Final Report of Research Proposal Submitted to:
The Pennsylvania Wine Marketing and Research Board
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TITLE: Evaluation of Botector for bunch rot control of wine grapes in Pennsylvania

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INTRODUCTION:

Bunch rot caused by *Botrytis cinerea* is an important disease of wine grapes in Pennsylvania. Chemical control programs for bunch rot typically consist of an application of a *Botrytis* spp.-specific fungicide at bloom, pre close, veraison, and pre-harvest However, bunch rot management programs that rely solely or primarily on chemical control do not always provide the highest level of bunch rot control and can hasten the development of resistance to the active ingredients we currently have. Therefore, there is an ongoing need for greater integration of non-chemical methods into bunch rot control programs.

A relatively new biological pesticide called Botector, has been labeled for use on grapes for the control of Botrytis bunch rot. Botector, a formulation of two strains of a common yeast (*Aureobasidium pullulans*) competes with the *Botrytis* fungus for nutrients on the surface of the vine and can limit the germination of spores and growth of *Botrytis*. Previous tests with Botector by various university researchers have had mixed results. Our previous tests have been somewhat disappointing, and a more thorough look at this product is warranted before Pennsylvania growers decide to try this material in their commercial vineyards. Westbridge Agricultural Products, the company that makes Botector, also recommends the use of a nutrient program applied by several spray applications throughout the growing season. Among other things, this nutrient program purportedly lengthens clusters of varieties with compact clusters, making them less compact and therefore, less susceptible to bunch rots. These 'non-chemical pesticide' programs were compared to, and combined with, trace bloom leaf removal, a well researched cultural treatment that has been shown to reliably reduce fruit set and reduce cluster compactness and bunch rot.

OBJECTIVES:

Our objective was to determine the efficacy of Botector and the Westbridge nutrient program on bunch rot development in solo, integrated, and rotational programs with chemical and cultural (trace bloom leaf removal) controls. The results will provide researchers and growers with information as to the best use of these products and options in grape disease and resistance management programs.

WORK STATEMENT:

Past research with bio-suppression of *Botrytis* in grapes tends to suggest that best results are obtained through the integration of biological control agents with cultural and chemical controls. Therefore, Botector treatments were applied alone, and as an integrated partner with chemical fungicides, Westbridge foliar nutrients, and/or leaf removal. This allowed us to examine Botector's efficacy alone and as a way to reduce the number of chemical sprays to manage resistance without compromising rot control. We anticipated the use of *Vitis vinifera* 'Chardonnay' in this trial, but severe winter cold killed the vast majority of the vines and we had to conduct the trial on *Vitis* interspecific hybrid 'Vignoles' instead. Like Chardonnay, Vignoles has a high susceptibility to bunch rots and is an ample substitute for Chardonnay. The trial was conducted with mature vines trained to a single-curtain, high-wire cordon

system at the Lake Erie Regional Grape Research and Extension Center in North East, PA. Treatments were applied to 3-vine plots in a randomized complete block design with four replications. All spray treatment applications were made with a Friend covered-boom plot sprayer at 100 psi and 50 (prebloom) or 100 (post bloom) gal/A. The leaf removal treatment involved the removal of the four most basal leaves of a fruit-bearing shoot at the beginning of bloom (trace bloom). Our treatment objectives were i) to evaluate and compare the individual efficacy of trace bloom leaf removal, chemical fungicides (Endura, Scala, Rovral, and Elevate), Botector, and the Triggrr/Westbridge foliar nutrient program, and ii) to evaluate combinations of these of these four programs, for control of *Botrytis* bunch rot. The trace bloom leaf removal treatment is designed to reduce fruit set, compactness, and susceptibility of clusters, and improve sunlight, air, and pesticide penetration into the cluster zone and has consistently provided good results in past trials. The chemical fungicide program is designed to protect the fruit from *Botrytis* infection at critical stages of fruit development. Botector, a formulation of yeast that colonizes the berry surface, controls rot by competing for resources with the *Botrytis* fungus. The Triggrr/Westbridge program contains nutrients and plant growth hormones designed to stretch clusters (to reduce cluster compactness and susceptibility) and to improve the integrity of berry skins, an important line of defense against *Botrytis.* Other diseases (i.e., powdery mildew, downy mildew, *Phomopsis* cane and leaf spot, and black rot) were controlled with standard fungicides applied with a Berthoud air blast sprayer. The incidence (percent infected) and severity (percent area infected) of *Botrytis* bunch rot were determined on 24-25 Sep from 30 clusters per plot, selected randomly from the center of the plot. Data were analyzed using analysis of variance (SAS Institute Inc., Cary, NC).

