2016 Hop Biofungicide Trial

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2016 HOP BIOFUNGICIDE TRIAL

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Downy mildew has been identified as the primary pathogen plaguing northeastern hop yards. This disease causes reduced yield, poor hop quality, and, in severe cases, plant death. Control measures that reduce disease incidence and have a low environmental impact are desperately needed for the region. Regular application of protectant fungicide sprays is an effective method for managing downy mildew pressure in hop yards. However, regular chemical applications can lead to residual toxicity in the soil and have a negative effect on beneficial organisms. Extended use of protectant and curative fungicides can also lead to pathogen resistance. The goal of this project was to evaluate the efficacy of organic approved biofungicides with a variety of active ingredients for control of downy mildew in hops.

MATERIALS AND METHODS

The replicated research plots were located at Borderview Research Farm in Alburgh, VT on a Benson rocky silt loam. The experimental design was a randomized complete block with 10' x 20' plots (each plot had 4 hills). Plots were replicated 3 times per treatment with two hop varieties: Cascade and Nugget. Cascade is a moderately resistant cultivar and Nugget is a downy mildew susceptible cultivar. Each block was split in to four fungicide treatments. The treatments were:

Cease

Cease (Bioworks Inc., EPA Reg. No. 264-1155-68539) contains a strain of *Bacillus subtilis* and uses multi-site modes of action to avoid the development of resistance. This product can be used to control a variety of fungal pathogens and bacterial diseases and is intended for use both as a foliar spray and a soil amendment. Cease is to be used at the onset of favorable disease conditions prior to the onset of symptoms.
http://www.bioworksinc.com/products/cease.php

Actinovate AG

Actinovate (Novozymes BioAg Inc., EPA Reg. No. 73314-1) contains a strain of *Streptomyces lydicus* and can be used to suppress or control foliar fungal, root rot, and damping off pathogens. This product is labeled for use against downy mildew and other pathogens. Actinovate AG contains live spores of the *Streptomyces* microbe and works best if it is used prior to disease onset.
http://www.monsantobioag.com/global/us/Products/Documents/actinovateag_biofungi_18oz_case_calif_4_13114.pdf

Regalia

Regalia (Marrone Bio Innovations, EPA Reg. No. 84059-3) is a broad spectrum bio-fungicide that is active against soil borne and foliar pathogens. The active ingredient is extracted from giant knotweed (*Fallopia sachalinensis*). Regalia works by stimulating the plant's natural defenses and has antifungal and antibacterial properties.
http://marronebioinnovations.com/ag-products/brand/regalia/

Champ WG

Champ WG (Nufarm Americas Inc., EPA Reg. No. 55146-1) is 77% copper hydroxide and works as a control measure against downy mildew in hops. When copper hydroxide is mixed with water, it releases copper ions, which disrupt the cellular proteins of the fungus.
http://www.nufarm.com/USAg/ChamprWG

All pesticides applied were OMRI-approved for use in organic systems. All pesticides were applied at rates specified by their labels. The treatments were applied using a Bellspray Inc. Model T4 backpack sprayer. This model had a carbon dioxide pressurized tank and a four nozzle boom attachment. It sprayed at a rate of 10 gallons per acre. Table 1 shows rates for the biofungicides used in this study.

Table 1. Biofungicide active ingredients and rates, Alburgh, VT, 2016.

Treatment	Active ingredient	Rate per acre
Champ WG	Copper Hydroxide	1.33 pounds
Regalia	Extract of *Reynoutria sacharensis*	2.40 quarts
Cease	QST 713 strain of *Bacillus subtilis*	2.40 quarts
Actinovate	*Streptomyces lydicus* WYEC 108	180 grams

This season, we calculated the number of days that had ideal downy mildew conditions using a Pacific Northwest forecasting model based on temperature and humidity, (Gent et al. 2010) (Figure 1). The model was calculated using data from an on-farm weather station at the data collection site at Borderview Research Farm, Alburgh, VT. The humidity data was collected from a nearby weather station in Chazy, NY. We found that 25 of the 183 days between April 1, 2016 and September 30, 2016 exhibited conditions considered likely for downy mildew infection. There was less downy mildew pressure in 2016 compared to 2015 where there were 38 days likely for downy mildew infection.

