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2015 Wild Blueberry Project Reports

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FOOD SAFETY AND NUTRITION

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1. TITLE: Increasing the food safety margin of wild blueberries through improved intervention measures.

OBJECTIVES:
1. To evaluate the effectiveness of different chemical sanitizers using short contact times in combination with freezing on microbial reduction; and
2. To develop an effective double spraying system for microbial reduction by combining two chemical sanitizers along with freezing.

METHODS: A bacterial cocktail with two strains each of *S.* Typhimurium (ATCC 6962 and ATCC 14028) and *L. monocytogenes* (ATCC 19115 and ATCC 49554) was used to inoculate the surface of blueberries by dipping method. Frozen blueberries (25g) without prior washing or decontamination were placed on sterile petri dish and inoculated with 2.5ml of bacterial cocktail suspension prepared for each pathogen. The inoculated blueberries were shaken for 2 minutes at 160rpm (Barnstead Thermolyne, Roto Mix-Type 50800) to allow the attachment of pathogens to blueberries. After 2 min the cocktail liquid is drained out and the berries were dried for about 1-2 hours in a laminar flow hood. The initial level of inoculum on surface of inoculated blueberries was approximately 7 log CFU/g for *S.* Typhimurium and *L. monocytogenes*. Fresh solutions of chemicals in distilled water were prepared the same day of each experiment. The treatments tested included: chlorine (Cl_2, 200ppm), aqueous chlorine dioxide (ClO_2, 15ppm), and lactic acid (2%). The control treatments include distilled water wash and un-treated inoculated blueberries. To imitate industrial setup similar to those used in blueberry processing, a portable conveyer belt with a fixed overhead spray modified with whirlijet nozzle was designed and used to do the treatments. Inoculated blueberry samples were spread on the conveyer belt and 150ml sterile distilled water (control) or different chemical solutions at different concentrations (ClO_2, 15ppm, Cl_2, 200ppm and lactic acid, 2%) were sprayed from a designated height while the berries were rotated and moved on the conveyer belt as shown in figure-1. The treated blueberries were left on the conveyer belt for different contact times (10sec, 1min and 3min). To initiate the end of each contact time, blueberries from wire screens after contact time are transferred to sterile stomacher bags. To evaluate the efficiency of these chemicals combined with frozen storage, after each treatment time, one set of blueberries was stored at -17°C for 1 week. Bacterial enumeration was conducted before and after freezing.

RESULTS: ***Objective 1*****:** Evaluating the effectiveness of different chemical sanitizers using short contact times in combination with freezing on microbial reduction:

The efficacy of all these sanitizers used in this study increased significantly ($p < 0.05$) in inactivating foodborne pathogens, when combined with freezing at -17°C for 1 week. Treatment

with sterile deionized water did not significantly reduce the levels of the pathogens ($p > 0.05$) as compared with all sanitizer treatments.

Efficacy of chemical sanitizers in reducing L. monocytogenes before freezing:

Log reduction of *L. monocytogenes* on blueberries treated with different chemical sanitizers (Cl_2, ClO_2, and lactic acid) before freezing is shown in figure-2. Cl_2 wash at 200ppm resulted in only < 2 log reduction. There is no significant difference in log reduction from 10sec to 1min and 3min contact times. Distilled water wash resulted in < 1 log CFU/g reduction in *L. monocytogenes* populations. Treatment with aqueous chlorine dioxide (ClO_2) for 3min resulted in 2.7 log reduction before freezing. Contact times of 10sec and 1min resulted in 2.2 log and 2.5 log CFU/g reduction respectively. Lactic acid treatment at 3min contact time showed 2.84 log CFU/g log reduction. Log reduction of 2.2 and 2.4 log CFU/g was achieved with 10sec and 1min treatment, respectively.

Efficacy of chemical sanitizers in reducing L. monocytogenes in combination with freezing at -17°C for 1 week:

The population reductions of *L. monocytogenes* from the surface of blueberries after treatment with different concentrations of Cl_2, ClO_2, and lactic acid sanitizers in combination with freezing is presented in figure-3. Cl_2 wash at 200ppm with 3min contact time when combined with freezing showed > 5 log reduction. Though there is increase in log reduction with increase in contact time, the 1min contact time did not shown any significant difference (($p > 0.05$) from 10sec treatment time. Treatment with aqueous chlorine dioxide (ClO_2) for 3min resulted in 2.7 log reduction before freezing and when combined with freezing the log reduction significantly increased to 5.5 log CFU/g. Exposure time of 1min with ClO_2 in combination with freezing showed > 5.0 log CFU/g reduction indicating shorter exposure times of ClO_2 are equally effective. With lactic acid treatment in combination with freezing, at 3min contact time, > 6.0 log CFU/g reduction was noted. Log reduction > 4.5 log CFU/g was achieved with lactic acid treatment in combination with freezing at shorter contact times indicating lactic acid is very effective even for shorter exposure times. *L. monocytogenes* populations were best reduced with lactic acid treatment in combination with freezing followed by ClO_2 and Cl_2 (LA> ClO_2 > Cl_2).

Efficacy of chemical sanitizers in reducing S. Typhimurium before freezing:

Log reduction of *S.* Typhimurium on blueberries treated with different chemical sanitizers (Cl_2, ClO_2, and lactic acid) before freezing is shown in figure-4. Around 1.5-1.8 log CFU/g reduction in population of *S.* Typhimurium was noted with 200ppm Cl_2 treatment. Though there is very little increase in log reduction with increase in contact time, there is no significant difference ($p > 0.05$) in log reduction from 10sec to 1min and 3 min contact times. Distilled water wash resulted in < 1 log CFU/g reduction. Aqueous chlorine dioxide (ClO_2-15ppm) treatment resulted in 1.8-2.5 log CFU/g reduction in *S.* Typhimurium populations. There is significant increase ($p < 0.05$) in log reduction with increase in contact times. Lactic acid (2%) treatment at 3min contact time showed 2.3 log CFU/g log reduction. Log reduction of > 2 log CFU/g was achieved with 10 sec and 1min treatment.

Efficacy of chemical sanitizers in reducing S. Typhimurium in combination with freezing at -17°C for 1 week:

The population reductions of *S.* Typhimurium from the surface of blueberries after treatment with different concentrations of Cl_2, ClO_2, and lactic acid sanitizers in combination with freezing is presented in figure-5. Treatment with Cl_2 wash (200ppm with 3min contact time) in combination with freezing resulted in 5.3 log CFU/g reduction. Contact times of 10sec and 1min resulted in only 3.7 and 4 log CFU/g reduction respectively indicating there is no significant difference ($p > 0.05$) between them. Treatment with aqueous chlorine dioxide (ClO_2-15ppm) for 3min when combined with freezing showed a log reduction of 4.8 log CFU/g. contact time of 1min with ClO_2 in combination with freezing showed 4.6log CFU/g reduction and 10sec showed only 4.4 log CFU/g reduction. Lactic acid treatment in combination with freezing, at 3min contact time resulted in 4.5 log CFU/g reduction. Though there is no significant difference in log reductions of 10sec and 1min after freezing, they resulted in < 4.0 log CFU/g reduction. *S.*Typhimurium populations were best reduced with chlorine treatment in combination with freezing followed by ClO_2 and lactic acid (Cl_2> ClO_2 >LA).

***Objective 2*:** To develop an effective double spraying system for microbial reduction by combining two chemical sanitizers along with freezing.
Study still in progress; no results at this time.

CONCLUSIONS: The efficacy of these sanitizers is increased significantly in inactivating foodborne pathogens, when combined with freezing. *L. monocytogenes* populations were best reduced with lactic acid treatment in combination with freezing followed by ClO_2 and Cl_2 (LA> ClO_2 > Cl_2) while *S.*Typhimurium populations were best reduced with chlorine treatment in combination with freezing followed by ClO_2 and lactic acid (Cl_2> ClO_2 >LA). Lactic acid (2%) treatment and 3min contact time resulted in greater than 6 log CFU/g reduction of *L. monocytogenes*. Maximum log reduction of *L. monocytogenes* obtained with chlorine dioxide (15ppm) and chlorine (200ppm) treatment in combination with freezing is 5.4 log CFU/g and 5.3 log CFU/g respectively. Greater than 5 log CFU/g reduction of *S.* Typhimurium was achieved with chlorine at 3min contact time. Chlorine dioxide and lactic acid treatments in combination with freezing are almost equally efficient in reducing *S.* Typhimurium. They resulted in >4.5 log CFU/g reduction. Our results conclude that, industrial treatment times when used with appropriate and low dosage chemical sanitizer, in combination with freezing can effectively reduce foodborne pathogens to 5 log CFU/g.

RECOMMENDATIONS: Our approach which imitates industrial conditions is worth to be considered by Maine Wild Blueberry industries, where these conditions can be easily incorporated to possibly eliminate pathogens more effectively.

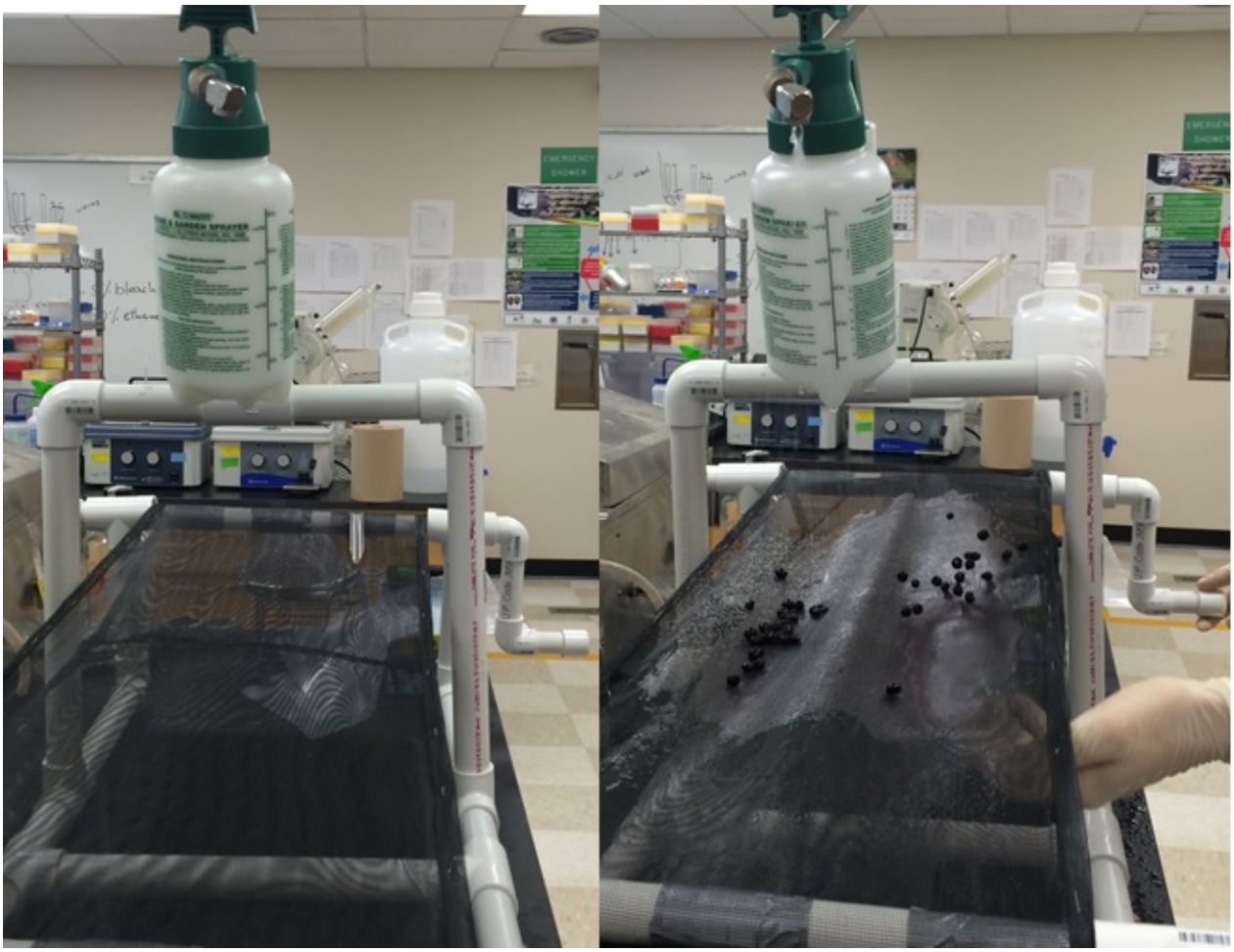


Figure 1: Application of sanitizers (A) Portable conveyer belt designed with a fixed overhead spray modified with whirlijet nozzle (B) Inoculated blueberries were spread on the conveyer belt and sprayed with 150ml sanitizer while the berries were moved on the conveyer bed while spraying

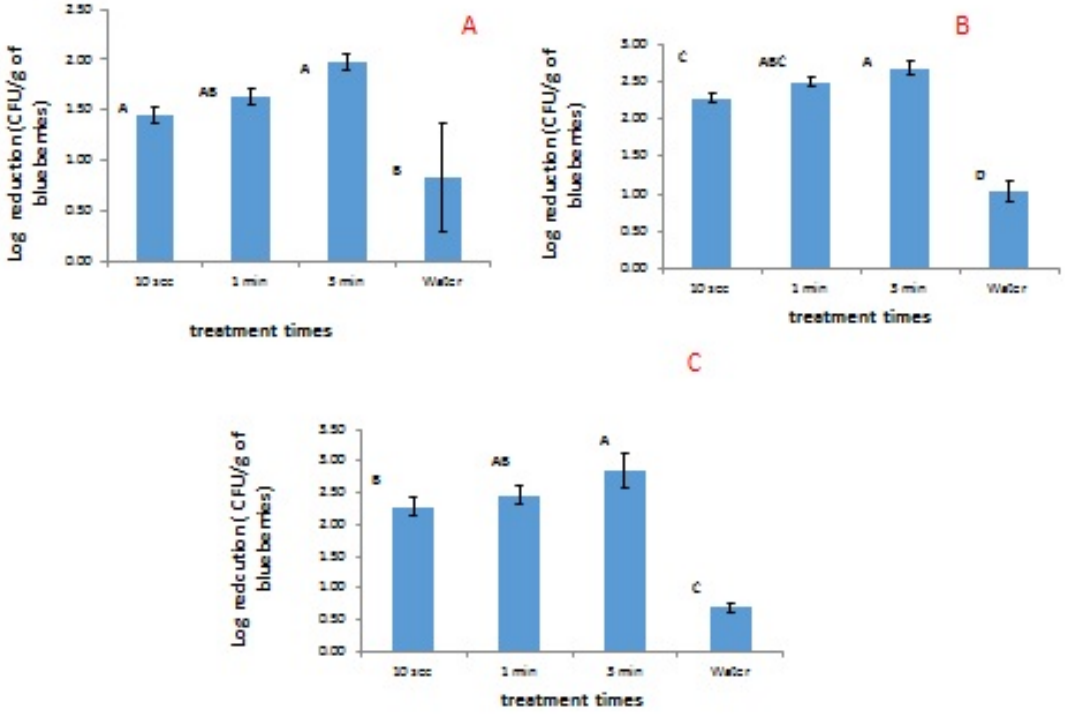


Figure 2: Log reduction of *L. monocytogenes* on blueberries treated with different chemical sanitizers before freezing.
(A) Chlorine (200ppm) (B) Chlorine dioxide (15ppm) (C) Lactic acid (2%). Mean values in the same row with different capital letters (A through D) are significantly different among various treatment times of the same concentration of the chemical treatment.

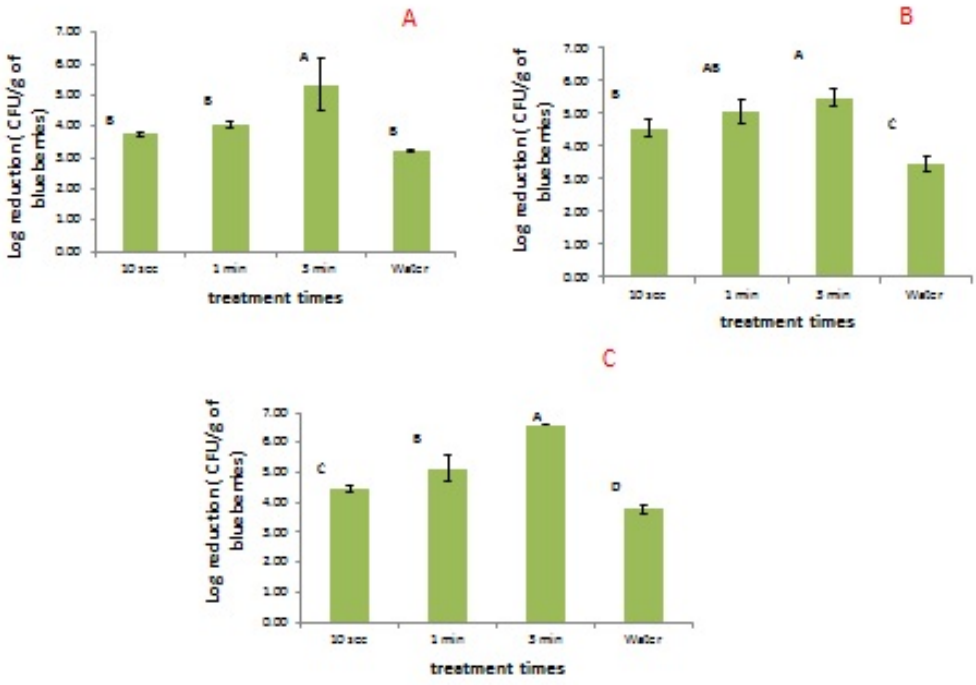


Figure 3: Overall log reduction of *L. monocytogenes* on blueberries treated with different chemical sanitizers in combination with freezing treatment at -17°C for 1 week.

