International Journal of Fruit Science, 9:23–34, 2009

ISSN: 1553-8362 print/1553-8621 online
DOI: 10.1080/15538360902801288

Susceptibility of Highbush Blueberry Cultivars to Cranberry Fruitworm and Japanese Beetle

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The susceptibility of ten highbush blueberry cultivars, Vaccinium corymbosum *L., to infestation by two key insect pests was compared. Over 5 years of assessing infestation by cranberry fruitworm,* Acrobasis vaccinii *(Riley), in a replicated field planting, "Duke" received the greatest infestation in 3 of the 6 years. Susceptibility of the 10 cultivars to Japanese beetle feeding was measured over 3 years in laboratory assays designed to measure relative feeding on fruit and foliage. "Brigitta" was the most susceptible cultivar to fruit feeding, while "Elliott" and "Legacy" were the least susceptible cultivars to foliage feeding. "Elliott" and "Legacy" also had the lowest level of Japanese beetle feeding on leaves of blueberry bushes in the field. These results suggest the presence of plant-based resistance to foliage and fruit feeding insect pests in some cultivars of* V. corymbosum*, which could be exploited within integrated pest management strategies.*

KEYWORDS *IPM,* Popillia japonica*,* Acrobasis vaccinii*, plant resistance, feeding assay*

We thank Anne Hanley, Marie Prescott, Joseph Prescott, and Robert Young for technical assistance with this project, and the staff of Trevor Nichols Research Complex for maintenance of the blueberry planting. This work was funded in part by Project GREEEN and the Michigan Agricultural Experiment Station.

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INTRODUCTION

Highbush blueberries, *Vaccinium corymbosum* L., are produced on over 50,000 acres in the United States with an annual value of approximately $516 million (United States Department of Agriculture, 2008). The largest concentration of this production is in Michigan where 18,000 acres are grown for the processing and fresh fruit markets. In Michigan and other regions of North America, this crop can suffer infestation by a complex of insects that feed directly on the fruit and foliage. 2 economically important direct insect pests of this crop in Michigan are the cranberry fruitworm, *Acrobasis vaccinii* Riley, and the Japanese beetle, *Popillia japonica* Newman (Szendrei and Isaacs, 2006a). There is zero threshold for contamination by these insects in fruit, and growers strive to meet these exacting standards using in-field and postharvest management techniques (Mallampalli and Isaacs, 2002; Isaacs et al., 2004; Szendrei et al., 2005; Szendrei and Isaacs, 2006b). Adult Japanese beetle feeding on foliage is also of concern to blueberry producers, particularly in young bushes that do not yet have fruit. Cranberry fruitworm adults typically emerge during blueberry bloom and lay eggs on the young blueberry fruit. Once the eggs hatch the larvae burrow into the berry and begin feeding, eventually webbing several berries together. Japanese beetle adults begin emerging in June or July depending on the latitude, and begin feeding on nearby plants and mating soon after emergence.

Integrated pest management (IPM) programs utilize a combination of biological, cultural, and chemical methods to control pests (Hollingsworth and Coli, 2001). Despite the exacting quality standards imposed upon the fruit industry, progress toward this goal has been made for many crops, leading to reduced dependence on broad-spectrum insecticides (Caltagirone and Doutt, 1989; Costello and Daane, 2003; Mills and Daane, 2005; Pickett and Gilstrap, 1986). Crop cultivars that limit the suitability of fields for insect growth and development can help maintain pest populations below economic thresholds (Pedigo et al., 1986). Plant resistance is one pest management approach that can reduce insect development and feeding injury on crop plants, thus helping to prevent pest outbreaks (Wiseman, 1994). This tactic has increased significance for organic crop producers who have a limited suite of management tools from which to choose (Zehnder et al., 2007).

In highbush blueberry breeding programs, resistance to insect pests is rarely a priority consideration. Other horticultural traits such as yield, ripening date, and cold hardiness are emphasized (Dale and Hancock, 2005; Finn et al., 2003), and there is also a long time-lag between breeding and commercial release, so pest challenges may change during that time. Another reason is that for the past fifty years, broad-spectrum insecticides have been available that could effectively control most insect pests. Increasingly, consumer choice and government legislation are leading to restrictions on the chemical management tools that blueberry growers are able to use, and cultivars with inherent resistance to insect pests may be considered more desirable for planting.

