



Acibenzolar-S-methyl is associated with yield reduction when used for managing bacterial wilt (*Erwinia tracheiphila*) in cantaloupe



Daniel S. Egel^{a,*}, Nathan M. Kleczewski^b, Fawzia Mumtaz^c, Rick Foster^d

^a Department of Botany and Plant Pathology, Purdue University, 915 West State Street, West Lafayette, IN 47907 United States

^b Department of Crop Sciences, University of Illinois, 1101 West Peabody Drive, Urbana, IL 61801 United States

^c Kabul University, Ministry of Agriculture, 0744121679 Afghanistan

^d Department of Entomology, Purdue University, 901 West State Street, West Lafayette, IN 47907 United States

ARTICLE INFO

Keywords:

Systemic acquired resistance
Induced systemic resistance
Cucumber beetle
Salicylic acid
Muskmelon

ABSTRACT

Bacterial wilt, caused by *Erwinia tracheiphila* (Smith), can result in serious yield loss of cantaloupe in the Midwest United States. Although neonicotinoid insecticides are an effective means of managing the cucumber beetles that vector *E. tracheiphila*, this management regime may cause toxicity to pollinators. An alternative method of managing this disease is to use products that induce resistance. Over four years, we have compared disease severity of bacterial wilt, cucumber beetle numbers, feeding damage and yield as influenced by products purported to induce Systemic Acquired Resistance (SAR) or Induced Systemic Resistance (ISR). Acibenzolar-S-methyl (Actigard[®]) effectively lowered disease severity of bacterial wilt in years when disease pressure was moderate or high. Actigard[®] was also associated with lower levels of cucumber beetle feeding in some years. However, when disease pressure was low, Actigard[®] at some rates and timings, caused yield loss. The other products, Regalia[®] and Serenade Max[®], are purported to elicit ISR and did not consistently lower disease levels or influence beetle feeding in the current study. The standard insecticide regime, imidacloprid used as a drench at planting and permethrin applications at weeks 3–6, was the most consistent method of lowering beetle feeding and thus managing bacterial wilt. Since Actigard[®] could be associated with yield loss, its use for the management of bacterial wilt in cantaloupe cannot be recommended.

1. Introduction

Bacterial wilt of cucurbits is a serious disease of cantaloupe in the Midwest United States (Brust and Rane, 1995). Affected plants wilt, lose vigor and may die. Yield losses, which may represent 20 percent of the crop (Brust and Foster, 1999), are caused when vines die before fruits mature. The disease is caused by the bacterium *Erwinia tracheiphila* (Smith) and is transmitted by the striped cucumber beetle (*Acalymma vittatum* (F.)) and the spotted cucumber beetle (*Diabrotica undecimpunctata howardi* Barber) (Saalau Rojas et al., 2015). The beetles carry the bacterium within their gut. After making feeding wounds on the plant, they defecate into the wound and, when free water is present, the bacteria enter the vascular system of the plant. The transmission process may take several hours of feeding and defecation. Once inside the vascular system of the plant, the bacterium multiplies, a process which causes blockage of the xylem (Watterson et al., 1971). In addition, the bacterium may move within the xylem and cause wilting in areas distal to the inoculation site (Watterson et al., 1972). Fruit on vines which are affected by bacterial wilt may not reach physiological

maturity or have reduced fruit quality which lowers marketable yields.

Management of bacterial wilt has focused on controlling the vector through the repeated use of insecticides, such as pyrethroids and carbamates. These insecticides rapidly kill the cucumber beetles, preventing transmission of the bacterium among plants (Foster et al., 2005). The use of insect thresholds will reduce the number of insecticide applications required and can result in higher yields than when weekly insecticide applications are made (Brust and Foster, 1999). Carbamates and the pyrethroid insecticides are toxic to beneficial insects and repeated use can result in outbreaks of secondary pests such as aphids.

More recently, neonicotinoid insecticides have been incorporated into pest management programs. These insecticides have systemic activity, which allows them to be used as seed treatments, in bedding tray treatments, in the transplant water at planting, through the irrigation system, or as foliar sprays. Neonicotinoids move through the plants and provide excellent control of cucumber beetles for up to 21 days with reduced impacts on beneficial insects compared to foliar applications of pyrethroids. Pollinator insects such as honeybees and

* Corresponding author.

E-mail address: egel@purdue.edu (D.S. Egel).

bumblebees, however, feed on pollen and nectar and may be harmed from exposure to neonicotinoid insecticides (Foster et al., 2016).

Alternative management options for bacterial wilt include the use of row covers and trap crops. Row covers have been used to physically prevent cucumber beetles from feeding on cantaloupe. Opening row cover ends at anthesis and removal 10 days later enhanced yields when bacterial wilt pressure was significant (Saalau Rojas et al., 2011). However, row covers increased production costs by 45 percent, which is an unacceptable cost when disease pressure was low. Trap crops may be used to lure insects to treated trap plants so that the amount of insecticide applied to the crop may be reduced. Blue Hubbard squash has been used to attract cucumber beetles away from a watermelon field to manage feeding damage (Pair, 1997).

