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# Efficacy of organic disease control products on common foliar diseases of tomato in field and greenhouse trials

D.S. Egel<sup>a,\*</sup>, L. Hoagland<sup>b</sup>, J. Davis<sup>c</sup>, C. Marchino<sup>a</sup>, M. Bloomquist<sup>c</sup>

<sup>a</sup> Department of Botany and Plant Pathology, Purdue University, 915 West State Street, West Lafayette, IN, 47907, USA

<sup>b</sup> Department of Horticulture and Landscape Architecture, Purdue University, 625 Agricultural Mall Dr., West Lafayette, IN, 47907, USA

<sup>c</sup> Department of Horticultural Science, 455 Research Drive, Mills River, NC, 28759, USA

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# ABSTRACT

Organic vegetable production continues to increase, while the knowledge about how to best manage foliar diseases in this system has lagged. Several combinations of organically certified products with different modes of action were evaluated for potential to manage foliar diseases of tomato in two field sites with different climatic conditions over two years, and in a greenhouse setting. Early blight, Septoria leaf spot and bacterial spot foliar diseases were naturally present in the field trials, while greenhouse plants were artificially inoculated with a virulent isolate of Alternaria solani, causal agent of early blight. The copper product Badge X2 (23.82% copper oxychloride and 21.49% copper hydroxide), significantly lowered disease levels in all field and greenhouse trials. The biological control product Prestop (Gliocladium catenulatum J1446), significantly lowered early blight disease levels in all field trials and two out of three greenhouse trials. Alternating Serenade Opti (B. amyloliquefaciens QST 713 (syn. B. subtilis QST713), with Regalia (extract of Reynoutria schalinensis) did not consistently lower disease levels compared to the untreated control in field trials. However, Serenade Opti alone lowered early blight disease levels in two out of three greenhouse trials. Sil-Matrix (potassium silicate 29%), and Regalia, applied alone and in combination were generally ineffective in both field and greenhouse trials. An alternation of Oxidate 2.0 (27.1% hydrogen dioxide and 2.0% peroxyacetic acid), which was used to lower the microbe populations on tomato foliage after which Sustane, an aerobically composted turkey litter, was applied to promote colonization by beneficial bacteria, did not consistently lower disease levels compared to the untreated control in greenhouse and field trials. Results of these trials indicate that the biological product Prestop is an effective alternative to copper for the control of early blight, but not necessarily other common foliar diseases in tomato.

#### 1. Introduction

The demand for organically grown produce has risen dramatically over the past decade: organic products are now available in almost 20,000 natural food stores and three out of four conventional grocery stores (Anonymous, 2017). In response, an increasing number of vegetable growers are using organic methods on all or a portion of their production. Organic production, however, presents unique problems, none more urgent than the management of foliar diseases of tomato (Hoagland et al., 2015). The most effective disease management strategies in organic systems rely on the integration of many cultural and biological practices such as planting resistant varieties, enhancing airflow and growing plants under plastic covers to minimize leaf wetness. In recent years, a wide variety of products with many different modes of action have become available to augment cultural and biological practices. However, many of these products lack independent third-party testing to verify efficacy, particularly in field environments. Consequently, the aim of this study was to provide an unbiased research-based evaluation of products reported by growers to effectively suppress diseases, along with new products with strong performances reported in company promotional material. In addition, we aimed to test the hypothesis that combining products with different modes of action would result in greater disease control.

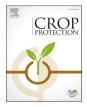
The types of products available for controlling foliar diseases in organic vegetable crops may be separated into different categories based on the type of active ingredient and potential mode of action (Table 1).

\* Corresponding author. *E-mail address:* egel@purdue.edu (D.S. Egel).

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The standard active ingredient that has been in use for decades in organic and conventional production is copper. Products with fixed copper as an active ingredient include copper hydroxide, copper sulfate, copper oxychloride and others. Copper ions kill pathogen cells on plant surfaces by bonding with sulfhydryl, phosphate, carboxyl and other ionogenic groups, thereby destroying enzymes critical for cell functioning (Somers, 1961). These products have a long history of effectiveness against foliar diseases (Morton and Staub, 2008). Although many copper products are certified for disease control in organic systems, copper is a heavy metal, and because it acts on contact rather than systemically, these products must be applied often, which can result in build-up of residues that negatively impact soil and water quality (Eijsackers et al., 2005). Copper accumulation has been demonstrated to negatively affect the composition and functional activity of soil microbial communities (Giller et al., 1998; Viti et al., 2008; Fernández-Calviño et al., 2008; Zhou et al., 2010). Therefore, the use of

# Table 1

Descriptions of	f all the	e products	trialed	in	this	study.
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Treatment	Active ingredient	Rate	Experimental	Comment
Badge X2	Copper oxychloride 28.82% Copper hydroxide 21.49%	2.0 kg// ha	Applied weekly	Standard organic product
Oxidate 2.0	hydrogen dioxide 27.1% Peroxyacetic acid 2.0%	1:100 with water v/v	Oxidate 2.0 was applied to plants and allowed to dry after which Sustane was applied. Treatment applied weekly.	Used by organic growers to sanitize surfaces (Oxidate) and add beneficial microbes via a compost tea (Sustane).
Sustane 4- 6-4	Aerobically composted turkey litter	378 g/ 37.8 L water		
Prestop	Gliocladium catenulatum strain J1446	0.05% w/v	Applied every 3 weeks.	Biological control agent that acts as a competitor to pathogens and a
Serenade Opti	$1.3 \times 10^{10}$ CFU/g Bacillus amyloliquefaciens QS T713 (syn. B. subtilis QST 713)	1.5 L/ ha	Alternated with Fracture, Regalia or Sil- Matrix in separate treatments; all applied	hyperparasite. Lipopeptides produced by bacteria are antifungal and may induce systemic resistance ( Lahlali et al.,
Fracture	BLAD polypeptide 20%	3.3 L/ ha	weekly. Alternated with Serenade Opti weekly.	2013). The plant derived active ingredient attacks the chitin in
Sil-Matrix	Potassium silicate 29%	1% v/v applied as drench	Alternated with Serenade Opti weekly.	fungal cells. Silicone is purported to add mechanical strength to leaves and/or induce resistance.
Regalia	Extract of Reynoutria schalinensis	7 L/ha	Alternated with Serenade Opti weekly.	May elicit phenolic compounds used in combating diseases.

