Pollination of commercial Cranberry (*Vaccinium macrocarpon* Ait.) in Newfoundland by native and introduced bees.

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Executive Summary

Cranberry flowers must be pollinated by insects for fruit to develop and bees are their main pollinators. This report outlines research conducted to determine the bee species important for the pollination of cranberry in Newfoundland, and the utility of importing commercial bumble bees to supplement pollination of the cranberry flowers. Several bumble bee and solitary bee species were indicated to be important pollinating species. The use of commercial bees did not increase pollination on the cranberry farms studied. We recommend that this practice be reviewed for its economic practicality. Future research should be undertaken in a multi-year project to determine whether year-to-year fluctuations in native bee abundance will impact yields if farmers depend solely on native species, and examine ways to minimize such impacts. Several native species were observed inside the colonies of the commercial bees. This sets up the potential for disease transmission to native species. Research should explore whether that is a real concern. Finally, the honey bee population of Newfoundland is a valuable resource. Future research should determine the efficacy of honey bees for pollinating cranberries in Newfoundland.

Background and Rationale

Many native insect species, especially bees, are important pollinators of commercial food crops. There are at least 60 species of bees occurring in Newfoundland and Labrador (Hicks pers. observation). The number of bee species recorded from Newfoundland differs considerable compared to mainland Atlantic Canada where 156 species have been recorded (Sheffield et al. 2003; Sheffield et al. 2008). With the increasing interest, investment and expansion in the cranberry industry in Newfoundland, it is of critical importance that we understand the pollination requirements of the plant in Newfoundland. Like many fruit crops, cranberry is pollinated by bees that visit the flowers to collect both pollen and nectar. These bees inadvertently carry pollen to other flowers, which provides the only mechanism for the plant to transfer its pollen. Only cursory studies on the pollination systems in Newfoundland cranberry have occurred. Newfoundland's unique climate and its isolation from mainland areas have resulted in a bee fauna that is much smaller than other areas. This isolation also provides us with healthy bee populations at a time when native bee populations in other areas are being threatened by diseases (Staffen-Dewenter et al. 2005; Goulson et al. 2008).

In Newfoundland, the cranberry industry began in 2002 in a provincial government initiative to get farmers interested in alternative crops. Since that time there has been an infusion of federal and provincial funding to help launch this fledgling industry here. $3 million allocated by the NL government in 2008 established commercial cranberry bogs in several areas throughout the province. In that year, 172,365 kilograms of cranberries were produced with a farm gate value of $325,000. That was an increase of 370% from the value of the crop in 2006 (Gov. of NL online).

Some Newfoundland cranberry producers have imported the non-native bumble bee, *Bombus impatiens*, to supplement the pollination activities of native species with the goal to increase berry yield. While anecdotal observations by farmers are mixed on whether the importation of bees affects production, observations within blueberry farms indicates that supplementation with *B. impatiens* does not increase blueberry production (Hicks 2011). Plus, there are indications from other areas that supplementation with bees for increased pollination may not be necessary

(Brown and McNeil, 2006). The importation of exotic bee species may have serious negative long lasting ecological consequences. The commercially available bumble bees are known to harbor serious bee diseases that, if transmitted to our presently unaffected native bees, will have disastrous consequences (Colla et al. 2006; Graystock et al. 2013).

The purpose of this study was to determine the native bee species that are important for cranberry pollination in NL and to determine the utility of importing commercial bumble bees (*Bombus impatiens*) for supplementing pollination on NL cranberry farms. Our hypothesis is that supplementation of NL cranberry fields with *Bombus impatiens* increases the fruit-set.

Funding and Partnerships

In addition to the funding ($8000) provided by ARI, Department of Natural Resources, Government of NL, the Research & Development Corporation (RDC) of Newfoundland and Labrador provided $30,000 under the Regional Collaboration Research Initiative between Grenfell Campus (Memorial University) and College of the North Atlantic.