Total rainfall for May, June, July, August, and September was 3.65, 4.3, 5.6, 3.1, and 3.3 inches, respectively. Weather conditions were relatively wet during the early fruit growth stages (June and July), providing favorable conditions for latent fruit infection of *Botrytis*. Conditions were less favorable for rot development during the beginning of the ripening period (late August), but turned wetter during the last three weeks of ripening in September. The majority of bunch rot was from *Botrytis.* None of the four programs when applied alone (trace bloom leaf removal, chemical fungicides, Botector, or the foliar Triggrr/Westbridge nutrient program), significantly reduced the incidence of *Botrytis* bunch rot (Table 1). However, two combinations of programs reduced the incidence of rot when compared to the untreated control: the combination of trace bloom leaf removal with Botector, and the fully integrated program (leaf removal + Triggrr/Westbridge + Botector (bloom and pre harvest) + chemical fungicides (Scala at pre close, Rovral at veraison)), the latter being superior to the Triggrr/Westbridge, Botector, or the full season chemical fungicide program (Endura, Scala, Rovral, and Elevate) alone.

Trace bloom leaf removal alone and in combination with other programs, and the full season chemical fungicide program, significantly reduced Botrytis bunch rot severity (Table 1) when compared to the untreated control. Combining leaf removal with the full season chemical fungicide program provided the highest level of rot reduction in the trial (78 %). Conversely, the Botector and Triggrr/Westbridge foliar nutrient program did not control rot severity and did little to increase the efficacy of other programs when combined with them. Cluster length measurements just before bloom (June 16) and at harvest showed that the Triggrr/Westbridge program did not increase cluster length as we had hoped, nor did it reduce compactness. In fact, vines sprayed with just the Triggrr/Westbridge program, had clusters that were significantly more compact than clusters collected from the check treatment.

Table 1. Incidence and severity of Botrytis bunch rot under various disease management programs.

Treatment and rate/A	Application timing[z]	Incidence	Severity[y]	% Control[x]
Foliar Triggrr 12 oz + Westbridge nutrient program[w]	1			
Foliar Triggrr 16 oz + Westbridge nutrient program	2, 3			
Foliar Triggrr 8 oz + Westbridge nutrient program	5			
Foliar Triggrr 2 oz + Westbridge nutrient program…….	7-11	93.3 ab[v]	30.5 a[v]	8
Botector 5 oz…….....	5, 6, 7, 10	94.2 a	29.3 a	11
Endura 8 oz	5			
Scala SC 18 fl oz	6			
Rovral 4F 1 qt + Induce 0.125%	7			
Elevate 50 WDG 1 lb…….....	10	90.9 ab	16.0 bc	52
Leaf removal (trace bloom)…….	4	79.2 abc	15.6 bc	53
Foliar Triggrr 12 oz + Westbridge nutrient program	1			
Foliar Triggrr 16 oz + Westbridge nutrient program	2, 3			
Foliar Triggrr 8 oz + Westbridge nutrient program	5			
Foliar Triggrr 2 oz + Westbridge nutrient program	7-11			
Botector 5 oz……..…….....	5, 6, 7, 10	90.8 ab	27.1 ab	18
Foliar Triggrr 12 oz + Westbridge nutrient program	1			
Foliar Triggrr 16 oz + Westbridge nutrient program	2, 3			
Foliar Triggrr 8 oz + Westbridge nutrient program	5			
Foliar Triggrr 2 oz + Westbridge nutrient program	7-11			
Leaf removal (trace bloom)	4			
Botector 5 oz…….....	5, 6, 7, 10	83.3 abc	12.6 c	62
Leaf removal (trace bloom)	4			
Botector 5 oz…….....	5, 6, 7, 10	73.3 bc	10.8 c	67
Leaf removal (trace bloom)	4			
Endura 8 oz	5			
Scala SC 18 fl oz	6			
Rovral 4F 1 qt + Induce 0.125%	7			
Elevate 50 WDG 1 lb…….....	10	76.7 abc	7.4 c	78
Foliar Triggrr 12 oz + Westbridge nutrient program	1			
Foliar Triggrr 16 oz + Westbridge nutrient program	2, 3			
Foliar Triggrr 8 oz + Westbridge nutrient program	5			
Foliar Triggrr 2 oz + Westbridge nutrient program	7-11			
Leaf removal (trace bloom)	4			
Botector 5 oz	5, 10			
Scala SC 18 fl oz	6			
Rovral 4F 1 qt + Induce 0.125%.....	7	69.2 c	15.6 bc	53
Untreated control.....		95.8 a	33.0 a	

[z]Timing: 1 = 30 May; 2 = 5 Jun; 3 = 10 Jun; 4 = 23 Jun (trace bloom); 5 = 24 Jun (bloom); 6 = 11 Jul (pre-close); 7 = 19 Aug (veraison); 8 = 26 Aug; 9 = 2 Sep; 10 = 9 Sep (pre-harvest); 11 = 15 Sep.