Another strategy to manage plant diseases is to use compounds that elicit induced resistance in the plant host. Induced resistance is a physiological state whereby the plant has an enhanced defensive capacity (van Loon et al., 1998). There are two types of induced resistance: Systemic Acquired Resistance (SAR) and Induced Systemic Resistance (ISR). SAR is a form of host resistance usually induced by exposure of the roots or foliage of the plant to abiotic or biotic stresses and involves the elicitor salicylic acid (Vallad and Goodman, 2004) or methyl salicylate (Park et al., 2007). SAR may develop in part of the plant well away from the area infected with a pathogen (Oostendorp et al., 2001). SAR has been shown to lessen the severity of plant diseases such as rice blast (Oostendorp et al., 2001; Vallad and Goodman, 2004) and/or the impact of insect feeding such as the common pine sawfly (Eyles et al., 2010). The application of acibenzolar-S-methyl, trade name Actigard[®], increased the levels of β -glucanase and peroxidase activity and reduced the severity of cucurbit anthracnose (Neher et al., 2009). The use of products that elicit SAR has the potential to lessen disease without any off-target effect on pollinators or beneficial insects (Vallad and Goodman, 2004).

Insect feeding and insect transmitted diseases may also be affected by the SAR pathway. Tomato spotted wilt severity was reduced by a combination of the application of Actigard[®] and reflective mulch. However, the populations of the thrips vector was not affected (Riley et al., 2012). The use of Actigard[®] on *Cucumis melo* increased the expression of pathogenesis-related 1a gene, which is associated with SAR, and decreased symptoms of *Cucurbit chlorotic yellows virus* (CCYV; Takeshita et al., 2013). CCYV is transmitted by white flies; however, this study did not find any effect on the whitefly vector. A salicylic acid analog, which induces SAR, reduced populations of the potato aphid on tomato (Cooper et al., 2004). Similarly, a salicylic acid analog reduced the weight gain of the larvae of *Spodoptera littoralis* on tomato (Sobhy et al., 2015).

Induced Systemic Resistance (ISR) is often elicited in response to specific strains of growth-promoting bacteria (Vallad and Goodman, 2004). A potting mix prepared with stabilized compost containing *Bacillus* sp. strains reduced the severity of bacterial leaf spot of radish (Krause et al., 2003). Strain QST 713 of *Bacillus subtilis*, the active ingredient of the product Serenade, was used to reduce the severity of clubroot on canola (Lahlali et al., 2013). A filtrate of the product, made with a 0.22- μ m membrane filter to exclude bacteria was not as effective as the product. Genes associated with jasmonic acid, ethylene and phenylpropanoid were upregulated with the use of Serenade, indicating that the ISR pathway might be involved (Lahlali et al., 2013). The application of rhizobacteria has been implicated in the reduction of cucumber beetle numbers and bacterial wilt severity in cucumbers via ISR (Zehnder et al., 2001). However, it is not known if SAR as elicited by the product Actigard[®] can also reduce cucumber beetle feeding or bacterial wilt.

The products trialed in this study for the management of bacterial wilt of cantaloupe are described here. Actigard induces systemic acquired resistance by causing an increase in methyl salicylate (Huang et al., 2012). Serenade[®] (*Bacillus subtilis* strain QST 713) has been shown to elicit ISR in canola (Lahlali et al., 2013). Regalia[®] (extract of

Reynoutria sachalinensis) induces a localized but translaminar resistance (Dayan et al., 2009). The product Copron-F[®] (a.i., copper and sulfur) is not known to produce SAR or ISR. Since metal hyper-accumulator plants tend to have less herbivory and insect feeding (Poschenrieder et al., 2006), it was thought that the copper in Copron-F[®] could help plants fend off insect feeding and thus bacterial wilt. These compounds were compared to a standard insecticide treatment and an untreated control in an attempt to determine if SAR or ISR inducing compounds could become a sustainable alternative to insecticide treatments in the management of bacterial wilt of cantaloupe.

The objective of this study was to determine if Actigard[®], which is known to elicit SAR, is a viable option for sustainable management of bacterial wilt in cantaloupe. Products purported to elicit ISR were compared to Actigard[®] in the field over four years. The products were compared to an untreated check, a standard insecticide treatment and a foliar nutrient application product not purported to elicit induced resistance.

2. Materials and methods

2.1. Field plot details

Experiments were conducted at the Southwest Purdue Agricultural Center (SWPAC) near Vincennes, IN from 2011 to 2013 and at the Throckmorton Purdue Agricultural Center (TPAC) near Lafayette, IN in 2014.

Experiments conducted at SWPAC used the cantaloupe variety Athena spaced 61 cm between plants and 1.8 m between rows. In all studies, the experimental unit was a row of 20 plants. However, only the interior 10 plants were used as treatment plants. The date of field transplanting was May 9, 2011, May 11, 2012, May 15, 2013. Each plot was on 1.2-m-wide beds with 0.8 mil black plastic mulch (Visqueen 4020) with drip irrigation. The experimental design was a randomized complete block with four replications.

In 2014 the experiment conducted at TPAC also used Athena and rows 6.1 m long with 61 cm between plants and 2.4 m between rows. The date of field transplanting was 28 May. Each experimental unit contained 10 plants spaced 61 cm between plants out of a total of 20 plants per row. Cultural practices and the experimental design were the same as those used at SWPAC.

2.2. Treatments

In this study, several commercially available compounds that have been purported to elicit SAR or ISR in host plants were tested (Table 1). The insecticide standard consisted of an application of Admire Pro[®] at 110 ml per ha (active ingredient, imidacloprid) drench at transplanting followed by Pounce 25 WP[®] at 896 g per ha (active ingredient, permethrin) applied weekly from week three to week six.

The trials in 2011, 2012 and 2013 indicated a possible yield reduction with the product Actigard[®]. Therefore, the 2014 trial was conducted to see if bacterial wilt and cucumber beetle feeding could be managed by varying rates and application sequence of Actigard[®] to overcome yield loss. In 2014, Actigard[®] was applied at two additional sequences.