Methods

Location. Two cranberry plots were chosen on two separate commercial farms near Stephenville, NL (Fig. 1.). The Lomond farm (48°27’14.20”N; 58°24’28.98”W) was supplemented with 16 colonies (4 quads) of *Bombus impatiens* (supplied by Biobest Canada Ltd, Leamington, Ontario) (Fig. 2 & 3). The McFatridge farm (48°34’16.82”N; 58°31’31.35”W), located 15 km away, was un-supplemented. The fields chosen on both farms had the Pilgrim variety of cranberry.

*Native bee diversity and abundance***.** On 17 July 2013 two 45m transects were established inside each cranberry farm. Nine cup traps (350ml plastic cup; Fig. 4) of alternating blue, yellow and white cups were placed along transects at 5m intervals inside the cranberry plot at each farm. A similar 45m transect with 9 cup traps was placed along the adjacent berm on each farm. The cup traps were ¾ filled with a solution containing water and Dawn® dishwashing detergent. The addition of the detergent caused any insects that entered the cup to easily break the surface tension of the solution and quickly drown. All traps were cleared weekly until 21 August (5 weeks). In addition, 30-minute sweep net samples were taken at each cranberry plot on three separate dates (23 July, 8 August, 13 August). All insects collected in the cup traps and sweep net samples were taken back to the lab and pinned for identification.

Figure 1. Location of the study farms near Stephenville, Newfoundland

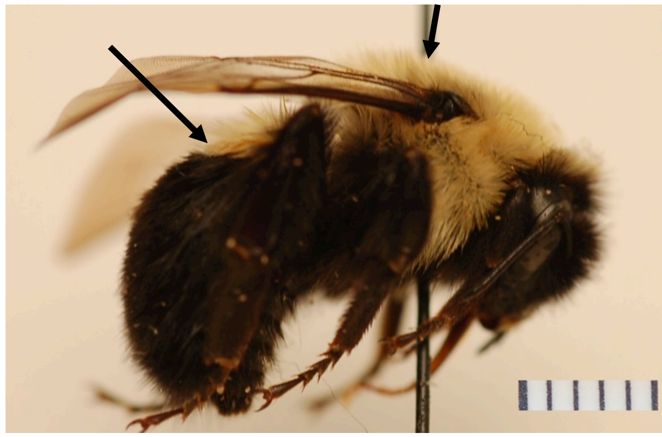

Figure 2. *Bombus impatiens* adult worker. Recognizing characters include: Abdomen entirely black except for yellow band in first segment and thorax without a black band. Scale = 5mm.

Figure 3. A "Quad" of *Bombus impatiens*. Four individual bee colonies make up a quad. The entrances of two colonies occur on opposite sides of the box.

Figure 4. A yellow cup trap placed in a cup stand located on the berm at McFatridge farm.

*Pollination***.** Starting on 23-24 July 2013, two 10m transects were established in the cranberry fields at each farm. At 50cm intervals, plants were tagged and their flowers counted. After the fruits had developed (21 September 2013), the fields were visited again and the number of fruit that developed on the tagged plants was counted. At the same time, 25 ripe fruit per plot were randomly sampled and taken back to the lab where their diameter, weight, and number of seeds was determined.

Pollination success by native bees. On 15 July 2013 two rows of four 1m^2 quadrat boxes were placed over the cranberry plants at 18.5m intervals. The quadrats were made of a wooden frame covered in fine mesh to exclude pollinators. During 80% bloom (McFatridge 29 July 2013; Lomond 1-2 August 2013), the mesh screens for each quadrat were removed for 1 hour and pollinator activity was observed directly in the quadrats. Visitors to the exposed un-pollinated flowers were identified to the lowest practical taxonomic level. Any flowers visited by a bee were individually covered with small screens to exclude future visits. Fruit that developed on these visited flowers was counted, removed and returned to the lab on 21 September, and the diameter, weight and number of seeds were determined for each fruit.

Density of native bee nests. The density of native solitary bee nests was determined by walking along a 10m transect on the berm of each farm and counting the number of nest entrances in a $1m^2$ area at 1m intervals (i.e. 10 observations per transect). Two different 10m transects per farm were observed on three separate dates (23 July, 8 August and 13 August).