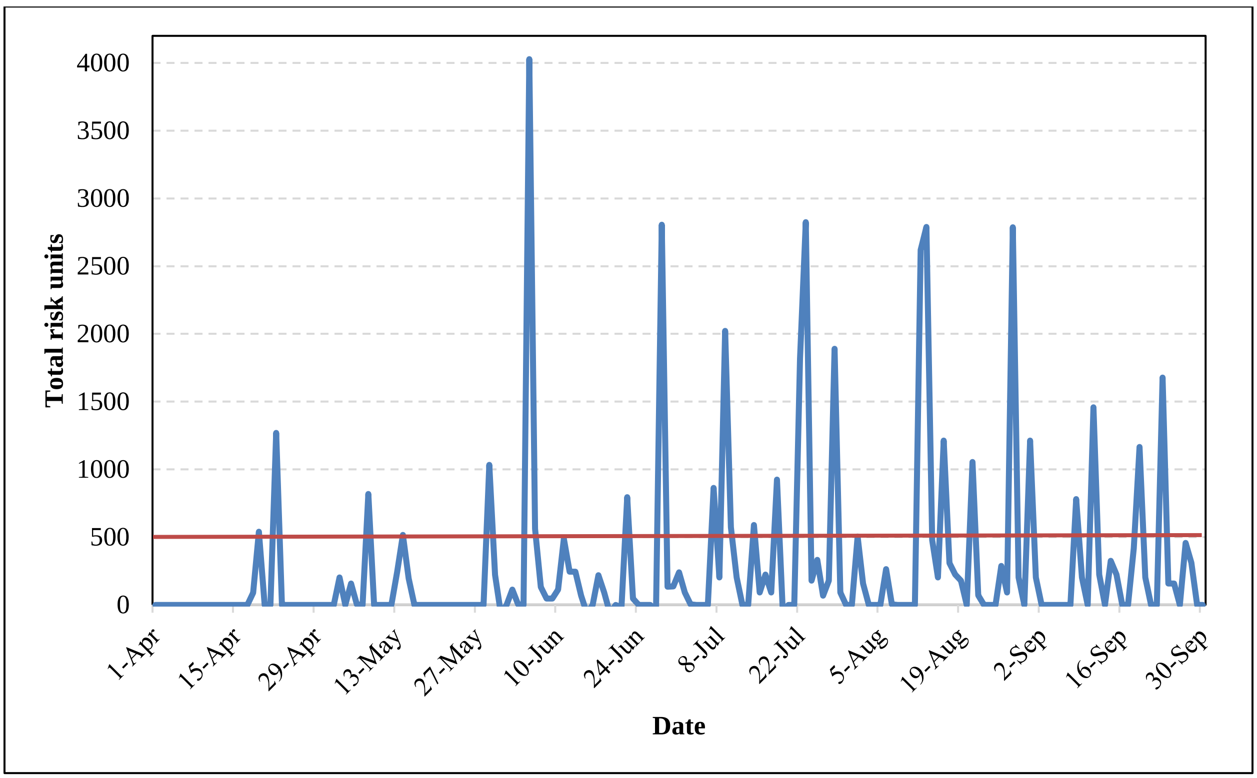

Figure 1. Number of risk units, (Gent et al. 2010), Alburgh, VT, 2016

The red line at 500 risk units indicates an increased likelihood of downy mildew infection.

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Predicting habitable conditions for downy mildew, using humidity and precipitation events, allowed us to determine optimal biofungicide application dates prior to periods of high infection risk. Given the climatic conditions of 2016, spray dates occurred weekly throughout most of the growing season. Table 2 shows fungicide application dates for the 2016 season.