A CO₂ pressurized backpack sprayer (R&D sprayer, Opelousas, LA) was used to apply pesticides at 414 kPa and 374 L/ha. The hand-held boom of the sprayer had four Teejet 8002VS nozzles. Each treatment was mixed in 3-L soda bottles that could be easily changed between treatments with a boom flush of water between applications.

2.3. Assessment

Disease severity was rated visually using the Horsfall-Barratt rating scale (Horsfall and Barratt, 1945). Leaves were observed for wilting; stems were observed for loss of turgor and collapse. The Area Under the

Table 1
Rate and timing of the products applied.

Trade name	Active ingredient	Rate (s)	Applications	Years
Actigard [®]	acibenzolar-S-methyl 50%	35 g/ha	Weeks 0–5	2011, 2012, 2013, 2014
Actigard [®]	acibenzolar-S-methyl 50%	35 g/ha	Weeks 0, 1 and 2	2014
Actigard [®]	acibenzolar-S-methyl 50%	35 g/ha	Weeks 0, 2 and 4	2014
Actigard [®]	acibenzolar-S-methyl 50%	70 g/ha	Weeks 0–5	2011, 2012, 2013, 2014
Actigard [®]	acibenzolar-S-methyl 50%	70 g/ha	Weeks 0, 1 and 2	2014
Actigard [®]	acibenzolar-S-methyl 50%	70 g/ha	Weeks 0, 2 and 4	2014
Actigard [®]	acibenzolar-S-methyl 50%	140 g/ha	Weeks 0–5	2012, 2013
Copron-F [®]	Sulfur 2.5%	4.7 L/ha	Weeks 0–5	2011, 2012
	Copper 5%			
	Derived from copper sulfate			
Regalia [®]	Extract of <i>Reynoutria sachalinensis</i> 5%	9.4 L/ha	Weeks 0–5	2011, 2012, 2013
Regalia [®]	Extract of <i>Reynoutria sachalinensis</i> 5%	2.8 L/378 L	Drench at planting	2013
Serenade Max [®]	QST 713 strain of <i>Bacillus subtilis</i> 14.6%	3.36 kg/ha	Weeks 0–5	2011, 2012, 2013
Admire Pro [®]	imidacloprid 42.8%	110 ml/ha	Drench at planting	2011, 2012, 2013, 2014
Pounce 25 WP [®]	permethrin 25%	896 g/ha	Weeks 3–6	

Disease Progress Curve (AUDPC) was determined by trapezoidal integration (Shaner and Finney, 1977). Statistical analysis of differences between means was determined by analysis of variance using general linear models procedure and Fisher's least significant differences test for mean separation (SAS version 9.1; SAS Institute, Cary, NC). Wilt severity data were arcsine square root transformed prior to analysis.

Vine growth was measured in 2011 by selecting two vines per experimental unit at random for measurement, which were marked so they could be measured at subsequent dates. In 2012, 2013, vines were selected randomly each time. The mean of the two measurements per experimental unit are used in the analysis.

Cucumber beetle feeding damage was estimated on each treatment plant in 2011, 2012 and 2013 by estimating the amount of damage using the Horsfall-Barratt rating scale. In 2013 only, the incidence of beetle feeding on cucumbers flowers was assessed. Cucumber beetle numbers were quantified by counting the number of live and dead beetles on the plant as well as on the ground directly underneath the plant in 2013 and 2014. Cucumber beetle numbers are presented per experimental unit (one row).

Harvest was conducted three times a week. Only fruit that were physiologically mature were harvested (full-slip). Fruit was weighed individually only from treatment plants.

3. Results

3.1. 2011 results

In 2011, a larger than expected influx of striped cucumber beetles and bacterial wilt were observed (Table 2). The standard insecticide treatment significantly reduced the amount of beetle damage. For example, on 7 June, the insecticide standard significantly reduced the amount of beetle feeding compared to any other treatment. Actigard[®] at 35 g and 70 g per ha and Regalia[®] also lowered beetle feeding compared to the untreated control. Similarly, bacterial wilt severity, as measured by AUDPC, was significantly less for the insecticide standard compared to all treatments except Serenade Max[®] and Actigard[®] at 70 g per ha (Table 2). The untreated control and Regalia[®] had significantly higher AUDPC than any other treatment. Vine growth was significantly greater for the insecticide standard than all other treatments on June 1 and 8 (Table 2). Actigard[®] at 70 g per ha had significantly less vine growth than any other treatment on June 1 and on June 8 was significantly less than any other treatment except for Regalia[®]. Despite differences in beetle and disease damage, yield was not significantly impacted by any of the treatments in 2011 (Table 3).

Table 2

Vine length, bacterial wilt severity and percentage of beetle feeding observed on Athena cantaloupe treated with one of the induced resistance products, standard insecticide treatment or left untreated. The experiment was conducted at the Southwest Purdue Agriculture Center in Vincennes, IN in 2011.

Treatments, rate per ha	Vine length (cm)		Bacterial wilt severity	% Beetle Feeding Damage ^y
	1 Jun	8 Jun	AUDPC ^z	7 Jun
Untreated check	25.5 b ^x	62.4 b	500.5 a ^x	3.3 a
Actigard [®] , 35 g	25.8 b	63.0 b	285.7 b	2.9 b
Actigard [®] , 70 g	20.5 c	53.1 c	130.1 bc	2.7 b
Serenade Max [®] , 3.36 Kg	24.4 b	61.4 b	154.3 bc	3.0 ab
Regalia [®] , 9.4 L	24.3 b	60.2 bc	583.2 a ^y	2.9 b
Copron-F [®] , 4.7 L	27.2 b	68.1 b	294.5 b	3.4 ab
Insecticide standard ^w	31.8 a	86.5 a	66.1 c	1.8 c
P-value	0.0003	0.0001	0.0001	0.0021

^wAdmire Pro[®] at transplant and Pounce WP[®] applied weekly starting at 3 weeks post-transplant.

