Response in different plant parts to contrasting *Xanthomonas campestris* pv. *phaseoli* strains is essential for breeding common bean with high levels of common blight resistance

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INTRODUCTION

Common blight caused by *Xanthomonas campestris* pv. *phaseoli* is an important disease in the Americas and worldwide causing yield losses over 40% (Singh and Schwartz, 2010). Use of genetic resistance is the most effective control, and more than 20 genes/QTL from the primary, secondary and tertiary gene pools confer common blight resistance (Miklas et al., 2006). Resistance QTL SAP6, BC420, and SU91 have been used in marker-assisted selection (Miklas et al., 2006). But, the resistance to common blight can vary according to the plant parts and bacterial strains inoculated (Arnaud-Santana et al., 1994; Viteri et al., 2014). Thus, the identification of genotypes with broad-spectrum resistance to different strains in different plant parts would be crucial. Our objectives were to determine: (1) the response of 28 genotypes in leaves and pods to two bacterial strains, (2) the presence or absence of SAP6, BC420, and SU91 resistance linked QTL, and (3) genotypes with high levels of resistance.

MATERIALS AND METHODS

Disease Response

Twenty eight common beans including the susceptible check pinto 'Othello' were planted in a randomized complete block design with three replications in the greenhouse in Idaho in 2014. Less-aggressive bacterial strain ARX08AC was inoculated in one ¾ th expanded primary leaf, and the more aggressive strain Xcp25 in the other at 1.7 x 10^8 CFU/ml. Similarly, the two lateral leaflets of the first trifoliolate leaf were inoculated with ARX8AC and the middle one with the Xcp25 bacterial strain. Also, two pods at mid-fill stage were inoculated with each strain at the same bacterial density. Disease severity was evaluated at 28 days post-inoculation in leaves and at 14 days in pods using a 1-9 scale, where 1= no symptoms, and 9=water soaking lesions extended to leaf or pod margins. Plants with scores of 1-3 were considered resistant; 4-6 were intermediate, and 7-9 were susceptible (Lema et al., 2007; Viteri et al., 2014). Also, the percentage of resistant plants, and range for common blight scores were calculate for each plant part.

Resistance Marker Assays

DNA extraction was carried out using the Dellaporta protocol (Dellaporta et al., 1983) and adjusted to 10 ug/ml. BC420 and SU91 QTL were run in a multiplex PCR (Duncan et al., 2011), while the protocol described by Viteri et al. (2014) was used for SAP6 QTL. PCR products were run in 1.4% agarose gel stained with 2% of ethidium bromide. The presence or absence of SAP6, BC420, and SU91 was recorded visually.

RESULTS AND DISCUSSION

Bacterial strain Xcp25 had higher mean common blight scores and lower percentage of resistant plants than ARX8AC, especially in leaves (Table 1 and 2). Similar results were observed in previous studies (Duncan el at., 2011; Lema et al., 2007; Viteri et al., 2014). Pinto Othello was susceptible (mean scores > 6.5) in all plant parts to both bacterial strains. ICB-3 and SE153, with SAP6 QTL, were resistant to ARX8AC in the trifoliolate leaf. But, ICB-3 had higher percentage of resistant plants (66.7%). Rexeter with the SU91 QTL was intermediate to both bacterial strains in all plant parts. However, higher percentage of resistant plants (22.1%) were observed in the trifoliolate leaf in response to Xcp25. Duncan et al. (2013) and Viteri et al. (2014) also found that SAP6 QTL had a better effect in response to less aggressive strain (e.g., ARX8AC), and SU91 QTL against more aggressive strains (e.g., Xcp25). USPT-CBB-5 and VAX 6, with SAP6 and SU91 QTL, were resistant to ARX8AC in both leaves. But, USPT-CBB-5 had higher scores in response to Xcp25. Thus, VAX 6 might possess the new Xa11.4 QTL that provides higher levels of resistance to more aggressive strains in leaves (Viteri et al., 2014). Andean AM101 had the lower mean common blight scores (≤ 2.4) and range (1-3) in both leaves and all plants (100%) were resistant in the trifoliate leaf to both strains. But, AM101, RCS63, and Wilkinson 2, with BC420 and SU91 QTL, were susceptible, and only VAX 6 had 11% of resistant plants (range 3-6) to both bacterial strains in pods. Thus, the identification of genes/QTL that confer resistance in pods from across *Phaseolus* species would be crucial for increasing the levels of common blight resistance in all plant parts.

REFERENCES

Arnaud-Santana, E., Coyne, D.P., Eskridge, K.M., and Vidaver, A.K. 1994. J. Am. Soc. Hortic. Sci. 119:116-121.

