AGRICULTURAL RESEARCH FOUNDATION
REPORT
FUNDING CYCLE 2018 – 2020

TITLE: Development of Epidemiological Models for IPM of Bacterial Blight in Carrot Seed Crops

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EXECUTIVE SUMMARY:
Bacterial blight of carrot, caused by the plant pathogenic bacterium *Xanthomonas hortorum* pv. *carotae* (*Xhc*), is a common disease of carrot and the seed-borne nature of *Xhc* makes it a major concern for Oregon's hybrid carrot seed industry. The biennial nature of carrot seed production requires at least 13 months from planting to harvesting, resulting in a potential "green bridge" that facilitates pathogen survival from year-to-year. Consequently, the crop is potentially exposed to bacterial blight inoculum from the seedling stage until harvest.

The presence of *Xhc* and other *Xanthomonas* species has been documented in air samples, demonstrating the potential for movement of plant pathogenic bacteria in dust and aerosols. Previous research using truck-mounted air samplers and a semi-selective medium demonstrated that airborne *Xhc* could be detected up to 1 mile downwind of crops being threshed. These studies provided some important initial insights into potential periods of *Xanthomonas* dispersal associated with cultural practices, but more information is needed to identify dispersal periods of the pathogen during all stages of carrot seed production (e.g., when thinning plants, removing winter row covers, irrigation events, and trimming, swathing, and burying male plants after pollination).

OBJECTIVES:
The objective of this research was to identify and quantify periods of *Xhc* dispersal during critical periods of the 13- to 14-month carrot seed cropping cycle. It was expected that these data can contribute towards the development of predictive models to inform growers of inoculum risk and improve control through better-timed bactericide applications, modified cultural practices, and increased knowledge of bacterial blight epidemiology in carrot seed crops.

PROCEDURES:
Burkard 7-day volumetric spore samplers were used to sample air continuously in three carrot seed-to-seed fields (A, B, and C) harvested in 2018 and two fields (D and E) intended for harvest in 2019. Fields B and C were located approximately 4 miles northeast and 11 miles southeast of field A, respectively. Fields D and E were located adjacent to fields A and B, respectively. Air samples were collected from fields A, B, and C from July 2018 until harvest (September or October 2018) and from fields D and E from stand establishment (August 2018) through

November 2018 and from April 2019 through harvest (October 2019). Fields were not sampled from December through March due to field accessibility issues.

Samples from Burkard spore traps were separated into daily segments and DNA was extracted from the Melinex tape using a previously published procedure (Calderon et al. 2002). For fields A, B, and C, subsamples of each daily DNA sample were pooled into weekly samples for initial analyses. Additional funding was acquired from the USDA-NIFA Western IPM Center for this project, allowing us to analyze DNA samples by individual days for fields D and E.

DNA of *Xhc* was quantified using a previously published quantitative polymerase chain reaction (qPCR) assay specific to *Xhc* (Temple et al. 2013). Leaf samples (100 leaves) were randomly collected from different plants on November 16, 2018 and on May 24, June 7, and July 3, 2019 to quantify *Xhc* populations in each field. Leaf samples were cut into 1-2 cm^2 pieces and subjected to a leaf wash assay (du Toit et al. 2005), followed by DNA extraction and qPCR as described by Temple et al. (2013).

To identify factors most significantly correlated with airborne levels of *Xhc*, principal component analyses was conducted using weather data collected by the MRSO AgriMet Weather station located at the Central Oregon Agricultural Research and Extension Center in Madras, OR. Daily weather data included minimum, maximum, and mean air and soil temperatures, solar irradiation, mean relative humidity, mean dew point, daily precipitation, wind speed, wind direction, and wind run. The timing of production practices in the sampled and surrounding fields was obtained from the seed contractor to determine if cultural practices such as trimming, thinning, or irrigation contribute to the dispersal of airborne *Xhc* inoculum.

SIGNIFICANT ACCOMPLISHMENTS:

During the 2017-2018 season, the bacterial blight pathogen was detected in 52 out of 54 weekly samples at levels ranging from 10^2 to 10^6 bacteria/week (Table 1). *Xhc* levels increased from 10^2 bacteria/week in July to 10^4 and 10^6 bacteria/week prior to harvest in fields A and C, respectively. Traps were pulled from fields after harvest (fields A, B, and C).

During the 2018-2019 season, air samples were collected on a total of 179 and 195 days in fields D and E, respectively, and *Xhc* was detected on 155 (86.6%) and 181 days (92.8%) of those days (Fig. 1). On average, 2.9 x 10^3 and 6.9 x 10^3 *Xhc*/day were detected in field D and E, respectively, with the maximum number of Xhc detected on a given day being 1.6 x 10^5 and 7.3 x 10^5 in each field.