^xMeans in columns followed by the same letter are not significantly different ($P = 0.05$) based on Fisher's protected LSD test.

^yBeetle feeding was estimated using the Horsfall-Barratt scale on the date indicated.

^zHorsfall-Barratt ratings were conducted 19 May, 31 May, 17 Jun, 22 Jun. Area under the disease progress curve (AUDPC) was calculated by trapezoidal integration.

Table 3

Yields of Athena cantaloupe treated with one of the induced resistance products, standard insecticide treatment or left untreated treated for bacterial wilt at the Southwest Purdue Agricultural Center Vincennes, IN 2011.

Treatment, rate per ha	Marketable fruit number (1000/ha)	Marketable fruit weight (metric tons/ha)	Mean weight marketable (kg)
Untreated check	5.38	16.15	2.99
Actigard [®] , 35 g	5.82	16.61	2.86
Actigard [®] , 70 g	2.68	7.68	2.86
Serenade Max [®] , 3.36 Kg	4.26	12.14	3.03
Regalia [®] , 9.4 L	4.93	14.35	2.90
Copron-F [®] , 4.7 L	5.60	16.68	2.95
Insecticide standard ^a	6.95	18.58	2.58
P-value	0.6220	0.6728	0.3529

^a Admire Pro[®] at transplant and Pounce WP[®] applied weekly starting at 3 weeks post-transplant.

Table 4

Vine length, beetle feeding and bacterial wilt severity as influenced by one of the induced resistance products, standard insecticide treatment or left untreated at the Southwest Purdue Agricultural Center in Vincennes, IN 2012.

Treatment, rate per ha	Vine length (cm)		% Beetle Feeding Damage ^z			Bacterial wilt severity
	30 May	5 Jun	22 May	29 May	5 Jun	
Untreated check	27.9	50.8	4.7 a ^y	11.3 a	9.4 a	0.6
Actigard [®] , 35 g	27.4	50.2	4.7 a	7.7 ab	5.4 bc	0.6
Actigard [®] , 70 g	27.9	47.0	3.3 ab	4.7 b	4.1 c	0.6
Actigard [®] , 140 g	27.8	49.8	0.6 b	4.7 b	4.7 bc	1.2
Serenade Max [®] , .36 Kg	28.3	44.8	2.9 ab	6.4 ab	6.4 ab	0.6
Regalia [®] , 9.4 L	29.5	46.0	2.9 ab	4.7 b	6.4 ab	1.2
Copron-F [®] , 4.7 L	28.3	44.5	5.4 a	9.4 a	5.4 bc	2.3
Insecticide standard ^x	32.5	56.6	0.6 b	2.3 c	0.5 d	0
P-value	0.8519	0.0873	0.0235	0.0030	0.0001	0.1838

^xAdmire Pro at transplant and Pounce WP[®] applied weekly starting at 3 weeks post-transplant.

^yMeans in columns followed by the same letter are not significantly different ($P = 0.05$) based on Fisher's protected LSD test.

^zBeetle feeding was estimated using the Horsfall-Barratt scale on the date indicated.

Table 5

Yields of Athena cantaloupe treated with one of the induced resistance products, standard insecticide treatment or left untreated treated for bacterial wilt at the Southwest Purdue Agricultural Center Vincennes, IN, 2012

Treatment per ha	Mean wt. (kg)	Total wt. (metric tons/ha)	Total no. (1000/ha)
Untreated control	2.08 bcd ^z	36.76 ab	17.70
Actigard [®] , 35 g	2.11 bcd	29.15 bc	13.89
Actigard [®] , 70 g	1.99 cd	27.37 bc	13.67
Actigard [®] , 140 g	1.96 d	23.64 c	12.10
Serenade Max [®] , 3.36 Kg	2.10 bcd	38.77 ab	18.60
Regalia [®] , 9.4 L	2.13 bc	32.92 bc	15.46
Copron-F [®] , 4.7 L	2.20 ab	33.07 bc	15.01
Insecticide standard ^y	2.33 a	47.07 a	20.17
P-value	0.0027	0.0167	0.0772

^yAdmire Pro[®] at transplant and Pounce WP[®] applied weekly starting at 3 weeks post-transplant.

^zMeans in columns followed by the same letter are not significantly different ($P = 0.05$) based on Fisher's protected LSD test.

3.2. 2012 results

Although a large number of beetles were again observed in 2012, little bacterial wilt was observed. On June 5, the insecticide standard was associated with significantly less beetle feeding than any other treatment (Table 4). On 22 May the Actigard at 140 g/ha had significantly lower levels of beetle feeding than any other treatment except for the insecticide standard (Table 4). On 29 May, Actigard at 70 and 140 g/ha and the Regalia treatment had significantly lower beetle feeding than the untreated control or the Copron-F treatment (Table 4). Actigard[®] at 140 g per ha had significantly lower beetle feeding than the untreated control on any date (Table 4). Despite the differences in beetle damage observed, there was no difference in bacterial wilt severity in 2012, perhaps due to the dry weather (Table 4). Additionally, no differences were observed in vine growth in 2012 (Table 4). Actigard at 140 g per ha had significantly lower total yields in metric tons/ha than the untreated control, insecticide standard or Serenade Max[®]

(Table 5). Actigard at 140 g per ha also had significantly lighter fruit than the insecticide standard, Copron-F[®] and Regalia[®].

