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Medium pH and Leaf Nutrient Concentration Influence Rust Pustule Diameter on Leaves of Dry Beans

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Abstract. **Nine bean cultivars/lines *(Phaseolus vulgaris* L.) were grown in three soils/rooting media at pH values of 7.9, 6.5, and 5.8 in greenhouse, growth chamber, and field experiments to evaluate the leaf reaction of the plants to a Nebraska bean rust *[Uromyces appendiculatus* (Pers.) Unger var. *appendiculatus]* isolate US85-NP-10-1. Significant differences were observed for rust pustule diameter between cultivars/lines grown in the three growth media. Plants grown in the medium at pH 5.8 showed significantly larger rust pustule diameters than those of plants grown at pH 6.5 or 7.9. A significant interaction occurred between growth medium and cultivars/lines for the rust reaction. Concentrations of Cl and Mn in leaves were positively correlated with rust pustule diameter. In contrast, concentration of K in leaves was negatively correlated with rust pustule diameter. Plant breeders attempting to improve beans for rust resistance must consider the growth medium pH in evaluating intensity and severity of rust symptoms on leaves.**

Even though plant disease response to host plant nutrition in horticultural and field crops has been reported (Forsyth, 1957; Gallegly and Walter, 1949; Platero and Tejerina, 1976; Walker, 1946), we found no reports on the effects of host plant nutrition, growth medium pH, and rust interaction in dry bean. Very little information is available about the effects of host nutrition on rust development, although other aspects of the disease have been studied thoroughly (Gersbeck and Schonbeck, 1988). The objectives in this research were to a) evaluate the effects of growth medium pH on rust development, and b) determine the relationship between the rust reaction of the plants and the nutrient concentrations of the leaf tissue.

Eight cultivars/lines of dry beans with known differences in reaction to one Nebraska isolate (US85-NP-10-1) of the bean rust pathogen were grown in three media in the greenhouse (Expt. I) and in the growth chamber (Expts. II and III). Medium 1 was Tripp sandy clay loam soil (coarse-silty, mixed, mesic Aridic Haplustolls) from the North Platte Research and Extension Center, Univ. of Nebraska, North Platte, with pH 7.9, 1.1% organic matter, 8.9 ppm P, 291 ppm K, 2.7 ppm NO_3, 0.5 ppm NH_4, 10.4 cmol Ca, 3.5 cmol Mg, 0.9 cmol K, and 0.8 cmol Na/kg of soil. Medium 2 was Sharpsburg silty clay loam (fine, montmorillonitic, mesic Typic Arguidolls) from the Dept. of Horticulture experimental garden, Univ. of Nebraska, Lincoln, with pH 6.5, 2.9% organic matter, 12.2 ppm P, 269 ppm K, 1.9 ppm NO_3, 0.6 ppm NH_4, 15.8 cmol Ca, 5.5 cmol Mg, 0.9 cmol K, and 0.6 cmol Na/kg of soil. Medium 3 was a potting mixture consisting of equal volumes of sand, soil (Sharpsburg silty clay loam), vermiculite, and peat moss, with pH 5.8, 2.7% organic matter, 5.8 ppm P, 157 ppm K, 0.5 ppm NO_3, 0.5 ppm NH_4, 5.3 cmol Ca, 2.8 cmol Mg, 0.4 cmol K, and 0.5 cmol Na/kg of soil.

A randomized complete-block design with a 3 × 8 factorial arrangement of growing media and cultivars/lines with four replications for Expts. I, II, and III was used. The abaxial surfaces of fully unfolded primary leaves of bean seedlings of the same age were inoculated with a water suspension of urediniospores 10^5 spores/ml 7 to 12 days after seeding, by use of a modified crown sprayer (Fisher Scientific, Pittsburgh, Pa.). Leaves were sprayed, without wounding, until completely wet. After inoculation, plants were held in high humidity by covering them with clear plastic for 18 h (overnight). Rust pustule diameter in micrometers was measured by use of 6 × edscorp pocket comparator (Edmund Scientific, Barrington, N.J.) 14 days after inoculation.

Plants were grown in 15-cm (1.5-liter) plastic pots (two plants per pot) using the three growing media in all experiments. The air was ≈25C (14-h day) and 22C (10-h night) in Expt. I and 27C (14-h day) and 20C (10-h night) in Expts. II and III. In the growth chamber experiments, the photosynthetic photon flux was 340 $\mu mol \cdot s^{-1} \cdot m^{-2}$ at plant height (110 cm below the light source). Lamps used to provide light were high-pressure sodium (General Electric, Lucalux, LU 4008) and metal halide (General Electric, Multivapor, MV 400).