Dellaporta, S.L., Wood, J., and Hicks, J.B. 1983. Plant Mol. Biol. Rep. 1:19-21.

Duncan, R.W., Singh, S.P., and Gilberson, R.L. 2011. Phytopathology 101:425-435.

Lema, M., Terán, H., and Singh, S.P. 2007. Crop Sci. 47:1367-1374.

Miklas, P.N., Kelly, J.D., Beebe, S.E., Blair, M.W. 2006. Euphytica 147:105-131.

Singh, S.P., and Schwartz, H. 2010. Crop Sci. 50:2199-2223.

Viteri D.M., Cregan, P.B., Trapp, J., Miklas, P.N., and Singh, S.P. 2014. Crop Sci. 54:1598-1608.

Table 1. Range for common blight scores and percentage of resistant plants for leaves and pods of nine common bean genotypes for two strains of *Xanthomonas campestris* pv. *phaseoli* evaluated in a greenhouse at University of Idaho, Kimberly in 2014.

Genotype	Primary leaf				Trifoliolate leaf				Pods	
	ARX8AC		Xcp25		ARX8AC		Xcp25		ARX8AC	Xcp25
	Range	%Rp[a]	Range	%RP	Range	%RP	Range	%RP	Range	Range
Othello	9	0.0	9.0	0.0	9.0	0.0	9.0	0.0	5-9	6-9
ICB-3	2-5	55.6	9	0.0	2-5	66.7	5-9	0.0	4-6	4-6
SE153	1-6	22.2	6-9	0.0	2-5	55.6	5-9	0.0	4-7	6-9
Rexeter	6	0.0	5-6	0.0	1-6	11.1	2-7	22.2	5-7	4-7
USPT-CBB-5	2-6	66.7	4-8	0.0	1-5	77.8	6-9	0.0	4-7	6-7
VAX 6	1-3	100.0	2-6	22.2	1-4	77.8	3-6	11.1	3-6[b]	3-6[b]
RCS63	2-6	11.1	4-7	0.0	1-2	100.0	1-3	100.0	6-8	6-8
Wilkinson 2	1-6	33.3	1-9	22.2	1-6	33.3	1-5	33.3	7-8	6-8
AM101	1-3	100.0	1-5	88.9	1-2	100.0	1-3	100.0	6-7	6-7
Mean	…	43.2	…	14.8	…	58.0	…	29.6	…	…

[a] Percentage of resistant plants

[b] Only VAX 6 had 11.1% of resistant plants for both strains in pods

Table 2. Marker composition and mean common blight scores in leaves and pods of nine common bean genotypes for two strains of *Xanthomonas campestris* pv. *phaseoli* evaluated in a greenhouse at University of Idaho, Kimberly in 2014.

Genotype	Marker	Primary leaf		Trifoliolate leaf		Pods	
		ARX8AC	Xcp25	ARX8AC	Xcp25	ARX8AC	Xcp25
Othello	None	9.0	9.0	9.0	9.0	6.9	6.9
ICB-3	SAP6	3.6	9.0	3.3	7.6	4.4	4.8
SE153	SAP6	3.8	7.9	3.3	7.1	5.8	7.6
Rexeter	SU91	6.0	5.8	4.5	5.1	6.4	6.0
USPT-CBB-5	SAP6, SU91	3.2	6.0	2.9	7.8	5.7	6.6
VAX 6	SAP6, SU91	2.3	4.4	2.2	4.8	5.0	4.8
RCS63	BC420, SU91	4.5	5.0	1.7	2.3	7.2	7.0
Wilkinson 2	BC420, SU91	4.3	4.6	3.7	3.2	7.7	7.3
AM101	BC420, SU91	1.6	2.2	1.6	2.4	6.8	6.9
Mean	…	4.3	6.0	3.6	5.5	6.2	6.4
LSD (P < 0.05)	…	1.0	1.0	1.0	1.0	0.7	0.8