Sampling of environmental variables. On 22 July, 5 August and 12 August 2013, 10 soil samples were taken along each transect occurring on the berms. A standard soil core sampler was used that extracted 78.5 cm^3 of soil to a depth of 25cm. Each soil sample was placed into a sealed plastic bag for transport to the laboratory. Upon arrival, the wet soil sample was weighed; dried in an oven at $60^{o}C$ for 5 days and then re-weighed to calculate the percentage soil moisture.

Data handing. Shannon-Wiener diversity indexes were calculated for each treatment using PAST online calculator (Hammer et al. 2001). PAST was used to compare the diversity indexes between the two farms using a t-test described by Poole (1974). A one way analysis of variance was used to compare means of variables (i.e. fruit-set and environmental measurements) after the variance was checked for normality. Proportional data and data that did not turn out to have the variance normally distributed, were transformed (arcsin and Log10). All statistical tests were performed in Minitab© version 15.

Results and Discussion

While the two farms that were chosen were spatially close, there were considerable differences in site characteristics. On the McFatridge farm, the cranberry plots were composed of organic soil (peat) with a sand layer over the top. The McFatridge berms retained the peaty soil and had a high diversity and abundance of herbaceous and woody shrubs (Alders and Blueberry) (Figure 3a). The Lomond fields appeared to have more sand over the top of the organic soil. The

Lomond berms were composed almost entirely of piled sand (see figure 3b) and because vehicles used the top of the berm the soil there was considerably compacted with little vegetative growth. The soil moisture measurements indicated that there were significant differences between the two farms. The soil moisture on McFatridge berms was 81.9% ± SE=3.42 (N = 30) compared to 22.6% ± SE=15.12 (N = 30) on the Lomond berms (F= 439.74, P = <0.001).

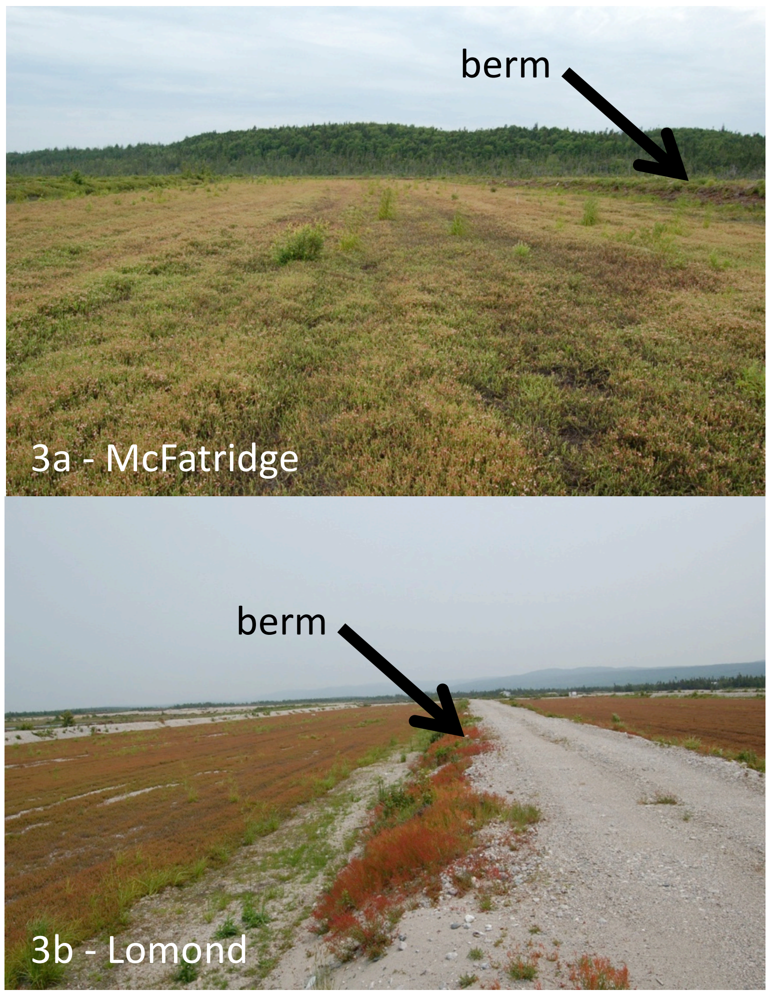

Figure 3. The cranberry farm located in western Newfoundland. A - McFatridge; B – Lomond. Note the difference in site characteristics between the two farms.