The eight cultivars/lines used were: Great Northern (GN) Belneb #1 and GN WM1-85-43 (race-specific resistance to rust); 'PC-50' and 'Jamaica Red' (race-specific and race-nonspecific resistance to rust); Pinto 'UI-114' and 'US-3' (highly susceptible to rust); and GN 'Harris' and GN 'Tara' (susceptible to rust).

Nine cultivars/lines of dry beans were grown in the field (Expts. IV and V) in specially designed wooden boxes (107 × 32 × 30 cm) in the media described. In addition to the eight lines used in the growth chamber studies, PI 165078, a line susceptible to Fe deficiency chlorosis (Zaiter et al., 1986), was included. A split-plot arrangement of treatments with soil medium as whole-plot and cultivars/lines as subplots was used with five replications in Expt. IV and four replications in Expt. V.

Rust pustule diameter in micrometers was recorded 14 days after inoculation, as described. The abaxial surfaces of fully unfolded first trifoliates of bean seedlings of the same age (14 to 16 days after seeding) were inoculated. Seeds sown in potting mix medium germinated 2 days earlier than in the Tripp or Sharpsburg soils.

Entire leaves were collected from plants at the early bloom stage in Expt. V for determination of nutrients. The trifoliate or single leaf from the top of each plant was sampled, dried at 60C for 48 h, weighed, ground to pass a 0.5-cm screen, and analyzed for Mg, Al, Si, P, Cl, K, Ca, Mn, Fe, Cu, and Zn by energy dispersive X-ray fluorescence (Knudsen et al., 1981).

A separate analysis of variance (ANOVA) was conducted for each of the five experiments. Appropriate $LSD_{0.05}$ values were calculated to make specific treatment comparisons in Expts. IV and V as described by Steel and Torrie (1960) (Table 1). Cultivars and breeding lines that expressed an immune reaction (rating = 0) (Belneb #1, 'PC-50', and WM1-85-42) were excluded from the ANOVA because the data were discrete.

Dry bean cultivars/lines differed markedly in all experiments in susceptibility to rust in response to soil medium pH (Table 1). Several cultivars/lines grown at pH 5.8 (potting

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Table 1. Mean leaf rust pustule diameter (μm)[z] on greenhouse- (Expt. I), growth chamber- (Expts. II and III), and field- (Expts. IV and V) grown dry bean cultivars/lines (inoculated with rust isolate US 85-NP-10-1) in three growing media.

	Mean leaf rust pustule diameter (μm)														
	Tripp soil (pH 7.9)					Sharpsburg soil (pH 6.5)					Potting mix (pH 5.8)				
Cultivar/line[y]	I	II	III	IV	V	I	II	III	IV	V	I	II	III	IV	V
GN Belneb #1 (U/N)	0	0	0	0	117	0	0	0	0	34	0	0	0	0	0
GN Harris (N)	575	400	450	780	625	600	650	600	640	625	900	700	550	840	825
Jamaica Red (J)	300	300	350	140	100	350	400	425	420	475	450	400	350	260	550
PC-50 (DR)	0	0	75	260	158	400	400	450	220	450	650	400	350	320	425
GN Tara (N)	300	350	300	580	500	575	500	525	440	350	550	400	575	740	775
Pinto UI 114 (I)	775	575	450	600	650	850	700	600	680	625	925	725	800	860	775
US-3 (U)	750	400	550	640	500	750	400	600	660	750	950	625	800	840	925
GN WM1-85-43 (N)	0	0	0	240	125	300	300	225	140	325	0	150	300	90	360
PI 165078 (T)	---	---	---	764	775	---	---	---	680	675	---	---	---	780	850
Pearson correlations between Expts. II & III and IV and V		+0.83**		+0.85**			+0.57**		+0.61**			+0.75**		+0.79**	

[z]Rust reactions: immune = 0, resistant = pustule size ≤300 μm, susceptible = pustule size >300 μm (Stavely and Pastor-Corrales, 1989).
[y]Origin: U/N = USDA/Nebraska; J = Jamaica; DR = Dominican Republic; I = Idaho; T = Turkey.
**Significant at $P = 0.01$. $LSD_{0.05}$ value to compare means for different factorial treatments = 59 for Expt. I; $LSD_{0.05}$ value to compare means for different factorial treatments = 76 for Expt. II and 98 for Expt. III; $LSD_{0.05}$ value to compare means for the same soil type for different cultivars/lines = 157 for Expt. IV and = 156 for Expt. V. $LSD_{0.05}$ value to compare means of an entry between different soil types = 166 for Expt. IV and 156 for Expt. V.