Table 2. Biofungicide application dates, Alburgh, VT, 2016.

Date
27-May
3-Jun
1-Jul
8-Jul
15-Jul
26-Jul
4-Aug

The hop yard was irrigated weekly in July and August at a rate of 3000 gallons of water per acre. Starting in late May, the hops received 4 lbs ac^{-1} of nitrogen (N) through the irrigation system on a weekly basis until side shoots were observed. At each fertigation application, 25 lbs of sodium nitrate organic fertilizer (16-0-0) was added to the irrigation lines. The fertilizer was distributed evenly through 3000 gallons of water using a Dosatron unit. In addition to the fertigation, hops were side dressed twice during the season. In mid-May, 150 lbs ac^{-1} of Pro Gro (5-3-4) and 50 lbs ac^{-1} of sodium nitrate were applied to the plants. 100 lbs N ac^{-1} was applied on 21-Jun using a combination of Pro Gro and Chilean Nitrate. Total N application (including fertigation) for the season was 158 lbs ac^{-1}. All fertilizers were OMRI-approved for use in organic systems.

Throughout the season, all plots were scouted weekly for leaves exhibiting characteristics of downy mildew infection. Plots were scouted for basal spikes and rated based on severity for two weeks at the beginning of the season. During subsequent weeks, aerial spikes on each plant were counted. Basal and aerial spikes were counted by total number per plant. Leaf scouting was performed by counting 10 leaves at random on the bottom 6 feet of each plant. Spikes and leaf infection are reported as an average per plot.

Hop harvest was targeted when cones were at 21-27% dry matter. At harvest, hop bines were cut in the field and were visually assessed for disease severity on a 1-5 scale, with 5 indicating the most severe infection. The plants were then brought to a secondary location to be run through our mobile harvester (Hopster5P, hopharvester.com). Picked hop cones were weighed on a per plot basis and moisture was determined using a dehydrator. 100 cones were separated from the plots and were assessed for incidence of disease by counting the number of diseased cones. Severity was assessed on a scale of 1-10, 10 being worst. A sample of wet cones taken was from each treatment and was brought to the UVM Plant Pathology lab to quantify disease presence. All hop cones were dried to 8% moisture, baled, vacuum sealed, and then placed in a freezer. Hop samples from each plot were analyzed for alpha acids, beta acids and Hop Storage Index (HSI) by the Northwest Crops and Soils team.

Yields are presented at 8% moisture on a per acre basis. Per acre calculations were performed using the spacing in the UVM Extension hop yard biofungicide trial section of 872 hills ac^{-1}. Yields were analyzed using the GLM procedure in SAS and brew values were analyzed using the PROC MIXED procedure in SAS with the Tukey-Kramer adjustment, which means that each cultivar was analyzed with a pairwise comparison (i.e. 'Cluster' statistically outperformed 'Cascade', 'Cascade' statistically outperformed 'Mt. Hood', etc.). Relationships between variables were analyzed using the GLM procedure.

RESULTS

Weather data was recorded with a Davis Instrument Vantage PRO2 weather station, equipped with a WeatherLink data logger at Borderview Research Farm in Alburgh, VT. Missing precipitation data from 17-Aug through 31-Oct was supplemented using data provided by the NOAA from Highgate, VT. March, May, August, and September had above average temperatures. Despite the lack of rain, June and July were close to the average temperature (Table 3). While March experienced more precipitation than usual, May through September was unusually dry, accumulating 7.27 inches less rain than in a usual year. Overall, there were an accumulated 2653 growing degree days (GDDs) this season, approximately 284 more than the historical 30-year average.

Table 3. Temperature, precipitation, and growing degree day summary, Alburgh, VT, 2016.