Liburd et al. (1998) demonstrated that cultivars of *V. corymbosum* vary in susceptibility to infestation by the blueberry maggot, *Rhagoletis mendax*. Additional studies have shown cultivars that exhibit some resistance to the sharp-nosed leafhopper, *Scaphytopius magdalensis* Provancher (Homoptera), as well as cultivars that affect survival and development of the lappet moth, *Streblote panda* Hübner (Lepidoptera) (Etzel and Meyer, 1986; Meyer and Ballington, 1990; Rooks et al., 1995; Calvo and Molina, 2004). Ranger et al. (2006) found lower survivorship and fecundity of blueberry aphid, *Illinoia pepperi* MacGillivray (Hemiptera), on different *Vaccinium* spp. However, there is little available information on relative susceptibility of this crop to other insect pests, including cranberry fruitworm and Japanese beetle. This study was conducted to determine whether commercially available cultivars of *V. corymbosum* vary in their susceptibility to these key insect pests.

MATERIALS AND METHODS

Several experiments were conducted in order to compare the susceptibility of *V. corymbosum* cultivars to cranberry fruitworm and Japanese beetle. Experiments were conducted at the Trevor Nichols Research Complex in Fennville, Michigan, using ten cultivars of *V. corymbosum* that were 4 years old at the start of the trials in 2002. The *V. corymbosum* cultivars planted in this site were "Bluecrop," "Bluegold," "Brigitta," "Duke," "Elliott," "Jersey," "Legacy," "Nelson," "Rubel," and "Toro". Cultivars were planted in a 0.15 hectare research planting in 4 completely randomized blocks with 3.05 × 1.2 m spacing, where each block contained 5 rows of 12 bushes. Every row contained 2 cultivars as contiguous groups of 6 bushes per cultivar. Plants were grown according to standard local management recommendations, but without insecticide or fungicide applications.

To determine the relative susceptibility of *V. corymbosum* cultivars to infestation by fruitworms, clusters on planted bushes were assessed for infestation during the course of the growing season. To assess susceptibility to Japanese beetles, laboratory bioassays were conducted using foliage and fruit from planted bushes to compare levels of feeding on leaves to feeding on ripe fruit of the same cultivar as well as to compare feeding levels on leaves of different cultivars. In addition, Japanese beetle leaf-feeding assessments were conducted on planted bushes at the end of *the growing season*.

Cranberry Fruitworm

The 10 cultivars were compared for susceptibility to infestation by cranberry fruitworm in the 2002–2007 growing seasons. The average number of infested clusters per bush was determined for all bushes containing more than 10 clusters. Infested clusters were identified as those with fruit infested by fruitworms made evident by larval penetration holes, webbing, or frass (Hutchinson, 1954). Due to low fruit set, bushes in only 2 of the 4 blocks were examined on July 12, 2002, whereas bushes in all 4 blocks were examined on July 16–23 2003, July 7–12 2004, July 11, 2005, July 12, 2006, and July 11, 2007. These timings were chosen based on captures of cranberry fruitworm adults in pheromone traps placed within 30m of the planting.

The average number of infested clusters per bush was analyzed using analysis of variance (ANOVA) followed by Fisher's Protected Least Significant Difference test (Fisher's PLSD) for post hoc comparison of means using Statview 4.57 (Abacus Concepts Inc., Berkeley, California). Data were analyzed without using a blocking factor because of low sample sizes in 2 of the blocks.

Japanese Beetle

Feeding on fruit and foliage

Fruit and foliage feeding experiments were conducted during the summers of 2003, 2004, and 2005 to compare the level of feeding on leaves to feeding on ripe fruit of the same cultivar. 5 *V. corymbosum* shoots with leaves and ripe fruit were cut from each cultivar in the planting described previously. Each shoot was trimmed so that it contained 10 ripe berries and 5 mature leaves, and individual shoots were each placed into 946 ml plastic cups (11 cm top diameter, 8.5 cm bottom diameter, 14.3 cm high) containing water-soaked floral foam (3–4 cm deep). After shoots were placed in the floral foam, paraffin wax was poured over the foam to 5mm depth to prevent moisture from escaping and beetles from burrowing into the foam. Assays took place at different times throughout the summer, depending on when berries were ripe on the bushes to take samples. Due to poor bush growth and fruit production in some cultivars, only 2 of the 4 blocks were used to collect shoots for these experiments.