copper products in organic production has become controversial due to possible environmental toxicity (Bruggen and Finckh, 2016). Moreover, after many years of use, some plant pathogens, such as *Xanthomonas* species that cause bacterial spot on pepper and tomato, have developed resistance to copper products (Marco and Stall, 1983). Soil quality is widely regarded as critical for productive and sustainable agriculture organic, with beneficial soil microbes helping to reduce pathogen infestations (Doran and Zeiss, 2000).

In addition, or instead of the use of copper containing products, organic growers may use biological control products, some with multiple modes of action. In this study, two microbial products, Prestop (AgBio, Loveland, CO) and Serenade Opti (Bayer, Research Triangle Park, NC), are evaluated. Prestop biological activity is based on a living fungus, Gliocladium catenulatum J1446. Gliocladium catenulatum is purported to suppress pathogens directly through a combination of competition and hyperparistism (Mcquilken et al., 2001). Serenade Opti relies on lipopeptides in the bacterium, Bacillus amyloliquefaciens QST 713. The lipopeptides produced by B. amyloliquefaciens strains have been reported to directly suppress pathogens by inhibiting lipid synthesis and transport/membrane integrity and function. In addition, B. amyloliquefaciens has been observed to activate an induced systemic resistance (ISR) process, which strengthens a plant's immune system and thereby reduces susceptibility to pathogen infection (Lahlali et al., 2013).

Botanical products Regalia (Marrone Bio Innovations, Davis, CA) and Fracture (FMC Agricultural Solutions, Philadelphia, PA) are also evaluated for their ability to suppress plant pathogens. The activity of botanical products varies as widely as the sources of the plants from which they are derived. Regalia, an extract of the plant Reynoutria schalinensis is, like Serenade Opti, purported to activate Induced Systemic Resistance (ISR) (Dayan et al., 2009). Fracture (FMC, Philadelphia, PA), a BLAD (Banda de Lupinus albus doce) polypeptide derived from germinating Lupinus albus plants, is a lectin which binds to chitin and possesses exochitinase and endochitosanase activities (Pinheiro et al., 2016). Although the mechanism is unclear, silicon has been used to reduce severity of rice diseases including blast and brown spot (Datnoff et al., 2001). Like copper, silicon is an inorganic ingredient, though while the effect of copper is directly on the pathogen, silicon appears to be involved in an ISR type of response. The silicon product evaluated in this study was Sil-Matrix (Certis USA, Columbia, MD).

Organic disease control products are often reported as effective in controlled greenhouse trials, but their efficacy under field conditions has been widely variable. One way to help overcome some of this variability might be to combine products with different modes of action (Guetsky et al., 2001). One of the combinations used here was to apply a product with the active ingredient hydrogen peroxide to lower the population of pathogens on the leaf surface, followed by compost tea to seed the leaf with beneficial microbes that can outcompete or directly suppress pathogens. Such 'priority effects' are well known to have long-term effects on microbial community structure (Tucker and Fukami, 2014), and the use of hydrogen peroxide has previously been found to increase the survival of microbes with biocontrol activity in turf systems (Mercier, 2006). Applying compost teas has reduced severity of early blight in tomato (Alternaria solani, (Ellis and Martin) Jones and Grout (Haggag and Saber, 2007), though in another study, compost extracts alone provided inconsistent control of late blight in potato (Ghorbani et al., 2005). There are many types of commercially available compost tea products; some compost teas are homemade (Islam et al., 2016). In this study, we applied a commercially available product manufactured from aerobically composted turkey manure, Sustane (Sustane Natural Fertilizers, Inc., Cannon Falls, MN) so that this research can be easily repeatable. This product, labeled as a fertilizer, contains a diverse assortment of microbial species (B. Gardener, personal communication). The hydrogen dioxide product used to lower the population of pathogens on the leaf (Fukami, 2015), was Oxidate 2.0 (BioSafe Systems LLC, East Hartford, CT).

Field trials evaluating efficacy of these products in managing foliar pathogens in tomato were conducted in organically certified experimental plots at Purdue University and North Carolina State University during the 2016 and 2017 summer growing seasons. In addition, three greenhouse trials were conducted at Purdue University. The field trials relied on natural spread of pathogens; the most prominent diseases observed in the two years and two locations of the trials were early blight, Septoria leaf spot (*Septoria lycopersici* Speg.) and bacterial spot (*Xanthomonas perforans* Jones). In greenhouse trials, plants were inoculated with a virulent isolate of Alternaria solani, causal agent of early blight, isolated from a commercial fresh market tomato leaf.