(A) Chlorine (200ppm) (B) Chlorine dioxide (15ppm) (C) Lactic acid (2%). Mean values in the same row with different capital letters (A through D) are significantly different among various treatment times of the same concentration of the chemical treatment.

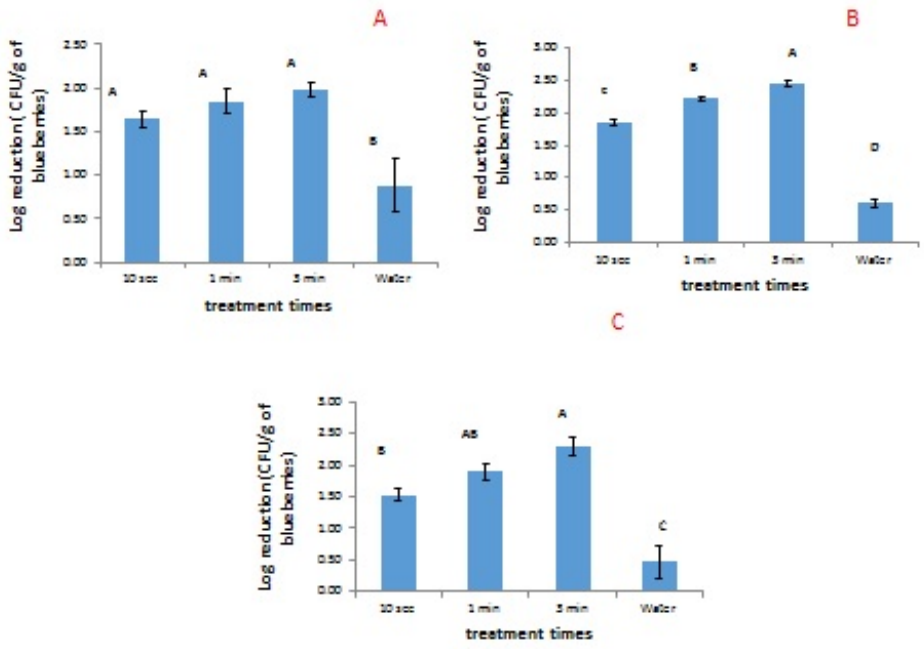


Figure 4: Log reduction of *S.* Typhimurium on blueberries treated with different chemical sanitizers before freezing.
(A) Chlorine (200ppm) (B) Chlorine dioxide (15ppm) (C) Lactic acid (2%). Mean values in the same row with different capital letters (A through D) are significantly different among various treatment times of the same concentration of the chemical treatment.

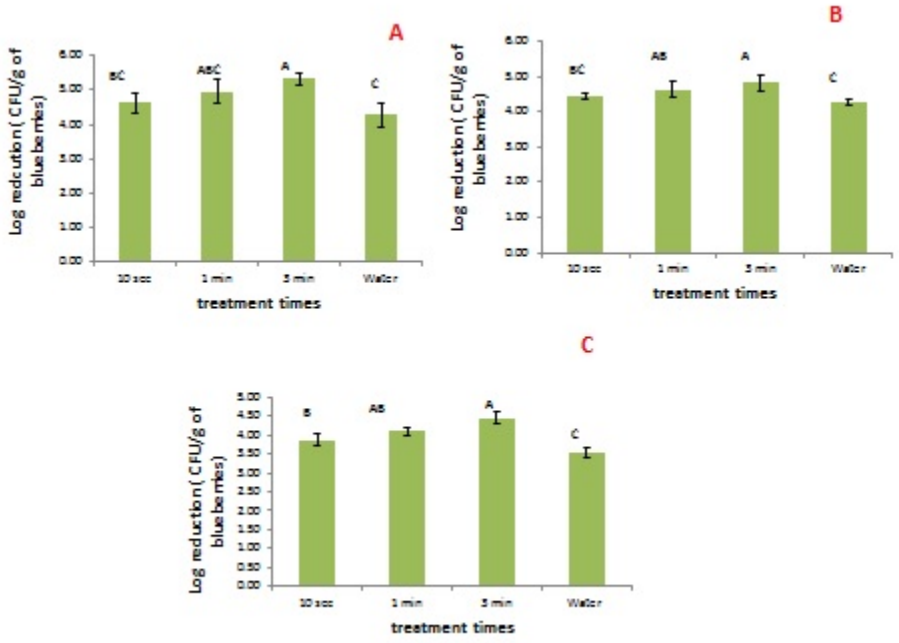


Figure 5: Overall log reduction of *S.* Typhimurium on blueberries treated with different chemical sanitizers in combination with freezing treatment at -17°C for 1 week.
(A) Chlorine (200ppm) (B) Chlorine dioxide (15ppm) (C) Lactic acid (2%). Mean values in the same row with different capital letters (A through D) are significantly different among various treatment times of the same concentration of the chemical treatment.

ENTOMOLOGY

INVESTIGATORS: F. A. Drummond, Professor of Insect Ecology/Entomology
J. A. Collins, Assistant Scientist of Insect Pest Management
E. Ballman, Research Associate in Invasive Species / Entomology

2. TITLE: I. Control tactics for blueberry pest insects, 2015.

Study 1. Field control of blueberry tip midge on wild blueberry

METHODS: Materials were applied in 25 gallons of water-mixture per acre with a CO_2-propelled, 80-inch boom sprayer (76-inch swath) equipped with four, flat-spray, 8002VS TeeJet® nozzles operating at 35 psi and at a slow walking speed. Walking speed for each application was regulated using a metronome.
On sample dates as indicated in the table, damage was assessed by counting the number of blueberry stems with and without tip midge damage as evidenced by curled leaves from each of three, ft^2 samples per treated plot. There were four replications of each material and four untreated checks. Plot size for all trials was 7 x 20 ft.
Rimon® 0.83EC (novaluron), Movento® SC (spirotetramat), Exirel® SE (cyazypyr), Sivanto® 200SL (flupyradifurone), and Assail® 30SG (acetamiprid) were applied on 27 May to a pruned-year field. A second application of each was made on 5 Jun. Blueberry stems were scattered and < 1 inch tall on 27 May and 1 to 1.5 inches tall on 5 Jun. Infestation was assessed on 12 and 25 Jun. No symptoms of phytotoxicity were observed in any plot.

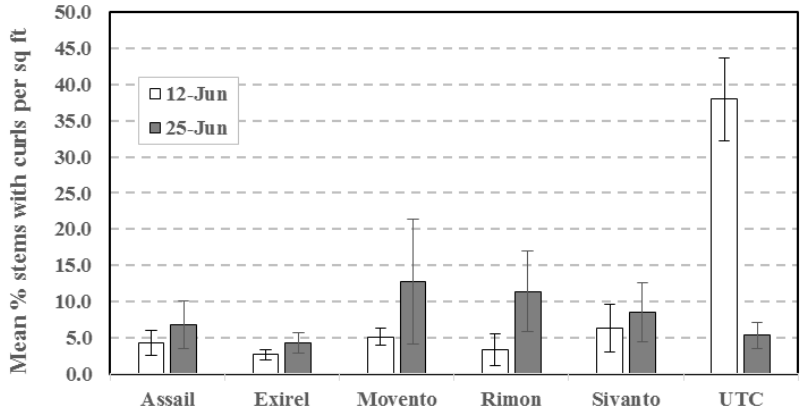
RESULTS: Subplots were pooled within main plots. Data were transformed by arcsin to stabilize variance prior to analysis. Analysis of Variance (ANOVA, RCB) and LSD ($P \leq 0.05$) were used to compare mean number of curls among the treatment plots. Rimon, Movento, Exirel, Sivanto, and Assail were all effective, at least initially, in suppressing tip midge populations. Significantly fewer damaged stems were found in the treated plots compared with the untreated checks (UTC) on the first sample date on 12 Jun ($F_{(5,15,)} = 19.31$, $P = < 0.0001$). By the second sample date on 25 Jun there appeared to be a trend towards increased numbers of damaged stems in treated plots compared with the non-treated controls; however, the difference was not significant ($F_{(5,15\ )} = 0.56$, $P = 0.7260$) (Table 1 and Fig. 1)

Table 1. Field control of tip midge with insecticides, summary, 2015.

Material	Amt. form./acre	Mean percent stems with curls/ft^2 (SE) 12 Jun	25 Jun
Assail 30SG	5.3 oz	4.3 (1.8) b	6.8 (3.3)
Exirel SE	20.5 oz	2.7 (0.7) b	4.3 (1.4)
Movento SC	10.0 oz	5.1 (1.2) b	12.8 (8.6)
+ Dyne-Amic	(0.5% v/v)		
Rimon 0.83EC	12.0 oz	3.4 (2.2) b	11.4 (5.6)
Sivanto 200SL	14.0 oz	6.4 (3.3) b	8.5 (4.0)
Non-treated check (UTC)	-	38.0 (5.7) a	5.3 (1.8)

Means within columns followed by the same letter are not significantly different (Tukey's, $P \leq 0.05$). Data were transformed by arcsin prior to analysis.

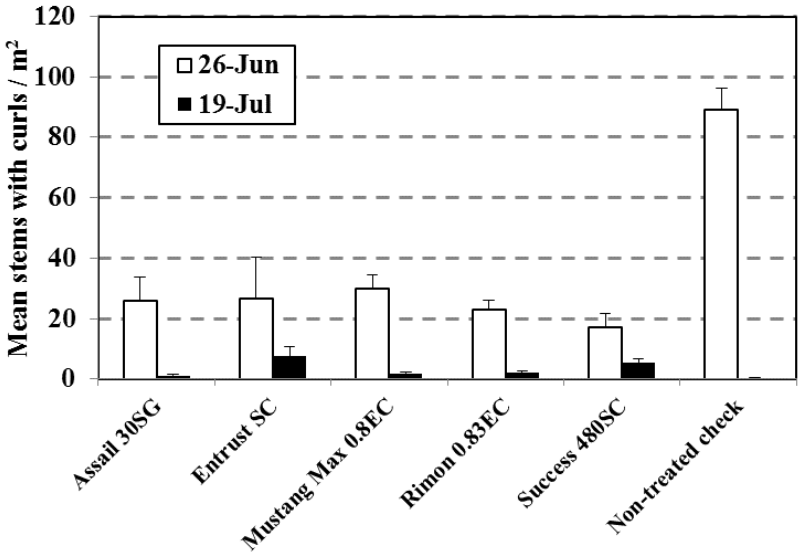
Fig. 1. Mean percent stems with curls/ft^2 (lines are standard error of the mean).



CONCLUSIONS: Very similar results were obtained from our 2014 trial (Fig. 2). In 2014 two applications of Rimon, Success® 480SC, and Entrust SC® or one application of Assail and Mustang Max® 0.8EC were all initially effective in suppressing tip midge infestation. Significantly fewer damaged stems were found in the treated plots compared with the untreated checks (UTC) on the first sample date on 26 Jun ($F_{(5,15)} = 7.50, P = 0.0010$). Although the number of damaged stems in all the plots was much lower by the second sample date on 18 Jul, it did appear that tip midge populations in the treated plots had rebounded somewhat. Plots treated with Entrust and Success (formulations of spinosad), Rimon, and Mustang Max (zeta-cypermethrin) all had significantly MORE damaged stems than the untreated check plots ($F_{(5,15)} = 7.16, P = 0.0013$).

These results continue to point to the possibility that tip midge populations are under some degree of biological control that contains their damage a small amount, but when insecticides are applied and these agents are eliminated then the sprayed plots receive higher degrees of damage.

Fig. 2. Mean number of stems with curls/m^2 (lines are standard error of the mean), data from 2014.



Study 2. <u>Efficacy and phytotoxicity of biopesticides for management of spotted wing drosophila (SWD) in wild blueberry</u>

METHODS: There were four blocks (replications) divided between two sites (Blocks 1 and 2 - TWP 18, ME; Blocks 3 and 4 - Blueberry Hill Farm, Jonesboro, ME). The site in TWP 18 is commercially managed land in the process of being reclaimed for wild blueberry production. Plot size for all treatment plots was 20 x 40 ft. There were no buffers between plots. Materials, rates, application timing, and dates of application are in Tables 1 and 2. Sivanto 200SL, Delegate® WG, and AzaSol® were added to the test substances for interest.

Each material was applied in 25 gallons of water-mixture per acre with a CO_2-propelled, 80-inch boom sprayer (76-inch swath) equipped with four, flat-spray 8002VS TeeJet® nozzles operating at 35 psi and at a slow walking speed. Speed was regulated using a metronome. Materials were applied in three passes, alternating the direction of each pass. Dates and weather at application are in Table 3.

Initial adult activity was determined from four, sugar-yeast baited traps placed at each site. Each trap consisted of a red, plastic Solo® cup with ca. 4 oz of standard yeast bait (1 tbsp yeast + 4 tbsp sugar + 12 oz water). First SWD were observed at Jonesboro on 10 Aug, and at TWP 18 on 4 Aug. Following first observed adult activity, red cup traps filled with ca. 3 oz apple cider vinegar were deployed in the center of each treatment plot, one per plot; traps were changed immediately prior to each application; and adults were collected by decanting the liquid through cheesecloth, then placing the cheesecloth in zip-lock bags and freezing for later evaluation. Traps were placed in each plot on 7 Aug at TWP 18 and 10 Aug at Jonesboro.

Larval infestation was evaluated by sampling fruit from each treatment plot. Each sample consisted of ca. 2/3 cup of fruit harvested from the center area of each plot using a commercial blueberry rake. Samples were processed for larvae using the Salt Extraction Method. Sample dates are in Table 4.

Table 1. Test/control substances, rates, and timing of first application.

Treatment [a]	**Rate**	**Timing of first application**
UTC	-	-
Delegate WG	6 oz	At first fruit infestation
Sivanto 200SL	10 oz/acre	At first fruit infestation
Sivanto 200SL	14 oz/acre	At first fruit infestation
Entrust SC	4-6 fl oz/acre	At first signs of adult activity
Grandevo	3 lbs/acre	At first signs of adult activity
Venerate XC	2 qts/acre	At first signs of adult activity
Entrust SC + Grandevo	4-6 fl oz/acre + 3 lbs/acre	At first signs of adult activity
Entrust SC + Venerate XC	4-6 fl oz/acre + 2 qts/acre	At first signs of adult activity
CimeXa Silica Gel Dust	5 lbs/acre	At first signs of adult activity
CimeXa Silica Gel Dust	10 lbs/acre	At first signs of adult activity
Entrust SC + CimeXa Silica Gel Dust	4-6 fl oz/acre + 5 lbs/acre	At first signs of adult activity
Venerate XC + CimeXa Silica Gel Dust	2 qts/acre + 5 lbs/acre	At first adult activity
AzaSol	6 oz/acre	At first fruit infestation
AzaSol	6 oz/acre	At first adult activity

[a] Veratran D was dropped from trial due to issues with the application. The material would not dissolve into solution and repeatedly clogged sprayer and nozzles.

Table 2. Application summary.

			Application dates						
Treatment	**Rate**	**Application summary**	**10-Aug**	**17-Aug**	**23-Aug**	**31-Aug**	**6-Sep**	**16-Sep**	**4-Oct**
UTC									
Delegate WG	6 oz	2 applications, at first infestation						x	x
Sivanto 200SL	10 oz/acre	2 applications, at first infestation						x	x
Sivanto 200SL	14 oz/acre	2 applications, at first infestation						x	x
Entrust SC	4-6 fl oz/acre	2 applications 7 days apart, at first sign of adult activity	x	x					
Grandevo	3 lbs/acre	5-7-day interval, at first sign of adult activity	x	x	x	x	x	x	x
Venerate XC	2 quarts/acre	Every 5-7 days, at first sign of adult activity	x	x	x	x	x	x	x
Entrust SC + Grandevo	4-6 fl oz/acre + 3 lbs/acre	Entrust every 7 days for first 2 applications. Followed by 2 applications of Grandevo every 5-7 days. Continue season long rotation.	x	x	x	x	x	x	x
Entrust SC + Venerate XC	4-6 fl oz/acre + 2 qts/acre	Entrust every 7 days for first 2 applications. Followed by 2 applications of Venerate every 5-7 days. Continue season long rotation.	x	x	x	x	x	x	x
CimeXa Silica Gel Dust	5 lbs/acre	Every 5-7 days, at first sign of adult activity	x	x	x	x	x	x	x
CimeXa Silica Gel Dust	10 lbs/acre	Every 5-7 days, at first sign of adult activity	x	x	x	x	x	x	x
Entrust SC + CimeXa Silica Gel Dust	4-6 fl oz/acre + 5 lbs/acre	Entrust every 7 days for first 2 applications. Followed by 2 applications of Silica every 5-7 days. Continue season long rotation.	x	x	x	x	x	x	x
Venerate XC + CimeXa Silica Gel Dust	2 quarts/acre + 5 lbs/acre	Tank mix between Venerate and Silica dust. Every 7 days	x	x	x	x	x	x	x
AzaSol	6 oz/acre	2 applications, at first infestation						x.	x
AzaSol	6 oz/acre	Every 5-7 days, at first sign of adult activity	x	x	x	x	x	x	x

Table 3. Application dates and weather.

