Bean Seedborne Bacterial Pathogens

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Bacterial Diseases of Bean

- Halo blight Pseudomonas syringae pv. phaseolicola
- Common bacterial blight Xanthomonas axonopodis pv. phaseoli
- Bacterial brown spot *Pseudomonas syringae* pv. syringae
- Bacterial wilt *Curtobacterium flaccumfaciens* pv. *flaccumfaciens*
- Wildfire *Pseudomonas syringae* pv. *tabaci* (in Brazil)

(Compendium of Bean Diseases, 2nd Edition. Schwartz et al. 2005. APS Press)

Washington State Bean Seed Quarantine

- WAC 16-301-380: Regulated diseases
- The following viral, bacterial and fungal diseases of beans, and any new strains or variations of these identified in the future on beans are regulated under the provisions of this chapter:

Halo blight

Common bacterial blight

Fuscous blight (variant of common blight)

Bean anthracnose

Brown spot

Bacterial wilt

Bean common mosaic virus (BCMV)

Halo blight

- Pseudomonas syringae pv. phaseolicola
- Aerobic bacterium
- Numerous races: differential host range
- Optimum: 20-23°C (68-73°F)
- Phaseolotoxin: optimum 20-23°C

Bacterial brown spot

- Pseudomonas syringae pv. syringae
- Aerobic bacterium
- Very wide host range
- Optimum: 28-30°C (82-86°F)
- Syringacin W-1 toxin

Common bacterial blight

- Xanthomonas campestris pv. phaseoli
- Aerobic bacterium
- Numerous races: differential host range
- Optimum: 28-32°C (82-90°F)



Pseudomonas syringae pv. phaseolicola

Halo blight, bacterial brown spot, & bacterial common blight

- **Contaminated seeds:** important inoculum source
- Infected seeds germinate, bacteria colonize emerging plants.
 Spread by:
 - splashing water (rain/irrigation)
 - windborne rain/irrigation, windborne soil
 - equipment, field crew, animals, insects
 - between adjacent, wet plants
- Infection & development favored by:
 - high humidity (stomata open), rain, wounds
 - 18-23°C for halo blight, 28-30/32°C for other two
 - asymptomatic ≠ non-infected! HB symptoms disappear >23°C
- Survival (= inoculum sources):
 - infested residues, volunteers, asymptomatic hosts (incl. weeds)



Halo blight

Pseudomonas syringae pv. phaseolicola



Halo blight

Pseudomonas *syringae* pv. phaseolicola



H.F. Schwartz, Colorado State University

Halo blightPseudomonas syringae pv. phaseolicola







Halo blight





Bacterial brown spot Pseudomonas syringae pv. syringae



Bacterial brown spot *Pseudomonas syringae* pv. *syringae*





Common bacterial blight

Xanthomonas campestris pv. phaseoli



Common bacterial blight *Xanthomonas campestris* pv. *phaseoli*





Common bacterial blight

Xanthomonas campestris pv. phaseoli

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Common bacterial blight

Xanthomonas campestris pv. phaseoli

2009 Washington State Bean Seed Crops: Harvested Seed Tested Positive for Halo Blight or Common Bacterial Blight Pathogens

					Stock			WSDA seed crop
Disease &			Bean		seed &	Previous	Adjacent crops	inspection
Field	County	Grower	cultivar	Irrigation	origin	crops	N, S, E, W	dates
Halo blight								
Field #1	Klickitat	Grower A	Cv. N	Circle	Z – WA	Beet, onion, grass, peas	Peas, green bean, grass, onion, beets	7-20, 8-18
Field #2	Grant	Grower B	Cv. N	Circle	Y – WA	Corn, peas, potatoes	Mint, corn, corn, bean seed	7-29, 8-18
Common blight								
Field #3	Grant	Grower C	Cv. N	Circle	X – WA	Wheat, alfalfa, alfalfa	Wheat, grass hay, wheat, wheat	8-7, 8-26
Field #4	Grant	Grower D	Cv. O	Circle	W – WA	Onion, wheat, potatoes	Grass seed, grass seed, alfalfa, onion	8-7, 8-26
Field #5	Grant	Grower E	Cv. P	Circle	U & V – ID	Corn, potatoes, corn	Cattle, beans, corn, canola, potatoes	8-11, 8-31

2012 Halo Blight in a Bean Seed Crop

- 17-acre bean seed crop in Grant Co., WA under center-pivot irrigation
- Diagnosis confirmed by a certified lab
- Crop destroyed immediately after confirmation



2014 Halo Blight in a Bean Seed Crop

- 40-acre cranberry bean seed crop in Grant Co., center-pivot irrigation
- Confirmed by a certified lab, crop destroyed promptly



L.J. du Toit, Washington State University

2016 & 2017 Bacterial Diseases in Bean Seed Crops

- 5 bean seed crops, all under center-pivot irrigation
- Confirmed by certified labs, & crops destroyed
- Samples also sent to Lindsey du Toit's lab for pathovar verification:
 - Pseudomonas syringae pv. phaseolicola = halo blight, but no halo (2016)
 - Pseudomonas syringae pv. syringae = bacterial brown spot (2017)