#### 2. Methods and materials

#### 2.1. Field experiments

Field trials were established in Vincennes, IN at the Southwest Purdue Agricultural Center, and the Mountain Organic Research and Extension Unit in Waynesville, North Carolina in 2016 and 2017. Both locations utilized certified organic land and followed all practices and products approved by the local organic certifying agencies. The soil types at the experimental locations in Vincennes, IN and Waynesville, NC were a Bloomfield fine loamy sand and an Evard-Cowee/Cullowhee-Nikwasi sandy loam, respectively. Rainfall totals for the duration of the experiments in Indiana in 2016 were 67.59 cm, and in 2017 were 51.51 cm. In North Carolina, rainfall totals in 2016 were 32.66 cm and in 2017 were 47.98 cm. Average temperature during the same time period in Indiana in 2016 was 21.5 °C and in 2017 was 21.36 °C; average temperature in North Carolina in 2016 was 18.7 °C and in 2017 was 18.4 °C (data taken from usclimatedata.com).

Tomato cv. Oregon Spring (Natural Gardening Company, Petaluma, CA) was planted in both locations in 2016; the tomato variety cv. Celebrity (Johnny's Seed, Fairfield, ME) was used in 2017. The variety Oregon Spring did not have adequate marketable yield in Indiana and North Carolina. In Vincennes, tomato seeds were sown in Johnny's Seed 512 Organic Mix soilless media on 12 Apr in 2016 and on 10 Apr in 2017. Seedlings were transplanted to the field at the Southwest Purdue Agricultural Center in Vincennes, IN on 24 May in 2016 and on 22 May in 2017. In North Carolina, seeds were sown in McEnroe Premium Light sowing mix (Millerton, NY) on 22 Apr in 2016 and 18 Apr in 2017 and seedlings were transplanted to the field on 8 Jun in 2016 and 23 May in 2017. In both locations, 10 plants were spaced 0.6 m apart within rows and rows were spaced 3.05 m apart. Each row was mulched with 1.22 m wide x 0.08 ml black plastic (Visqueen 4020). Nature Safe 10-2-8 (Irving, TX), a composted manure product, was added at the rate of 1.68 kg per 6.1 m plot in Indiana. True 7-5-7 (AgCare, Candler, NC) (1.905 kg/ ha) and lime (2241.7 kg/ha) were incorporated pre-plant in North Carolina. Brown's Fish Hydrolysate (Andrews, NC) was applied July 23 and Aug 5, 2017 at the rate of 23 L per hectare in North Carolina through drip irrigation. Boron was added as Solubor (AgCare, Candler, NC) to the plots in North Carolina at the rate of 1.1 kg/Hectare through drip irrigation on July 21, 2018. Plants were irrigated as needed with drip-tape under the plastic throughout each growing season. Tomato plants were staked and maintained using the Florida weave method. Each row was an experimental unit and the experimental design was a randomized complete block with four replications.

#### 2.2. Treatments

Treatments, active ingredients, rates, experimental details were determined from product labels and in consultation with industry representatives (Table 1). Sustane 4-6-4 (Sustane.com; Cannon Falls, MN) was prepared by adding 100 g to 10 L of nonchlorinated water in a polypropylene bucket and mixing briefly with a wooden spoon. An aquarium bubbler was used to constantly agitate the solution (Aqua, pump MK-504; 25-inch aquarium stone) for 24 h. The solution was

decanted to spray bottles to avoid nozzle clogging. Fracture was approved by an organic certifier in Indiana for 2016 only.

In 2016, products were applied approximately weekly from 24 May to 12 Aug in Indiana and 8 Jun to 3 Aug in North Carolina. In 2017, applications were made from 14 Jun to 30 Aug in Indiana and 26 May to 11 Aug in North Carolina. Applications were made with a CO<sub>2</sub> pressurized backpack sprayer (R&D sprayer, Opelousas, LA) at 414 kPa and 374 LHa1. The hand-held boom of the sprayer had four Teejet 8002VS nozzles.

Field plots were not inoculated with any pathogen; disease incidence and severity in tomato was dependent on natural inoculum and environmental conditions. Disease severity was rated visually using the Horsfall-Barratt rating scale (Horsfall and Barratt, 1945) over the entire plot, which was an experimental unit. Ratings were conducted on 25 Jul, 3, 11, 18 Aug in Indiana (2016); in North Carolina ratings were conducted 12, 26 Jul, 1, 12, 17, 25 Aug in 2016. In 2017, the rating dates were 9, 17, 24 Aug, and 4 Sep in Indiana; in North Carolina ratings were conducted 5, 12, 19, 24 Jul, 2, 9, 16, 23, 30 Aug, and 8 Sep. Area under the disease progress curve (AUDPC) was calculated using trapezoidal integration (Shaner and Finney, 1977). Tomato fruit in 2016 were harvested on 19, 26 Jul, 2, 9, and 17 Aug in Indiana; 1, 4, 8, 12, and 17 Aug in North Carolina. In 2017, fruit were harvested 11, 18, 25 Aug, 1, and 8 Sep in Indiana; 16, 23, 30 Aug, 8 Sep in North Carolina. Total yield was determined by combining all harvests.

# 2.3. Greenhouse trials

The three greenhouse experiments were conducted in the fall of 2017 at the Southwest Purdue Agricultural Center. While Serenade Opti and Regalia were alternated in the same treatment in field trials, in the greenhouse, these products were separated into two treatments. Likewise, the Sil-Matix alternated with Regalia treatment was separated into two treatments. All treatments were applied 24 h prior to inoculation. A virulent isolate of *Alternaria solani*, designated SWPAC 16–33, which was used in the greenhouse experiments was sequenced to confirm identity. Genomic DNA was extracted for amplification of the internal transcribed spacer (ITS) region of the ribosomal DNA (GenBank accession no. MK684179) using primers ITS 1 and ITS 4 (White et al., 1990) and sequenced. The resulting sequences were trimmed and aligned using Geneious<sup>®</sup> software v. 9.1.8. The consensus sequence produced a 99.82% similarity to GenBank accession CP022025 (Wolters et al., 2018).