Table 2. Mineral element concentrations (MEC) in leaves of nine dry bean cultivars/lines grown in three media (Expt. V) and correlations between MEC and rust pustule size of susceptible (S) cultivars/lines.

	Cultivars/lines											
Ions/pH/media[z]	GN Belneb #1	GN Harris(S)	Jamaica Red	PC-50	PI 165078(S)	GN Tara(S)	Pinto UI 114(S)	US-3(S)	GN WM1-85-43	$LSD_{0.05}$[y]	$LSD_{0.05}$[x]	r
					g·kg⁻¹							
Cl												
L (PM)	14.8	15.4	16.5	17.9	16.8	14.0	1.70	14.6	12.5	0.38	0.43	+0.52
I (S)	2.3	2.9	4.5	6.4	3.4	3.2	0.41	2.0	3.0			
H (C)	2.2	2.6	2.5	2.4	4.1	2.7	0.25	4.2	2.5			(P = 0.0001)
K												
L (PM)	21.1	22.3	24.5	19.5	25.9	23.0	25.3	22.9	23.4	0.59	0.68	−0.43
I (S)	31.2	36.2	40.8	34.6	32.2	34.5	35.1	33.7	32.6			
H (C)	41.2	46.7	44.8	37.3	40.7	43.3	40.9	37.4	42.9			(P = 0.0004)
					mg·kg⁻¹							
MN												
L (PM)	250	220	224	332	227	213	233	194	214	37.0	49.3	+0.44
I (S)	71	71	58	80	60	71	58	61	69			
H (C)	78	79	78	96	51	74	63	116	67			(P = 0.0005)

[z]L = low pH (5.8, PM = Potting mix); I = intermediate pH (6.5, S = Sharpsburg soil); H = High pH (7.8, C = Tripp soil).
[y]$LSD_{0.05}$ values to compare means for the same soil type from different cultivars/lines.
[x]$LSD_{0.05}$ values to compare means of an entry between different soil types.

mix) showed significantly larger rust pustule diameters than plants grown on the soils at pH 6.5 or 7.9. The highest mean pustule diameter on leaves was 950 μm on 'US-3' plants grown on the low pH medium. GN Belneb #1 (immune except in Expt. V on high and intermediate pH medium where pustule diameter was <300 μm) was the most resistant entry and 'US-3' (pustule diameter means ranged from 400 to 950 μm over all experiments) was the most susceptible entry over all experiments. Growing media × cultivars/lines interactions were detected (F value significant at $P < 0.01$). GN 'Harris', 'Jamaica Red', 'PC-50', GN 'Tara', and Pinto 'UI-114' were more susceptible (larger rust pustule diameter) when grown in the low pH medium than in the high pH medium, but no differences in pustule diameter were noted for PI 165078 on these two media. 'Jamaica Red' and 'PC-50', either produced an immune reaction (rating = 0) or small pustules (flecking with no sporulation in pustule ≤ 300 μm) when grown on the high pH medium while sporulating pustules >300 μm (except in two instances) were produced in all experiments on the other two media. WM1-85-43 showed an immune reaction (rating = 0) in three out of five experiments on the high pH medium but in only one out of five experiments in the low pH medium. Pustule diameters ≤300 μm occurred on leaves of WM1-85-43 in all other experiments on all media except in the case of Expt. V at pH 5.8 and 6.5 (> 300 μm). Moderately high positive correlations were noted between the pustule diameter on plants grown in the two growth chamber experiments (II and III) and in the two field experiments (IV and V), indicating good agreement of the data over experiments (Table 1).

Higher concentrations of Cl and Mn were noted in leaves of plants grown at pH 5.8 than in plants grown at 7.9 or 6.5 (Table 2). In contrast, K concentration in leaves on plants grown in the low pH medium was about half that found in plants grown at high or intermediate pH. Concentrations of the above nutrients were usually similar in resistant and susceptible cultivars/lines (Table 2). Concentrations of Cl and Mn in leaves were positively correlated with rust pustule diameter. In contrast, concentration of K in leaves was negatively correlated with rust pustule diameter (Table 2). Concentrations of Mg, Al, Si, P, Ca, Fe, Zn, and Cu in leaves were not correlated with pustule diameter (correlations not shown).

The degree of rust infection on dry beans was influenced by pH and genotype. Because of the interactions of rust development on cuhivars/lines grown on different growth media, plant breeders and plant pathologists now need to take this into consideration in evaluating and reporting on the degree of resistance and susceptibility of bean germplasm. Differences in pH of media may be primarily involved, but this was not proved in these experiments since the media also varied in other properties. Further experiments need to be conducted under controlled pH, nutrient, and environmental conditions to determine the influence of pH, each nutrient, and nutrient interactions on rust development in beans.

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