Alburgh, VT	March	April	May	June	July	August	September
Average temperature (°F)	33.9	39.8	58.1	65.8	70.7	71.6	63.4
Departure from normal	2.90	-4.90	1.80	0.00	0.10	2.90	2.90
Precipitation (inches)	2.50	2.60	1.50	2.80	1.80	3.00	2.50
Departure from normal	0.29	-0.26	-1.92	-0.88	-2.37	-0.93	-1.17
Growing Degree Days (base 50°F)	32	59	340	481	640	663	438
Departure from normal	32	-16	74	7	1	82	104

Based on weather data from a Davis Instruments Vantage Pro2 with WeatherLink data logger. Historical averages are for 30 years of NOAA data (1981-2010) from Burlington, VT. Alburgh precipitation data from 8/17/16-10/31/16 was missing and was replaced by data provided by the NOAA for Highgate, VT.

Overall, there was little difference between Cascade treatments, (Table 4). The highest yielding treatment was Actinovate at 613 lbs ac^{-1}; however, it was not statistically different from the other treatments. The lowest yielding treatment, Regalia, yielded 478 lbs ac^{-1}. The 100 cone weight, plant disease severity, cone disease incidence, and cone disease severity were also not statistically significant between treatments. Treatments did differ by dry matter at harvest, however, there is no set value for best performance, and all treatments were harvested within the optimal range of 21-27%.

Table 4. Impact of biofungicides treatments on Cascade hop yield and disease, Alburgh, VT, 2016.

Treatment	Harvest dry matter		100 cone weight		Yield at 8% moisture		Plant disease severity		Cone disease Incidence		Cone disease severity	
	%		g		lbs ac^{-1}		1-5		%		1-10	
Actinovate	21.6	B	36.6	A	613	A	2.33	A	48.0	A	1.50	A
Cease	23.4	A	36.6	A	490	A	2.00	A	43.0	A	2.00	A
Champ	22.2	AB	36.5	A	480	A	2.33	A	44.3	A	2.00	A
Regalia	22.2	AB	36.6	A	478	A	2.00	A	46.7	A	1.50	A
Trial mean	22.4		37.1		516		2.17		45.5		1.71	
p-value	0.230		0.739		0.763		0.645		0.980		0.836	

Within a column, values followed by the same letter are not significantly different from each other.

The highest yielding Nugget treatment was Regalia with 773 lbs ac^{-1}, (Table 5). All other treatments were not statistically different from Regalia. The only characteristics that had significant values were plant disease severity and cone disease incidence. Regalia yielded plants with the lowest visual disease severity; Actinovate and Champ treatments were not statistically significant from Regalia. Champ had the least browned cones at 37.7. This value was not statistically significant from Cease and Regalia treatments. There were no statistical differences for harvest dry matter, 100 cone weight, and cone disease severity.

Table 5. Impact of biofungicides treatments on Nugget hop yield and disease, Alburgh, VT, 2016.

Treatment	Harvest dry matter		100 cone weight		Yield at 8% moisture		Plant disease severity		Cone disease Incidence		Cone disease severity	
	%		g		lbs ac^{-1}		1-5		%		1-10	
Actinovate	23.2	A	42.1	A	687	A	2.00	AB	68.0	B	2.17	A
Cease	22.2	A	44.9	A	655	A	2.50	B	49.0	AB	1.67	A
Champ	21.4	A	41.3	A	673	A	2.17	AB	37.7	A	1.83	A
Regalia	22.5	A	44.3	A	773	A	1.67	A	51.0	AB	1.83	A
Trial mean	22.3		43.1		697		2.08		51.4		1.88	
p-value	0.417		0.491		0.692		0.096		0.163		0.599	

Within a column, values followed by the same letter are not significantly different from each other.

After harvest, cones were examined under a microscope for the presence of disease spores; alternaria, downy mildew, and fusarium were found on the hop cones, (Image 1, Table 6). Alternaria was identified on the majority of Nugget cones, and on all Cascade cones that were examined. Downy mildew was much more prevalent on Nugget cones than on Cascade cones. Nugget is susceptible to downy mildew, and Cascade is moderately resistant. Fusarium was not found on Nugget cones, but was found in small quantities on Cascade cones.