Inoculum for greenhouse experiments was produced for the September 2017 trial using host extract agar from tomato (Koley and Mahaptra, 2015). However, subsequent experiments revealed that 2 L of a  $1 \times 10^3$  conidial suspension could be produced with 8–10 plates of potato dextrose agar (Difco Laboratoires, Sparks, MD) (data not shown); therefore, inoculum for the two October trials were produced using PDA. To make healthy tomato extract media, healthy, fully expanded tomato leaves were collected from greenhouse plants and dried using a food dehydrator for approximately 12 h. The leaves were then crushed to a powder and stored for future use at 4 C. To make host extract agar (HEA) media, 3.75 g of the powdered leaves were diluted in 250 ml RO water. The solution was then brought to a boil. After cooling, the suspension was filtered through cheesecloth and adjusted to 500 ml with RO water. 7.5 g of agar were added to the solution which was then autoclaved, cooled and poured into petri plates.

A single-spored isolate of *A. solani* was transferred to a fresh culture. The isolate was allowed to grow under 12-h lights at 22 C for 7–10 days. To stimulate the production of conidia, the cultures were then incubated in the dark for 24 h. Conidia were harvested from cultures using 10 mM MgSO<sub>4</sub> buffer and 1 ml Tween 80 in 250 ml of solution. Approximately 10 ml of buffer was used per plate. The conidial suspension was vortexed and filtered through cheesecloth. Conidia concentrations were adjusted using a hemocytometer to  $5 \times 10^3$  conidia per ml.

While field trials were conducted in an organically certified manner,

no attempt was made to certify the greenhouse trials. Tomato cv. Oregon Spring was used in all greenhouse experiments. Seeds were planted into Metro Mix 360 (Sungro, Agawam, MA) in polystyrene transplant trays for approximately five weeks, before being transplanted into 250 cc polystyrene pots in Metro Mix 360. After an additional two weeks of growth, plants were treated with products or controls as described and challenged with Alternaria solani. After inoculation, plants were stored in clear plastic containers for approximately 36 h to maintain leaf wetness. After the incubation period, plants were moved to a greenhouse where plants were rated for disease using the Horsfall-Barratt rating scale or other methods as described. Each plant was an experimental unit and each was rated in its entirety. The first experiment (Fig. 1) was rated 7 days post inoculation; the second experiment was rated 4- and 7days post inoculation (Fig. 2); the third experiment (Fig. 3) was rated 3 and 6 days post inoculation. An individual plant was an experimental unit and each trial was arranged in a completely randomized design, with four replications per treatment.

### 2.4. Statistical analysis

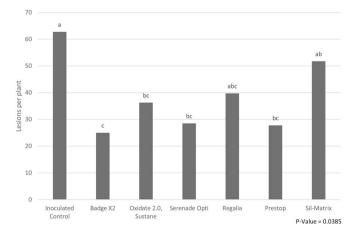
Statistical analysis of data from all disease and plant growth parameters was conducted by analysis of variance using general linear models procedure and Fisher's least significant difference test for mean separation (SAS version 9.1; SAS Institute, Cary, NC).

#### 3. Results

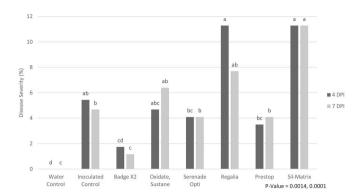
#### 3.1. 2016 field results

Early blight, the primary disease observed in Indiana in 2016, first appeared in the plots on 25 July (Table 2). Septoria leaf spot was also observed in Indiana in 2016 and accounted for less than 10 percent of the symptoms. In North Carolina, early blight was first observed in the plots on 12 July; Septoria leaf spot was first observed in North Carolina on 12 Aug (Table 3).

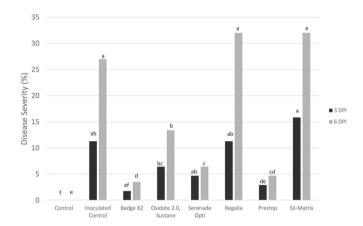
In 2016 in Indiana, plants treated with Badge X2 and Prestop had significantly reduced AUDPC values. The final disease rating for plants treated with Badge X2 and Prestop also had significantly reduced percent disease compared to untreated control plants; however, only the plants treated with Badge X2 had a significantly reduced final disease rating compared to plants treated with Prestop. In addition, plants treated with the Serenade Opti and Fracture alternation had a final rating significantly reduced compared to the untreated control plants.



**Fig. 1.** A trial conducted in the greenhouse of products for the management of early blight of tomato. Treatments were applied 24 h before inoculation with *Alternaria solani*. An experimental unit was one tomato plant in a pot and was replicated 4 times. Lesions were counted 7 Days Post Inoculation (DPI). Means within each column with a letter in common are not significantly different (Fisher's Protected LSD), P = 0.05.



**Fig. 2.** A trial conducted in the greenhouse of products for the management of early blight of tomato. Treatments were applied 24 h before inoculation with *Alternaria solani*. An experimental unit was one tomato plant in a pot and was replicated 4 times. Disease severity was rated on 4 and 7 Days Post Inoculation (DPI) using the Horsfall-Barratt scale and converted to percent using the ELANCO tables. Means within each column with a letter in common are not significantly different (Fisher's Protected LSD), P = 0.05.



**Fig. 3.** A trial conducted in the greenhouse of products for the management of early blight of tomato. Treatments were applied 24 h before inoculation with *Alternaria solani*. An experimental unit was one tomato plant in a pot and was replicated 4 times. Disease severity was rated on the Day Post Inoculation (DPI) indicated using the Horsfall-Barratt scale and converted to percent using the ELANCO tables. Means within each column with a letter in common are not significantly different (Fisher's Protected LSD), P = 0.05.