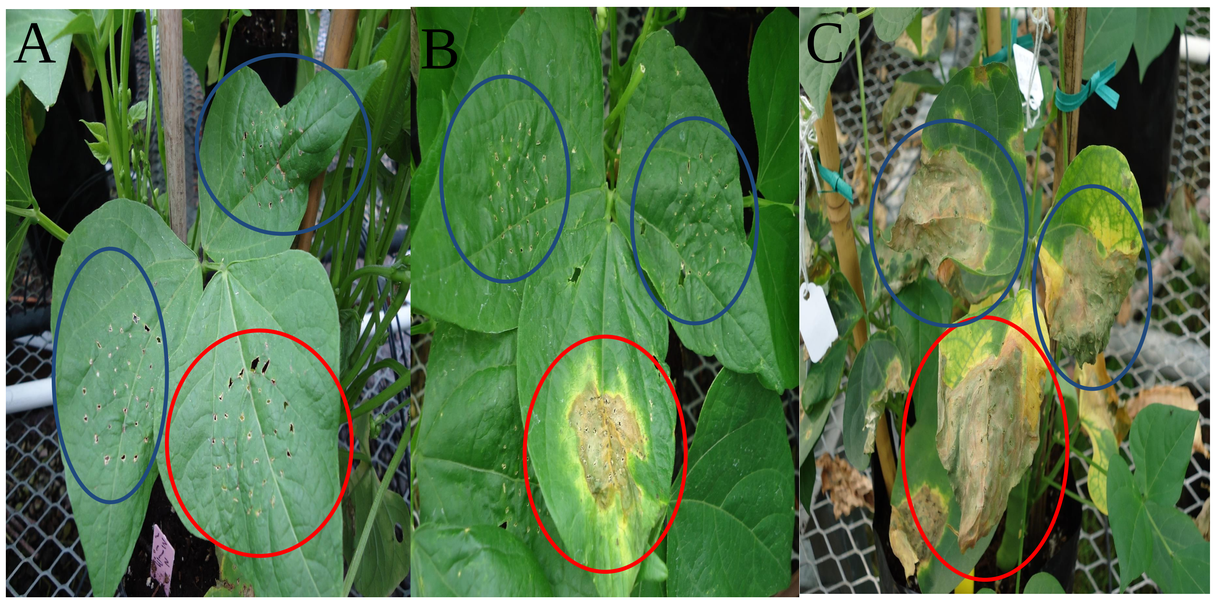

Figure 1. (A) RCS63 resistant to ARX8AC and Xcp25, (B) USNA-CBB-3 resistant to ARX8AC and susceptible to Xcp25, (C) Othello susceptible to ARX8AC and Xcp25.

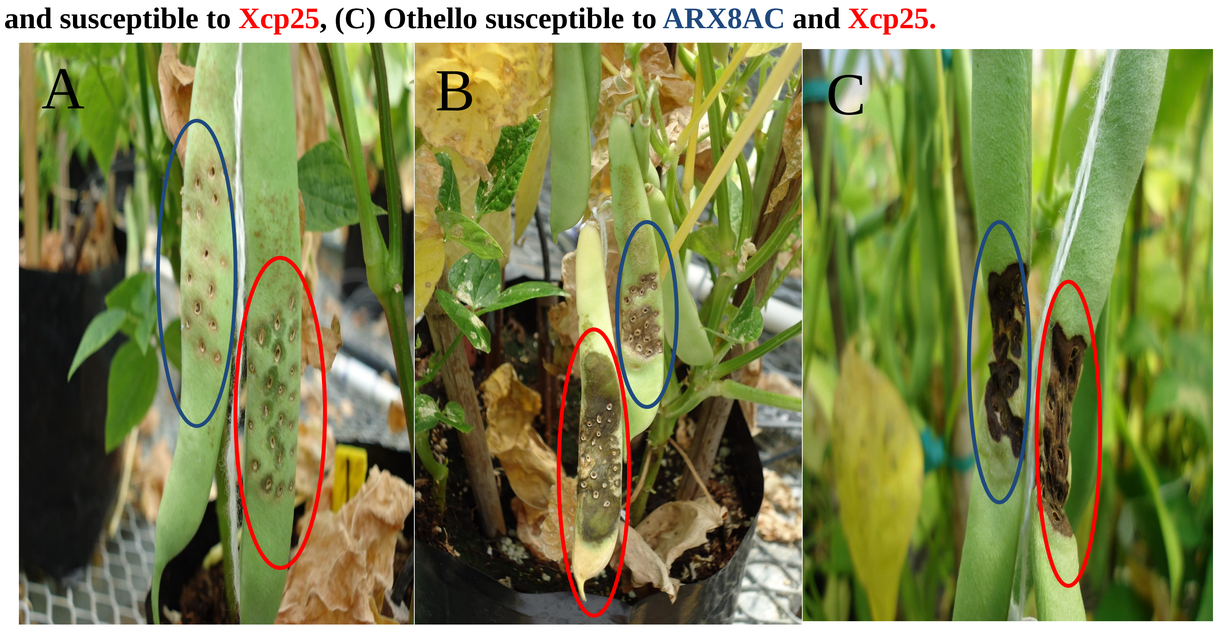

Figure 2. (A) VAX6 resistant to ARX8AC and Xcp25, (B) USNA-CBB-2 intermediate to ARX8AC and susceptible to Xcp25, (C) Wilkinson 2 susceptible to ARX8AC and Xcp25.