2017 FINAL PROJECT REPORT

PROJECT TITLE: Hive Density and Pollinator Supplementation in Washington Highbush Blueberry

INVESTIGATORS:
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OBJECTIVES:
The main objective of this project is to evaluate practices that have the potential to promote pollination, fruit set, yield, and fruit quality attributes in pollination-limited western Washington blueberry. Specific practices that we evaluated include: increasing honey bee hive density, application of commercial pheromones (“Bee Scent”), and pollination supplementation with a native species of bumble bee. Additionally, we measured the cost effectiveness of these evaluated practices in commercial blueberry production so growers can assess cost/benefits.

PROCEDURES:
Hive Density Experiment. Six commercial, conventionally managed field sites of ‘Duke’ blueberry grown in western Washington were used in this experiment conducted in 2016 and 2017. Hives were placed in the study sites at approximately 5% bloom. To maintain independence, sites were a minimum of one mile apart. To minimize variation, we acquired honey bees from the same source (Bellevue Bees). The treatments were as follows, with each treatment replicated three times per year (i.e., 3 sites received treatment #1 and 3 sites received treatment #2).

Treatments:
1. 4 hives/acre of honey bee (control)
2. 8 hives/acre of honey bee

Bumble Bee Supplementation Experiment. Six commercial, conventionally managed field sites of ‘Draper’ blueberry grown in western Washington were used in this experiment. To maintain independence, sites were a minimum of one mile apart and honey bees were acquired from the same source (Bellevue Bees). Honey bee hives were placed in the study sites at approximately 5% bloom. Native yellow-faced bumble bee (Bombus vosnesenskii) was our supplemental bumble bee pollinator, which we obtained through a commercial propagator locally. The treatments were as follows, with each treatment replicated three times (e.g, 3 sites received treatment #1 and 3 sites received treatment #2). Data for 2016 are only presented, as our source of bumble bees became unexpectedly unavailable in 2017. Despite best attempts, we were unable to identify another source of B. vosnesenskii or alternative native bumble bee species to use in this experiment.

Treatments:
1. 4 hives/acre of honey bee (control)
2. 4 hives/acre honey bee + 3 hives/acre commercially-raised bumble bees (Yellow-faced bumble bee, *B. vosnesenskii*)

**Pheromone Experiment.** Commercial “Bee Scent” pheromone was evaluated in ‘Draper’ blueberry grown in Whatcom County, Washington in 2017 only. Seven plant plots were treated with or without Bee Scent pheromone and plots were replicated four times in order to allow statistical analysis. Bee Scent was applied according to the label starting at 5% bloom and extending to ~75% bloom (2 qt/acre; 3 applications total). Control plots were treated with water only.

**Data Collection – Hive Density and Bumble Bee Experiments.** Three 90-m long transects going down the length of the row were established within each experimental field site. Transects began at the natural edge of a field and proceeded towards the interior of the planting. Ten bushes spaced evenly along the length of each transect were flagged and used to measure pollinator activity and abundance (N = 30 bushes per site; 10 bushes per transect; described below). Initial bloom and yield component data were collected from the flagged bushes. Procedures for the collection of pollinator activity and abundance data were similar to those described by Courcelles et al. (2013). Honey bee and bumble bee activity was assessed at each site from 10:00 AM to 4:00 PM at approximately 0 days after full bloom (DAFB), 3 DAFB, and 6 DAFB or when weather permitted (i.e., air temperatures > 55 °F, clear to partly cloudy skies, and no precipitation). Measures of activity only considered “legitimate” pollination events, which occurs when a species forages within the flower and enters through the corolla (i.e., no “nectar robbing”). Abundance, or total number of bees on a bush (both pollinating and not), were also recorded. Activity and abundance data were collected from tagged bushes within one-minute intervals. Counts were replicated three times within a day for a minimum of three days per site.

Honey bee and bumble bee colony strength was evaluated the same day in which activity and abundance measurements occur. These measurements were collected by counting the number of incoming bees returning to their hives within a one-minute interval (or in cases of inclement weather, the next honey bee active day), replicated three times per day and site. Temperature, radiation (light), and wind data were also collected from local weather stations through the WSU AgWeatherNet program ([http://weather.wsu.edu/awn.php](http://weather.wsu.edu/awn.php)).

Fruit set, estimated berry number, and estimated yield were determined from all tagged plants. Thirty randomly selected ripe berries were sampled from each transect for assessment of berry size, firmness, and seed number. Seeds were extracted according to a method provided by Strik (unpublished). Number of fertile seeds > 1.7 mm in length was determined, which is indicative of fertilization and can serve as a proxy for effective pollination in blueberry (Dogterom et al., 2000).