In North Carolina in 2016, plants in any treatment had a significantly reduced AUDPC compared to the untreated control plants (Table 3). Disease severity over the season (AUDPC) was lower in plants treated with Prestop than the untreated control plants, plants in the Oxidate alternated with Sustane treatment and plants in the Serenade Opti alternated with Regalia treatment. Plants treated with Badge X2, Prestop and Sil-matrix alternated with Regalia had a significantly reduced final disease rating compared to the untreated control plants.

There were no significant differences in any yield parameter for Indiana or North Carolina in 2016 (Tables 2 and 3).

#### 3.2. 2017 field results

Bacterial spot was first observed in field plots in 2017 in Indiana on 9 August (Table 4). Early blight and Septoria leaf spot were both observed in the North Carolina plots in 2017. Early blight was first observed in North Carolina on 12 July and Septoria leaf spot on 5 July (Table 5).

In Indiana during 2017, the severity of bacterial spot on tomato plants, as measured by AUDPC, was significantly reduced by Badge X2 as compared to the untreated control plants (Table 4). An isolate of *Xanthomonas* spp. was isolated from an infected tomato fruit and was

#### Table 2

Efficacy of several organically certified products against early blight and Septoria leaf spot of tomato and yields in trials conducted at the Southwest Purdue Agricultural Center in Vincennes, Indiana, 2016.

		Disease Severity		Marketable Yield/ Hectare <sup>a</sup>	
Treatment, rate	Treatment dates <sup>b</sup>	AUDPC <sup>c</sup>	Final rating <sup>d</sup> (Percent)	Weight (lb)	Number
Untreated Control	NA	223.9 a <sup>e</sup>	43.0 a	11088	72894
Badge X2, 2.0 kg//ha	B, C, D, E, F, G, H, I	63.5 c	6.4 d	13256	90110
Oxidate 2.0 1:100 with water + Sustane 1% w/v	B, C, D, E, F, G, H, I	172.4 abc	22.5 abc	11610	80023
Prestop, 0.05% w/v	B, E, G	114.3 bc	15.8 c	10241	69666
Serenade Opti 1.5 L/ha Fracture, 3.3 L/ ha	C, E, G, I B, D, F, H	165.9 abc	18.8 bc	12479	78946
na Sil-Matrix, 0.24 L/plant, Fracture, 3.3 L/ ha	A, C, E, G, I B, D, F, H	261.0 a	37.5 ab	9449	65227
P-Value		0.0451	0.0009	0.1632	0.3026

<sup>a</sup> Marketable yields calculated from total of all harvest dates.

<sup>b</sup> Fungicide application dates were: A = 24 May; B = 16, C = 24 Jun; D = 1, E = 8, F = 15, G = 22 Jul; H = 5, I = 12 Aug. The first application date was for a Sil-Matrix application as a root drench only. Other products applied with a CO<sub>2</sub> backpack sprayer.

<sup>c</sup> Disease severity is recorded as Area Under the Disease Progress Curve (AUDPC) or percent disease on the final rating date. Horsfall-Barratt ratings were completed 25 Jul, 3 Aug, 11 Aug, 18 Aug in Indiana. Area under the disease progress curve (AUDPC) was calculated by trapezoidal integration. Disease severity is presented as combined early blight and Septoria leaf spot; the latter disease accounted for less than 10 percent of the symptoms.

<sup>d</sup> Disease severity is presented as observed on the final rating date. Data was converted from Horsfall-Barratt values to percent using the ELANCO tables.

 $^{\rm e}$  Means within each column with a letter in common are not significantly different (Fisher's Protected LSD), P = 0.05.

found to be sensitive to copper hydroxide at 50 ppm (data not shown). There were no significant differences in yield in this trial.

In the 2017 North Carolina trial, early blight and Septoria leaf spot were rated separately (Table 5). Plants treated with Badge X2, Oxidate alternated with Sustane and Serenade Opti alternated with Regalia had significantly reduced Septoria leaf spot AUDPC values compared to the untreated control plants. The AUDPC for plants in the Badge X2 treatment was significantly reduced compared to plants in any other treatment. For the 2017 North Carolina early blight data, plants treated with Badge X2, Prestop and Serenade Opti alternated with Regalia all had significantly reduced AUDPC compared to the untreated control plants. Plants treated with Badge X2 had a lower AUDPC than plants in any of the other treatments. However, there were no significant differences for yield data for the 2017 North Carolina trial.

#### 3.3. Greenhouse trials

In the greenhouse trial conducted during September 2017, plants in all treatments had significantly fewer lesions of early blight compared to the inoculated control plants except for Regalia and Sil-matrix treated plants (Fig. 1). Plants treated with Badge X2, Oxidate 2.0 alternated with Sustane, Serenade Opti and Prestop all had significantly fewer lesions per plant than the inoculated control plants. However, the lesions per plant did not differ significantly among the plants so treated. Plants treated with the Badge X2 had significantly reduced disease severity

#### Table 3

Efficacy of several organically certified products against early blight and Septoria leaf spot of tomato and yields in trials conducted at the Mountain Organic Research and Extension Unit, Waynesville, North Carolina, 2016. Disease severity is recorded as Area Under the Disease Progress Curve (AUDPC) or percent disease on the final rating date.