Date	Time	% Cloud Cover	Wind Speed (mph)	Direction	Temp (F)	Site
10-Aug	7:00 AM	<10%	calm		70	TWP 18
17-Aug	7:00 AM	clear	1.4	SW	76	Jonesboro
17-Aug	9:30 AM	clear	2.8	S	78	Jonesboro
23-Aug	9:00 AM	100	0.7	N	70	TWP 18
23-Aug	11:30 AM	50	calm		83	Jonesboro
31-Aug	9:00 AM	25	2.2	WSW	73	TWP 18
31-Aug	12:45 PM	25	3.9	S	86	Jonesboro
6-Sep	9:15 AM	25	3.5	N	71	TWP 18
6-Sep	12 noon	25	1.3	NW	88	Jonesboro
16-Sep	11:00 AM	25	5 to 8	SW	65	Jonesboro
4-Oct	7:00 AM	25	5 to 8	SW	43	Jonesboro

Table 4. Sample dates; adults and larvae (fruit samples).

Sample date	TWP 18		Jonesboro	
	Adults	Fruit	Adults	Fruit
7-Aug		X prespray		
10-Aug				
14-Aug				X Prespray
17-Aug	X		X	
23-Aug	X	X		
31-Aug	X		X	X [a]
6-Sep	X	X	X	X
16-Sep	X	X	X	X
21-Sep			X	X
25-Sep	X	X		
28-Sep			X	X
5-Oct			X	X

[a] Collected 100 berries from the ground in order to access infestation levels in dropped fruit. Samples collected from both sites.

RESULTS:

Maggot infestation

The first larvae were detected in fruit samples from Jonesboro on 21 Sep. Infestation levels were very low at TWP 18. No larvae were found in any fruit samples and the trial on those two blocks were ended due to a lack of fruit after the 25 Sep sample date. The lack of fruit was due to a severe drought that occurred during the month of August and September. Even though the TWP

18 field was irrigated, fruit drop occurred by the end of September. In addition, the drought greatly delayed the onset of SWD attack in the remaining two blocks in Jonesboro (blocks 3 and 4). The experiment, therefore, was reduced to two statistical blocks (blocks 3 and 4, in the Jonesboro field) and the densities of larval infestation were only moderate. Data were transformed by the square root prior to analysis by ANOVA, CRD. Means were separated by LS Means Differences Student's t as a liberal measure of mean separation since we only had two blocks.

Although maggot infestation was low, an analysis of the two replications did indicate some potential differences (ANOVA. $F_{(14,15)} = 1.16$; $P = 0.034$) (Table 5). The low rates of CimeXa Silica Gel dust (5 lbs/acre) and Sivanto 200SL (10 oz/acre) reduced infestation levels in comparison with the untreated checks (UTC).

Table 5. Results of analysis for SWD larvae.

Treatment [a]	**Rate**	**Total larvae**	**Mean**	**SE**	**Mean separation**
UTC		14	7.0	2.0	a
Delegate WG	6 oz	17	8.5	7.5	ab
Sivanto 200SL	10 oz/acre	0	0.0	0.0	b
Sivanto 200SL	14 oz/acre	3	1.5	1.5	ab
Entrust SC	4-6 fl oz/acre	12	6.0	1.0	ab
Grandevo	3 lbs/acre	13	6.5	5.5	ab
Venerate XC	2 qts/acre	17	8.5	2.5	a
Entrust SC + Grandevo	4-6 fl oz/acre + 3 lbs/acre	1	0.5	0.5	ab
Entrust SC + Venerate XC	4-6 fl oz/acre + 2 qts/acre	8	4.0	3.0	ab
CimeXa Silica Gel Dust	5 lbs/acre	0	0.0	0.0	b
CimeXa Silica Gel Dust	10 lbs/acre	11	5.5	5.5	ab
Entrust SC + CimeXa Silica Gel Dust	4-6 fl oz/acre + 5 lbs/acre	8	4.0	2.0	ab
Venerate XC + CimeXa Silica Gel Dust	2 qts/acre + 5 lbs/acre	5	2.5	1.5	ab
AzaSol	6 oz/acre	8	4.0	3.0	ab
AzaSol	6 oz/acre	5	2.5	2.5	ab

Adult abundance

We did not perform an analysis on the adult abundance because the controls had the lowest abundance (Table 6). In general, adults move around through the treatment plots greatly and so adult efficacy is very difficult to measure unless treatment and control plots are several acres in size. However, the monitoring of adults is used to initiate applications and to determine the levels of SWD pressure.

Table 6. Summary of adult SWD captures over the season.

					Sample date						
Treatment											**Cumulative**
		17-Aug	**24-Aug**	**31-Aug**	**6-Sep**	**16-Sep**	**21-Sep**	**25-Sep**	**28-Sep**	**5-Oct**	**mean**
UTC		0.5	1.0	0.5	1.0	0.8	1.0	1.0	8.0	6.5	20.3
Delegate WG		1.0	2.0	1.0	1.3	1.5	0.5	2.0	4.5	9.0	22.8
Sivanto 10 oz		1.5	3.0	1.5	1.8	2.5	0.5	3.0	5.5	8.5	27.8
Sivanto 14 oz		2.0	4.0	2.5	4.5	3.3	2.0	4.0	14.5	16.5	53.3
Entrust SC		2.5	5.0	2.5	3.5	4.5	1.0	5.0	48.5	37.0	109.5
Grandevo		3.0	6.0	3.5	3.5	5.3	4.5	6.0	56.5	31.5	119.8
Venerate XC		3.8	7.0	3.8	6.0	8.0	5.0	7.0	72.0	59.5	172.0
Entrust SC + Grandevo		4.0	8.0	4.3	4.3	5.3	0.5	8.0	17.5	9.0	60.8
Entrust SC + Venerate XC		4.5	9.0	5.0	6.0	6.0	2.5	9.0	35.5	12.5	90.0
CimeXa Silica Gel Dust 5 lbs		5.0	10.0	5.0	5.0	6.3	4.0	10.0	45.5	7.5	98.3
CimeXa Silica Gel Dust 10 lbs		5.5	11.0	5.8	7.0	6.0	2.5	11.0	26.0	12.5	87.3
Entrust SC + CimeXa Silica Gel Dust		6.3	12.0	6.3	8.5	7.8	1.0	12.0	21.5	13.5	88.8
Venerate XC + CimeXa Silica Gel Dust		6.5	13.0	6.5	6.5	6.8	1.0	13.0	6.5	6.0	65.8
AzaSol at infestation		7.0	14.0	7.0	7.0	8.8	1.5	14.0	21.0	20.0	100.3
AzaSol at first adults		7.5	15.0	7.5	8.3	10.3	1.5	15.0	20.0	12.5	97.5

CONCLUSIONS: Because only two replicates ended up in the experiment, the ability to detect differences was low. However, we were able to see greater maggot infestation in the control plots compared to the Sivanto treatments at 10 oz and CimeXa Silica Gel Dust at 5 lbs/acre. However, in both the Sivanto and CimeXa treatment the higher rates were not significantly different from the control which suggests that the significant differences observed may be due to chance. Another treatment that had low larval infestation, but NOT different from the control was Entrust SC + Grandevo. Our conclusions are that in 2016 the number of treatments should be greatly reduced in order to increase the number of replications.

Study 3. Control of blueberry spanworm (SW) larvae on wild blueberry with Belt® SC insecticide (Laboratory study)

METHODS: Each treatment was applied on 21 May in 25 gallons of water-mixture per acre with a CO_2-propelled, 80-inch boom sprayer (76-inch swath) equipped with four, flat-spray 8002VS TeeJet® nozzles operating at 35 psi and at a slow walking speed. Speed was regulated using a metronome. The materials were allowed to dry on the foliage. Field-collected, mid- to late instar spanworm larvae were placed in plastic cups with petri dish lids. There were four replications of ten SW each. One treated stem was cut and placed in each cup. The cups were held at room temperature and assessed for mortality at daily intervals for four days.

RESULTS: Analysis of Variance (ANOVA, RCB) and LSD ($P \leq 0.05$) were used to compare percent survival of SW larvae among the treatments. Data were transformed by SIN^{-1} SQRT (proportion) to stabilize variance prior to analysis. The standard Delegate® WG provided excellent control of blueberry spanworm larvae within 24hr of application. Belt SC (both rates) was also highly effective and significantly reduced SW survival within 48hrs of the application.

Table 1. Laboratory control of spanworm with insecticides, summary.

Material	Amt. form./acre	Mean percent survival 21 May	22 May	23 May	24 May
Belt SC	1.5 oz	82.50 ab	35.00 b	22.50 b	17.50 b
Belt SC	2.4 oz	77.50 ab	45.00 b	12.50 b	12.50 b
Delegate WG	6.0 oz	52.48 b	0.00 c	0.00 b	0.00 b
UTC	-	95.00 a	95.00 a	92.50 a	90.00 a
P value		0.0551	0.0002	< 0.0001	< 0.0001

CONCLUSIONS: This was the first time we have tested the efficacy of Belt insecticide against blueberry spanworm larvae. The material proved highly effective in this laboratory trial and warrants further investigation. If a suitable infestation of SW larvae can be located in 2016, we will evaluate effectiveness in the field.

Study 4. Testing the efficacy of Beauveria bassiana on mitigating spotted wing drosophila (Drosophila suzukii) infestations in Maine wild blueberry
Report from Gabriel Al-najjr (Master's student), Dr. Eleanor Groden, and Dr. Frank Drummond

The spotted wing drosophila (SWD) is a recent introduction into Maine (2012). We have been pursuing several tactics for its control. One approach was to look at potential for biological control of this pest using insect pathogenic fungi. Laboratory studies revealed that the fungus, *Beauveria bassiana* (strain GHA), has potential for killing this insect pest. We conducted a field-cage study to test this fungus under field conditions.

METHODS: Nine cages were deployed on 6 Aug in Jonesboro, Maine. The following week 2,000 live adult SWD were released in six of the cages, the three other cages being negative control cages. Initial estimations were carried out with culture tubes of known fly abundances of 50, 100, 150 and 200. About one hour prior to SWD release on the evening of 13 Aug, three of the six release cages were sprayed with Mycotrol® at the recommended rate of 10^{13} conidia/acre. A 140 square foot area was also sprayed and divided into three equal sampling areas in an uncaged section of wild blueberry to assess the impact of light shading on conidia viability. Six leaves were plucked randomly from each cage as well as the three uncaged spray areas at 0, 24, 48 and 96 hours following the *B. bassiana* application. A tissue sampler was used to cut out one, 11mm diameter disc from each leaf. All six subsamples were aggregated in a labeled centrifuge vial and then placed in a dark cooler. Immediately after arriving back at the lab, conidia from leaf discs were vortexed into a suspension with 0.01% tween and plated on dodine agar.
Following the initial 0 hour leaf collection, 2,000 flies each were released in three sprayed and three unsprayed cages, while the remaining three cages served as no-release controls. The next day, culture tubes were removed and any live flies were tapped out onto a dry petri dish without media. All remaining flies were counted and subtracted from the release total.
Twelve days after the SWD release, blueberry samples were collected from each of the nine cages. Five cups of fruit were collected from stems and one cup of fruit was collected from berries on the ground. Sampling was randomized in each cage and care was taken to collect berries from different positions on stems. Each sample was processed within one week of the collection date. Berries were crushed in a plastic bag and mixed with 10% saline solution for thirty minutes to induce dissociation of larvae from the pulp. Samples were then strained and placed in a black tray where SWD larvae counts were taken.
After collection of berry samples, a single trap was positioned within each cage in order to assess impacts of spraying on SWD adults. Each trap consisted of a red, 16 oz cup positioned on a 2½ ft tall post and was filled with approximately 2 in. of liquid mixture consisting of the following ratio: 1 tbsp yeast: 4 tbsp sugar: 12 oz. of water. SWD adults were removed from traps and counted six days later.

RESULTS: All control cages were free of larvae. Larval abundances were transformed into ordinal data (0 = 0 larvae, 1 = 0-10 larvae, 2 = 11-100 larvae, 3 = N > 100 larvae) and analyzed via an ordinal logistic model. Releasing flies within cages had a significant effect on the number of larvae observed infesting fruits ($P = 0.0008$). However, spraying GHA did not appear to mitigate infestations ($P = 0.096$) as shown in figure 1.

Since adult flies were captured in no-release control cages, an analysis of variance (ANOVA) was utilized to detect in adult abundance among treatments. Release cages were associated with a higher abundance of adult SWD in comparison to control cages ($F_{(6,8)} = 5.46$, $P = 0.045$). However, according to a Tukey's post-hoc test, there were no discrepancies in the number of adult flies captured in sprayed and unsprayed release cages (Fig. 1). An additional model was constructed with release estimates factored in as a covariate, but this did not provide any additional explanation for the variation observed in larval abundance ($F_{(3,8)} = 0.41$, $P = 0.81$) or adult abundance ($F_{(3,8)} = 0.47$, $P = 0.78$) within cages. Lastly, a power analysis was conducted in order to assess the statistical power entailed in this study due to the limitations of small sample sizes. It was determined that the experiment could detect a difference of 30 and 90 larvae between the experimental treatments with statistical power of 12 and 58%, respectively. A difference of 30 and 90 adult flies could be detected between sprayed and unsprayed cages with statistical power of 7 and 21%, respectively.

Fig. 1. Average SWD adult (shaded) and larval (black) abundance from cages in each treatment. Statistical analyses show no differences in the abundance of either life stage as a result of *B. bassiana* application.

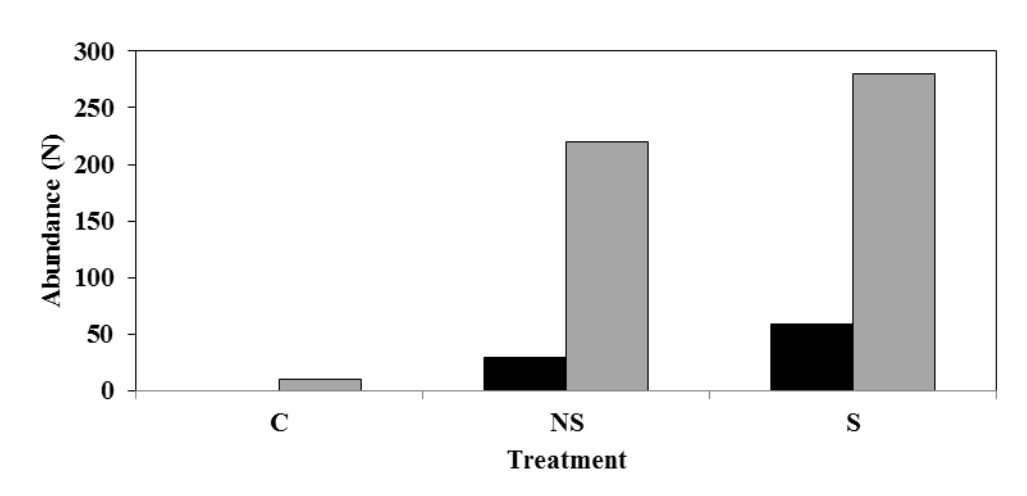


Table 1. Data for all nine cages including estimated fly release, stem larvae counts, weighted average larval counts of stem and ground larvae, and fly capture data for control (C), no spray release (NS), and spray release (S) treatments (TMNT).

TMNT	Cage	Estimated # SWD released	Wght avg larvae	Ordinal Wght avg larvae	Fly capture
C	1	0	0	0	3
C	2	0	0	0	2
C	3	0	0	0	25
NS	1	1384	7	1	60
NS	2	1577	41	2	335
NS	3	1604	38	2	264
S	1	1578	131	3	280
S	2	1565	17	2	392
S	3	1777	30	2	168

CONCLUSIONS: The results of this experiment suggest that releasing flies within cages resulted in blueberry infestations as no maggots were detected in any of the no - release control cages (Table 1). *B. bassiana* applications were not effective at mitigating larval infestations of fruit or reducing the abundance of SWD adults within cages. This may be attributed largely to the abnormally dry month of August at the study location. Previous laboratory assays have shown that, under controlled conditions, SWD adults are susceptible to mycosis by *B. bassiana*. However, during these assays relative humidity was maintained near 100%. As such, future field experiments should work to address this variable. Currently, spore viability data are being processed and *B. bassiana* has positively been identified on dodine agar plated from leaf disc suspensions, suggesting that the conidia tested in this experiment are viable. As such, it is evident that more research is required to assess the efficacy of *B. bassiana* on mitigating SWD infestations in small fruit crops.

Study 5. Potential synergistic activity between the insecticide Assail® (acetamiprid) and the fungicide Tilt® (propiconazole) on colonies of the common Eastern bumble bee (Bombus impatiens)

Report from Gabriel Al-najjr (Master's student) and Dr. Frank Drummond

Both the insecticide acetamiprid, Assail; and the fungicide, propiconazole, formulated as several products; are commonly used in insect and disease pest management in wild blueberry just prior to bloom. Therefore, the chance of exposure of both these pesticides to bumble bees is likely. Laboratory studies conducted by Japanese researchers a little over a decade ago suggested that simultaneous exposure of both these pesticides to honeybee larvae could synergize the toxic reaction response. The result was a fairly low toxic response to acetamiprid when this was the sole exposure, but an extremely high toxic result with simultaneous exposure. We conducted a lab / field study to test the hypothesis of synergy in bumble bees using the commercial bumble bee, *Bombus impatiens.*

METHODS: Twenty *Bombus impatiens* colonies were obtained during the spring of 2015. Colonies arrived as quads (four independent colonies per large hive box) and were numbered in accordance with the box in which they arrived. Based on an estimate of 200 adult *B. impatiens* per colony, LD_{50} values were used to calculate the necessary acetamiprid (LD_{50} = 14.5 ug/bee) and propiconazole (LD_{50} = 100 ug/bee) exposure concentrations. Four hives each were fed either: 1) syrup containing no pesticide, 2) syrup with acetamiprid alone at either 1.0x, 3) or 0.1x the LD_{50}, 4) syrup with propiconazole alone at 0.1x the LD_{50}, 5) or a syrup mixture consisting of both pesticides each at 0.1x the LD_{50}. Prior to mixing of pesticides with syrup, an empty bag was weighed and used as a standard bag weight. Syrup was also weighed following the addition of pesticides to determine how much of the mixture was given to each hive.
Untreated syrup was removed from hives and replaced with the treated syrup and bees were not allowed to forage for 14 days. Following the two-week exposure period, hives were placed in the field in Orono (University of Maine campus) and allowed to forage for four weeks. Prior to data collection, hives were closed and placed in a cold room for 24 hours. In addition to hive weights and pictures, syrup solutions were weighed to determine the amount of syrup consumed by the colony. Mortality levels of adult bees were categorized as such: none (0 dead), low (1-5 dead), medium (6-25 dead) and high (> 25 dead). Immediately after collection of data, hives were placed in a boom freezer for two days so larvae and pupae could be counted. Adults were categorized as being small, medium or large based on qualitative observation.
Mortality data were analyzed via an ordinal logistic model. Raw percent data were evaluated utilizing an analysis of variance (ANOVA). All remaining count data were square root transformed and analyzed via ANOVA. Models that showed a significant interaction between treatment and the variable of interest were further analyzed with the Tukey post-hoc test.