3.3. 2013 results

In 2013, there were no significant differences in bacterial wilt severity as measured by AUDPC or vine length (Table 6). Beetle feeding damage on flowers in 2013 was significantly lower for the insecticide standard than any treatment except for Serenade Max[®] or the Actigard[®] 35 g per ha treatment (Table 6). The number of dead beetles was significantly different only for the insecticide standard compared to all other treatments (Table 6). The total yield in weight was less for the Actigard at 35 g and 140 g per ha than the untreated control, the insecticide standard, Serenade Max[®] or Regalia[®] applied on foliage (Table 7). Yield in total numbers of fruit was more for the untreated control than for Actigard[®] at 140 g per ha.

3.4. 2014 results

There was moderate bacterial wilt pressure in 2014. The untreated control had more disease on 31 July than the insecticide standard and Actigard[®] applied six times at 35 g and 70 g per ha and Actigard[®] at 70 g per ha applied three times applied in sequence (Table 8). The AUDPC for the insecticide standard was significantly lower than any other treatment except for Actigard at 70 g/ha applied six times (Table 8). The insecticide standard had fewer beetles than any other treatment (Table 8). The insecticide standard resulted in the greatest yield in terms of total weight and number of fruits compared to any other treatment (Table 9). Actigard at 70 g per ha at 6 applications resulted in lighter fruit than the untreated check, Actigard at 70 g per ha applied 3 times applied in sequence, Actigard at 35 g per ha applied 3 times applied in sequence or alternated (Table 9).

4. Discussion

In all four years of trials, the insecticide standard resulted in the lowest amounts of beetle feeding or beetle populations. However, in 2012, the insecticide standard did not result in significantly less bacterial wilt because disease severity was low in all treatments, including the untreated control. There are a variety of factors that may have contributed to low disease incidence. Leaf wetness is critical for *E. tracheiphila* to cause infection (Saalau Rojas and Gleason, 2012) and 2012 was a dry year. In 2013, a similar level of bacterial wilt was observed as in 2011 (Tables 2 and 6), however, the tendency of beetles to aggregate on a plant (Darlington, 2006) may lead to unacceptable variability in the data. For example, in 2013, the highest number of beetles observed on 4 June was in the low rate of Actigard, whereas on 11 June the highest number of beetles occurs on the Regalia drench treatment (Table 6); the amount of variation affects live beetle counts and probably affects the distribution of bacterial wilt among treatments.

The effect of Actigard[®] on beetle feeding seems to be more consistent than the effect of Actigard on bacterial wilt. Actigard[®] treatments resulted in significantly less beetle feeding compared to the untreated control in 2011 and 2012, suggesting that Actigard may be used to manage beetle feeding. Similar to the insecticide standard, however, lower beetle numbers did not always result in lower amounts of bacterial wilt. In 2014, in contrast, Actigard resulted in lower levels of bacterial wilt, but not necessarily beetle numbers compared to the untreated control. In 2014, beetle numbers were quantified, however beetle feeding was not. It may be that Actigard[®] may affect beetle feeding, but not the number per plant.

In 2011, 2014, there was no yield reduction due to the use of Actigard[®]; in both years, the severity of bacterial wilt was significantly affected by treatments. In 2012, 2013, in contrast, Actigard[®] caused yield reduction and no treatments resulted in significant reduction in

Table 6

Cucumber beetle feeding damage, cucumber population numbers, vine length and bacterial wilt disease severity on Athena cantaloupe as influenced by one of the induced resistance products, standard insecticide treatment or left untreated at the Southwest Purdue Agricultural Center in Vincennes, 2013.

Treatment, rate per ha	30 May Incidence ^z	4 June		11 June		Vine length (cm) ^y	AUDPC ^x
		# alive	# dead	# alive	# dead		
Untreated Check	4.3 b ^w	7.5	0.5 b	8.3	1.5 b	42.8	665.1
Actigard [®] , 35 g	3.3 bc	14.8	0.0 b	6.5	2.0 b	43.8	227.2
Actigard [®] , 70 g	3.8 b	11.8	0.0 b	8.3	1.8 b	48.1	364.4
Actigard [®] , 140 g	3.8 b	8.5	0.0 b	5.0	0.8 b	49.2	242.2
Serenade Max [®] , 3.36 Kg	3.0 bc	8.5	0.3 b	7.0	2.3 b	46.9	227.2
Regalia [®] , 9.4 L	7.3 a	14.3	0.3 b	6.8	1.3 b	48.8	306.8
Regalia [®] 2.8 L ^y	5.0 ab	11.5	0.0 b	11.3	2.3 b	51.0	180.4
Insecticide standard ^u	0.8 c	8.8	22.5 a	6.5	59.0 a	45.4	157.4
P-value	0.0030	0.6220	0.0001	0.4365	0.0001	0.7838	0.2815

^uAdmire Pro[®] at transplant and Pounce WP[®] applied weekly starting at 3 weeks post-transplant.

^yRegalia Drench at planting per 378 L of water.

^wMeans in columns followed by the same letter are not significantly different ($P = 0.05$) based on Fisher's protected LSD test.

^xHorsfall-Barratt ratings were conducted 3, 13 and 19 Jul and 2 Aug. Area under the disease progress curve (AUDPC) was calculated by trapezoidal integration.

^yThree vines per plot were measured on 29 May.

^zNumber of flowers with observed cucumber beetle feeding damage on the 10 plants in an experimental unit.