Leaf samples taken November 2018 from fields D and E showed *Xhc* populations were 4.9 x 10^3 and 9.6 x 10^3 bacteria/g leaf tissue, respectively, demonstrating that considerable *Xhc* populations can be established on seed-to-seed carrot fields prior to winter. *Xhc* was not detected in leaf samples collected from field D in May 2019, but the pathogen was recovered in June (1.7 x 10^3 bacteria/g) and July (4.0 x 10^3 bacteria/g). The pathogen was recovered at high levels from leaf samples collected from field E in May (1.2 x 10^7 bacteria/g), June (1.4 x 10^8 bacteria/g), and July (1.1 x 10^8 bacteria/g) of 2019. Xhc levels in the harvested seed were 2.7 x 10^5 and 3.5 x 10^6 in fields D and E, respectively.

Principle component analysis identified three components with eigenvalues greater than one and the scree plot also indicated that only the first three components were informative (data not shown). The first component was associated with temperature variables (daily mean, maximum, and minimum air and soil temperatures) and accounted for 53.2% of the total variance. The second component was associated with wind (wind speed, direction, and run) and 19.1% of the total variance. Factors related to moisture (daily precipitation, relative humidity, and dew point) were associated with the third component and accounted for 13.0 % of the total variance. Results were consistent for both fields, so data were combined and summarized in Table 2 and Fig. 2.

BENEFITS & IMPACT:

This research provides a foundation for the development of a forecasting model to aid in the management of bacterial blight in carrot seed crops in central Oregon. Current bactericide applications are scheduled to coincide with other production practices (e.g. applications of fertilizers, herbicides, insecticides, and/or fungicides). Although some of this is due to logistics or convenience, there is also a large knowledge gap concerning the timing and causes of bacterial blight inoculum dispersal. By identifying weather factors and cultural practices that promote the reproduction or dispersal of *Xanthomonas*, growers can potentially time their bactericide applications to occur prior to the development of favorable conditions or practices, maximizing the protective effect of copper-based bactericides. Benefits and impact from this project include: 1) the identification of weather factors and production practices that contribute to the spread of *Xanthomonas* among carrot seed fields; 2) data that can contribute to a disease forecasting model that can be used to time bactericide applications when they will be most effective; 3) presentations at Extension meetings, field days, and farm fairs discussing the use and integration of disease forecasting models in IPM programs; and 4) publications in Extension papers, station reports, and scientific journals describing the aerobiology of bacterial blight in carrot seed production systems.

ADDITIONAL FUNDING RECEIVED DURING PROJECT TERM:

Additional funding ($29,747) for this project was received through the Western IPM Center 2019 Competitive Grants Program.

FUTURE FUNDING POSSIBILITIES:

These data will be included in a proposal to be submitted to the USDA-NIFA Specialty Crops Research Initiative in March 2020.

Table 1. Weekly levels of airborne *Xanthomonas hortorum* pv. *carotae* bacteria captured in carrot seed fields using Burkard air samplers[z]

Start	End	Field A	Field B	Field C	Field D	Field E
7/8/18	**7/14/18**	*	*	1.4×10^2	*	*
7/15/18	**7/21/18**	5.8×10^2	7.6×10^4	2.3×10^2	*	*
7/22/18	**7/28/18**	0	1.8×10^5	1.3×10^3	*	*
7/29/18	**8/4/18**	4.6×10^2	7.3×10^2	9.3×10^2	*	*
8/5/18	**8/11/18**	1.2×10^4	4.0×10^2	8.9×10^3	*	*
8/12/18	**8/18/18**	4.2×10^3	2.5×10^3	6.7×10^3	*	*
8/19/18	**8/25/18**	7.9×10^4	4.1×10^2 (H)	2.9×10^4	*	*
8/26/18	**9/1/18**	MD (H)	6.6×10^3	9.8×10^2	2.1×10^5	8.0×10^3
9/2/18	**9/8/18**	MD	*	5.7×10^4	1.2×10^5	3.4×10^3
9/9/18	**9/15/18**	*	*	6.4×10^3	6.2×10^2	7.5×10^3
9/16/18	**9/22/18**	*	*	3.7×10^3	5.2×10^2	3.1×10^3
9/23/18	**9/29/18**	*	*	1.6×10^6	1.4×10^4	2.2×10^3
9/30/18	**10/6/18**	*	*	7.8×10^4 (H)	1.6×10^3	7.2×10^2
10/7/18	**10/13/18**	*	*	*	2.7×10^3	6.8×10^2
10/14/18	**10/20/18**	*	*	*	2.6×10^3	1.9×10^3
10/21/18	**10/27/18**	*	*	*	1.5×10^4	3.0×10^3
10/28/18	**11/3/18**	*	*	*	7.1×10^3	9.7×10^2
11/4/18	**11/10/18**	*	*	*	2.7×10^3	0
11/11/18	**11/17/18**	*	*	*	2.4×10^3	8.2×10^3
11/18/18	**11/24/18**	*	*	*	5.9×10^3	7.3×10^3
11/25/18	**11/30/18**	*	*	*	3.4×10^3	6.2×10^5