The abundance of bees collected in the cup traps from transects located on the cranberry plots (field) was similar between the supplemented and un-supplemented farms (Table 1). The abundance of the bees collected was higher on the Lomond berms than on the McFatridge berms. We feel that this is an anomaly because the Lomond berms had considerably reduced vegetative cover compared to McFatridge berms. The increased diversity and abundance of plants on the McFatridge berms should be more attractive to native bees and provide greater forage and nesting habitat. The higher abundance of bees collected in the cup traps from the Lomond berms is probably the result of the cups being more conspicuous to the bees on the bare berms.

Table 1. Abundance of bees collected in cup traps on Cranberry farms in western Newfoundland during July and August 2013.

Date	Lomond		McFatridge	
	Field	Berm	Field	Berm
Jul 25	10	42	13	16
Jul 31	15	82	16	34
Aug 7	42	108	26	11
Aug 14	21	71	30	20
Aug 21	9	16	23	16
Totals	97	319	108	97
% of total	23.3	76.7	52.7	47.3

While the abundance of bees appeared to be greater on the Lomond farm compared to the McFatridge farm, this was not reflected in either the bee species richness, the Shannon diversity index or the % fruit set (Table 2). There was no difference in the numbers of species collected between the two farms. While Lomond had 20 species and McFatridge had 16, when you take into consideration that 2 of the recorded species from Lomond were introduced species (*Bombus impatiens* and *Apis mellifera*), there really is no difference in species richness. With regard to fruit set, the un-supplemented farm (McFatridge) had a greater percentage fruit set (62.2%) when compared to the supplemented farm (Lomond) (53%). Comparison of the means using ANOVA showed that they were marginally non-significantly different (F = 3.87, P = 0.053). Whether yield at the unsupplemented field was greater or the same, it suggests that supplementation of cranberry fields in Newfoundland is not effective in increasing fruit set. This supports Hicks (2011) who showed that supplementation of Newfoundland blueberry farms with *B. impatiens* did not increase fruit set. Moreover, the farmer-reported yields from the experimental fields supported the fruit set data and suggest that supplementation is not effective (McFatridge = 14,545 lbs/acre: Lomond = 12,500 lbs/acre).

Table 2. Species richness, Shannon diversity index, abundance of bees and fruit set on two farms of Cranberry located on the west coast of Newfoundland during Summer 2013.

Location	Richness	Diversity	Abundance on farm (Field + Berm)	% Fruit set (n)
Lomond (supplemented)	20†	1.432a‡	416*	53 (40)a
McFatridge (un-supplemented)	16	1.525a	205	62.2 (39)a

† This number includes the 2 non-native (introduced) bees, *B. impatiens* and *A. mellifera*
‡ Values in columns followed by the same letter are not statistically significant (p = 0.05)
*This value includes 25 specimens of the introduced bee, *B. impatiens* and *A. mellifera*.

The most common bumble bee species collected in both farms by cup traps (Table 3) and sweep netting (Table 4) was *Bombus ternarius*. This is a medium sized bee that has an orange band across the middle of its abdomen (Fig. 4). It is similar in appear to another bumble bee (*B. sylvicola*) but can be distinguished by its all black second last abdominal segment and with a mixture of black and yellow hairs on the top of its head. Hicks (2010) did not collect any *B. ternarius* on blueberry farms on the Avalon Peninsula. *Bombus ternarius* thus appears to be an important bumble bee species in the cranberry farms studied here.

Table 3. Bee species and total abundance collected in cup traps over a 5-week period (July 18-Aug 21, 2013) in cranberry farms in Western Newfoundland.