		Disease Severity		Marketable Yield/ Hectare <sup>a</sup>	
Treatment, rate	Treatment dates <sup>b</sup>	AUDPC <sup>c</sup>	Final rating <sup>d</sup> (Percent)	Weight (KG)	Number
Untreated Control	NA	428.6 a <sup>e</sup>	32.0 a	19627	108131
Badge X2, 2.0 kg/ha	В—І	178.4 bc	3.5 с	20837	113242
Oxidate 2.0 1:100 with water,	В—І	231.5 b	13.4 ab	17002	104903
Sustane 1% w/v Prestop, 0.05% w/v	B, E, G, I	49.0 c	11.3 b	21482	128842
Serenade Opti 1.5 L/ha, Regalia, 7 L/ ha	C, E, G, I B, D, F, H	216.8 b	13.4 ab	22058	136373
Sil-Matrix, 0.24 L/plant, Regalia, 7 L/ ha	A, C, E, G, I B, D, F, H	191.6 bc	7.7 bc	18845	112972
P-Value		0.0025	0.0079	0.9803	0.8980

<sup>a</sup> Marketable yields calculated from total of all harvest dates.

 $^{\rm b}$  Fungicide application dates were: A = 8, B = 15, C = 22, D = 28, Jun; E = 6, F = 12, G = 20, H = 26, Jul; I = 3 Aug. The first application date was for a Sil-Matrix application as a root drench only. Other products applied with a CO<sub>2</sub> backpack sprayer.

<sup>c</sup> Horsfall-Barratt ratings were conducted on 12, 18, 26 Jul, 1, 12, 17, 22, 25 Aug. Area under the disease progress curve (AUDPC) was calculated by trapezoidal integration.

<sup>d</sup> Disease severity is presented as observed on the final rating date. Data was converted from Horsfall-Barratt values to percent using the ELANCO tables.

 $^{\rm e}$  Means within each column with a letter in common are not significantly different (Fisher's Protected LSD), P=0.05.

compared to plants in the inoculated control and the Sil-matrix treatments.

In the two greenhouse trials conducted in October 2017, there were no symptoms on the water controls (Figs. 2 and 3). Plants treated with Badge X2 were the only plants that had significantly reduced percent disease severity compared to the inoculated control plants in the early October trial on both DPI (Fig. 2). Plants treated with Prestop on both DPI had reduced percent disease compared to Sil-Matrix treated plants. Plants treated with Badge X2 had significantly reduced percent disease on 3 DPI compared to all treatments except for the water and Prestop treated plants (Fig. 3). The other treatments that had significantly reduced percent disease compared to the inoculated control plants was Serenade Opti on 6 DPI and Oxidate alternated with Sustane on 6 DPI.

## 4. Discussion

Effectively managing diseases caused by foliar pathogens is the biggest challenge in organic tomato production (Hoagland et al., 2015). The primary goal of these studies was to provide an unbiased evaluation of products for organic disease management. The copper product, Badge X2, consistently provided the best disease control for the fungal diseases observed in field trials in Indiana and North Carolina in 2016, in North Carolina during 2017, and for bacterial spot management in Indiana in 2017. In addition, Badge X2 provided good control of early blight in the three greenhouse trials. Copper products have a long history of use in the management of plant diseases, dating back to the 1700's. More recently, copper products have become a standard for disease management in

#### Table 4

Efficacy and yield of several organically certified products against bacterial spot of tomato in trials conducted at the Southwest Purdue Agricultural Center in Vincennes, Indiana 2017.

					Yield/ hectare <sup>a</sup>
					kg
Treatment, Rate	Treatment Dates <sup>b</sup>	AUDPC <sup>c</sup>	Final Date <sup>d</sup>	Red Fruit	Bacterial Spot
Untreated Control	N/A	696.7 ab <sup>e</sup>	43.0 a	23,360	8608
Badge X2, 2.0 kg/ha	B-M	208.9 c	11.3 b	33,558	4483
Oxidate 2.0, 1:100 + Sustane, 1% w/v	B-M	838.2 ab	49.0 a	36,046	4259
Prestop, 0.05% w/v	В, Е, Н	575.3 b	27.0 a	28,559	8070
Serenade Opti, 20 oz/A Regalia, 7 L/ha	C, E, G, I, K, M B, D, F, H, J, L	752.0 ab	32.0 a	33,580	5693
Sil-Matrix, 0.24 L/plant Regalia, 7 L/ha	L A, C, E, G, J, L B, D, F, H, I, K, M	924.6 a	49.0 a	22,887	4976
P-Value	-	0.0073	0.0396	0.0999	0.1905

<sup>a</sup> Marketable yields calculated from total of all harvest dates.

<sup>b</sup> Fungicide application dates were: A = 31 May; B = 16, C = 24 Jun, D = 1, E = 8, F = 15, G = 22, H = 26 Jul, I = 1, J = 8, K = 15, M = 22, N = 30 Aug. The first application date was for a Sil-Matrix application as a root drench only. Other products applied with a  $CO_2$  backpack sprayer.

<sup>c</sup> Horsfall-Barratt ratings were conducted on 9, 17, 24 Aug and 4 Sep. Area under the disease progress curve (AUDPC) was calculated by trapezoidal integration.

<sup>d</sup> Percent disease on final rating date.

 $^{e}$  Means within each column with a letter in common are not significantly different (Fisher's Protected LSD), P = 0.05.

organic agriculture. However, alternative disease control approaches are needed, as repeated applications of products containing copper can negatively affect soil and water quality, and some pathogens have evolved resistance to copper. Moreover, excessive applications can be phytotoxic to plants (Dias, 2012). Newer formulations contain copper compounds that are more readily available on the leaf surface than older fixed copper products, and combine soluble and insoluble compounds to improve disease control efficacy and prevent negative potential side effects. Badge X2, used here as a standard for comparison against alternative organic products, is one such product, which combines copper hydroxide and copper oxychloride.