[z] *: No data (no trap installed); MD: Missing data due to equipment malfunction; H: Harvest

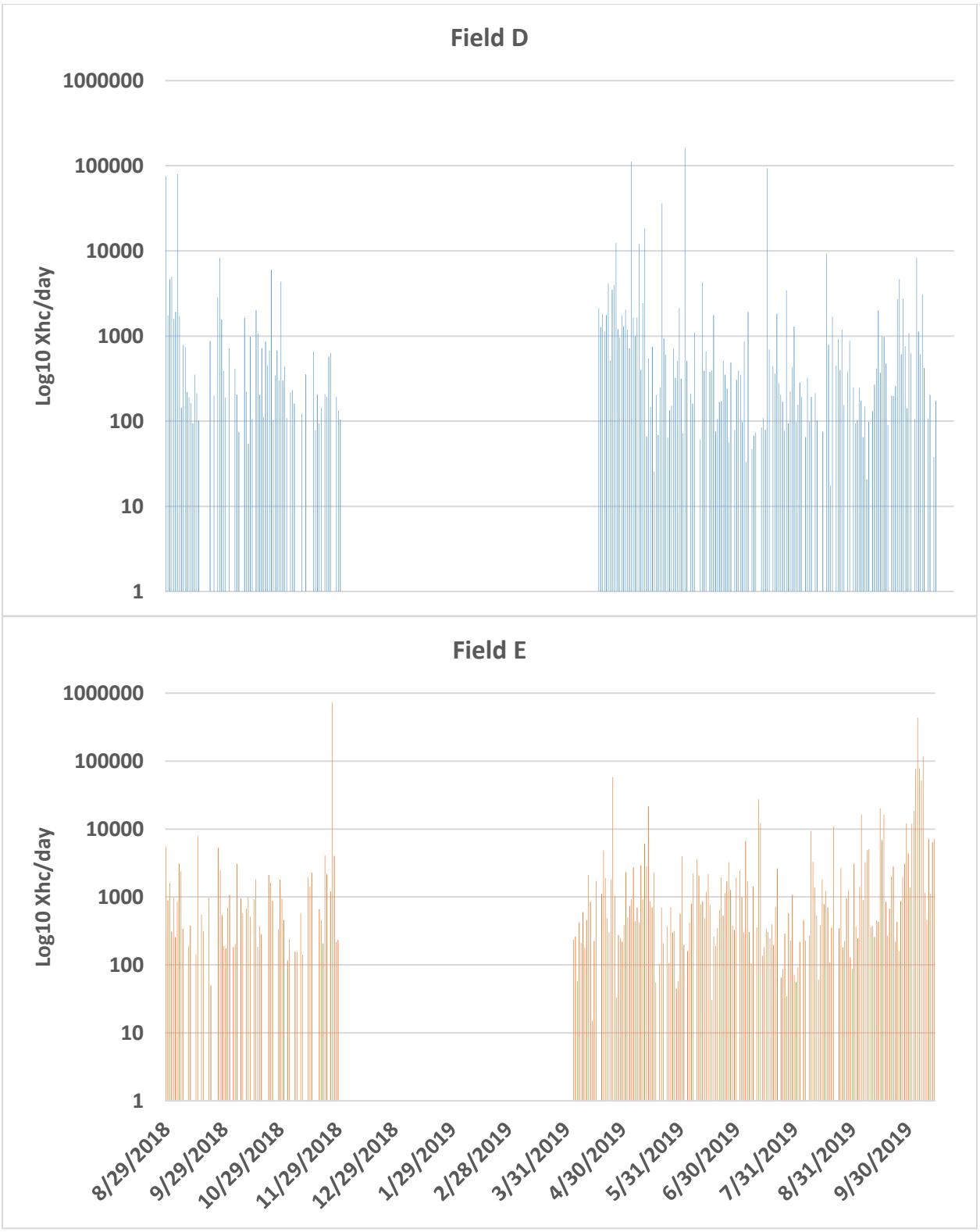

Fig. 1. Daily levels of airborne *Xanthomonas hortorum* pv. *carotae* bacteria captured in two Madras, OR carrot seed fields using Burkard air samplers. Fields were not sampled from December 2018 through March 2019.

Table 2. Principal components (PC) and eigenvector values of predictor variables for airborne *Xanthomonas hortorum* pv. *carotae* in two carrot seed fields between November 2018 and October 2019.