[y]Severity was rated using the Barratt-Horsfall scale (0-11) and was converted to % area infected (0-100 %) using Elanco conversion tables.

[x]Percent control = control of disease severity on clusters relative to the untreated control.

[w]Westbridge nutrient program consisted of Organic Biolink (OBL) Cal Plus + OBL 3-3-3 + OBL Micro, all at 32 oz/A.

[v]Means followed by the same letter within a column are not significantly different according to Tukey-Kramer ($P \leq 0.05$).

We also examined the efficacy of Botector through laboratory inoculation of field treated berries with spores of *Botrytis* and examination of subsequent levels of rot per treatment. Samples of healthy berries were collected from the Full Botector program, 1, 3, 7, and 14 days after the pre-harvest application (when berries are most susceptible to invasion through the skin) to determine the period of optimum efficacy after application (how long it takes for the yeast population in Botector provide maximum protection against *Botrytis*). Samples were also collected from the full chemical program and from clusters in the check treatment for comparison. Before inoculation with Botrytis, half the berries in each sample were wounded with a sterile needle to assess the ability of the yeast in Botector and the fungicide in the chemical program (Elevate at 1 lb/A) to protect the wound from *Botrytis*.

On unwounded, intact berries, inoculation with Botrytis at one day after treatment application produced almost no infection in any treatment (Figure 1). Infection did increase to a small extent in subsequent samples at 3, 7, and 14 days after application, but remained at very low levels regardless of treatment and there were no significant treatment effects. This suggests that an intact berry skin is a formidable obstacle to Botrytis infection and preservation of skin integrity should be an important objective in bunch rot control programs.

Wounding greatly increased infection in all treatments (Figure 2). Unfortunately, when compared to the check, Botector appeared to do little to reduce infection of wounded berries at any time after application (1, 3, 7, or 14 days after application), whereas application of Elevate significantly improved control of rots that developed upon inoculation of wounded berries at 7 and 14 days after application. Therefore, protection of wounded berries by Botector was poor in all samples and did not appear to change at any point within the 14 days after the application. On the other hand, protection of wounded berries by Elevate, although modest (average of about 66% rot reduction), was significantly more effective than Botector at 3, 7, and 14 days after application. With these results and those of the field trial, we cannot recommend the use of Botector or the Westbridge nutrient program for control of bunch rot of grapes and have decided not to request further funding of this project.

Figure 1: Rot Incidence 1, 3, 7, and 14 days after application (daa); uninjured berries