Prestop (Gliocladium catenulatum J1446) was the most effective treatment in controlling foliar disease in tomato of all the alternatives to copper products evaluated in this study. Prestop was effective in lowering early blight disease severity in all field trials where this disease was prominent. This included Indiana in 2016 and in North Carolina in both 2016 and 2017. Prestop did not reduce the severity of other diseases in the field (Septoria leaf spot or bacterial spot), which is expected as no diseases caused by Septoria species or any bacteria are listed on the Prestop label. Gliocladium. catenulatum J1446, the active ingredient in Prestop, has been shown to be a hyperparasite of fungi via appressoriumlike structures formed by G. catenulatum and production of B-1,3-glucanase and chitinase (Chatterton and Punja, 2008). It is not clear why Septoria lycopercisi, causal agent of Septoria leaf spot of tomato, would not be parasitized in the same way as A. solani. Consistent with the field trials, Prestop reduced disease severity of early blight in the greenhouse trials conducted in September and late October when compared to the inoculated control (Figs. 1 and 3). In the greenhouse trial where Prestop did not significantly lower disease severity compared to the inoculated control, Prestop was not significantly different than Badge X2 (Fig. 2). It is possible that fewer differences were noted in the later trial due to overall reduced levels of disease. The percent disease in the inoculated control in Fig. 2, five percent, is much less than the level of disease for the inoculated control in Figs. 3 and 25 percent.

The similar performance of the product Prestop in the field and the greenhouse for early blight suggests that the greenhouse trials, although not organically certified, may be used to simulate or perhaps refine

#### Table 5

Disease severity and yield of tomatoes treated with a combination of several organically certified products against early blight and Septoria leaf spot of tomato in trials conducted at the Mountain Organic Research and Extension Unit, Waynesville, North Carolina, 2017.

		AUDPC <sup>a</sup>	Final rating Date <sup>b</sup> AUDPC		Final rating date	Marketable Yield <sup>c</sup>
Treatment, rate	Treatment Dates <sup>d</sup>	Septoria leaf spot	Septoria leaf spot	Early blight	Early blight	kg/ha
Untreated Control	NA	2903.2 a <sup>e</sup>	2903.2 a	569.5 a	49.0 a	40,020
Badge X2, 2.0 kg/ha	B-M	1290.0 c	1290.0 c	149.4 c	11.3 c	47,210
Oxidate 2.0 1:100 +	B-M	2416.9 b	2416.9 b	446.2 ab	27.0 b	37,839
Sustane 1% w/v Prestop,	B, D, F, H, J, L	2609.9 ab	2609.9 ab	403.2 b	27.0 b	37,705
0.05% w/v Serenade Opti	C, E, G, I, K, M	2446.6 b	2446.6 b	350.9 b	22.5 b	43,242
20 oz/A Regalia <sup>a</sup> ,	B, D, F, H, J, L	2440.00	2440.00	330.9 0	22.30	73,272
7 L/ha Sil-Matrix, 0.24 L/plant/plant	C, E, G, I, K, M B, D, F, H, J, L	2567.9 ab	2567.9 ab	425.5 ab	27.0 b	47,950
Regalia, 7 L/ha P-Value		<.0001	<.0001	0.0019	0.0019	0.2507

<sup>a</sup> Area under the disease progress curve (AUDPC) was calculated by trapezoidal integration.

<sup>b</sup> Percent disease on final rating date.

<sup>c</sup> Marketable yields calculated from total of all harvest dates.

<sup>d</sup> Fungicide application dates were: A = 23, B = 26 May; C = 2, D = 9, E = 16, F = 23, G = 30, Jun; H = 7, I = 14, J = 21, K = 28, Jul; L = 4, M = 11 Aug. The first dates is for Sil-Matrix application as a root drench at transplant only. Other products applied with a CO<sub>2</sub> backpack sprayer.

<sup>e</sup> Means within each column with a letter in common are not significantly different (Fisher's Protected LSD), P = 0.05.

results of field trials. In contrast, Serenade Opti was more consistent in controlling early blight in greenhouse trials than in the field. Perhaps this result is due to the alternation of Serenade Opti in the field with other ineffective products whereas in greenhouse trials, this product was compared directly to other products without alternation. We can't rule out the possibility that the non-organically certified greenhouse trials reacted differently than the organically certified field trials. However, the major non-organic input in the greenhouse trials was the conventional soilless mix which seems unlikely to have caused any differences.

Rather than directly suppressing pathogens on contact, an alternative approach for mitigating damage by foliar diseases is to stimulate a plant's immune system via ISR. This phenomenon was first defined by van Loon in 1998, as a state of increased defensive capacity developed by a plant when appropriately stimulated through the activity of latent resistance mechanisms, and induced by diverse agents including beneficial bacteria and fungi. The protection conferred by ISR is generally non-specific, and once the plant's natural defense mechanisms are activated, increased defensive capacity can be maintained for prolonged periods against multiple pathogens (Pieterse et al., 2014). Many commercial products are purported to stimulate ISR-like defense responses in plants including Serenade Opti (Lahlali et al., 2013) and Regalia (Daayf et al., 1997). However, while microbially-mediated induction of systemic resistance in tomato against foliar pathogens including early blight, late blight and Septoria leaf spot has been demonstrated in multiple studies under greenhouse conditions (Fritz et al., 2006), as with many biocontrol products, efficacy in field trials is variable. Silicon has also been reported to induce an ISR like response in plants against pathogens (Cai, 2009).