PC	Min. air T	Max. air T	Mean air T	Min. soil T	Max. soil T	Mean soil T	Wind speed	Wind direction	Wind run	Daily precipitation	Relative humidity	Dew point
1	0.3375	0.3604	0.3717	0.3649	0.3703	0.3698	0.0082	0.0856	0.0082	-0.0593	-0.2258	0.2608
2	0.1819	-0.0884	0.0217	0.0055	-0.0522	-0.0274	0.5237	0.4044	0.5237	0.2767	0.2536	0.2681
3	0.1549	-0.0771	-0.0059	0.0832	0.0490	0.0624	-0.4152	0.0567	-0.4152	0.4513	0.4685	0.3790
4	0.1137	0.0653	0.1037	0.0278	-0.0069	0.0100	0.1748	-0.7949	0.1749	0.4742	-0.1503	-0.0098

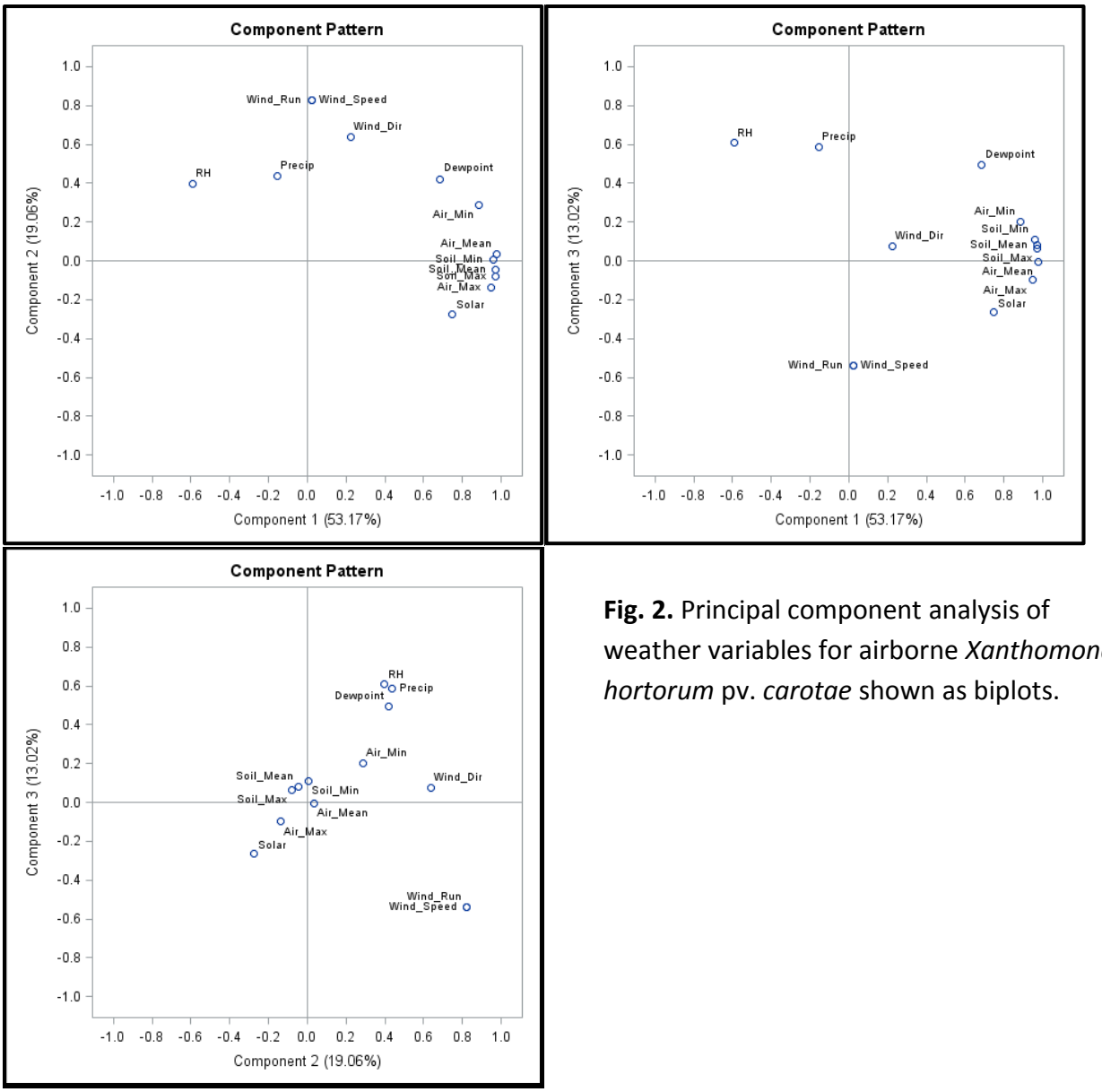

Fig. 2. Principal component analysis of weather variables for airborne *Xanthomonas hortorum* pv. *carotae* shown as biplots.