Table 7

Yield of Athena cantaloupe as influenced by one of the induced resistance products, standard insecticide treatment or left untreated treated for bacterial wilt at the Southwest Purdue Agricultural Center Vincennes, IN, 2013

Treatment, rate per ha	Mean wt. (kg)	Total wt. (metric tons/ha)	Total no. (1000/ha)
Untreated check	2.29	28.05 ab ^z	12.32 abc
Actigard [®] , 35 g	2.18	20.66 dc	9.64 cd
Actigard [®] , 70 g	2.11	21.13 bdc	9.86 bcd
Actigard [®] , 140 g	2.02	17.50 d	8.51 d
Serenade Max [®] , 3.36 Kg	2.27	29.29 a	13.00 ab
Regalia [®] , 9.4 L	2.22	28.87 a	13.00 ab
Regalia [®] 2.8 L ^y	2.22	26.29 abc	11.88 abc
Insecticide standard ^x	2.30	32.44 a	14.12 a
P-value	0.2160	0.0044	0.0241

^xAdmire Pro[®] at transplant and Pounce WP[®] applied weekly starting at 3 weeks post-transplant.

^yRegalia Drench at planting per 378 L of water.

^zMeans in columns followed by the same letter are not significantly different ($P = 0.05$) based on Fisher's protected LSD test.

bacterial wilt severity. When the yield reduction from bacterial wilt severity was low, as in 2012, or randomly assigned to treatments as in 2013, the yield reduction which was caused by Actigard[®] was observable and, at times, significant. The inconsistent effect of Actigard[®] on yield reduction would seem to preclude the use of this product for management of bacterial wilt.

Actigard[®] effect on yield and vine length do not seem to be related. In 2011, there was no yield differences due to Actigard[®] and yet vine length was significantly reduced due to Actigard[®]. In 2012, 2013, Actigard was associated with yield reduction and yet there was no difference in vine length. The effect of Actigard on yield may be cumulative. Negative effects late in the season may be more likely than vine growth in the early season. Actigard has been implicated with yield reduction in other crops. The use of Actigard helped to reduce the disease severity of bacterial spot on pepper, however, yield losses were observed when Actigard was applied in the absence of bacterial spot (Romero et al., 2001). Actigard has been implicated with yield reduction in grafted tomatoes (Kunwar et al., 2017). In a study to examine the efficacy of Actigard[®] on tomato seedlings in a transplant greenhouse, Actigard[®] was associated with significantly lower dry weight of tomato seedlings (Louws et al., 2001).

Regalia[®] effects on beetle feeding were not consistent. Regalia[®] reduced beetle feeding damage compared with the untreated control in

Table 8

Bacterial wilt severity, presented as percent on 31 July and AUDPC, and beetle numbers on 19 June in 2014 Meigs Lafayette IN.

Treatment, rate per ha	Spray timing ^z	Disease Severity		
		AUDPC ^y	31 Jul (%) ^x	Live beetles/plot ^w
Untreated	–	1199.0 a	68 a	37.5 ab
Insecticide standard ^v	0,1,2,3,4,5	61.2 c ^u	1.7 d	1.5 c
Actigard [®] , 35 g	0,1,2,3,4,5	564.6 b	32 bc	26.3 ab
Actigard [®] , 70 g	0,1,2,3,4,5	457.5 bc	22.5 c	27.3 ab
Actigard [®] , 35 g	0,2,4	590.9 b	49 ab	22.5 b
Actigard [®] , 70 g	0,2,4	654.6 b	43 abc	44.3 a
Actigard [®] , 35 g	0,1,2	609.1 b	49 ab	38.0 ab
Actigard [®] , 70 g	0,1,2	773.4 b	37.5 bc	30.5 ab
P-value		0.0013	< .0001	0.0072

^uMeans followed by the same letters are significantly different at the $P > 0.05$ level Fisher's LSD.

^vImidacloprid (Admire Pro[®]) was applied as a drench immediately after planting at the rate of 3.8 ml/8.8 L per treatment. Lambda cyhalothrin (Warrior IEC[®]) was applied as a foliar spray on weeks 1–5.

^wNumber of live striped cucumber beetles were counted on 19 Jun.

^xDisease severity rating converted from the Horsfall-Barratt ratings using the ELANCO tables.

^yCantaloupe were rated for disease severity on 3, 7, 9, 21, 28 and 31 Jul using the Horsfall-Barratt rating scale. AUDPC was calculated using trapezoid integration.

^zWeeks after planting on 28 May.

2011 and on one date in 2012. However, Regalia[®] did not significantly reduce bacterial wilt severity compared to the untreated control in 2011. In 2013, Regalia[®] applied as a foliar spray resulted in higher beetle feeding than any other treatment except for the Regalia[®] drench. In no year did Regalia[®] result in significant differences from the untreated control in yield.

Serenade Max[®] had little effect on the variables measured here. However, in 2011, Serenade Max[®] was associated with an AUDPC lower than the untreated control although beetle feeding was not affected.

Copron-F[®], a nutrient supplement containing copper and sulfur, did not lower beetle feeding in 2011, yet was associated with a lower level of bacterial wilt. It is intriguing to think that the copper portion of the product helped to reduce the number of bacteria in the plant, thus lowering disease severity. However, Copron-F[®] effects on beetle feeding in 2012 were mixed and there were no differences in bacterial wilt severity.

Although SAR inducing products have been shown to reduce insect

Table 9

Weight and number of cantaloupe per plot in (kg) and weight per fruit in 2014 Meigs Lafayette IN.