One potential way to increase the efficacy of biocontrol products could be to combine products with different modes of action. For example, such an approach has previously been demonstrated to enhance the biological suppression of *Botrytis cinerea* on strawberry leaves (Guetsky et al., 2001), and the control of late blight in tomato (Lourenco Junior et al., 2006). In fact, because it has recently become clear that different microbial taxa can act together in a consortium, much research is currently underway to develop products with an aim to mix microbial taxa, botanicals from extracted from different plants and inorganic products to help enhance the efficacy of pathogen biocontrol (Thakkar and Saraf, 2015; Sarma et al., 2015). In our field trials, we used in alternation a botanical product, Regalia, with a biological product, Serenade Opti, and, in another treatment, we alternated Regalia with an inorganic product, Sil-Matrix.

Results of our study indicate that the efficacy of combining commercial products derived from different sources and potential modes of action depends on the pathogen and field location. For example, plants treated with an alternation of Regalia with Serenade-Opti had AUDPC values significantly reduced compared to the untreated control plants when fungal diseases were the major pathogens involved (Tables 3 and 5). However, the Serenade Opti and Regalia alternation was not effective against bacterial spot (Table 4). Regalia also failed to lower disease severity of a mixed bacterial spot and bacterial speck infection when applied alone (Trueman, 2015). Although Serenade Opti is labeled for management of bacterial spot, trials of this product have produced inconsistent results (Abbasi and Weselowski, 2015; Roberts et al., 2008). When separated in greenhouse trials, we also observed variability in the efficacy of Serenade Opti, which was effective against early blight in two of three greenhouse experiments. Abbasi and Weselowski (2014), observed that Serenade Opti was inconsistent in lowering early blight of tomato disease levels below the untreated control in two out of three years. Given the results presented here, additional trials evaluating Serenade-Opti, alone and in combination with other products for the control of early blight should be completed to better understand factors contributing to this variability.

Plants treated with Sil-matrix alternated with Regalia was one of a number of treatments that had a significantly reduced level of disease compared to untreated control plants in the North Carolina field trial in 2016. However, the Sil-matrix alternated with Regalia treatment was not effective compared to the untreated control in any other field trial. When Serenade Opti, Regalia and Sil-Matrix treatments were trialed independently from each other in the greenhouse as opposed to alternation treatments in field trials, neither the Regalia nor the Sil-matrix treatments had significantly different disease levels compared to the inoculated control. The performance of Serenade Opti versus Sil-Matrix in field and greenhouse trials, leads one to conclude that Serenade Opti was a more effective alternation partner compared with Sil-Matrix. To our knowledge, this is the first published account of a silicon product used to manage early blight, Septoria leaf spot or bacterial spot. While some studies have observed disease suppression by silicon, results are often variable. For example, silicon was tested against Botrytis gray mold of tomato in a hydroponic system and also had little effect (Pozo et al., 2015).

In 2016 in Indiana only, we compared Fracture, a polypeptide derived from germinating *Lupinus albus* plants, which is purported to inhibit chitin synthesis in fungi. Since this product was not certified in 2017 and therefore not trialed, the results were not replicated and no conclusions can be drawn. It is interesting, however, that regardless of whether Serenade Opti or Sil-Matrix was alternated with Fracture or Regalia, there was no significant difference in disease control between these treatments over four field trials (Tables, 2, 3, 4 and 5).

The hypothesis behind the Oxidate/Sustane treatment was to apply Oxidate (active ingredient hydrogen dioxide) to lower the population of microbes on the surface of the foliage, followed by application of the Sustane product, a turkey manure compost tea, in an effort to increase colonization by beneficial microbes on the leaf surface and thereby inhibit pathogen growth. The order of microbial species arrival, or socalled 'priority effects', has previously been shown to result in large differences in community structure (Braun-Kiewnick et al., 2000; Toju et al., 2018). Early colonizers have an advantage because they can occupy space and use resources earlier, produce physical barriers such as biofilms, or produce antibiotics that slow colonization of subsequent microbial taxa (Fukami, 2015; Toju et al., 2018). In this study, the combined treatment was effective in lowering disease levels compared to the untreated control against early blight in North Carolina in 2016 and Septoria leaf spot in North Carolina in 2017. In addition, this treatment was effective in significantly lowering early blight disease levels compared to the inoculated control in two out of three greenhouse experiments. However, it was not effective during the field trials conducted in Indiana. Thus, the effectiveness of this treatment has been inconsistent. We did not evaluate these two products alone in either the field or greenhouse trial due to insufficient space, so we cannot judge if one product was providing more efficacy over another. For example, the hydrogen dioxide portion of the treatment (Oxidate) is commonly used alone in field environments, and this treatment combination might have been more effective if used multiple times per week since the product does not provide residual control. However, such an approach would disrupt the potential for beneficial microbes in the compost tea treatment to colonize the leaf and prevent pathogen growth. While the efficacy of compost tea to suppress foliar diseases has been variable in other trials (Evans and Percy, 2014), results of these trials along with testimonials from growers indicate that this approach deserves further study.

Results of this study confirm that copper products such as Badge X2 can effectively control foliar pathogens in tomato. However, growers should consider using copper products sparingly if possible to protect soil and water quality, and rotate with crops that are not susceptible to tomato foliar pathogens and thus do not require organic fungicide applications to reduce overall use. *Gliocladium catenulatum* J1446 appears to be an acceptable alternative to copper products as a management option for early blight of tomato. Applied to foliage, this product (Prestop) consistently lowered the disease level of early blight to the level provided by a copper product. This product, however, did not lower disease levels of bacterial spot or Septoria leaf spot of tomato. Additional field or greenhouse work will be required to determine what,

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if any, products may be used with *G. catenulatum* J1446 to manage other common diseases of tomato such as bacterial spot and Septoria leaf spot.

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