Adult Japanese beetles were obtained on the day of the experiments using traps baited with sex pheromone and floral lures (Trécé Inc., Adair, Oklahoma). Trapped beetles were held in plastic bins (40.6 × 28 × 14 cm) and fed untreated wild grape foliage (*Vitis riparia* Michx.) or sassafras leaves (*Sassafras albidum* (Nutt.) Nees) *ad libitum* until the start of the experiments. A female beetle was placed at the bottom of each container to start the experiment. Only female beetles were used because they feed for longer periods of time than males (Smith, 1923). The location and state of each beetle on fruit, on foliage, on stem, off the foliage, or dead were recorded after 1, 24, and 48 hours. After 48 hours, the level of feeding damage was visually assessed and the amount of tissue removed from fruit and foliage was recorded in 5% increments.

Percent feeding damage data were arcsine square root ($p^{1/2}$) transformed and compared between cultivars with ANOVA, followed by Fisher's PLSD test for *post hoc* comparison of means. In addition, fruit and leaf feeding percentages were compared within replicates using paired t-tests.

Foliage feeding choice assay

Feeding choice bioassays were performed in August 2003, 2004, and 2005. 10 fully expanded, undamaged leaves per cultivar were randomly collected from each of the 2 blocks used in the fruit and foliage feeding trials for a total of 20 leaves per cultivar. 1 leaf from each blueberry cultivar was randomly placed in a circle inside a Petri dish (100 × 15 mm), such that the leaf petioles were facing inward. Each leaf was glued to the bottom of the Petri dish with a hot glue gun to prevent displacement by the beetles. A cotton ball moistened with distilled water was placed in the center of the Petri dish to provide moisture for the Japanese beetles. Beetles were captured from the field using the same traps and were held as described above. At the start of the experiment, one female Japanese beetle was placed in the center of the Petri dish. After 7 days, feeding damage (percentage of leaf area removed) on each leaf was recorded using 5% increments, with trace feeding damage recorded as 1% damage. Beetle mortality was also recorded where present.

Percent feeding damage data were arcsine square root transformed and compared among cultivars before being subjected to ANOVA with time and cultivar as variables, followed by Fisher's PLSD test for post hoc comparison of means.

Foliage feeding on bushes

Japanese beetle feeding on the leaves of all the bushes of each cultivar in the planting described above was measured in October 2005 and September 2007. The top cane on each bush was selected and the total number of leaves showing evidence of Japanese beetle feeding was counted. The damaged leaf data were compared among cultivars using ANOVA followed by Fisher's PLSD test for post hoc comparison of means.

RESULTS

Cranberry Fruitworm

Cranberry fruitworm infestation varied significantly among the 10 blueberry cultivars in 4 of the 6 years of assessments (2002: $F = 5.30$, $df = 9{,}99$, $P < 0.0001$; 2003: $F = 8.68$, $df = 9{,}142$, $P < 0.0001$; 2004: $F = 5.13$, $df = 9{,}171$, $P < 0.0001$; 2005: $F = 1.51$, $df = 8{,}96$, $P = 0.16$; 2006: $F = 1.18$, $df = 9{,}147$, $P = 0.31$; 2007:

$F = 7.06$, $df = 9{,}136$, $P < 0.0001$) (Table 1). There were no significant differences among cultivars in 2005 or 2006. The early ripening cultivar, "Duke" had significantly higher levels of infestation than all the other cultivars in both 2002 and 2003, and had higher levels of infestation than all but 2 cultivars in 2004. "Toro" and "Rubel" bushes had the lowest levels of infestation in 2002 and 2004, while bushes of the "Legacy" cultivar had the lowest infestation levels in 2003. In 2007, "Elliott" and "Jersey" had the highest levels of infestation while "Brigitta" and "Legacy" had the lowest levels of infestation.

Japanese Beetle Feeding on Fruit and Foliage

In 2003, foliage feeding was highest in "Bluegold" and lowest in "Elliott" cultivars ($F = 3.03$, $df = 9{,}90$, $P = 0.0033$) in these no-choice trials (Table 2). This trend was similar in 2004, although the differences were not significant ($F = 0.23$, $df = 9{,}90$, $P = 0.99$). "Elliott" also had the lowest level of foliage feeding in 2005, but again this cultivar was not significantly different from the other cultivars ($F = 1.38$, $df = 8{,}81$, $P = 0.22$).