RESULTS: Including original box packaging as a covariate did not provide a better explanation for the variance observed across hives in either total adult (queens, workers, and males) abundance ($F_{(8,19)} = 0.70$, $P = 0.72$). There did not appear to be any detectable differences between treatments in the abundance of total individuals (larvae and adults) ($F_{(15,19)} = 2.01$, $P = 0.14$); although, the P value suggests that caution should be exercised with this conclusion. Nor did the addition of pesticides to syrup result in reduced syrup consumption ($F_{(15,19)} = 0.32$, $P = 0.86$) or hive weight ($F_{(15,19)} = 1.47$, $P = 0.26$) as shown in Table 1, respectively. However, there was a difference observed in the number of gynes (potential queens for following year) ($P = 0.049$) between hives fed syrup containing 0.1x propiconazole and 1.0x acetamiprid (Fig. 1).

Fig. 1. Queen production by end of experiment (error bars are se), treatments sharing the same letters are not significantly different from one another (Tukey post-hoc test, $P \leq 0.05$).

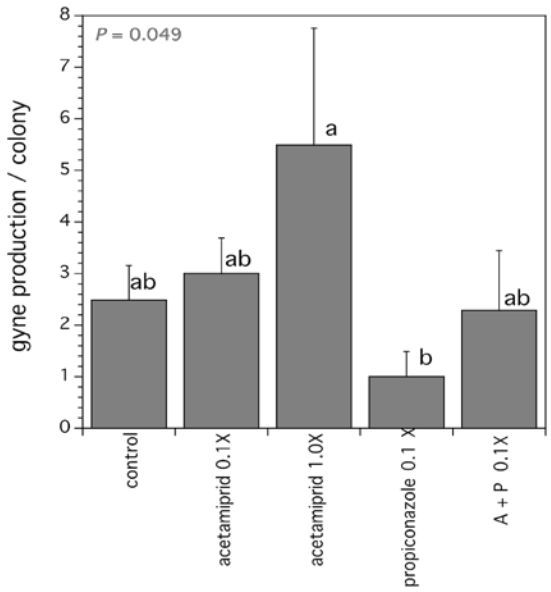


Table 1. Mean ± standard deviation for each dependent variable represented by treatment (A: acetamiprid, P: propiconazole). Only one statistically significant difference was detected in the abundance of gynes between hives fed syrup containing 1.0x Assail and hives a fed syrup mixture containing 0.1x Assail with 0.1x Tilt.

TMT	Total bees	Adults	*Gynes	Med Adults	Sm Adults	Pupae	Larvae	Honey Pots	% Δ Weight	% Δ Syrup
Control	180 ± 26	616 ± 103	2.5 ± 1.3	115 ± 25	62 ± 44	211 ± 101	225 ± 95	62 ± 19	52% ± 24%	67% ± 15%
0.1x A	157 ± 53	693 ± 234	3 ± 1.4	110 ± 42	44 ± 14	290 ± 118	246 ± 112	68 ± 20	42% ± 18%	69% ± 17%
1.0x A	172 ± 51	638 ± 97	*5.5 ± 4.5	117 ± 47	49 ± 10	225 ± 34	241 ± 88	72 ± 41	46% ± 11%	58% ± 16%
0.1x P	248 ± 45	1021 ± 260	*1 ± 0.8	173 ± 55	74 ± 11	369 ± 108	405 ± 112	79 ± 36	70% ± 27%	58% ± 37%
0.1x A + P	230 ± 92	938 ± 480	2.3 ± 0.5	174 ± 64	54 ± 29	420 ± 262	288 ± 149	49 ± 9	72% ± 30%	72% ± 22%

CONCLUSIONS: Oral exposure of *B. impatiens* to pesticides did not seem to have an impact on the overall abundance of bumblebees when exposure was through syrup food. All hives experienced positive weight growth and adults did not appear deterred from feeding on syrup containing pesticides (Table 1). The only treatment effect observed was in newly produced queen abundance where hives fed syrup containing 1.0x acetamiprid appeared to have more large adults than those fed syrup with 0.1x propiconazole (Table 1). However, more research is required to determine whether acetamiprid exposure consistently results in a greater abundance of large adult bees (queens at the end of the season). Consequently, these results indicate that controlled oral exposure of *B. impatiens* to propiconazole may pose an immediate threat to the overall health of the colony in terms of production of reproductives in the following year. However, we did not find any evidence for synergy. These results are based upon a feeding

experiment and so they need to be followed up by a more realistic field exposure study involving spraying the crop just prior to bloom.

Study 6. ***<u>The effects of foraging on imidacloprid-treated wild blueberry (Vaccinium angustifolium) on the development of Bombus impatiens colonies.</u>***

<u>Report from Kalyn Bickerman (Ph.D. candidate) and Dr. Frank Drummond</u>

METHODS: We evaluated the effects that Admire® Pro Systemic Protectant (42.8 wt% imidacloprid) had on the development of commercial *Bombus impatiens* colonies. Four flight cages, each approximately 6 meters long, were set up in a wild blueberry field in the beginning stages of bloom on 18 May. On 27 May the blueberry clones in three of the flight cages were sprayed with Admire Pro at the rate of 2.8 oz/acre and allowed to dry. One flight cage was kept untreated as the control (Fig. 1).

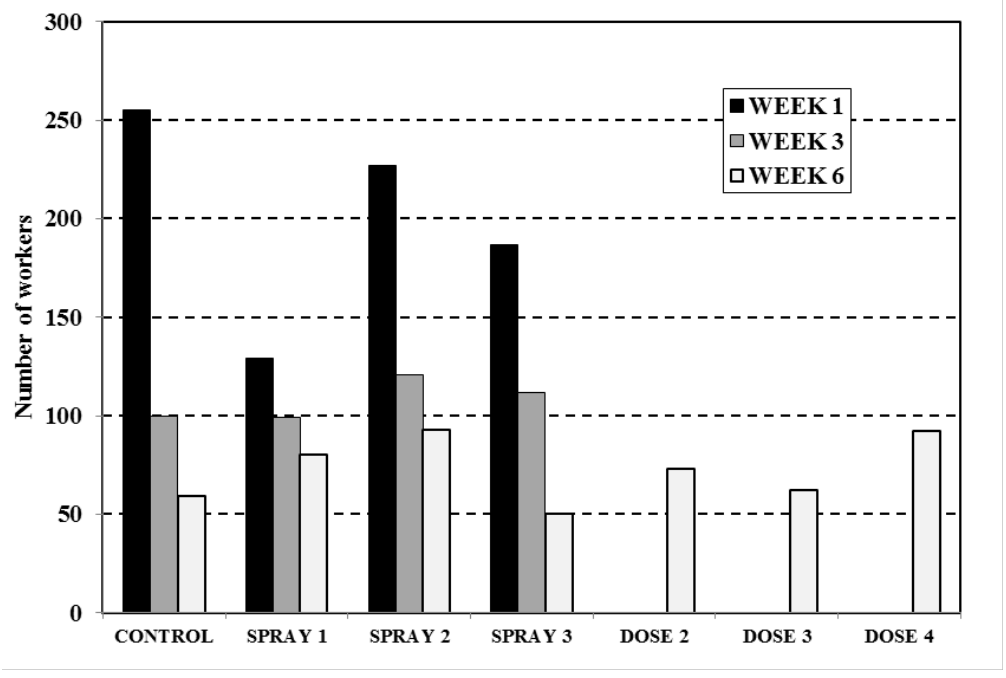
Fig. 1. The field set up of the flight cages and spray regimen.

That same day, 12 colonies of commercial *B. impatiens* (Koppert Biological Systems, Romulus, MI) were split into four groups of three and placed inside the flight cages. The boxes were opened and bees were allowed to forage on the blueberry flowers. One week post treatment (5 Jun), one colony and flower samples were collected from each flight cage and immediately frozen. Three weeks post treatment (18 Jun), one colony was again collected from each flight cage and frozen. Due to blueberry bloom ending, the remaining four colonies (one from each cage) were transported back to the Rogers Farm Forage and Crop Research Facility in Stillwater,

ME and allowed the forage there. These colonies were collected and frozen six weeks post spray (7 Jul).
Additionally, on 4 Jun we removed the bags containing the Bee Happy® food from four extra colonies and used the Admire Pro to create a concentration of 20 ppb imidacloprid in each food bag. We weighed and replaced the food bags and kept the colonies outside on the University of Maine campus but closed, so the only option for the bees would be to consume the sugar syrup as their energy source. At the end of a two-week dosing period (18 Jun), the dosed food bags were removed and weighed from the colonies and the bees were taken to Rogers Farm to forage until being frozen six weeks post-dosing (14 Jul).
All colonies were removed from the freezer, photographed from above, and all parts (adults, pupae, larvae, pollen, nectar, and wax) separated into Falcon tubes for chemical analysis at the Connecticut Agricultural Experiment Station to ascertain concentrations of imidacloprid throughout the colony components through time.

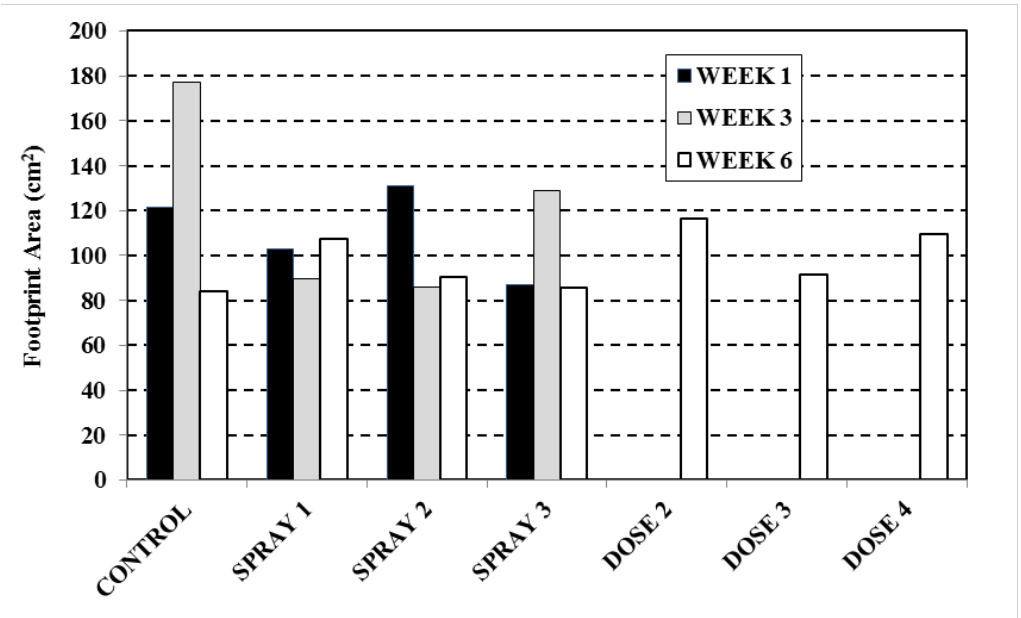
RESULTS: The number of adults visible in each of the colonies was significantly different between Week 1 and Week 3 and between Week 3 and Week 6. Week 1 had significantly more adults present; although, no significant difference was detected between Weeks 3 and 6 ($F_{(2,6)}$ = 17.08; $P < 0.003$). There was no significant difference between the number of adults in each colony in the sprayed flight cages and the control flight cage ($P = 0.207$) and there was no week by cage interaction, which would have suggested that the colonies in each treatment are declining at different rates ($P = 0.360$) (Fig. 2). The number of adults left in the dosed colonies was not significantly different at the end of the six weeks when compared to the flight cage colonies ($P = 0.970$).

Fig. 2. Number of adult bumble bees visible in each colony from each flight cage by week due to exposure by spraying flowers or by feeding contaminated syrup.



Measurements of the colonies' areas (the "footprint" of the brood clump that represents the size of the colony) showed no significant difference between control and sprayed treatments (P = 0.540), between weeks (P = 0.110), or between flight cages by week (P = 0.176). There was also no difference at the end of the six weeks between the sizes of the sprayed colonies in the flight cages and the dosed colonies (P = 0.324) (Fig. 3).

Fig. 3. The relationship between the area of the footprints of the colonies' brood and exposure to imidacloprid by spraying flowers or by feeding contaminated syrup.



Dose 1 was omitted from all analyses due to evidence of a severe earwig infestation. No adults or even comb were left inside of the colony box at the time it was brought in and frozen.

CONCLUSIONS: There was a significant difference between weeks in the number of adults present in each colony, but this is to be expected due to the natural decline of commercial colonies that should be at their peak at the time they are received and then taper off in numbers. There was, however, no significant difference between the control flight cage as compared to the three flight cages treated with imidacloprid, formulated as Admire Pro. No significant effect of week or flight cage was observed in terms of colony brood clump area. Colonies that fed on the food treated with 20 ppb imidacloprid for two weeks were also not observed to be significantly different in number of adults or brood area after six weeks had passed from the initial treatment as compared to the sprayed colonies in the flight cages. This is not surprising because we purposely selected a fairly low dose of imidacloprid so as not to kill the colony in order to trace the insecticide spread within the hive. Once the chemical analysis on the colony components is completed, we will have a better idea how (at what levels) imidacloprid is incorporated into a colony by foraging bees and how long it stays in the colony.

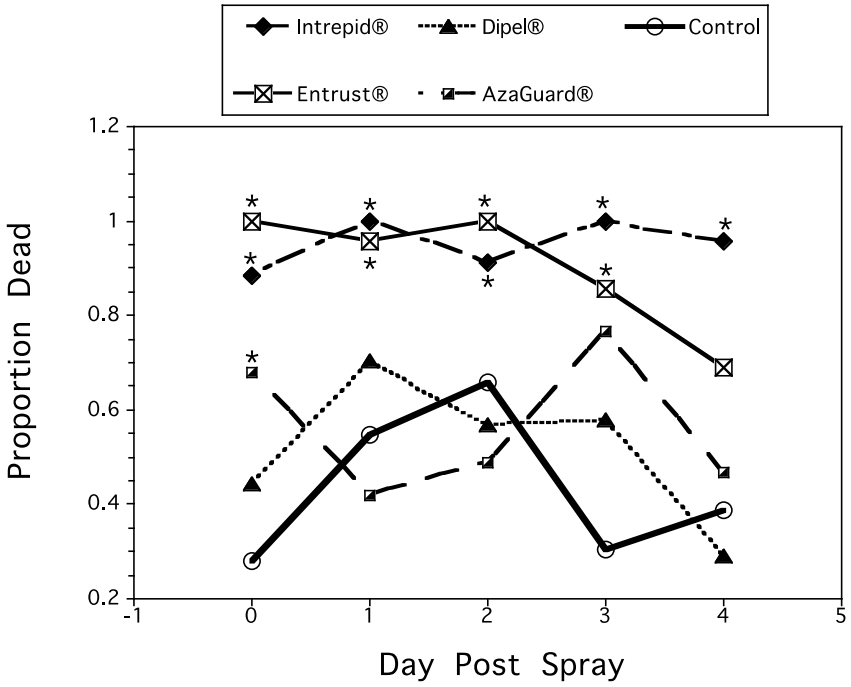
Study 7. ***<u>Control of winter moth (WM) larvae on wild blueberry.</u>***
<u>Report from Dr. Eleanor Groden, Dr. Frank Drummond, and Ms. Judith Collins</u>

Winter moth, *Operophtera brumata*, is an emerging new invasive insect pest. It is a European moth species that was accidently introduced into Nova Scotia several decades ago. It was detected in Maine in 2012 and has spread throughout most of the central coastal and coastal Downeast blueberry growing regions of Maine. It feeds upon many shrub and tree species in Maine, wild blueberry being one of them. In anticipation of it becoming problematic in Maine wild blueberry, we initiated a control trial in 2015. We selected biorational insecticides that have a low impact on bees, since this spring defoliator can occur during bloom.

METHODS: Winter moth larvae were collected in Harpswell, ME on 18 May. Each treatment was applied in 20 May in 25 gallons of water-mixture per acre with a CO_2-propelled, 80-inch boom sprayer (76-inch swath) equipped with four, flat-spray 8002VS TeeJet® nozzles operating at 35 psi and at a slow walking speed. Speed was regulated using a metronome. The materials were allowed to dry on the foliage. The following treatments were applied: 1) Dipel® DF - 16 oz/acre, 2) Entrust® SC - 2 oz/acre, 3) AzaGuard® - 16 oz/acre, 4) Intrepid® 2F - 16 oz/acre, and 5) a non-sprayed control. Foliage from each treated plot was collected 4 hours post spray when all foliage had dried. Foliage was added to cups (1 per plot within each block equaling 3 cups per treatment) and 15 winter moth larvae were added. Cups were held in an environmental chamber at 18^{o}C for 5 days. Dead larvae were removed and counted daily. The assay was repeated daily with field sampled foliage at 1, 2, 3, and 4 days post spray in order to assess the persistence of treatment effects over time after the spray.