Species (Bowl traps)	Lomond field	Lomond berm	McFatridge field	McFatridge berm
Bombus ternarius	43	129	33	9
B. impatiens	6	15	0	0
B. terricola	4	9	1	0
B. frigidus	2	7	0	0
B. vagans bolsteri	4	9	3	1
B. borealis	1	1	4	2
B. rufocinctus	1	0	1	1
B. (Psithyrus) fernaldae	0	1	1	0
Apis mellifera	0	4	0	0
Lasioglossum (Dialictus) spp.	31	137	38	66
Lasioglossum leucozonium	1	1	21	9
Lasioglossum athabascense	0	2	0	1
Lasioglossum quebecense	1	0	0	3
Halictus rubicundus	0	1	0	0
Andrena carolina	1	0	1	1
A. rufosignata	0	1	0	0
A. frigida	0	1	0	3
A. hirticincta	1	0	2	0
A. vicina	0	1	0	0
Hylaeus modestus	0	0	2	0
Megachile frigida	1	0	0	1
M. melanophaea	0	0	1	0
Total Bees	97	319	108	97

Table 4. Bee species and total abundance collected by sweep netting in cranberry farms. One 30 minute sweep net at each farm per week for 5-weeks (July 18-Aug 21, 2013).

Species (Sweep net)	Lomond	McFatridge
Bombus ternarius	4	26
B. terricola	1	3
B. frigidus	2	2
B. vagans bolsteri	0	2
B. borealis	0	1
Lasioglossum (Dialictus) spp.	1	2
Andrena carolina	0	2
Osmia bucephala	0	1
Total Bees	8	39

Figure. 4. *Bombus ternarius*. Note the red band on the abdomen; black on the second last abdominal segment and the mix of black and yellow hairs on the top of the head. Scale = 5mm.

Bees in the *Lasioglossum* subgenus *Dialictus* (Halictidae) were also commonly collected (Fig. 5). These soil nesting solitary bees were common on Avalon Peninsula blueberry fields (Hicks 2010). Table 5 shows the efficiency of selected bee species for pollinating cranberry during a single flower visit. The Halictidae were the most efficient, pollinating 36.8% of the flowers that they visited once. Native *Bombus* species had a slightly lower but similar success rate (34.8%). It is interesting to point out in Table 5 that the imported *B. impatiens* observed had the lowest pollination success rate during a single flower visit (14.3%). The number of observations that we made was low so we must use caution in interpreting this data.

Figure 5. *Lasioglossum (Dialictus)* spp. (Halictidae). These bees are very small but important pollinators. Scale = 5mm.

Table 5. Pollinator success of selected bee species observed in cranberry farms in Western Newfoundland. Flowers were isolated once the bee visited them and later the fruit that developed was counted and measured.

Bee type	# of obs.	# of fruit	% Fruit set	Mean diameter (mm)	Mean Weight (g)	Mean # seeds/fruit
Bombus spp.	46	16	34.8	11.8	0.613	12.8
B. impatiens	14	2	14.3	11.9	0.654	14.0
Halictidae	19	7	36.8	10.4	0.421	9.5
Andrenidae	4	1	25.0	9.1	0.368	9.0

Because of the small number of observation, the mean diameter, mean weight and # of seeds/fruit were not significantly different among the bees observed (Table 5). Generally the larger bees that visited a flower appeared to produce larger fruit with a larger number of seed than the smaller bees. This trend, of larger bees pollinating flowers that then produce larger fruit has been observed in other agro-ecosytems (Javorek et al. 2002; Ratti et al. 2008).

The small species of soil nesting solitary bees collect pollen from the cranberry flowers to provide for their offspring. The female generally digs vertical galleries into soil and builds cells off from the main tunnel. In these cells, she lays an egg and then collects pollen and nectar that she places inside the cell. When she determines that enough provisions are supplied that will allow the bee larvae to grow to the pupal stage, she seals the cell and continues to construct other cells. The entrances to the main tunnels can be easy observed on the soil surface (Fig. 6) and the density of the nest entrances can be high (Fig. 7). The berms are the best habitat for nest construction mainly because these areas are not sprayed directly with insecticides or water during irrigation. A count of nest abundance on the berms of the farms showed that both farms had similar abundances along transects sampled (McFatridge = 1.3 nests/m^2; Lomond = 1.0 nests/m^2). Hicks (2010) suggested that people often overlook the smaller bees of the Halictidae and Andrenidae as important pollinators in general. The data presented in this study provides further evidence that the small bees are abundant in the fields and contribute to cranberry pollination.