Image 1. Cones infected with alternaria, fusarium, and downy mildew

Table 6. Cone diseases by cultivar and treatment, Alburgh, VT, 2016.

Cultivar	Treatment	Cones with disease present		
		Alternaria	Downy Mildew	Fusarium
Cascade	Actinovate	100%	0%	30%
	Cease	100%	40%	20%
	Champ	100%	0%	0%
	Regalia	100%	0%	20%
Nugget	Actinovate	80%	80%	0%
	Cease	90%	50%	0%
	Champ	100%	40%	0%
	Regalia	90%	30%	0%

Cones were grouped by treatment rather than plot, so no statistics are available.

Leaf infection is indicative of in-season downy mildew infection (Table 7). There were no statistically significant treatments; however Champ was the top performing treatment for both hop varieties. There were no statistically significant differences between treatments for aerial and basal spike presence (data not shown).

Table 7. Leaf infection by treatment and variety, Alburgh, VT 2016.

Treatment	Leaf infection			
	Cascade		Nugget	
Actinovate	0.002	A	0.001	A
Cease	0.002	A	0.001	A
Champ	0.000	A	0.000	A
Regalia	0.000	A	0.004	A
Trial mean	0.001		0.002	
p-value	0.281		0.897	

Within a column, values followed by the same letter are not significantly different from each other.

Table 8 shows hop brewing quality values for Cascade plants by fungicide treatment. Ideal Cascade acid content ranges between 5.5-9.0% for alpha acid and between 6.0-7.5% for beta acid. No treatment fell in the range for optimal alpha acid content. Actinovate, Champ, and Regalia treatments fell in the optimal range for beta acid; plots treated with Cease did not. Overall, the treatments did not differ significantly in quality.

As the hops "age" with exposure to time, heat and oxygen, the α-acids are converted into a variety of unflavored and off-flavored compounds, collectively referred to as "oxidized α-acids." As the oxidation takes place, the absorbance of UV light at the wavelength corresponding to the α-acids decreases, while the absorbance at the wavelength corresponding to the oxidized α-acids increases. The ratio of one absorbance value to the other gives a unit-less number known as the Hop

Storage Index. The HSI values run from a low of about 0.15 to a high of about 2.5. On this scale, lower numbers are "good" and higher numbers are "bad." Overall, the HSI values for the treatments did not differ significantly.

Table 8. Cascade alpha and beta acid content and hop storage index values, Alburgh, VT, 2016.

Treatment	Alpha acids %		Beta acids %		Hop storage index	
Actinovate	5.30	A	7.43	A	0.203	A
Cease	5.12	A	4.16	A	0.189	A
Champ	4.94	A	6.71	A	0.201	A
Regalia	5.44	A	7.45	A	0.181	A
Trial mean	5.17		7.14		0.194	
p-value	0.914		0.851		0.728	

Within a column, values followed by the same letter are not significantly different from each other.

Nugget brewing quality values are shown in Table 9. The fungicide treatments did not significantly differ in alpha or beta acids. Ideal Nugget acid content ranges from 13.5-16.0% for alpha acid, and between 4.40-5.50% for beta acid. No treatment fell in the range for optimal alpha or beta acid content. Overall, the alpha, beta, and HSI values for the treatments were not statistically different from one another.

Table 9. Nugget alpha and beta acid content and hop storage index values, Alburgh, VT, 2016.

Treatment	Alpha acids %		Beta acids %		Hop storage index %	
Actinovate	11.1	A	3.93	A	0.219	A
Cease	10.2	A	3.62	A	0.210	A
Champ	10.5	A	3.75	A	0.222	A
Regalia	12.3	A	4.21	A	0.231	A
Trial mean	11.0		3.88		0.220	
p-value	0.347		0.345		0.681	

Within a column, values followed by the same letter are not significantly different from each other.