RESULTS: Immediately following treatment (Day 0), WM larvae fed field sprayed foliage experienced significantly higher mortality on foliage treated with Entrust or Intrepid compared with the control. This significant mortality continued for four days post spray with Intrepid and three days post spray with Entrust. Neither Dipel nor AzaGuard resulted in significantly higher mortality than the controls at any time post spray (Fig. 1).

Fig. 1. Proportion of winter moth larvae dead after 5 days of exposure to foliage collected from field plots between 0 and 4 days after being sprayed with insecticides. The symbol "*" indicates treatments that are significantly different from the control of no treatment at $P = 0.05$.



CONCLUSIONS: Clearly, the winter moth being a new threat to wild blueberry, apple, highbush blueberry and raspberry, needs management options. We have demonstrated that Azaguard and *Bt* do not fit the bill. Entrust and Intrepid appear to work quite well. Recommendations to growers in the near future will include these two insecticides. However, it is interesting that *Bt* might be affected by the acid soil of the wild blueberry environment. This has never been shown before, but could have great implications in a state like Maine where the soils are highly acidic.

RECOMMENDATIONS: Several insecticides appear to provide some control for the blueberry tip midge; although, it seems to provide only temporary control with populations rebounding later in the season and not different than the non-treated controls. Because of this we are still not recommending specific insecticide control of this pest. However, we are also recommending that if growers do treat for tip midge they DO NOT USE Rimon (novaluron) as this has been shown to be a very potent egg laying disruptor of queen honeybees and is thought (by some researchers) to have caused the 2014 summer collapse of several thousand honeybee colonies.

Several new insecticides were evaluated for the SWD in 2015, but as of yet there are none to recommend. Unfortunately, a two–year study on the biological control of SWD did not demonstrate any promise of the fungus, *Beauveria bassiana*, for effective control. It was also apparent during our insecticide trial that our previous recommendation of Entrust only having a 3-day residual was borne out. Using it at 7-day intervals did not provide acceptable control.

Growers that encounter high defoliating levels of the winter moth now have a control. Entrust® and Intrepid® insecticides work effectively against this new insect pest.
For several years we have been cautioning growers about using Assail for spring insect control when a sterol-inhibiting fungicide is also used. This caution was based upon a Japanese laboratory study that showed synergy between these two pesticides. A laboratory and field study suggests that for bumble bees this is not the case, they are both fairly non-toxic. A comprehensive study with honeybees will be conducted in 2016.

ENTOMOLOGY

INVESTIGATORS: F. A. Drummond, Professor of Insect Ecology/Entomology
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3. TITLE: II. Pest biology and IPM, 2015.

Study 1. Long-range, within-field, movement of blueberry maggot fly in wild blueberry: A release/recapture study

This trial is the continuation of a study begun in 2013 to assess the long-range movement patterns of blueberry maggot fly (BMF). The central question being…how far does an isolated field need to be from a larger blueberry production area with multiple fields to enjoy the benefit of reduced blueberry maggot fly pressure?

METHODS: BMF were collected as pupae from infested blueberries in 2014. Collection and wintering procedures are outlined in Bio. Study 1 of this report. The wintering cups of pupae were separated into four equal groups. A small paintbrush was used to layer orange, DayGlo® dye on top of the vermiculite in each of the cups. The cups were placed in cages and flies were allowed to emerge. Following emergence, the flies were fed honey and yeast for one week prior to release.
A line transect of 50 baited, yellow, Pherocon® AM traps was set along one edge of a pruned year blueberry field in Beddington, ME (Pork Brook) with 10 ft between traps. Ammonium acetate superchargers were attached to every other trap to enhance attractiveness. On 30 Jun, the marked blueberry maggot flies were released at a point 1800 ft (549 m) across the pruned field from trap number 25 (the middle of the perpendicular transect); ca. 200 flies were released. Traps were checked daily for seven days and periodically for an additional seven days; thereafter. All BMF were removed from the traps, brought back to the laboratory, and examined under UV light (Black-Ray® Longwave Ultraviolet Lamp) for the presence of fluorescent dye.

RESULTS: In 2015, we recaptured a total of seven marked flies (3.5%); no flies were recaptured after 10 days from release; the first fly was recaptured four days after release. The furthest any BMF traveled was 1813 ft for a fly captured on day 4 after release. The mean distance traveled by the seven recaptured flies was 1806.8 ft.
In a similar trial in 2014 we recaptured a total of 33 marked flies (4.7%) that were released at a point 200 ft (61 m) from the trap transect; no flies were recaptured after seven days from release;

23 of those were recaptured within the first four days after release. The furthest distance that any marked fly traveled was 352.3 ft on day 3. On day 1, two BMF traveled 320.2 ft. Mean travel distance was 266.4 ft.
In 2013 we recaptured seven of 1000 marked flies (0.7%) that were released 328 ft (100 m) from a similar trap transect. Three flies were recaptured within two days (one on day 1 and two on day 2); four additional flies were recaptured six (n=3) or seven (n=1) days after release. The mean distance traveled by the seven recaptured flies was 397 ft. The furthest any BMF traveled was 472.4 ft for a fly captured on day 3 after release. In 2013, we also released 1000 BMF at a point 1312 ft (400 m) from the transect. Only one fly (0.1%) was recaptured (on day 7 after release); that fly traveled 1380.7 ft.

Table 1. Recapture rates and average distance traveled by marked flies

Release Distance	**Trial year**	**Fly recapture rate (%)**	**Average distance traveled (ft) (n)**
1800 ft (549 m)	2015	3.5	1806.8 (7)
200 ft (61 m)	2014	4.7	266.4 (33)
328 ft (100 m)	2013	0.7	397.0 (7)
1312 ft (400 m)	2013	0.1	1380.7 (1)

CONCLUSIONS: Our data are fairly low in sample size and so little can be deduced from the results, except: 1) flies moved and were recaptured at the distance of the perpendicular transect, and 2) recapture rates are fairly low (< 5 %) and so a more definitive study will require several thousands of flies. We hope (depending upon the number of flies that we collected for release next year) to conduct an experiment in 2016 that will allow us to set up parallel transect from the release with traps set at 400, 800, 1200, 1600, 2000, 2500, and 3000 ft from a release point. If we can catch enough flies along this gradient we hope to estimate the furthest likely movement distance.

Study 2. Long-term trends (18 years) in parasitism of the blueberry maggot fly

METHODS: Diet cups containing blueberry maggot fly (BMF) pupae (128 cups of 50 pupae each) were maintained in the laboratory for a minimum of four weeks following the last observed emergence of BMF adults. The pupae were collected from infested fruit as described in Bio. Study 1 of this report. Parasitic wasps were observed in the rearing cages. The wasps were collected and an estimate was made of percent parasitism. An estimate of relative size of blueberry maggot populations from year to year was obtained from both collections of pupae from fruit and from trap captures of adult flies and added to our database of the relationship between BMF population increase from year to year and parasitism.

RESULTS: Figure 1 shows the time series of blueberry maggot percent parasitism from 1998 to 2014 (2014 parasitism is estimated one year later in 2015). Parasitism rates dropped (11.95%) following the sharp increase in parasitism of pupae collected in 2013 (23.0%). However, there does not appear to be a tight linkage between fly trap captures and the parasitism rates over time (Fig. 2). Although upon visual inspection of Figure 2, one can see that whenever parasitism rates peak, usually a decline in fly number occur the year or two following. Modeling fly rate of increase (annual fly population growth) as a function of parasite density suggests that a possibility exists that a parasitic wasp (presumably *Opius sp.*) is important in regulating fly numbers and that steps should be taken to conserve its numbers ($F_{(1,15)} = 7.637$, $P = 0.015$). Also, based upon data collected from 1998 through 2015 and plotted in Figure 3 it appears that parasitism behaves as a density dependent factor that controls fly abundance from one year to the next. One can see that at as parasitoid numbers increase fly reproduction falls precipitously and that only a small window of very low parasitoid density allows positive blueberry fly increase in numbers from one year to the next; although, only 33.7 % of the variation in fly increase is explained by parasitoid numbers. Figure 4 shows the relationship between the logarithm of fly abundance in year t versus the log rate of increase from year t to year t+1 (Log(Nt+1 /Nt)). The linear relationship suggests that a density dependent relationship exists between fly abundance and the next year's increase or decrease in the blueberry maggot fly population ($F_{(1,15)} = 17.739$, $P = 0.0008$). In addition, inspection of Figure 4 suggests that a seasonal fly abundance of 10 is the threshold for increase. Below a density of 10 the population will increase and above a seasonal density of 10 the population will decrease. What is particularly interesting about this threshold is that this is the threshold that we suggest is used for making decisions regarding insecticide control of blueberry maggot fly.

Fig. 1. Percent parasitism of blueberry maggot fly pupae.

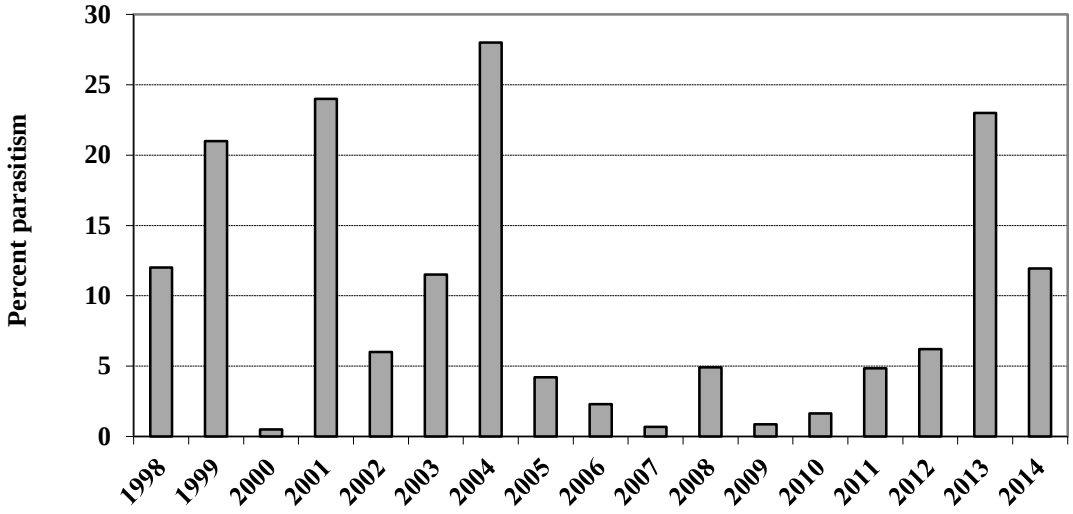


Fig. 2. Relationship between relative density of flies and % parasitism over time. Horizontal line depicts the average fly abundance over the period from 1998 – 2015.

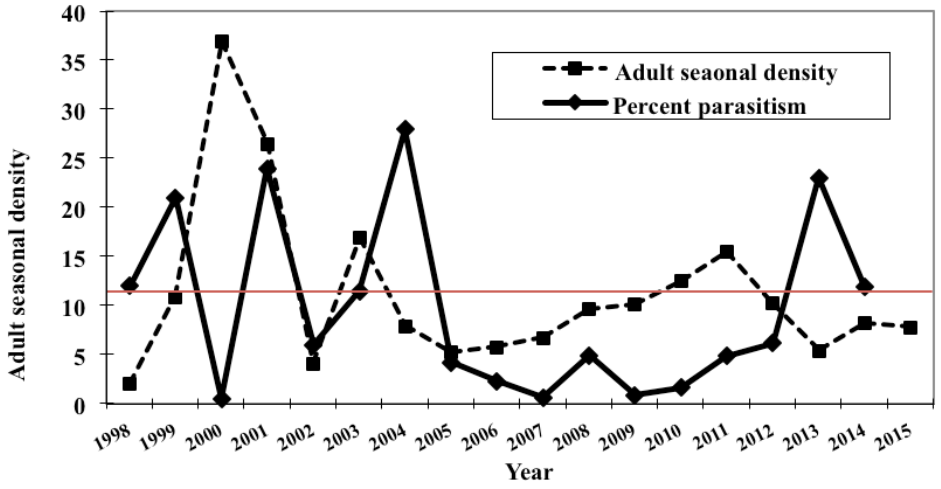


Fig. 3. Relationship between fly population increase and parasitoid density the previous year (1998-2014).

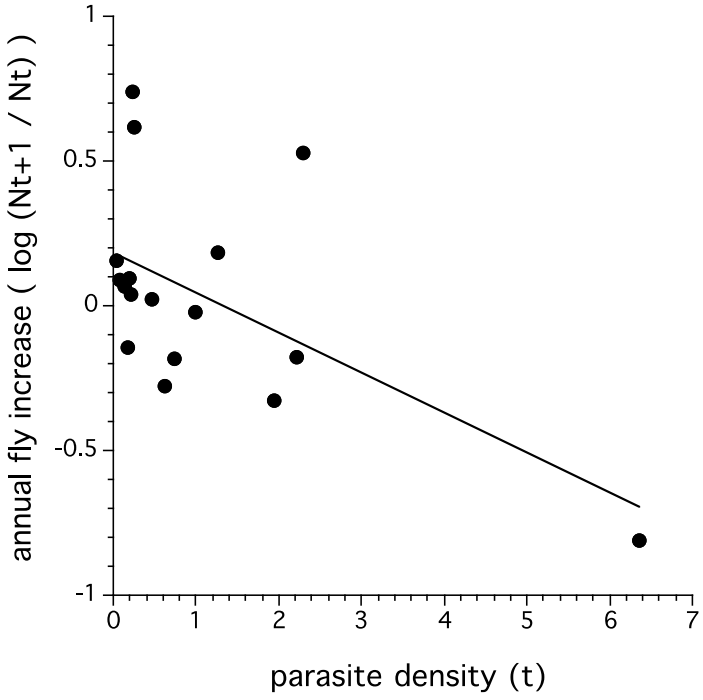


Fig. 4. Relationship between fly population reproduction and fly density the previous year. Dotted line demarks point of zero population increase (1998-2015).

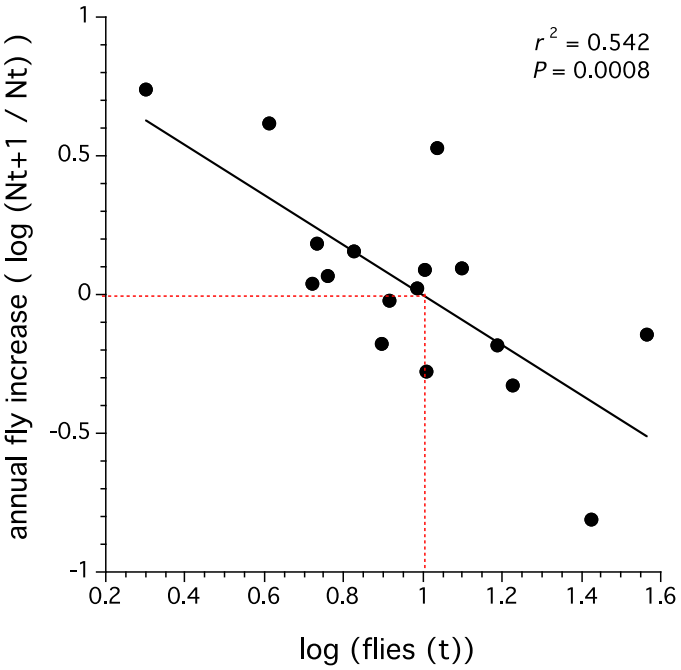
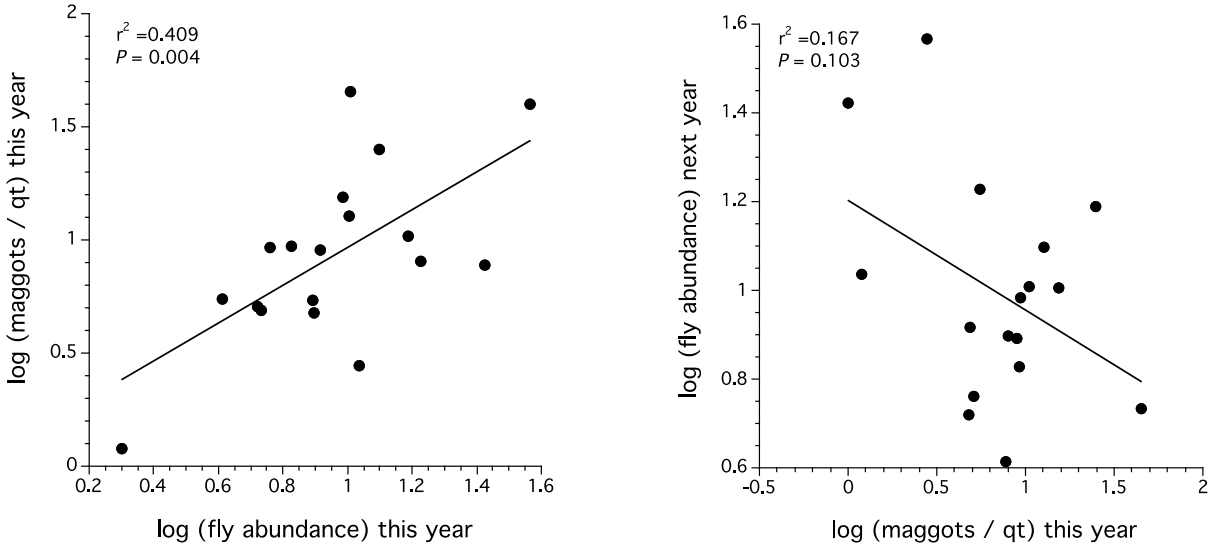


Fig. 5. Relationship between this year's flies and this year's maggots in fruit (left), and the relationship between this year's maggots in fruit and next year's fly population.