Treatment, rate per ha	Spray timing ^z	Total Weight (metric tons/ha)	Number of Fruit (1000/ha)	Weight/Fruit kg
Insecticide standard ^y	0,1,2,3,4,5	65.32 a ^x	22.54 a	2.89 a
Actigard [®] , 35 g	0,1,2,3,4,5	22.54 bc	9.22 bc	2.45 bc
Actigard [®] , 70 g	0,1,2,3,4,5	30.09 b	12.98 b	2.32 c
Actigard [®] , 35 g	0,2,4	24.82 bc	9.56 bc	2.70 ab
Actigard [®] , 70 g	0,2,4	24.52 bc	9.90 bc	2.47 bc
Actigard [®] , 35 g	0,1,2	18.56 bc	6.49 c	2.86 a
Actigard [®] , 70 g	0,1,2	23.81 bc	8.54 bc	2.90 a
Untreated	–	12.90 c	4.95 c	2.63 ab
P-value		< .0001	< .0001	0.0021

^x Means followed by the same letters are significantly different at the P > 0.05 level Fisher's LSD.

^y Imidacloprid (Admire Pro[®]) was applied as a drench immediately after planting at the rate of 3.8 ml/8.8 L per treatment. Lambda cyhalothrin (Warrior IEC[®]) was applied as a foliar spray on weeks 1–5.

^z Weeks after planting on 28 May.

feeding, insect weight gain or insect fecundity (Cooper et al., 2004; Kessler and Baldwin, 2002; Sobhy et al., 2015) to our knowledge, this is the first time that cucumber beetle feeding has been shown to be reduced by this particular SAR inducing product.

Since *E. trachiphiela* does not require cell death to cause infection, has a relatively narrow host range and is not easy to culture, it may be considered a biotroph (Glazebrook, 2005). In general, ISR has been shown to be more effective against necrotrophs and SAR more effective against biotrophs (Vallad and Goodman, 2004). It is interesting that the application of plant growth promoting bacteria elicited ISR in cucumber and successfully reduced cucumber beetle feeding and bacterial wilt (Zehnder et al., 2001). In this study, SAR induced by Actigard[®] was more successful in reducing bacterial wilt than Regalia[®] or Serenade Max[®], which are more often associated with ISR. It is possible that the use of growth-promoting rhizobacteria (Zehnder et al., 2001) was superior in eliciting ISR than either Regalia[®] or Serenade Max[®].

Since Actigard has been shown here to reduce beetle feeding and bacterial wilt severity, it is tempting to begin using this product to manage bacterial wilt of cucurbits. The yield loss observed here, although inconsistent, is sufficient reason to avoid the use of this product for bacterial wilt management. Future studies may be able to show that specific rates and timings of Actigard[®] can be used that will be useful for management of bacterial wilt in cantaloupe without yield loss.

Funding

This work was supported by USAID SAAF Agreement No: AID-306-A-00-11-00516 Grant No 105252 and USDA, National Institute of Food and Agriculture (Grant No: 10512824).

References

Brust, G.E., Foster, R.E., 1999. New economic threshold for striped cucumber beetle (Coleoptera: chrysomelidae) in cantaloupe in the Midwest. *J. Econ. Entomol.* 92, 936–940.

Brust, G.E., Rane, K.K., 1995. Differential occurrence of bacterial wilt in muskmelon due to preferential striped cucumber beetle feeding. *Hortscience* 30, 1043–1045.

Cooper, W.C., Jia, L., Goggin, F.L., 2004. Acquired resistance against the potato aphid in tomato. *J. Chem. Ecol.* 30, 2527–2542.

Dayan, F.E., Cantell, C.L., Duke, S.O., 2009. Natural products in crop protection. *Bioorg. Med. Chem.* 17, 4022–4034.

Darlington, M., 2006. Sexual Selection in the Spotted Cucumber Beetle, *Diabrotica undecimpunctata howardi* (Coleoptera: Chrysomelidae): the Benefits of Female Choice. Ph.D. dissertation. The Pennsylvania State University, University Park, PA, USA.

Eyles, A., Bonello, P., Ganley, R., Mohammed, C., 2010. Induced resistance to pests and pathogens in trees. *New Phytol.* 185, 893–908.

Foster, R., Brust, G., Palumbo, J., 2005. Watermelon, muskmelon, and cucumber. In: *Vegetable Insect Management*. Meister Media Worldwide, Willoughby, OH, pp. 198–214.

Foster, R., Bordelon, B., Hirst, P., Clingerman, V., O'Donnell, M., Ballard, R., 2016. *Protecting Pollinators in Fruit and Vegetable Production*. Purdue University POL-2. 8 pp.

Glazebrook, J., 2005. Contrasting mechanisms of defense against biotrophic and necrotrophic pathogens. *Annu. Rev. Phytopathol.* 43, 205–227. <https://doi.org/10.1146/annurev.phyto.43.040204.135923>.

Horsfall, J.G., Barratt, R.W., 1945. An improved grading system for measuring plant diseases. *Phytopathology* 35, 655 (abstr.).

Huang, C., Vallad, G.E., Zhang, S., Wen, A., Balogh, B., Figueiredo, J.F.L., Behlau, F., Jones, J.B., Timur Momol, M., Olson, S.M., 2012. Effect of application frequency and reduced rates of acibenzolar-S-methyl on the field efficacy of induced resistance against bacterial spot on tomato. *Plant Dis.* 96, 221–227.

Kessler, A., Baldwin, I.T., 2002. Plant responses to insect herbivory: the emerging molecular analysis. *Annu. Rev. Plant Biol.* 53, 299–328. <https://doi.org/10.1146/annurev.arplant.53.100301.135207>.

Krause, M.S., De Ceuster, T.J.J., Tiquia, S.M., Michel, F.C., Madden, L.V., Hoitink, H.A.J., 2003. Isolation and characterization of rhizobacteria from composts that suppress the severity of bacterial leaf spot of radish. *Phytopathology* 93, 1292–1300.