DISCUSSION

Hop downy mildew is present in most, if not all, hop yards in the Northeast. The pathogen has been systemic in our research hop yard in Alburgh since 2012. It is possible to manage downy mildew in the northeast; however it requires a multi-faceted approach that can include crowning, using disease forecasting models, applying fungicides, and removing infected plant materials.

During the wet 2015 season, fungicides containing copper were the most effective at managing downy mildew. In 2016, however, there was no clear advantage to using one fungicide treatment over another. It is possible downy mildew was unable to sporulate prolifically and infect plants due to the dry conditions. The main transport mechanisms for downy mildew is rain splash; while some plants exhibited severe downy mildew leaf sporulation, due to the lack of rain, it is likely these spores were unable to travel and infect adjacent plants. We hope to explore these organic fungicide options again during the 2017 growing season.

While downy mildew can have major implications on plant productivity and survival, it does not play as large a role in cone browning as we had originally anticipated. Over the past two growing seasons, Alternaria has played an important role in cone browning and post-harvest cone aesthetic. Alternaria is a secondary pathogen able to infect cones late in the season after fungicide spray applications have stopped. In order to produce the highest quality crop, we hope to research this pathogen in the future and decrease late season infection. There are few conventional products labeled for this disease, so further research will need to be conducted to determine the efficacy of organic methods of control.

This trial provides insight to how much variability can occur from season to season regarding environmental conditions, yield, and disease prevalence. The amount of infection in the research hop yard was dramaticly lower in 2016 than in 2015; this difference can be largely attributed to the discrepancies in growing conditions between the two years. While there was little downy mildew in 2016, it is important to acknowledge this only reflects one year of data collection. Good scouting habits, disease forecasting models, and other tools are imperative to maintain a healthy and productive hop yard.

WORKS CITED

Gent, D., Ocamb, C., & Farnsworth, J. (2010). Forecasting and Management of Hop Downy Mildew. Plant Disease (94), 425-431.

Other fungicides registered for use in Vermont are listed in Appendix A. Regulations change frequently. Always read the label on the product for accurate information on where the product can be applied and how to use it safely. The following PNW handbook provides more information about fungicide options: https://pnwhandbooks.org/plantdisease/host-disease/hop-humulus-lupulus-downy-mildew

ACKNOWLEDGEMENTS

The UVM Extension Crops and Soils Team would like to thank Borderview Research Farm and staff for their generous help with the trials. We would like to thank Nate Brigham, Erica Cummings, Kelly Drollette, Hillary Emick, Abha Gupta, Lindsey Ruhl, Xiaohe "Danny" Yang, and Sara Ziegler for their assistance with data collection and entry. This work is made possible through funding provided by the USDA Hatch Initiative.

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APPENDIX A: OTHER FUNGICIDES

Fungicides registered for use in Vermont are listed in the table below. Regulations change frequently. Always read the label on the product for accurate information on where a product can be applied and how to use it safely. The following handbook provides more information about fungicide options: **http://pnwhandbooks.org/plantdisease/hop-humulus-lupulus-downy-mildew**

Brand name	Active ingredient	Restricted use	Certified Organic
Champ Formula 2	Copper hydroxide	No	No
Champ WG	Copper hydroxide	No	Yes
C-O-C-S WDG	Basic copper sulfate, copper oxycholoride	No	No
Cueva	Copper octanoate (copper soap)	No	Yes
Cuprofix Ultra 40D Disperss	Basic copper sulfate	No	No
Kocide 2000	Copper hydroxide	No	No
Curzate 60DF	Cymoxanil	No	No
Tanos	Famoxadone, Cymoxanil	No	No
Agri-Fos	Mono- and di-potassium salts of phosphoric acid	No	No
Aliette WDG	Aluminum tris (o-ethyl phosphate)	No	No
Flint	Trifloxystrobin	No	No
Pristine	Pyraclostrobin	No	No
Sonata	*Bacillus pumilus*	No	No
Regalia	*Reynoutria sachalinensis*	No	Yes
Actinovate	*Streptomyces lydicus*	No	Yes