Two other relationships have emerged from the long-term BMF data. First, the data documents that while an increase in the number of flies result in an increase in maggot infestation of fruit (Fig. 5), there is a noisy relationship suggesting that other factors affect infestation levels of fruit. The most obvious factor is insecticide application. The other relationship shows that the current

year's infestation of fruit is related to the subsequent year fly abundance. However, this relationship is an inverse relationship suggesting that parasites, predators, and fly dispersal from out of the field that they originated in to new fields are density dependent processes and as maggot infestation increases the following year's flies are reduced. However, this relationship is also very noisy and barely significant at the 10% level ($P = 0.103$).

CONCLUSIONS: This study is the only long-term monitoring of the blueberry maggot fly in North America. It shows that in 18 years there have been roughly three explosive outbreaks of the blueberry maggot fly in Downeast Maine. Each outbreak has 2-3 years to peak and then fly densities fall precipitously after the peak. This study is not directly applicable to management of the blueberry fly, but it does help explain what regulates its densities such as parasitoids, the frequency of outbreaks, and the relationship of threshold fly captures and the population growth of this very important pest. I plan on continuing this study for another two years so that a sound basis of this pest's population dynamics can be acquired after 20 years of study.

Study 3. Impact of blueberry tip midge (BTM) on flower-buds and subsequent flower development. 2010 - 2015

METHODS: On 15 Jul 2015 at Jonesboro, we marked 100 stems per site, 50 with tip midge infestation (pink flags) as evidenced by leaf curls and 50 without infestation (yellow flags). On 5 Oct 2015 we cut 25 stems from each treatment, brought them into the laboratory and counted the number of flower-bud clusters on each stem. Twenty-five marked stems of each treatment were left in the field at each site. In May of 2016 the remaining stems from our 2015-2016 trial will be cut and brought into the laboratory to determine the number of flowers that develop from individual flower-bud clusters.

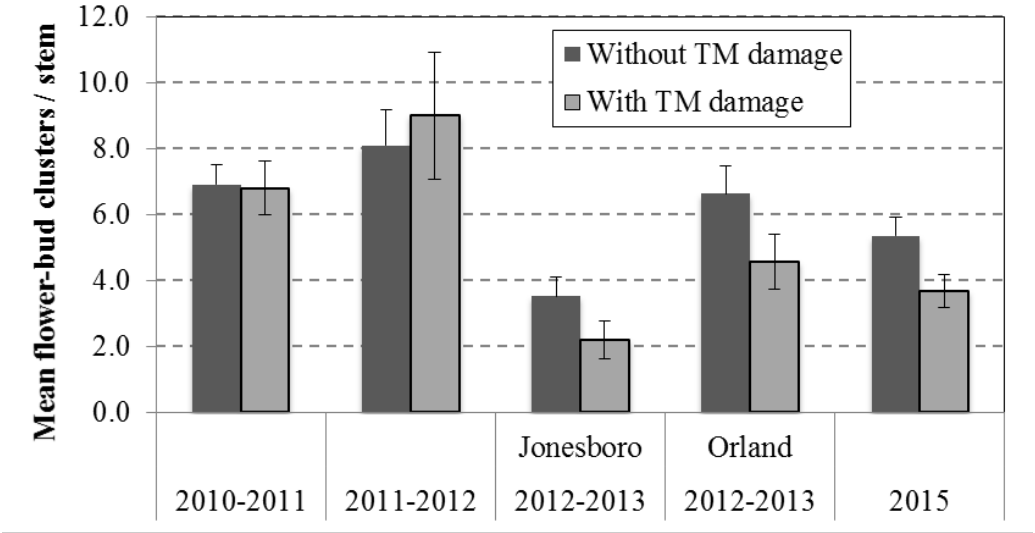
Another focus of our investigations has been the development of economic thresholds. Based upon the amount of crop loss from BTM infestations estimated over previous year, an estimate of the percent of stems infested relative to the cost of crop loss and the cost of insecticide control was determined to assess economic thresholds. These relationships will be updated in 2016, when the latest crop loss estimates (see above) are available.

RESULTS: Data were transformed by the square root prior to analysis. The count of flower-bud clusters showed there was a significant difference in the number of flower-bud clusters (ANOVA, CRD; $F_{(1,48)} = 5.56$, $P = 0.0225$) per stem between stems that had been infested by blueberry tip midge in the prune cycle compared to non-infested stems of the same clone (Fig. 1).

Previous studies demonstrated that blueberry plant response in flower-bud production can be quite variable. In 2010-2011 trial we found NO difference in flower-bud clusters per stem due to blueberry tip midge ($F_{(1,48)} = 0.01$, $P = 0.9054$) (Fig. 1); however, stems with blueberry tip midge infestation developed significantly fewer flowers then those without tip midge infestation (Fig. 2) ($F_{(1,48)} = 17.46$, $P < 0.0001$)

There was no significant difference in the number of flower-bud clusters ($F_{(1,48)} = 0.16$, $P = 0.6897$) in our 2011-2012 trial (Fig. 1.). When individual flowers were counted in 2012, there appeared to be a trend towards more flowers on tip-midge damaged stems; however, the difference was not significant ($F_{(1,48)} = 2.83$, $P = 0.0967$) (Fig. 2).

Fig. 1. Bar graph comparing mean number of flower-bud clusters between stems with and without tip-midge damage. Data collected from trials conducted in 2010-2011, 2011-2012, 2012-2013, and 2015.



In both our trials begun in 2012 there was a significant difference in the number of flower-bud clusters (ANOVA, CRD; $F_{(1,48)} = 5.0$, $P = 0.03$, Jonesboro; $F_{(1,48)} = 4.22$, $P = 0.0454$, Orland) per stem between stems with and without tip midge damage (Fig. 1). Stems without damage had significantly more flower-bud clusters. And, stems with tip-midge damage developed fewer flowers than undamaged stems; although, at our Jonesboro site the difference was not significant ($F_{(1,48)} = 2.73$, $P = 0.1050$, Jonesboro; $F_{(1,48)} = 6.18$, $P = 0.0164$, Orland) (Fig. 2).
The results of our economic threshold analysis are shown in Figure 3. The graphs show economic thresholds if, on average, crop loss due to infested stems is 46%. It can be seen that for the average level of production currently in Maine, 3,000 lbs/acre, the economic injury level is about 7-10% infestation (the level where the cost of control equals the cost of crop loss).

Fig. 2. Bar graph comparing mean number of individual flowers per stem between stems with and without tip-midge damage. Data collected from trials conducted in 2010-2011, 2011-2012, and 2012-2013.

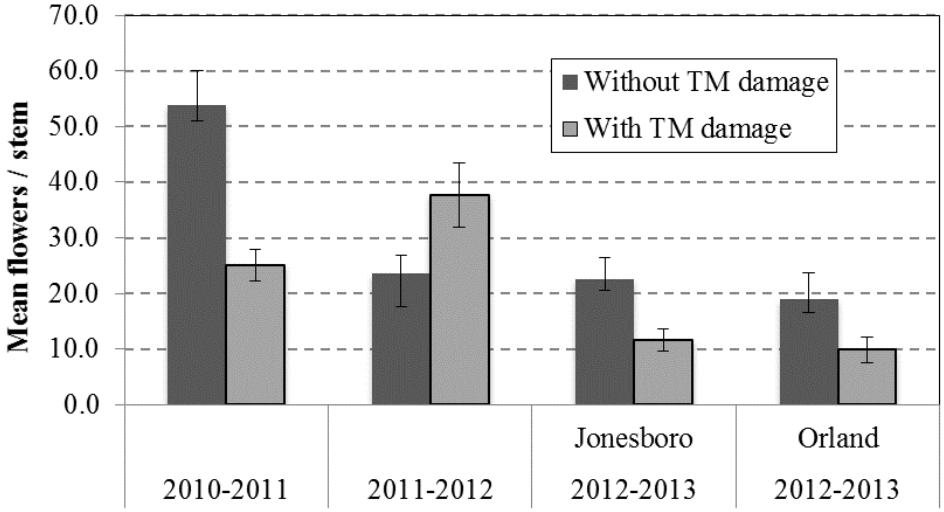
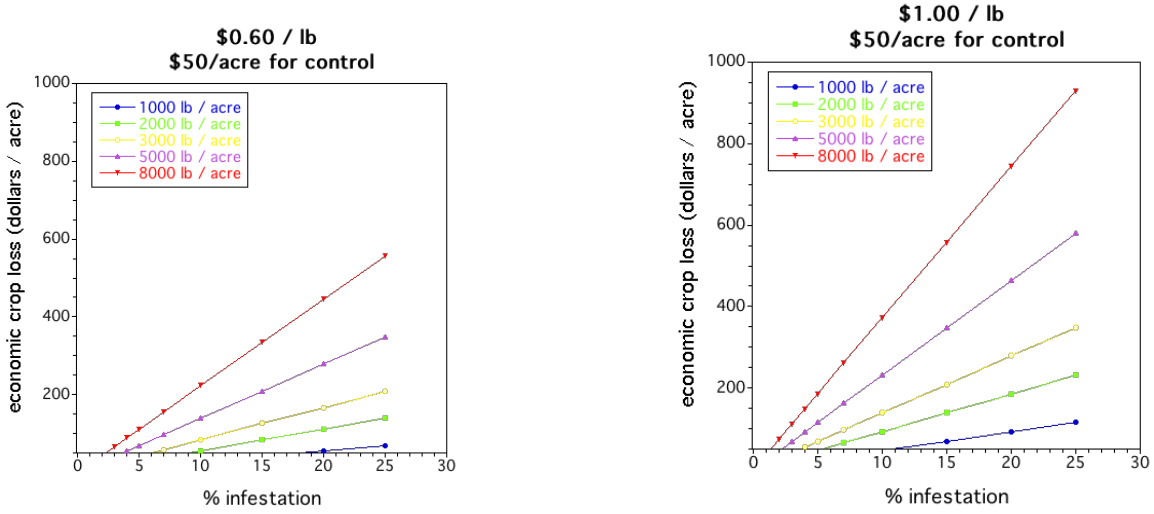


Fig. 3. Economic threshold calculations for control costs of $50.00/acre and two prices for the crop ($0.60/lb and $1.00/lb) under varying levels of production.



CONCLUSIONS: At this point it is not possible to conclude that blueberry tip midge is a consistent pest of concern. The results from our research suggest that tip midge can be damaging and that potential yield loss can be as high as 50% on any one infested stem. However, because stem infestation rate (or the number of infested stems in a field) is usually less than 5%, there is no evidence that control of blueberry tip midge is a justified expense in most cases. This may not be true if tip midge infestation continues to increase as it appears to have done over the past decade.

We plan on continuing research in order to assess the risk that growers face when this pest builds up to noticeable levels. Economic thresholds suggest that a level of infestation ranging from 7-10% should be used for decision making on whether to apply an insecticide for control. Our 2016 data on crop loss will be added to the economic threshold model.

Study 4. Use of yellow pan traps in blueberry tip midge IPM

METHODS: Four specific aspects of the biology and ecology of the blueberry tip midge have been investigated. First, the use of yellow pan traps to monitor adult populations of blueberry tip midge (BTM) were addressed in 2015. We deployed eight yellow pan traps in April 2015 in a wild blueberry field (Jonesboro, Maine) with a prior history of blueberry tip midge infestation. Twice a week traps were monitored along for adults and plants were observed for signs of leaf-bud attack by the blueberry tip midge. Association between the occurrence and progression of attack and cumulative capture of tip midge adults was analyzed and compared to results from 2014.

RESULTS: A good monitoring tool for BTM would be for adult trap captures in yellow bowl traps to precede the onset of damage. We did not find this to be the case in 2014 or in 2015. In 2014 adult trap capture preceded damage by almost three weeks and in 2015 adult trap capture lagged behind the onset of visible infestation.

CONCLUSIONS: First, we conclude, that unlike the situation in cranberry production, yellow bowl traps do not provide a consistent early warning of infestation and timing for insecticide applications in wild blueberry. In addition, adult BTM are extremely small and difficult to taxonomically identify. There are several species of Cecidomyiidae where the adults can be confused with those of BTM. Therefore, at present we are not recommending that growers monitor for BTM by deploying yellow bowl traps.

Study 5. Influence of fertility and disease management practices on sap-feeding insects, premature flowering, stem characteristics, leaf spot, leaf retention, and foliar nutrients

OBJECTIVE: This experiment was designed to evaluate potential effects that summer applications of fungicide might have on the insect sap guild that feed on wild blueberry. This study was initiated by the increased feeding damage by sap feeding insects observed in New Brunswick, Canada.

METHODS: The experiment was setup at the University of Maine Blueberry Hill Farm. Five replicated plots (20 x 20 ft) were deployed in a field that was in the prune cycle. The fungicides applied were Pristine® (18.5 oz/acre), Pristine (18.5 oz/acre) + DAP fertilizer (200 lbs/acre), and Bravo® (56 oz/acre). All fungicides and DAP were applied on 24 Jun at recommended rates. Materials were applied in 25 gallons of water-mixture per acre with a CO2-propelled, 80-inch boom sprayer (76-inch swath) equipped with four, flat-spray, 8002VS TeeJet® nozzles operating at 35 psi and at a slow walking speed. Walking speed was regulated using a metronome. DAP

was applied using a shaker can to spread the material evenly over the plot. A non-treated control was also included in the experiment.
During the first year of this experiment, leaf spot fungi were sampled and evaluated on 3 Aug and 2 Oct. Sweep sampling for sap-feeding insects was conducted on 2 Jul. Ten sweeps with a standard 12-inch diameter sweep net were taken systematically through the center area of each plot avoiding plot boundaries. Leaf spot fungi were sampled and evaluated on 3 Aug and 2 Oct, and leaf drop was evaluated on 2 Oct. Premature flowering in the prune crop was assessed on 25 Sep. Plots were rated as either with or without stems with flowers present. Stems were collected on 9 Oct to assess stem density, length, branching and number of flower bud clusters. All stems within two, sq ft quadrats per plot were cut and brought into the laboratory. Samples of foliage was also collected from each plot. Ten stems were randomly collected, flowers were removed and the leaves were collected and dried for analysis.

RESULTS: Four species of sap feeding insects were collected in sweep samples on 2 Jul. We did not observe any effects of the fungicide treatments ($F_{(3,12)} = 2.294$, $P = 0.129$). It is interesting, that although not significant, there were more of the four species of sap feeding insects in the non-treated control plots than the fungicide treated plots. As far as leaf retention, the fungicide plots had significantly more leaves in October than the non-treated control ($F_{(3,12)} = 7.049$, $P = 0.006$). The differences were that fungicides had retained more than 90% of their leaves compared to only 75% of the non-treated controls. As far as actual fungal induced leaf spot, we did observe a treatment effect ($F_{(3,12)} = 3.419$, $P = 0.0527$). Figure 1 shows that Pristine + DAP fertilizer resulted in more leaf spot than either Bravo or Pristine without fertilizer. The control was not significantly different from any of the other treatments. We also observed no fungicide effects on premature flowering ($X^2 = 2.331$, $P = 0.507$).
Analysis of Variance (ANOVA, CRD) and LSD ($P \leq 0.05$) were used to compare stem density, length, branching and number of flower bud clusters. Subplots were pooled within main plots. The only significant difference was in the number of flower bud clusters/stem. Plots treated with Pristine + DAP fertilizer or Bravo had significantly more flower-bud clusters than stems treated with Pristine alone or the untreated checks ($F_{(3,16)} = 4.12$, $P = 0.024$) (Fig. 2). There were no significant differences in stem density ($F_{(3,16)} = 1.30$, $P = 0.308$), branching ($F_{(3,16)} = 1.65$, $P = 0.219$), or stem length ($F_{(3,16)} = 1.06$, $P = 0.394$) among the treatments.

The results of the foliage analysis are in Table 1. There were significant differences in levels of nitrogen, calcium, potassium, phosphorus, and aluminum among the treatments. Pristine + DAP treatments had significantly more nitrogen ($F_{(3,16)} = 6.28$, $P = 0.005$) and phosphorus ($F_{(3,16)} = 8.60$, $P = 0.001$) then the other treatments. There was also a significant difference in levels of potassium in Bravo-treated plots compared with plots treated with Pristine ($F_{(3,16)} = 4.78$, $P = 0.015$) and mean separation indicated more aluminum in Bravo-treated plots compared with plots treated with Pristine + DAP ($F_{(3,16)} = 2.46$, $P = 0.101$) and higher levels of calcium in the Pristine + DAP treatment then Pristine alone ($F_{(3,16)} = 2.22$, $P = 0.125$) (Fig. 3).

Fig. 1. The effects on the fungicide treatments applied in 2015 on fungal leaf spot incidence. Letters which are different denote significant differences ($P < 0.05$).

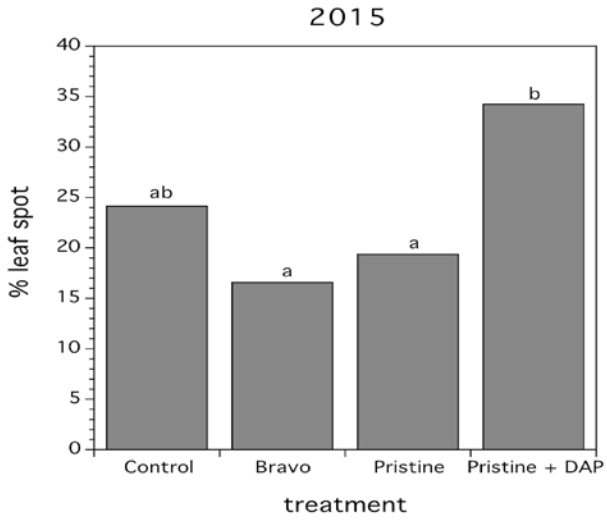


Fig. 2. Mean flower-bud clusters per stem. Lines are standard error of the mean.