In 2003, "Brigitta" and "Elliott" fruit received the highest level of fruit feeding, while "Toro" fruit had the lowest level of feeding compared to the other cultivars ($F = 3.35$, $df = 9{,}90$, $P = 0.0014$) (Table 2). In 2004, "Brigitta" had significantly higher levels of fruit feeding than the lowest 6 cultivars, while "Bluegold" had significantly lower fruit feeding than any of the other cultivars ($F = 6.37$, $df = 9{,}90$, $P < 0.0001$). In 2005, there was no significant difference among cultivars ($F = 0.96$, $df = 8{,}81$, $P = 0.47$).

Japanese beetle foliage choice assay

Japanese beetle feeding preferences resulted in significant differences among the cultivars in the area of leaf removed in the choice tests ($F = 2.37$, $df = 9{,}570$, $P = 0.0012$) (Fig. 1A). In the tests conducted from 2003 to 2005, Bluecrop received significantly higher leaf feeding damage than the 5 cultivars with the lowest damage. The 2 cultivars with the lowest feeding damage, the late-ripening "Elliott" and "Legacy" cultivars, received significantly less leaf damage than all but one of the other cultivars.

Japanese beetle leaf feeding on bushes

Japanese beetle feeding preferences on bushes in the field were significantly different among the cultivars ($F = 6.43$, $df = 9{,}149$, $P = 0.0001$) (Fig. 1B). "Bluegold" had significantly higher levels of leaf feeding than the 6 lowest cultivars. Similar to the laboratory bioassays, "Elliott" had significantly lower

TABLE 1 Average (±S.E.) Number of Clusters from Ten Blueberry Cultivars Infested by Cranberry Fruitworm from 2002 to 2007

Cultivar	Mid-Fruiting date	2002	2003	2004	2005[1]	2006	2007
Duke	192	2.7 ± 0.5 a	4.4 ± 0.8 a	1.9 ± 0.5 ab	1.3 ± 0.6 a	4.2 ± 1.4 a	2.2 ± 1.2 bc
Toro	204	0.0 ± 0.0 c	0.8 ± 0.3 bc	0.2 ± 0.1 e	0.0 ± 0.0 a	2.7 ± 0.8 a	1.2 ± 0.4 c
Bluegold	208	1.0 ± 0.5 bc	1.5 ± 0.7 bc	0.4 ± 0.2 e	0.3 ± 0.3 a	3.0 ± 0.9 a	2.1 ± 1.1 bc
Bluecrop	209	0.7 ± 0.4 bc	0.8 ± 0.4 bc	0.5 ± 0.2 de	0.9 ± 0.3 a	5.6 ± 1.0 a	1.8 ± 0.5 c
Rubel	220	0.0 ± 0.0 c	0.7 ± 0.2 bc	0.2 ± 0.1 e	1.1 ± 0.6 a	2.8 ± 0.8 a	1.8 ± 0.4 c
Nelson	222	0.6 ± 0.3 bc	0.5 ± 0.2 bc	0.8 ± 0.2 cde	0.5 ± 0.2 a	3.0 ± 0.7 a	1.7 ± 0.4 c
Jersey	225	0.6 ± 0.2 bc	0.5 ± 0.2 bc	0.5 ± 0.2 de	2.0 ± 0.7 a	2.8 ± 0.6 a	4.2 ± 1.1 b
Brigitta	226	1.6 ± 0.6 b	0.2 ± 0.1 bc	1.4 ± 0.5 bcd	0.0 ± 0.0 a	3.1 ± 1.2 a	0.7 ± 0.2 c
Elliott	236	0.4 ± 0.2 c	0.9 ± 0.3 bc	2.4 ± 0.5 a	0.9 ± 0.4 a	5.3 ± 1.4 a	7.2 ± 1.0 a
Legacy	237	0.2 ± 0.2 c	0.0 ± 0.0 c	1.5 ± 0.5 abcd		3.2 ± 0.7 a	0.5 ± 0.2 c

[1]No clusters present on any Legacy bushes.

Infested clusters were counted on bushes with more than ten clusters. Cultivars are arranged by Julian date of mid-fruiting. Values within a column followed by the same letter are not significantly different at $\alpha = 0.05$.