Toxigenic vs. non-toxigenic strains of halo blight pathogen



- Pseudomonas syringae pv. phaseolicola
- Phaseolotoxin causes chlorotic halo
- *Tox*⁺ isolates = gene cluster present & functional
- *Tox* isolates = gene cluster absent or non-functional, were considered "minor epidemiological importance"
- 2016 WA isolates: *Tox* but phaseolotoxin gene cluster present, highly virulent



Inoculation of cv. Bush Blue Lake 274

P. syringae pv. glycinea

Atoxigenic P. syringae pv. phaseolicola

P. syringae pv. phaseolicola *P. syringae* pv. syringae

Control

Derie et al. 2018

Phaseolotoxin production: Inhibition of E. coli



Phylogenetic tree based on multi-locus sequence analysis of gapA, gltA, gyrB, and rpoD genes of Pseudomonas strains from WA bean seed crops in 2016-17



Derie et al. 2018

2018 Bacterial Brown Spot in a Bean Seed Crop in Columbia Basin

- Seed crop under center-pivot irrigation
- Confirmed from: 1) phytosanitary field sample, 2) harvested seed
- Foliar & seed samples sent to Lindsey du Toit's lab for diagnosis:
 - *Pseudomonas syringae* pv. *syringae* = bacterial brown spot pathogen
 - Isolated from leaves
 - Isolated from each of two 1-kg samples of harvested seed
- Additional seed sample sent to Eurofins STA Labs:
 - Suspect bacterial colonies isolated from seed
 - Transfer to King's B agar medium = atypical, no further tests completed
 - Negative seed test result

DNA fingerprinting & pathogenicity tests of bacterial isolate from leaves of a 2018 bean seed crop

Pathogenicity test of bacterial isolates from foliage & harvested seed of 2018 bean seed crop





Pathogenicity test of bacterial isolates from harvested seed of a 2018 bean seed crop



2019 Bacterial Brown Spot in a Lima Bean Seed Crop in the Columbia Basin

- Center-pivot irrigation
- Foliar samples sent to CA Seed & Plant Lab:
 - *Pseudomonas syringae* pv. *syringae* isolated = bacterial brown spot pathogen
 - Confirmation: DNA sequencing, pathogenicity tests on bean plants & pods



Widespread Reports of Bean Leaf Spots in July 2019?



Widespread Bean Leaf Spots in 2019: Chemical Phytotoxicity?

- Almost 100% incidence in most fields
- Very rapid appearance
- During hottest time of season
- Widespread across the Columbia Basin
- 1,000's of acres, particularly lima beans
- ~7 d after aerial or ground tank-mix applications:
 - Copper (e.g., Badge)
 - Fungicide (e.g., thiophanate-methyl)
 - Fertilizer (e.g., sulfur, ammonium) + micronutrients
 - Insecticide
 - Adjuvant(s)
 - Other ingredients?



Photos: C.H. Wohleb, Washington State University

Ozone/fire smoke/PAN air pollution?

Adzuki beans highly sensitive?

Photos: L.J. du Toit, Washington State University

Spider mites & thrips

2020 Bacterial Diseases in Bean Seed Crops in the Columbia Basin

- 6 bean seed crops, center-pivot irrigation, 3 counties
- Brown spot and halo blight
 - 3 fields planted with same seed lot, grown in WA for ~5 years
 - DNA sequencing, pathogenicity tests on bean pods





Bacterial Disease Management in Bean Crops

- Certified, 'pathogen-free' seed produced in semi-arid regions
- Antibiotic seed treatment: e.g., streptomycin
- Resistant cultivars
- Crop rotation: 3+ years
- Eliminate weed hosts & volunteer bean plants
- Furrow or drip vs. overhead irrigation: reduce spread
- Limit personnel or equipment in crop when canopy is wet: dews, irrigation, high humidity, & optimal temperatures
- Copper-based foliar spray program: preventative/protectant
- Incorporate bean crop residues soon after harvest

Application of a pesticide to a crop or site that is not on the label is a violation of pesticide law and may subject the applicator to civil penalties up to \$7,500.

Such an application may also result in illegal residues that could subject the crop to seizure or embargo action by WSDA and/or the U.S. Food and Drug Administration.

It is your responsibility to check the label before using the product to ensure lawful use and obtain all necessary permits in advance.

Quarantine Bacterial Diseases in Bean Seed Crops in the Columbia Basin: Considerations

- Incidence of infection, distribution of symptomatic plants
- Shipping samples <u>overnight</u> to a lab with relevant expertise & resources, keep soil off leaves/pods
 - ISDA Seed Lab, ID
 - Eurofins STA Labs, CO; Iowa State University Seed Lab, IA; CA Seed & Plant Lab, CA
- Plant & seed assays
 - Standardized seed assays for halo blight & common bacterial blight, not for bacterial brown spot
- Confirmation of suspect bacterial colonies
 - Pathogenicity tests
 - DNA fingerprinting
- 18+ fields in WA infected since 2009, 0 from 1968-2008?
- Overhead irrigation increases risk of bacterial diseases
- Stock seed testing?