**Data Collection – Pheromone Experiment.** All data were collected as described above with the exception that pollinator activity and abundance, fruit set, estimated berry number and yield, berry size, firmness, and seed number were sampled from the inner five plants per seven plant plot.
RESULTS:

_Hive Density Experiment:_ All honey bee hives were in similar health and with good uniformity in brood production and colony strength when hives were opened with the assistance of a beekeeper. Flower number/bush was the same across sites, indicating the sites were comparable with regards to bloom characteristics (data not shown). When honey bee hive density was increased to 8 hives/acre, honey bee visitation rates, estimated yield, and berry characteristics (weight and seed number) were increased (Table 1 and Figs. 1-2). Berry firmness was not affected by year and was the same across the treatments (Table 1). While we did observe an increase in the number of legitimate honey bee visits in the 8 hives/acre treatment compared to the control of 4 hives/acre, fruit set was not found to concurrently increase. Average berry weight was increased in the high hive density treatment (22% and 20% increase in 2016 and 2017, respectively). Seed number/berry also increased in the high hive density treatment relative to the control (20% and 31% increase in 2016 and 2017, respectively). Estimated yield increased among plants treated with the high density stocking rates and were 22% and 40% greater than the control stocking rates in 2016 and 2017, respectively (Fig. 2).

We performed regression analysis to evaluate the relationship between our explanatory and outcome variables. Honey bee visitation rate was positively related to seed number/berry ($R^2 = 0.61$ and $P$-value $< 0.001$). Honey bee visitation rate was also positively related to berry weight ($R^2 = 0.47$ and $P$-value $< 0.001$). Additionally, the number of seeds/berry was positively related to berry weight ($R^2 = 0.50$ and $P$-value $< 0.001$). These regression analyses were statistically significant and demonstrate that increased pollination and seed number explains a significant amount of the variation associated with berry weight.

We conducted basic economic analyses using grower-reported costs and price data from 2017. We assumed 1,450 plants/acre, $60/honey bee hive, and $0.78/lb for harvested ‘Duke’ blueberry. Scaling up our estimated yield results, our data suggests a grower can achieve 3.8 tons/acre and 5.9 tons/acre using honey bee stocking densities of 4 and 8 hives/acre, respectively. Using a grower revenue spreadsheet provided by one of our grower cooperators, we estimated an approximately $360/acre net increase by increasing honey bee stocking densities from 4 to 8 hives/acre. These data demonstrate that increasing honey bee stocking density is a promising practice to promote honey bee visitation, pollination, berry size, and yield in western Washington blueberry. Further research evaluating modified honey bee stocking densities in other cultivars of blueberry and evaluation of the point of diminishing returns is recommended.
Table 1. Fruit set, berry weight, seed number/berry, and berry firmness of ‘Duke’ blueberry stocked with honey bee hives at 4 and 8 hives/acre in 2016 and 2017.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Fruit set (%)</th>
<th>Berry weight (g/berry)</th>
<th>Seed no./berry</th>
<th>Firmness (g·mm⁻¹ of deflection)</th>
</tr>
</thead>
<tbody>
<tr>
<td>4 hives/acre</td>
<td>93.9</td>
<td>96.2</td>
<td>1.4 b</td>
<td>1.6 b</td>
</tr>
<tr>
<td>8 hives/acre</td>
<td>95.2</td>
<td>99.0</td>
<td>1.8 a</td>
<td>2.0 a</td>
</tr>
<tr>
<td><strong>P-value</strong></td>
<td>0.2512</td>
<td>0.4131</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
</tr>
</tbody>
</table>

^ Means separations were performed with Tukey’s Honest Significant Difference (HSD) test; means with the same letter are not different at \( P \leq 0.05 \); means were combined across both years when analyses revealed no significant interaction due to year.

Figure 1. Honey bee activity rates in ‘Duke’ blueberry in 2016 and 2017.

Figure 2. Estimated yield in ‘Duke’ blueberry in 2016 and 2017. Note yield is presented in kg/bush (1 kg = 2.2 lbs).
**Bumble Bee Experiment.** The bumble bee colonies, which are propagated from captured wild queens in late winter, arrived late in 2016 due to propagation difficulties that were beyond our control. As a result, our bumble bee colonies were only present during 90-100% full bloom in 2016. That year, we did not see any significant differences in pollinator activity (Fig. 3), fruit set, estimated yield, berry size, seed number, or fruit firmness (Table 2; firmness and seed data are not presented).