TABLE 2 Average (± S.E.) Percent Feeding Damage to Blueberry Foliage and Fruit by Japanese Beetles After 48 Hours of Exposure to Fruit and Foliage of Individual Blueberry Varieties

		Foliage			Fruit		
Cultivar	Mid-Fruiting date	2003	2004	2005[1]	2003	2004	2005[1]
Duke	192	3.5 ± 2.0 ce	9.5 ± 3.1 a	6.0 ± 2.3 a	16.0 ± 4.8 bcd	25.5 ± 5.1 abc	15.5 ± 5.1 a
Toro	204	12.0 ± 2.6 abd	7.5 ± 1.5 a	8.5 ± 1.7 a	8.0 ± 3.2 e	20.0 ± 3.2 bc	22.0 ± 5.1 a
Bluegold	208	19.0 ± 3.9 a	11.5 ± 2.5 a	9.0 ± 2.1 a	13.0 ± 3.4 ce	4.0 ± 1.5 d	12.5 ± 4.2 a
Bluecrop	209	6.5 ± 2.4 be	8.5 ± 1.8 a	12.5 ± 3.7 a	19.0 ± 3.6 ac	15.0 ± 4.1 c	12.5 ± 4.6 a
Rubel	220	6.5 ± 1.5 be	9.0 ± 2.8 a	11.5 ± 1.8 a	15.5 ± 3.3 bcd	26.0 ± 2.2 ab	19.5 ± 3.8 a
Nelson	222	6.0 ± 2.3 bde	10.5 ± 2.6 a	10.5 ± 1.6 a	22.5 ± 2.0 ad	15.0 ± 3.0 c	9.5 ± 3.8 a
Jersey	225	11.0 ± 3.9 abc	7.5 ± 1.9 a	7.5 ± 1.1 a	21.0 ± 3.8 ac	14.5 ± 3.0 c	15.0 ± 4.8 a
Brigitta	226	8.0 ± 2.0 be	9.5 ± 2.0 a	9.0 ± 1.9 a	27.5 ± 3.9 ab	31.5 ± 2.6 a	12.0 ± 3.5 a
Elliott	236	2.5 ± 1.5 e	6.5 ± 1.5 a	5.5 ± 1.7 a	27.5 ± 2.8 a	18.5 ± 4.2 b	11.5 ± 2.5 a
Legacy	237	11.0 ± 2.9 ab	7.5 ± 1.5 a		21.0 ± 3.7 ac	27.0 ± 3.6 ab	

[1]No clusters present on any Legacy bushes.

Cultivars are arranged by Julian date of mid-fruiting period. Values within a column followed by the same letter are not significantly different at $\alpha = 0.05$.

