epidemic and in this case the greater the c value, the greater the Y_f . With regard to the FDR coefficients, a linear term in the regression model tends to limit the rate of disease increase relative to the effect of a quadratic term when $B_2 > 0$; thus the negative intercorrelation between Y_f and B_1 and the positive intercorrelation between Y_f and B_2 . Also, the WDF scale parameter is inversely proportional to the rate of disease increase (13); thus, the negative intercorrelation between b and Y_f . The intercorrelation between b and the FDR coefficients is

TABLE 1. Mean, standard deviation, and range values^a for eight variables used to characterize bean root rot epidemics induced by *Rhizoctonia solani* and *Fusarium solani* f. sp. *phaseoli*

Variable ^y	Mean	Standard deviation	Range
b	50.42	7.95	35.42 to 81.54
c	6.83	2.91	1.90 to 14.28
ADPC	3.37	0.90	1.37 to 5.95
Y_f	0.32	0.12	0.11 to 0.64
B_1	-0.01	0.01	-0.0279 to 0.0467
B_2	0.00024	0.00018	0.00 to 0.00073
PPI	88.60	12.70	41.60 to 100.00
X_0	14.10	8.66	5.00 to 35.000

^aBased on values obtained from 100 disease progress curves.

^yb = Weibull distribution function scale parameter; c = Weibull distribution function shape parameter; ADPC = area under the disease progress curve; Y_f = final disease severity; B_1 = first-difference regression linear coefficient; B_2 = first-difference regression quadratic coefficient; PPI = percentage of plants infected; X_0 = time of disease onset.

TABLE 2. Correlation of variables^x used to characterize epidemics of bean root rot induced by *Rhizoctonia solani* and *Fusarium solani* f. sp. *phaseoli*

Variable ^y	c	ADPC	Y_f	B_1	B_2	PPI	X_0
b	-0.76	-0.29	-0.64	0.60	-0.62	-0.25	0.01
c		0.01	0.55	-0.67	0.72	0.15	0.11
ADPC			0.72	-0.17	0.36	0.42	-0.21
Y_f				-0.68	0.86	0.54	-0.07
B_1					-0.89	-0.32	-0.07
B_2						0.43	-0.00
PPI							-0.12

^xBased on values obtained from 100 disease progress curves.

^yb = Weibull distribution function scale parameter; c = Weibull distribution function shape parameter; ADPC = area under the disease progress curve; Y_f = final disease severity; B_1 = first-difference regression linear coefficient; B_2 = first-difference regression quadratic coefficient; PPI = percentage of plants infected; X_0 = time of disease onset.

explained by similar reasoning for the intercorrelation between Y_f , B_1 , and B_2 . Factor 1 can be interpreted as an overall description of the curve including shape (c), rate (b, B_1 , B_2), and level (Y_f).

Factor 2 accounted for 20% of the variance and signified an expected high positive intercorrelation between ADPC and Y_f . This factor related the area (ADPC) and level (Y_f) of the epidemic.

The third factor, accounting for 12.5% of the variance, revealed the absence of an intercorrelation between X_0 , the time of disease onset, and any of the seven other variables. Factor 3 can be interpreted as the epidemic location factor which identifies the uniqueness of the time of disease onset (X_0) with respect to the other curve elements.

Factor 4 accounted for 15% of the variance and indicated an intercorrelation among PPI, B_2 , and Y_f . This factor identified the relationship between disease severity (Y_f) and disease incidence (PPI).

DISCUSSION

The eight disease progress curve elements investigated in this bean root rot study were reduced to four unobservable, common factors which could be assigned plausible epidemiological interpretations. The first factor was interpreted as an overall description of disease progression including curve shape, rate of disease progression, and the estimated disease severity level 50 days after planting. This factor accounted for the largest proportion of the total variance and thus may be labeled as the "main" factor for this analysis. Madden and Pennypacker (7,8), using principal component analysis of disease severity values, also found that level, rate, and shape of disease progress curves were the main underlying factors of epidemics for several pathogen-suscept combinations.

The second factor related the area under the disease progress curve and the disease level or final disease severity of the bean root rot epidemics. The higher a disease severity value, the greater its influence on the total area under the curve. Final disease severity for bean root rot and most unilateral epidemics is usually the highest disease level encountered during the growing season. Thus, the strong relationship in this host-pathogen system between Y_f and ADPC is reasonable and not unexpected. The evidence of such a relationship is strengthened by the work of Kranz (5), who showed a similar association.

The third factor (ie, epidemic location) identified the uniqueness of the estimated time of disease onset in this epidemic system. This result was unexpected, since Kranz (5) had previously shown a strong relationship between the time of disease onset and the consequences of the epidemic for 40 different host-pathogen

TABLE 3. Principal Axis Factor Analysis (Varimax rotation) utilizing eight curve elements to characterize 100 naturally occurring bean root rot epidemics induced by *Rhizoctonia solani* and *Fusarium solani* f. sp. *phaseoli*

Variable ^x	Factor			
	1	2	3	4
b	-0.83^y	-0.29	.05	.07
c	.93	-0.05	.06	-0.03
ADPC	.07	.96	-0.11	.20
Y_f	.64	.65	-0.01	.35
B_1	-0.85	-0.03	-0.07	-0.33
B_2	.84	.24	.02	.38
PPI	.14	.25	-0.07	.90
X_0	.04	-0.09	.99	-0.06
Variance Explained	3.4	1.6	1.0	1.2
variance (%):	42.5	20.0	12.5	15.0

^xb = Weibull distribution function scale parameter; c = Weibull distribution function shape parameter; ADPC = area under the disease progress curve; Y_f = final disease severity; B_1 = first-difference regression linear coefficient; B_2 = first-difference regression quadratic coefficient; PPI = percentage of plants infected; X_0 = time of disease onset.

^yItalicized values are considered significant as their loading is ≥ 0.35 (5).

combinations investigated over 2 yr. Kranz (5), however, studied only systems in which foliar pathogens were involved.

The identified uniqueness of the estimated time of disease onset for this bean root rot system may have important epidemiological consequences as far as breeding for resistance to this disease is concerned. If the uniqueness of this curve element is confirmed, the type of bean root rot resistance which would delay the onset of the epidemic would not be useful because such a resistance would not significantly affect the other curve elements delineated in this study.

The fourth factor indicated a relationship between disease severity and disease incidence. This relationship should be further investigated, with the possible consequence that a type of resistance to bean root rot reducing disease severity would also reduce disease incidence. The reverse also would be true.

The specification of interrelationships between or among curve elements may lead to the identification of epidemiologic relationships not previously suspected by mere inspection of disease progress curves and may suggest new approaches to breeding strategies for disease resistance. The accurate identification of the associations among, or uniqueness of, curve elements also aids in the judicious selection of elements to provide the most realistic and yet parsimonious representation of an epidemic for analytical purposes. Interrelationships among the curve elements of disease progression should be determined prior to the initiation of modeling, forecasting, predicting yield loss, or breeding for resistance involving any disease.

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