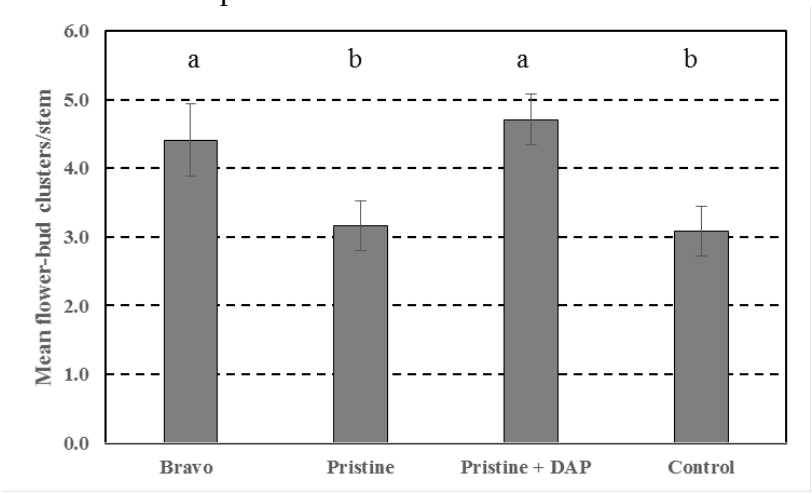


Fig. 3. Foliar nutrients. Lines are standard error of the mean and bars with different letters are significantly different ($P \leq 0.05$).

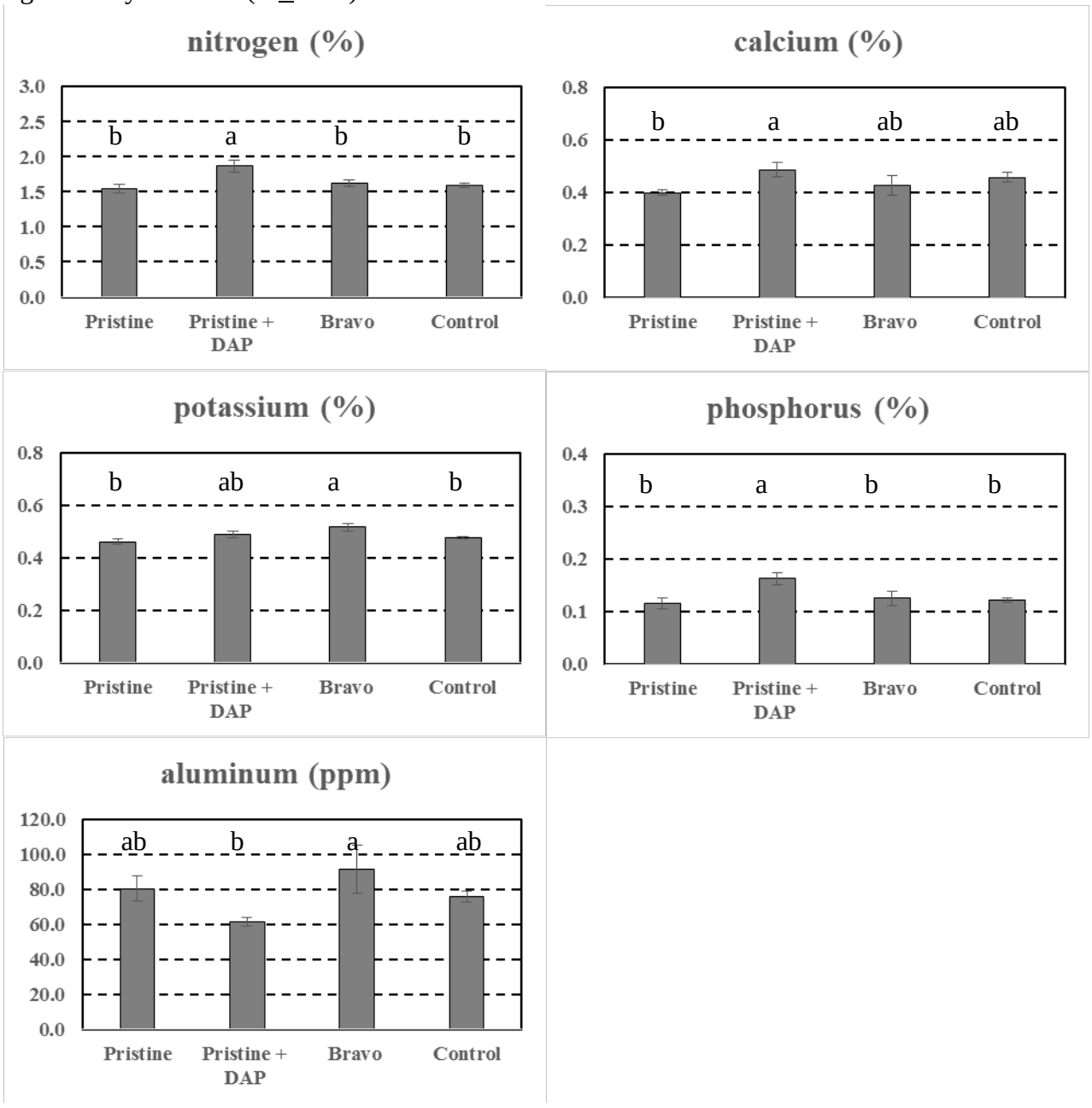


Table 1. Foliage analysis.

	(%)	(%)	(%)	(%)	(%)	(ppm)	(ppm)	(ppm)	(ppm)	(ppm)	(ppm)
Treatment	**N**	**Ca**	**K**	**Mg**	**P**	**Al**	**B**	**Cu**	**Fe**	**Mn**	**Zn**
Pristine	1.54	0.40	0.46	0.20	0.12	80.46	25.40	4.04	36.70	917.40	12.60
Pristine + DAP	1.87	0.49	0.49	0.19	0.16	61.42	23.54	3.72	40.84	1040.20	14.02
Bravo	1.62	0.43	0.52	0.18	0.13	91.52	25.72	4.30	44.24	1197.20	15.10
Control	1.59	0.46	0.48	0.21	0.12	75.82	25.26	4.18	38.12	947.60	14.56

CONCLUSIONS: We have not found any evidence that fungicides applied during the prune year in wild blueberry result in a hazard by increasing sap-feeding insects or stimulate premature flowering. We will continue sampling for sap feeding insects in 2016. Fungicides do result in significantly greater leaf retention, but not relief from fungus attack to the foliage. Sampling potential yield, measured as flower buds in the spring of 2016 will enable us to evaluate the economic benefits of applying fungicides; although, our analysis of flower-bud clusters suggest that Bravo might increase potential yield, but that Pristine will only increase potential yield when nitrogen fertilizer is applied. However, this treatment combination also resulted in increased leaf spot. The main question is whether fungicides, by resulting in leaf retention, increase potential yield the following year.

Study 6. Insect survey on the weed, St. John's wort, in wild blueberry fields

OBJECTIVE: St. John's wort (SJW) in an invasive noxious weed that has been recorded in all counties across Maine. In portions of its invasive range, SJW can build up in very dense aggregations. We sought to quantify the number of fields with flowering SJW and which insects are associated with this weed.

METHODS: We examined 25 wild blueberry fields for the presence of flowering SJW. Fourteen of the fields were in the crop cycle and 11 were in the prune cycle. We only used crop fields which had not yet been harvested since harvesting equipment is likely to pull up or damage SJW plants. Our survey fields were located in the Downeast blueberry growing region and ranged from Aurora to Jonesboro, Maine. At five of our sites, we sampled the insects on one stalk per SJW plant. At the remainder of the sites, we collected 20 stems from each field into gallon Ziploc® bags. We quickly and carefully bagged each stem and then froze the samples to collect and count any associated insects. We examined a total of 132 stalks of SJW for insects. At each field with SJW, we characterized the percentage of the field with SJW and the number of SJW plants.

RESULTS: Of the 14 crop fields we examined, seven contained SJW and of the 11 pruned fields, only one field had SJW. In this particular field, the SJW was growing in an area with a rocky edge which did not appear to be routinely mowed. St. John's wort flowers its second year

so fields that have been mowed within a year (such as in pruned blueberry fields) are unlikely to have flowering SJW. In fields with flowering SJW, there was an average of 6.36 SJW plants per field. SJW was well under 5% of the total vegetation in all fields examined and most plants were found along field edges.

The most commonly associated insects were those in the order Hemiptera with 62 specimens found on 40 plant stalks (Table 1). The vast majority of these were non-predatory with aphids being the most common Hemipteran. Spiders were also commonly collected on stalks of SJW with a total of 40 spiders. We also collected 17 beetles, but it is interesting to note that 14 of them were in the genus *Chrysolina*. This genus contains biocontrol agents that were specifically released to control SJW. Further examination is needed to verify if they are the same species released for control. In general, it can be seen from Table 1 that St. John's wort did not appear to support a high diversity of pollinators, but it did support predator species that might also benefit wild blueberry production by resulting in consumption of blueberry insect pests. Herbivores noted in Table 1 suggest that this weed is under considerable attack by herbivorous insects.

Table 1. Insects found on flowering St. John's wort.

Insect	Number of Specimens	Number of Stalks	Herbivores	Pollinators
Araneae	40	17	NA[1]	NA
Coleoptera	17	10	14	T[2]
Diptera	5	5	T[2]	3
Hemiptera	62	40	58	0
Hymenoptera	10	5	T[2]	9
Lepidoptera	2	2	2	0[3]
Orthoptera	3	2	3	0

[1] NA: there are no spider herbivore or pollinator species, all are predators.
[2] Trace: this refers to the assumption that only few species would be in this order of insects
[3] while moths are significant pollinators, the stage collected in this survey was the caterpillar stage which is an herbivore.

CONCLUSIONS: St. John's wort is an exotic flowering weed found in half of the fruiting fields we examined. It was rarely found in pruned fields probably because of its biannual flowering cycle. St. John's wort is known to form very dense stands in its invasive range; however, it was never found in dense aggregations and most fields had only a few plants. A number of insects and arachnids are associated with this new invasive plant including Hemipterans and spiders. We suspect we would have found more hymenoptera if we had sampled earlier in the year when flowers were at peak bloom. Most of the beetles sampled were in the genus *Chrysolina*, but additional work is needed to verify that these are the same species released for SJW biological control.

Study 7. <u>Potential insect pest problems for the future</u>

Over the past five years, the invasion of new insect pests or resurgence of old pests in Maine wild blueberry has been increasing. We have been vigilant in conducting detection surveys for the past five years. Even though we did not detect spotted wing drosophila previous to its detection in November 2012, we had been conducting surveys for two years previous to its detection by Dr. James Dill. Other recent newly emerging insect pests are the winter moth and the blueberry tip midge.

METHODS: Much of our survey is confined to the blueberry fields that we conduct research in, but growers often bring samples in of uncommon pests. In 2015 we surveyed for the Brown Marmorated Stinkbug (BMS), with a sweep net in both the Coastal and Downeast growing areas while Dr. David Handley conducted surveys in southern and western Maine. The African fig fly (AFF) was surveyed for in 16 blueberry fields when the spotted wing drosophila (SWD) was sampled since it is also attracted to the same trap bait. In 2015, insect samples were sent to us by the blueberry companies: G.M. Allen, Cherryfield Foods, and Wyman & Sons as well as various independent growers. Microscope observations along with consulting taxonomic keys were used to determine species.

RESULTS: Dr. Drummond did not find any BMS in crop fields during his sweeps for late season insect pests and St. John's wort sampling. Dr. David Handley also conducted surveys in southern and western Maine and also did not detect any BMS. This insect pest is quite devastating in the mid-Atlantic region on many fruit crops. A photo is included below (Fig. 1). We also did not see any AFF in the SWD traps this year. It is currently found in eleven U.S. states and like the SWD has potential to be a devastating pest to wild blueberry (Fig. 2).
We did find evidence of two uncommon insect pests in Maine wild blueberry, the black headed fireworm and the cranberry weevil or blueberry blossom weevil (Fig. 3-5). Both of these insect pests are native to the U.S. and may be increasing in abundance in Maine due to climate change. However, both of these pests have also been traditionally observed as cranberry pests. So, is a phenomenon occurring where cranberry pests are adapting to wild blueberry? This has also been the case for the blueberry tip midge. Black headed fireworm was seen in very high numbers in the Downeast blueberry barrens in 2015. We are not sure if just a single early generation of caterpillars occurs or if two generations of caterpillars occurs, the second during bloom. The cranberry weevil was sent to us from the Orland region in 2015. We are not sure if this was just an isolated incident or if this pest is becoming more widespread. Fortunately, there is much known about the biology and control of both of these pests as they are serious pests of cranberry.

Fig. 1. The brown marmorated stinkbug.

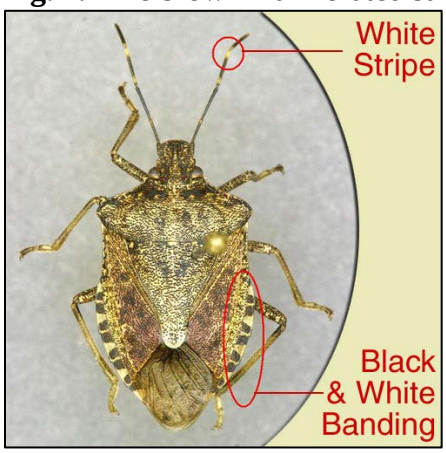


Fig. 2. The African fig fly.

Fig. 3. Cranberry weevil damage

Fig. 4. Cranberry weevil adult

Fig. 5. The black headed fireworm adult and caterpillar (on cranberry)

CONCLUSIONS: In 2015, we did not find evidence that the two new invasive pests that are becoming established in states south of us are in Maine yet. However, it may just be a matter of time before the brown marmorated stinkbug and the African fig fly are part of our pest complex. We plan to investigate the biology of the black headed fireworm in 2016 and continue to track its population growth.

ENTOMOLOGY

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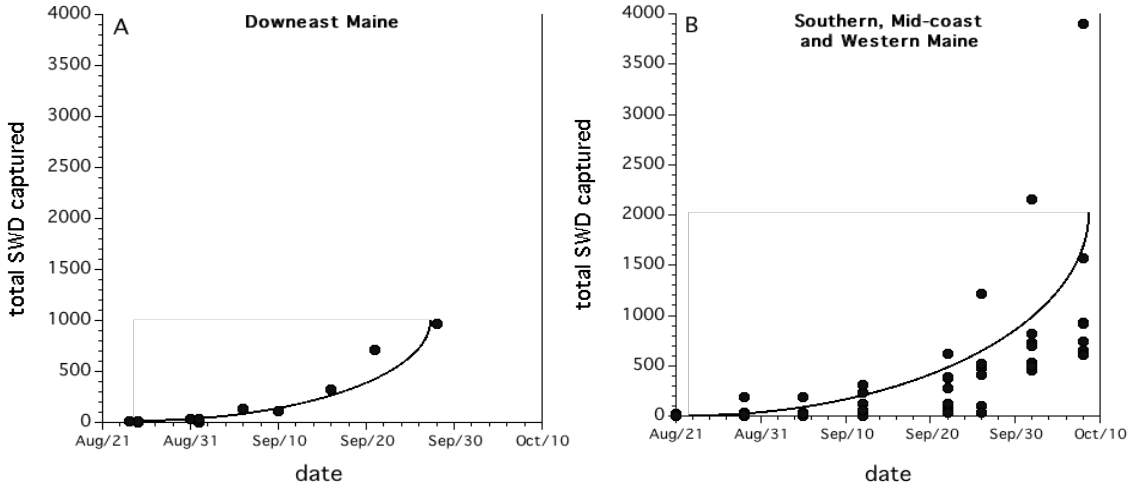
4. TITLE: III. Biology of Spotted Wing Drosophila, 2015.

Study 1. Comparison of adult abundance of spotted wing drosophila with larval fruit infestation

METHODS: To assess SWD adult infestation early in the season (17 Jun), three traps were set in each of four fruit-bearing blueberry fields. After one week the traps were removed and evaluated for the presence of spotted wing drosophila (SWD) adults. Beginning in mid-Jul and continuing until fields were harvested, traps were placed in 17 wild blueberry fields in Downeast and mid-coast, Maine; and Dr. David Handley monitored adult SWD in 27 highbush blueberry and raspberry fields in southern and western Maine. Traps were monitored at 3 to 7 day intervals for the presence of SWD adults. Four traps were placed at each site. All traps were constructed from Solo®, 16 fl. oz, red polystyrene cups with light-blocking lids. Seven to 10, 3/16-inch holes were punched on the side of each container near the top, evenly spaced around the rim. Bait consisted of live yeast (1 tbsp) + sugar (4 tbsp) + 12 oz water (makes enough for four traps). The traps were hung 1-2 ft above the top of the canopy using 36' plant stands. Throughout the study, and on each sample date, traps set the previous week were collected and returned to the laboratory where male, female, and total abundance of SWD adults were determined and recorded. Using this data we calculated the mean SWD males per trap captured from each site for each date and the mean cumulative number of males over the collection period.
To compare adult abundance with larval infestation, fruit samples were taken from the 17 wild blueberry fields on various dates from mid-Jul until mid-Sep and processed using the Salt Extraction Method described in the new factsheet published by Drummond and Yarborough (http://umaine.edu/blueberries/factsheets/insects/210-spotted-wing-drosophila/).
Each sample consisted of 2/3 cup samples collected from the vicinity of the adult trap. There were ca. 359 berries per 2/3 cup sample. Using this data we calculated the mean number of maggots collected from each site on each sample date. This data was compared with the adult abundance data collected over the same time period.