Kunwar, S., Paret, M.L., Freeman, J.H., Ritchie, L., Olson, S.M., Colee, J., Jones, J.B., 2017. Foliar Applications of acibenzolar-s-methyl negatively affect the yield of grafted tomatoes in fields infested with *Ralstonia solanacearum*. *Plant Dis.* 101, 890–894.

Lahlali, R., Peng, G., Gossen, B.D., McGregor, L., Yu, F.Q., Hynes, R.K., Hwang, S.F., McDonald, M.R., Boyetchko, S.M., 2013. Evidence that the biofungicide Serenade[®] (*Bacillus subtilis*) suppresses club- root on canola via antibiosis and induced host resistance. *Phytopathology* 103, 245–254.

Louws, F.J., Wilson, M., Campbell, H.L., Cuppels, D.A., Jones, J.B., Shoemaker, P.B., Sahin, F., Miller, S.A., 2001. Field control of bacterial spot and bacterial speck of tomato using a plant activator. *Plant Dis.* 85, 481–488.

Neher, O.T., Johnston, M.R., Zidack, N.K., Jacobsen, B.J., 2009. Evaluation of *Bacillus mycoides* isolate BmJ and *B. mojavensis* isolate 203-7 for the control of anthracnose of cucurbits caused by *Glomerella cingulata* var. *orbicularis*. *Biol. Contr.* 48, 140–146.

Oostendorp, M., Kunz, W., Dietrich, B., Staub, T., 2001. Induced disease resistance in plants by chemicals. *Eur. J. Plant Pathol.* 107, 19–28.

Pair, S.D., 1997. Evaluation of systemically treated squash trap plants and attractorial baits for early-season control of striped and spotted cucumber beetles (Coleoptera: chrysomelidae) and squash bug (Hemiptera: coreida) in cucurbit crops. *J. Econ. Entomol.* 90, 1307–1314.

Park, S., Kaimoyo, E., Kumar, D., Mosher, S., Klessig, D.F., 2007. Methyl salicylate is a critical mobile signal for plant systemic acquired resistance. *Science* 318, 113–116. <https://doi.org/10.1126/science.1147113>.

Poschenrieder, C., Tolra, R., Barcelo, J., 2006. Can metals defend plants against biotic stress? *Trends Plant Sci.* 11, 288–295. <https://doi.org/10.1016/j.tplants.2006.04.007>.

Riley, D.G., Joseph, S.V., Srinivasan, R., 2012. Reflective mulch and acibenzolar-S-methyl treatments relative to thrips (Thysanoptera: thripidae) and tomato spotted wilt virus incidence in tomato. *J. Econ. Entomol.* 105, 1302–1310. <https://doi.org/10.1603/EC11179>.

Romero, A.M., Kousik, C.S., Ritchie, D.F., 2001. Resistance to bacterial spot in bell pepper induced by acibenzolar-S-methyl. *Plant Dis.* 85, 189–194.

Saalau Rojas, E., Gleason, M.L., 2012. Epiphytic survival of *Erwinia tracheiphila* on muskmelon (*Cucumis melo* L.). *Plant Dis.* 96, 62–66.

Saalau Rojas, E., Gleason, M.L., Batzer, J.C., Duffy, M., 2011. Feasibility of delaying removal of row covers to suppress bacterial wilt of muskmelon (*Cucumis melo*). *Plant Dis.* 95, 729–734.

Saalau Rojas, E.S., Batzer, J.C., Beattie, G.A., Fleischer, S.J., Shapiro, L.R., Williams, M.A., Bessin, R., Bruton, B., Boucher, T.J., Jesse, L.C.H., Gleason, M.L., 2015. Bacterial wilt of cucurbits: resurrecting a classic pathosystem. *Plant Dis.* 99, 564–574. <https://doi.org/10.1094/PDIS-10-14-1068-FE>.

Shaner, G., Finney, R.E., 1977. The effect of nitrogen fertilization on the expression of slow-mildewing resistance in Knox wheat. *Phytopathology* 67, 1051–1056.

Sobhy, I.S., Mandour, N.S., Sarhan, A.A., 2015. Tomato treatment with chemical inducers reduces the performance of *Spodoptera littoralis* (Lepidoptera: noctuidae). *Appl. Entomol. Zool.* 50, 175–182.

Takeshita, M., Okuda, M., Okuda, S., Hyodo, A., Hamano, K., Furuya, N., Tsuchiya, K., 2013. Induction of antiviral responses by acibenzolar-S-methyl against *Cucurbit chlorotic yellows virus* in melon. *Phytopathology* 103, 960–965.

Vallad, G.E., Goodman, R.M., 2004. Systemic acquired resistance and induced systemic resistance in conventional agriculture. *Crop Sci.* 44, 1920–1934.

van Loon, L.C., Bakker, P.A.H.M., Pieterse, C.M.J., 1998. Systemic resistance induced by rhizosphere bacteria. *Annu. Rev. Phytopathol.* 36, 453–483.

Watterson, J.C., Williams, P.H., Durbin, R.D., 1971. Response of cucurbits to *Erwinia tracheiphila*. *Plant Dis. Rep.* 55, 816–819.

Watterson, J.C., Williams, P.H., Durbin, R.D., 1972. Multiplication and movement of *Erwinia tracheiphila* in resistant and susceptible cucurbits. *Plant Dis. Rep.* 56, 949–953.

Zehnder, G.W., Murphy, J.F., Sikora, E.J., Kloepper, J.W., 2001. Application of rhizobacteria for induced resistance. *Eur. J. Plant Pathol.* 107, 39–50.