Figure 6. The nest entrance of *Lasioglossum leucozonium* (Halictidae) on the berm of the McFatridge farm.

Figure 7. Four nest entrance located on the berm of the McFatridge farm. A 35mm lens cover is in the photo for size comparison and the nest entrance are indicated.

At the end of the flower period the quads of *B. impatiens* were frozen at -20^{o}C to kill all bees in the colonies. The colonies were opened and the contents searched to see the number of queens and workers. 58% of the colonies that were examined contained native bees. It is unknown why these native bees were inside the colonies of *B. impatiens*. The native species observed included *B. ternarius, B. terricola, B. frigidus,* and *B. vagans bolsteri*. In addition specimens of the honey bee (*A. mellifera*) were also observed inside the *B. impatiens* colonies. This is the first record, that we are aware of, showing the mixing of native bees inside of the commercially supplied bee hives.

Presently, Newfoundland is in an enviable position regarding its honey bee (*Apis mellifera)* population. The province has strict importation regulations and because of its geographical isolation it does not harbor the same parasites that plague honey bees in other areas worldwide (Williams *et al.* 2010). The parasites and diseases of native *Bombus* spp. have not been studied and we are unsure of their impact on native populations. The commercial *B. impatiens* harbor several pathogens and parasites that have been documented to spillover and infect native species (Colla *et al.* 2006). Recently Greystock et al. (2013) showed that the diseases occurring in commercial bumble bees can be transmitted to native bumbles and to honey bees. Until now, we did not think that bumble bee diseases could cross over to honey bees. With the observation of several native bumble and honey bee specimens inside the colony boxes of *B. impatiens*, it sets up a perfect avenue for the transmission of diseases to the native species and honey bees. While it is unclear what impact that the parasites and diseases may have on native bee species, they have been implicated as the cause of the decline of important bee pollinators in North America (Berenbaum *et al.* 2007).

Communication and Outreach

The data collected were presented at the Atlantic Cranberry Growers association conference in St. John's, March 19-21, 2014. Title of the invited talk was "Is the importation of Commercial bumble bee practical for NL cranberry pollination?" There were approximately 100 people including farmers, government officials and technicians in attendance.

Conclusion and Future Recommendations

Supplementation of cranberry fields with commercially supplies *B. impatiens* bees did not increase fruit set. The practice of buying non-native bees for boosting yields is not supported by the data and should be reconsidered in the future.

Bombus ternarius was the most abundant bumble bee species on the cranberry farms and appears to be an important pollinator of this crop. In addition, *Lasioglossum (Dialictus)* spp. (Halictidae), a small native bee, is also abundant and important as a pollinator of cranberry.

The limitations of this study stem from the fact that it occurred over one field season. Further research should be undertaken to verify the claims outlined in this report.

The transmission of bee disease from commercially supplied bees to native bees is a possibility and would be a disastrous consequence that would undoubtedly negatively impact Newfoundland's already small bee fauna. Moreover, the possible transmission of diseases to honey bees will limit the unrealized importance that these bees have on a global scale. It is because the Newfoundland honey bee population is presently healthy that further research should be initiated to grow the industry as a global source of clean queen bees.

The decline in native species for various reasons (see Colla and Packer 2008) may open up new niches that may be filled by exotic species. With global warming, Newfoundland may be at greater risk of having non-native bees establish, as the climate becomes milder allowing exotic bees to survive here when historically they were excluded. In Newfoundland, cranberry plants rely on native bee species for their pollination. The loss of native species by diseases and competition with exotic species may result in significant changes to the island's ecosystem.

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