Bacterial Seed Health Assays for Bean Seed Crops: Stock Seed Lots

• WAC 16-301-380: Regulated diseases of bean

Halo blight * 2009, 2012, 2014, 2016, 2017 (toxigenic & a~), 2020
Common bacterial blight * 2009
Fuscous blight (variant of common blight)
Brown spot * 2016, 2017, 2018, 2019, 2020
Bacterial wilt
Bean anthracnose
Bean common mosaic virus (BCMV)

- 2019:
 - 46 stock seed lots sent to commercial seed testing lab
 - 11 tested positive for 1 or 2 bacterial pathogens
 - Subsamples of 8 lots tested at WSU & commercial lab

		X. axonopodis	<i>P. syringae</i> pv.	P. syringae pv.	<i>C. flaccumfaciens</i> pv.
Lot	Lab	pv. phaseoli	phaseolicola	syringae	flaccumfaciens
1	WSU	0	0	0	0
	Lab X	1.55 x10 ⁵ cfu/ml	0	0	0
	Lab Y	0	0	0	0
2	WSU	0	0	0	0
	Lab X	0	0	0	20 cfu/ml
	Lab Y	0	0	0	0
3	WSU	0	0	0	0
	Lab X	20 cfu/ml	0	0	0
	Lab Y	0	0	0	0
4	WSU	0	0	0	0
	Lab X	0	0	0	1 x 10 ² cfu/ml
	Lab Y	0	0	0	0
5	WSU	0	0	0	Positive
	Lab X	0	0	0	10 cfu/ml
	Lab Y	0	0	0	Positive
6	WSU	0	0	0	Positive
	Lab X	0	0	0	40 cfu/ml
	Lab Y	0	0	0	0
7	WSU	0	0	3.80 cfu/g	Positive
	Lab X	0	0	0	60 cfu/ml
	Lab Y	0	0	0	0
8	WSU	0	0	15.6 cfu/g	Positive
	Lab X	0	0	1.14 x 10 ⁴ cfu/ml	8 x 10 ⁶ cfu/ml
	Lab Y	0	0	0	Positive

Bacterial Wilt of Bean?!

- Curtobacterium flaccumfaciens pv. flaccumfaciens
- Systemic vascular wilt: bean, soybean, pea, Vigna
- 1922 = 1st report in South Dakota
- Spread throughout Central Plains & Midwest, central Canada, Mexico in 1960's – 1970's
- All but disappeared until early 2000's, increasing prevalence again
- Widely distributed in USA and other countries but quarantine pest in EU (EPPO A2 pest)
- Seedborne & seed transmitted
- Flaccid leaves, wilting, interveinal foliar necrosis ('firing'), stunting, death, reduced yield
- Seed may be discolored, shriveled, or asymptomatic
- High temperatures (>80°F)
- Survives:
 - in seed >20 years
 - Infected crop debris
- Pathogenic & non~ strains
- Asymptomatic hosts?

Corn, sunflower, melon, sugar beet, potato, canola



APS Press

Quarantine Bacterial Diseases in WA Bean Seed Crops

- Infected stock seed lots planted inadvertently despite testing?
 - Destructive seed assays, uneven distribution in seed lots
 - Probability of detection, field conditions, type of irrigation
- Variation in seed health protocols among labs, & technical seed pathology lab expertise, need for ring tests & lab proficiency testing
- Seed health assay limitations, interpretations, lack of standardized protocols for some pathogens
- Bean Bacterial Disease Working Groups:
 - 1. Rules & Regulations
 - 2. Cultural Best Practices & In-Field Treatments
 - 3. Pathology, Testing, & Detection Technology
- WSDA Bean Seed Quarantine: Revisions proposed to phytosanitary inspections, regulated area (*add western WA?*), crop destruction (partial vs. whole fields), seed testing protocols, gaps & nuances in wording & technical aspects for 7 pathogens & multiple host species, compatible with ID State Dept. of Ag rules
- WSDA Seed Program Manager:
 - Paula Moore <u>Pmoore@agr.wa.gov</u>, 509-314-1032

Management: Role of Crop Consultants

- Bacterial pathogens:
 - Spread rapidly
 - Don't discriminate seed vs. production crops
- Success of PWW bean industry depends on cooperation of bean seed & production growers
- Seed crops: inspected 3x/season
- Production crops: consultants/growers = 'inspectors'
 - Submit suspect samples to diagnostic lab
 - Management practices implemented before further spread
 - Avoid planting seed crops in proximity to infected fields
 - Equipment moved between fields potentially spread bacteria