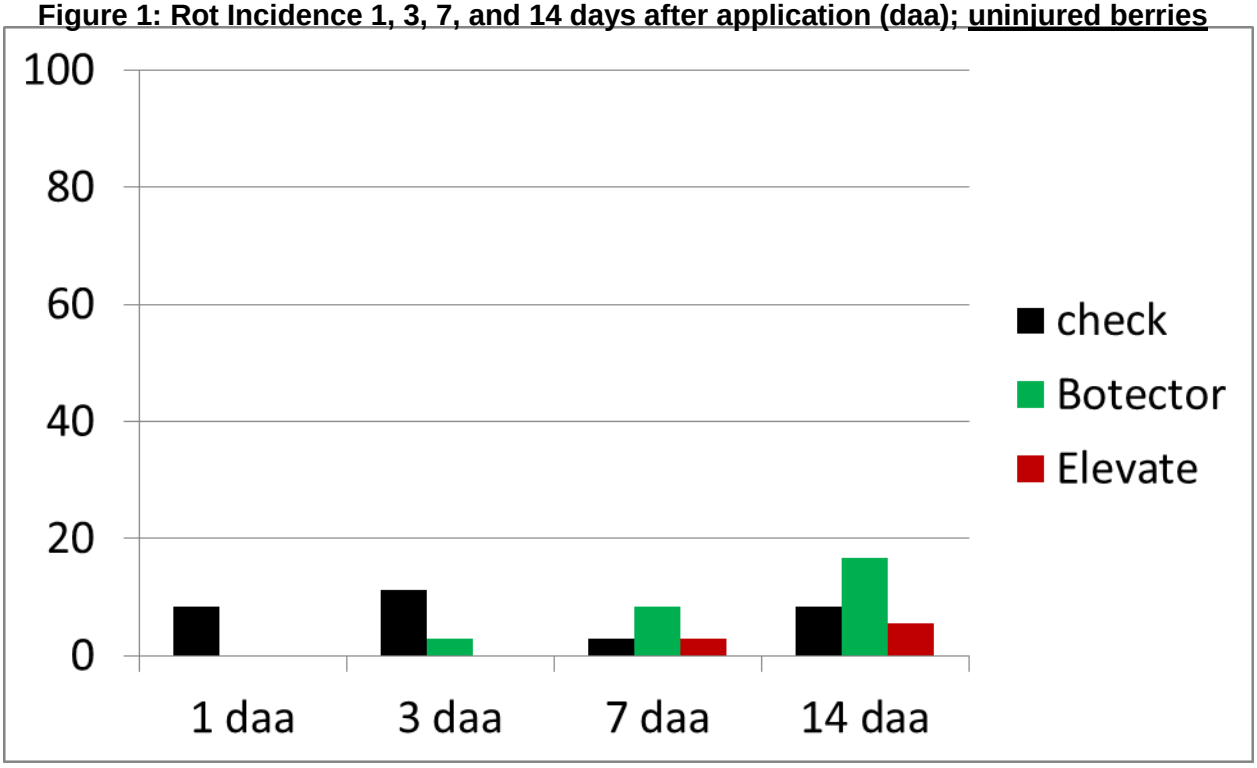
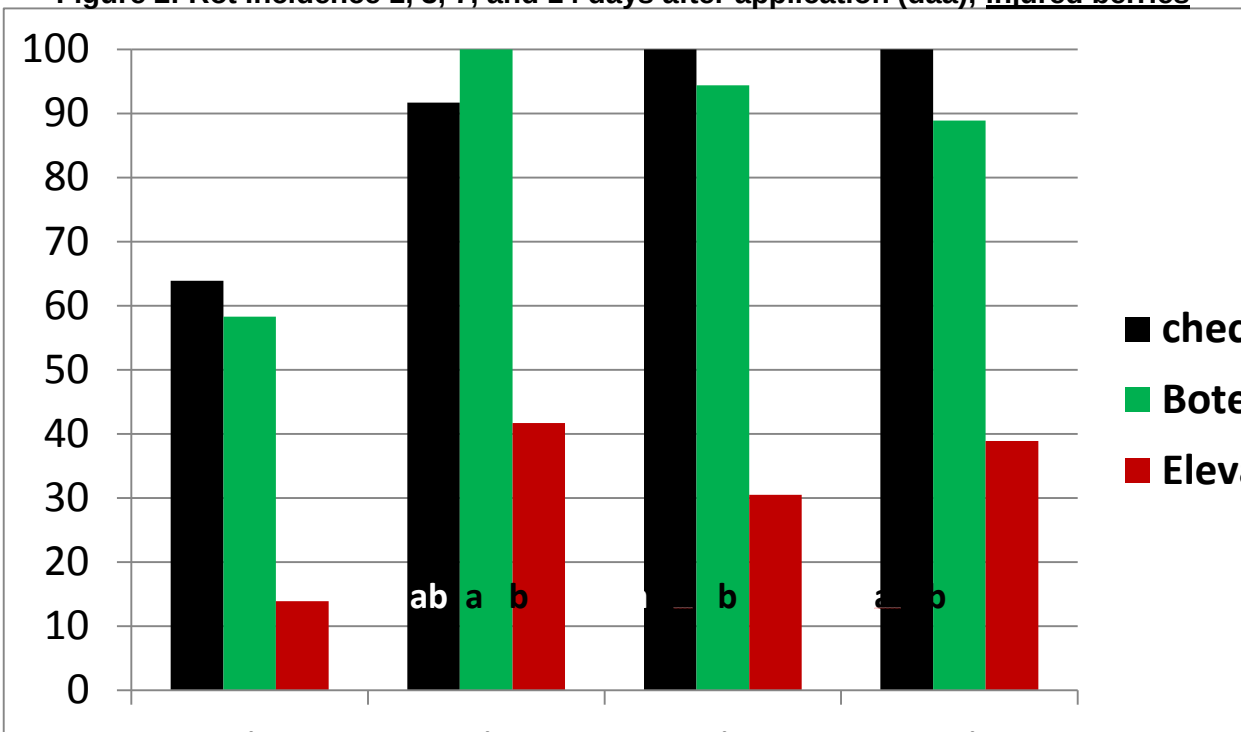


Figure 2: Rot Incidence 1, 3, 7, and 14 days after application (daa); injured berries



BOTTOM LINE: The most effective programs integrated the use of early leaf removal and chemical fungicides.

BUDGET: Funds were requested for labor for sampling, plot maintenance, production of fungal inoculum, inoculation of berries in the lab, and travel for presentation of information. The following is an itemization for how the funds were spent.

		Amount ($)
Wages:		
Category III @ $1,200		$1,200
+ Fringe Benefits $96		$96
	Total	$1,296
Other costs:		
Supplies: petri plates, microbiological media, sampling bags, moist chambers		$400
Travel: for presentation of results at Mid Atlantic Fruit and Vegetable Convention		$300
Total:		**$1,996**