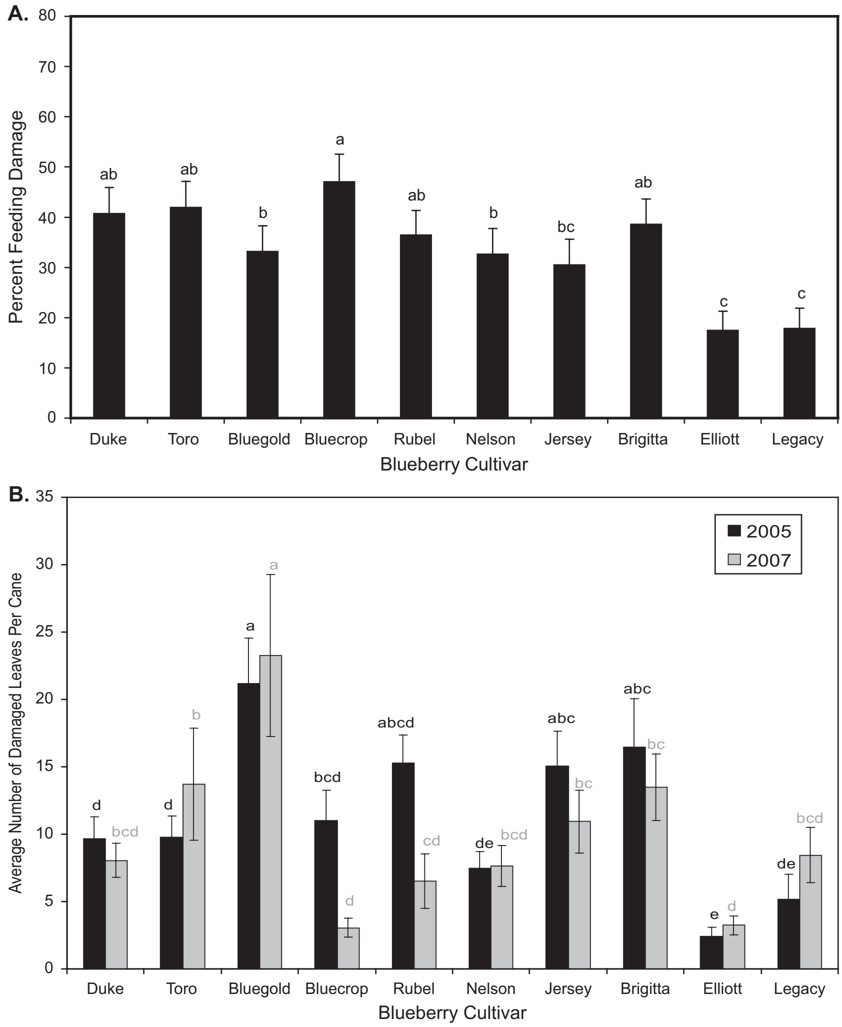

FIGURE 1 Japanese beetle feeding damage to leaves of different highbush blueberry, *V. corymbosum* cultivars in (A.) leaf choice feeding bioassays from 2003 to 2005 and (B.) on field-planted bushes of the ten cultivars, assessed for damaged leaves in 2005 and 2007. Values with the same letter within each year are not significantly different at $\alpha = 0.05$.

leaf feeding levels than the 7 highest cultivars, and "Legacy" had the second lowest feeding damage, significantly lower than the highest 3 cultivars.

DISCUSSION

The development of IPM programs for pest control in commercial blueberry production involves exploration of various methods to achieve effective pest control. Selection of disease and insect resistant cultivars based on potential pest pressure can aid with maintaining low pest populations. This also has the potential to reduce management costs significantly over the life of a blueberry planting, thus making those fields more profitable in the long run.

The results of this study indicate that there are some differences in susceptibility of blueberry cultivars with regards to both cranberry fruitworm and Japanese beetle feeding on fruit. Cranberry fruitworm cluster assessments revealed that "Duke" had the highest infestation levels in 3 of the 6 years of assessments. One potential reason is that "Duke" is an earlier fruiting cultivar than the other cultivars tested, so fruit development would be more advanced, allowing higher rates of oviposition and making fruit more suitable for oviposition. Japanese beetle feeding assays demonstrate that beetles fed on "Brigitta" fruit more than the other cultivars in 2 of the 3 years of testing, with "Toro" and "Bluegold" showing the lowest level of feeding in 2003 and 2004, respectively. These trials were conducted in a small planting under choice test conditions, and it would be expected that these measured effects would be less evident if a grower plants a large contiguous field of the same cultivar. Understanding whether reduced feeding translates into lower fecundity in Japanese beetle would be a valuable next step in this research.

With regards to feeding on foliage, beetles fed less on "Elliott" and "Legacy" cultivars as indicated by both laboratory assays and assessments on blueberry bushes. As late ripening cultivars, these two have fruit that remain green for longer than the others tested which could potentially minimize attraction of beetles to fruit volatiles, but certain blueberry cultivars have been found to have some resistance to many of the common diseases in blueberries, including mummy berry, *Monilinia vaccinii-corymbosi* (Reade), phomopsis, *Phomposis vaccinii* (Sheare), and anthracnose fruit rot, *Colletotrichum* spp. (Stretch et al., 1995; Beard Baker, 1995; Stretch and Ehlenfeldt, 2000; Stretch et al., 2001) with "Elliott" being consistently more resistant to diseases than other cultivars (Ehlenfeldt and Stretch, 2002).

These differences among cultivars can potentially be utilized by commercial blueberry growers as another tool in an IPM program. In addition to consideration of factors such as yield, fruit quality, and ripening dates, growers can take plant resistance into account when deciding what cultivar to plant. This study provides important information on blueberry cultivar resistance to cranberry fruitworm infestation and Japanese beetle feeding. Further research is needed to determine the mechanism(s) underlying differences among cultivars found in this study, to understand whether it can be exploited in future breeding programs.

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