We hoped to re-test supplementation with bumble bees in 2017 and obtain a better assessment of their potential benefits for promoting pollination in blueberry. However, this sourcing issue continued in 2017 and the initial source we used for supplying bumble bees became unavailable with no warning. Despite best attempts, were unable to locate another source of native bumble bee species and, as a result, were unable to duplicate this portion of the project in 2017. Based on our 2016 results and the challenges we experienced obtaining a reliable supply of bumble bees for experimentation, we do not recommend supplementation with bumble bees at this time. A reliable source of native bumble bees must first become available. Supplementation with non-native bumble bee species is also not recommended (e.g., *B. impatiens* and *B. occidentalis*) due to concerns that they may displace native species and/or vector viruses that could be detrimental to native pollinating species. However, on-farm practices that promote native bumble bee species and other pollinators in the farm landscape may bring growers pollination benefits and is a potential area of future study.

**Table 2.** Fruit set, berry weight, and estimated yield of ‘Draper’ blueberry with and without supplementation from bumble bees, 2016.

<table>
<thead>
<tr>
<th>Treatments</th>
<th>Fruit set (%)</th>
<th>Berry weight (g/berry)</th>
<th>Estimated yield (lbs/plant)</th>
</tr>
</thead>
<tbody>
<tr>
<td>4 hives/A (control)</td>
<td>81.77</td>
<td>3.04</td>
<td>3.37</td>
</tr>
<tr>
<td>4 hives/A + 3 hives/A <em>vosnesenskii</em></td>
<td>81.83</td>
<td>2.98</td>
<td>3.97</td>
</tr>
<tr>
<td><em>P</em>-value</td>
<td>ND(^z)</td>
<td>NS</td>
<td>NS</td>
</tr>
</tbody>
</table>

\(^z\) Denotes not statistically different at *α* = 0.05.

**Figure 3.** Total honey bee and bumble bee visitation in 2016. Sites 1-3 were stocked with 3-4 hives/acre of honey bee (control), whereas sites 4-6 were stocked with 4 hives/acre honey bee +
3 hives/acre commercially-raised bumble bees.

Pheromone Experiment. No differences in honey bee visitation rates (Fig. 4), fruit set, estimated yield, or berry characteristics (berry weight, seed number, and firmness) were observed relative to untreated plants (data not presented). As is standard for field experiments and because pheromone application is such a widespread practice in commercial blueberry production in western Washington, additional years of field trails and experimentation with more commercially available pheromones is recommended.

![Figure 4](image-url) Honey bee visitation rate in ‘Draper’ blueberry treated with Bee Scent pheromone and a water control, 2017.

ANTICIPATED BENEFITS AND INFORMATION TRANSFER
We have been active in sharing project information among growers and the scientific community (please see the list of publications and presentations below). The WSU Small Fruit Horticulture Program has been focusing on techniques to promote pollination, fruit set, and corresponding yield since its beginning under DeVetter’s leadership in 2014. She had a PhD student focusing on this specific issue (Matt Arrington, who graduated December 2017) and has recruited another MS student who will continue studying this important topic (Weixin Gan).

Combined with other studies, we have shown that reduced pollinator activity under climactic conditions that do not favor their foraging limits yield in western Washington highbush blueberry. We have also shown in related research funded by the Washington Blueberry Commission that foliar applications of boron and calcium applied in the spring as a pre-bloom spray does not improve fruit set and yield components when plants are not deficient in these nutrients (however, collaborative work led by Gerbrandt has demonstrated that foliar calcium applications can reduce premature fruit drop in ‘Draper’). Augmenting honey bee hive density has shown to increase overall honey bee visitation rates and corresponding yield components. Combined, these results show yield deficits experienced during the bloom and fruit set window are primarily driven by a lack of insect-mediated pollination, not deficiencies in boron and calcium (barring plants are not deficient in these nutrients). Results from this and related projects give growers the information and tools to consider modifying honey bee hive density in order to promote pollination and corresponding yield. Additional research evaluating the impacts of modified honey bee hive
density and placement in other commercially important cultivars and assessment of the honey bee hive density in which the point of diminishing returns is reached is recommended and will be pursued in future project proposals. Also, developing an improved understanding of the biology and environmental constraints to pollen across several commercially important cultivars of blueberry is recommended and will be pursued in future project proposals. Tools to promote pollination, such as pheromones, have been trialed in 2017 and to date show no benefit. However, future research is recommended in order to allow a robust evaluation of these potential tools that could have application to Washington blueberry growers.

PUBLICATIONS AND PRESENTATIONS

Publications:


Presentations:


We also have been and will continue to upload project information to the WSU Small Fruit Website (http://smallfruits.wsu.edu/). The site is currently undergoing a re-structuring and re-organization.

REFERENCES


ACKNOWLEDGEMENTS
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