RESULTS: No SWD adults were captured at any site during the mid-June sample week. Figure 1A shows the trap captures in 10 wild blueberry fields in Downeast Maine. The build-up of the SWD population started at the end of August and reached higher populations in southern and mid-coast Maine (Fig. 1B).

Fig. 1. Adult SWD captures for 2015 in 10 wild blueberry fields in Downeast, Maine (A) and 27 highbush blueberry, wild blueberry, and raspberry fields in Southern, Mid-coast, and Western Maine (B).



Only two of the 17 wild blueberry fields that were sampled for SWD in 2015 ended up having infested fruit before they were harvested (Fig. 2 and 3).

Fig. 2. Relationship between sample date, cumulative male fly trap capture and SWD infestation of fruit (Blueberry Hill Farm, Jonesboro, ME).

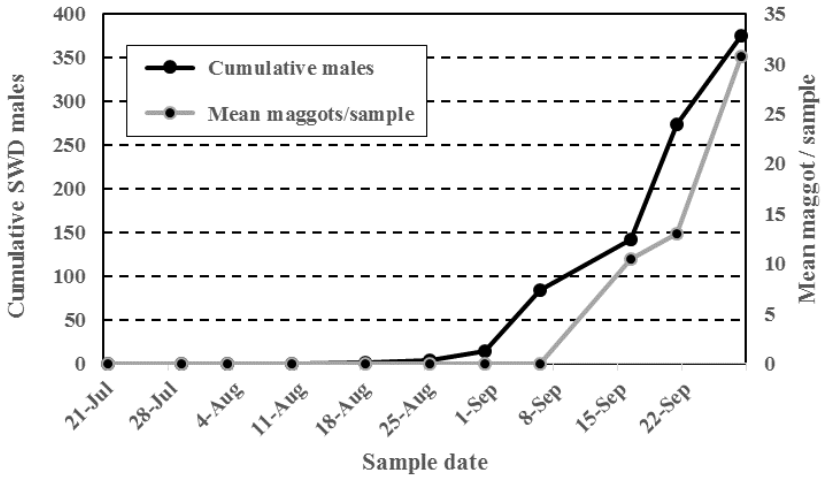
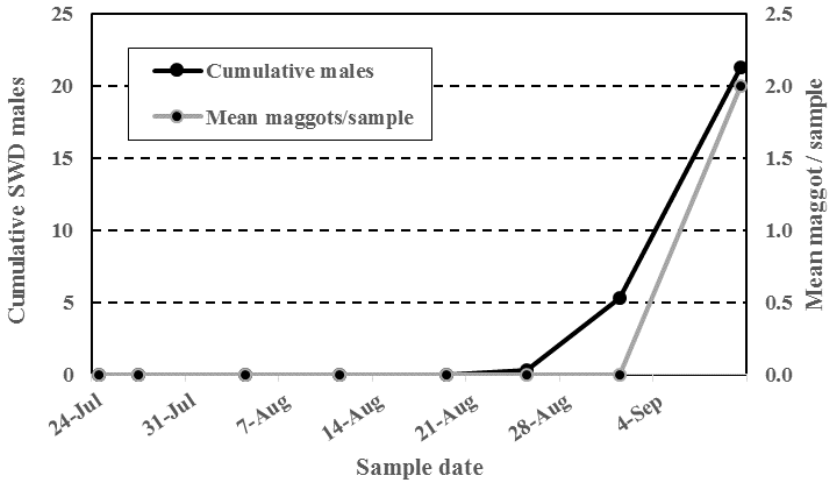


Fig. 3. Relationship between sample date, cumulative male fly trap capture and SWD infestation of fruit (Cherryfield, ME).



There was a wide range of variation between the two fields where maggots were detected in 2015 (Table 1). Cumulative SWD males captured in traps at first detection of infestation ranged from 16.0 to 141.9 males per cup. The range prior to first detection of maggots in fruit was 5.0 to 84.0 males per cup.

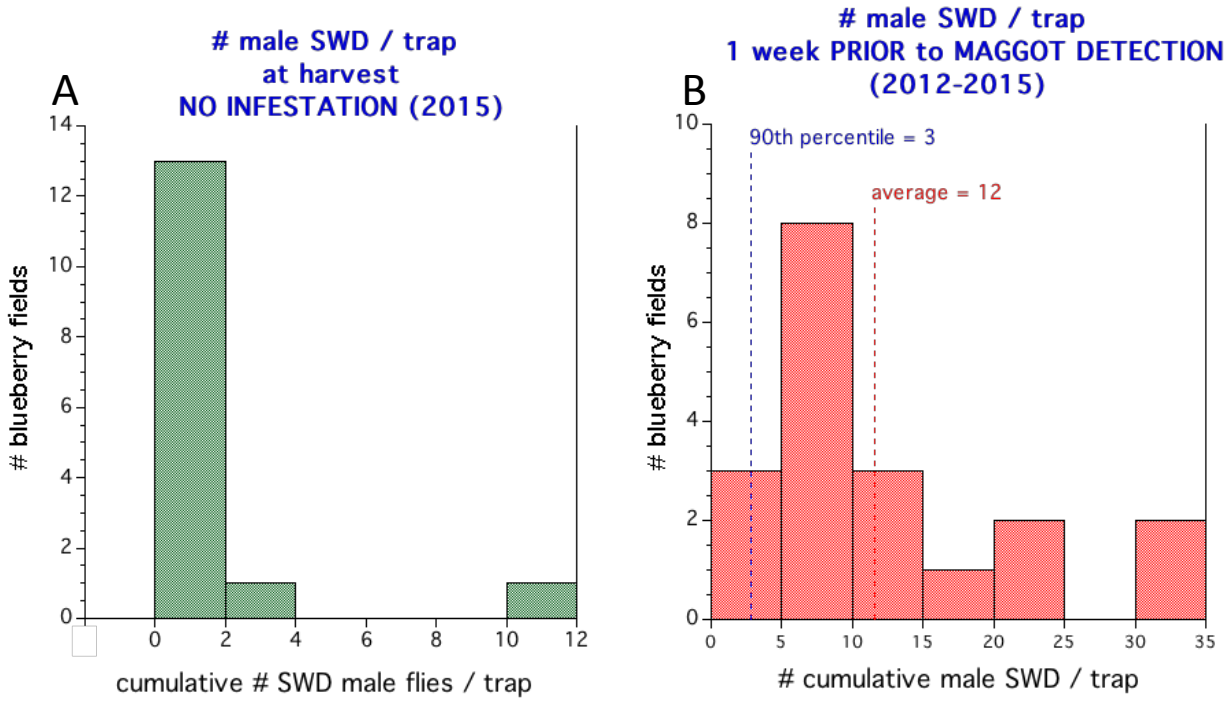
Table 1. Those fields that had maggot infested fruit prior to harvest in 2014 and 2015.

	Sample date[1]	Cumulative SWD males with infestation	Sample date[2]	Cumulative SWD Males prior to infestation
2014				
BBH Garden	2-Sep	16.8	10-Sep	10.5
BBH Sec 4	17-Sep	39.0	23-Sep	30.0
BBH Sec 8	2-Sep	19.5	10-Sep	9.5
Jonesboro	26-Aug	9.7	26-Aug	6.7
Cherryfield	26-Aug	14.7	26-Aug	8.7
Sedgwick	25-Aug	26.0	25-Aug	17.0
Otis	26-Aug	12.3	26-Aug	2.7
Orland	19-Aug	6.7	19-Aug	4.3
Stockton Springs	25-Aug	2.7	25-Aug	1.7
Penobscot	19-Aug	17.0	19-Aug	9.0
2015				
Cherryfield	2-Sep	141.9	10-Sep	84.1
BBH	17-Sep	16.0	23-Sep	5.0

[1] Date of first maggots in fruit samples.
[2] Date prior to first infestation.

Figure 4A shows graphically, the distribution of male SWD trap captures at harvest for those fields that did not have infested fruit prior to harvest in 2015. The majority were characterized by very low male trap captures (≤ 2 SWD males/trap). However, one field had more than 10 male SWD and still did not have infested fruit. Figure 4B shows the distribution of SWD males per trap the week prior to discovering maggot infestation in the fruit for all fields that were infested prior to harvest in 2013, 2014, and 2015. It can be seen that the median male trap capture is 12 males/trap the week prior to first detection of maggots in the fruit. This could be used as a very liberal threshold where there is a 50% likelihood that using 12 males/trap will have no infestation for a week, but on the other hand a 50% chance that the field is already infested. A threshold of three male SWD is more conservative and suggests that there is a 90% chance that with this threshold the field will have no infestation for a week and only a 10% chance that the field is already infested. A threshold of a single male SWD is the most conservative and has the least risk to use. It suggests that there is a 99% chance that when this threshold is reached the field will not be infested until the following week; likewise, a 1% chance exists that the field is already infested by the time the first male fly is captured.

Fig. 4. The frequency distribution of fields with various levels of male SWD/trap not infested by the time of harvest in 2015 (A) and the frequency distribution of fields with various levels of male SWD trap capture the week prior to detection of the first maggot infestation in the fruit (B).



CONCLUSIONS: In 2013, 16 blueberry fields were monitored for SWD fly captures and fruit infestation. In 15 of 16 cases the fields were harvested prior to infestation of the fruit. The one field where SWD fruit infestation was detected had a cumulative male SWD trap capture of 20 flies just prior to fruit infestation. Ten of 14 fields that were sampled for male SWD in 2014 ended up having infested fruit before they were harvested. The results of the 2014 research showed that the 20.0 male SWD may be too high and that 10.0 male SWD was possibly a better

estimate. A comparison of cumulative number of male SWD with date of first maggots in fruit samples resulted in an average of 15.75 SWD males. Our analysis of all infested fields from 2013-2015 suggest that a liberal threshold is 12 male SWD per trap, a more conservative threshold is 3 males per trap, and the threshold with the least risk is 1 male SWD per trap that suggests there is only a 1% chance that a field will be infested by the time the first fly is captured. We need several more years of data to quantify accurately the probability of maggot infestation in a field under a range of thresholds.

Study 2. Within-field movement of spotted wing drosophila

Spotted wing drosophila (SWD) is an invasive insect in Maine that lays its eggs in wild blueberries and is a major concern for blueberry growers. Since its first detection in Maine in 2011, it has spread rapidly across the state and is found in virtually all blueberry growing regions. The wooded edges that commonly border blueberry fields in this region are thought to be places of refuge for the flies and are generally the areas where flies are first detected. In Maine, SWD first appears during the late summer in late July or early August. Previous studies sought to quantify how far and how quickly these flies can travel. We repeated these studies to see if results were similar across years. We conducted this study before any wild SWD were present so that we could avoid the use of dyes which have the potential to confound results. We hope the results of this study can be used to further understand the risk of SWD re-infesting a field after control materials are applied or for predicting how quickly SWD may move into a new area.

METHODS: The trial was run at the University of Maine's Wild Blueberry Farm in Jonesboro, ME. A monitoring cup was placed at each of the four corners of a fruiting blueberry field and checked for wild SWD one week prior to the start of the release trial. The monitoring cups were made from red Solo® cups (16 oz) and contained 4 oz standard yeast bait (4:1:20 sugar:yeast:water). Because no wild SWD were detected in the field, undyed flies were released. Five recapture transects (labeled A through E) were set up in the field with each transect 50 feet from the next one. Within each transect, red Solo® cups were filled with 4 oz standard SWD bait, set on a plant stand 2.5 feet tall, and placed 12.5, 25, 50, 100, 200, and 400 feet from the start of the transect. The Solo® cups had holes punched around the rim of the cup and had a red shade square glued to the lid to prevent overheating inside the cup. On 14 Jul, 3,000 flies were tapped out into Petri dishes at the start of transect C. All flies that had not flown away after 10 minutes were sealed into the dishes and brought back to the lab to count. Flies in these dishes were subtracted from the total number of flies placed in Petri dishes to give an accurate number of flies released. The cups were replaced daily with new cups filled with fresh bait. The cups were brought back to the lab where flies were strained out of the bait and counted. The cups were checked and replaced daily through 21 Jul.

RESULTS: A total of 2,820 flies were released. The first trial had an overall recapture rate of 2% with a recapture of 47 flies. Thirty recaptured flies were female and 17 were male. In this trial, the furthest distance flies flown was 112 feet by males in two days and 400 feet by females in four days (Table 1, Fig. 1.).

Fig. 1. The distance dispersed in a four-day period of marked and released SWD.

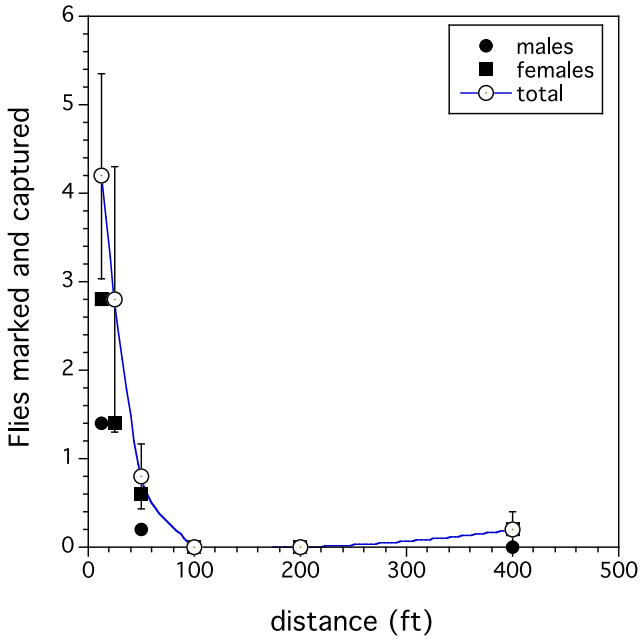


Table 1. The number of SWD marked flies recaptured at each sample point over a four day period in 2015.

Distance (ft)	A	B	C (release point)	D	E
12.5	1 male, 1 females	1 male, 5 females	3 males, 4 females	1 male, 4 females	1 male
25	1 male	1 female	7 male, 4 females	2 females	
50	1 females	1 females		1 male, 1 female	
100					
200					
400			1 female		

CONCLUSIONS: We observed a low recapture rate in our trial this summer. However, this recapture rate is similar to the recapture rates we saw last year during our release recapture study using dyed flies which ranged from 0.24% to 6.14% recapture. These continually low recapture rates highlights the relatively poor attractive nature of the red cup trap and bait. Surprisingly, our recapture rates when using non-dyed flies were not higher than the recapture rates compared to dyed flies.

Last year we were unable to set up a full recapture grid in a fruiting field. We wanted to determine if flies would travel the same distances in a fruiting field compared to a pruned field. This trial proved that flies are still traveling great distances even across fruiting fields. Last year, we observed that a fly was able to travel 400 feet within four days in a pruned field and this year we recorded this was also true in a fruiting field. We did not set up any baited cups past the 400 foot mark. It is likely that flies traveled distances greater than 400 feet; although, capturing them would be a very low probability. This trial has given us additional baseline physiological possibilities of the fly's flight capabilities. Future studies using much greater number of flies will help us determine if perimeter treatments for this pest are worth pursuing.

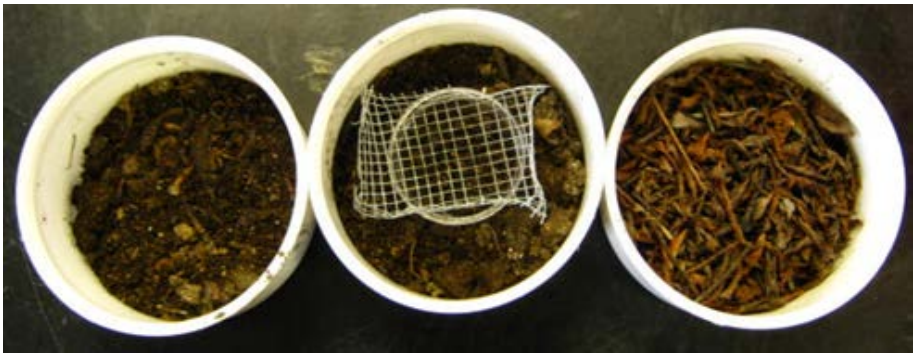
Study 3. Spotted wing drosophila pupation behavior, field and laboratory experiments

In 2014 we ran a study to determine where spotted-wing drosophila (SWD) pupate. We wanted to determine if they were more likely to pupate in the fruit or the substrate. Their pupation location might have major implications for control methods and predation rates by natural enemies. In 2015, we used similar methods as our 2014 study and ran both a lab and field trial simultaneously. In 2014, the lab and field trials had very different results so we hoped running both trials simultaneously could eliminate any confounding factors that may contribute to these differences.

METHODS: We collected wild lowbush blueberries from the University of Maine's Blueberry Hill Farm in Jonesboro. The berries were dipped briefly in water to provide moisture and humidity to the flies during oviposition. Eighteen berries were placed inside of several 9.3 x 2.6 cm clear plastic drosophila culture tubes and 20 adult SWD were added to each tube. The flies remained in the tubes where they were able to oviposit for six days. After the six day oviposition period, the flies were discarded and the berries were added to one of three treatment arenas. All treatment arenas consisted of a round, 9 x 4 cm plastic cup filled with 28.3 g (1 ounce) of blueberry soil by volume and were lightly misted with water. The first treatment consisted of blueberries sitting directly on top of soil. The second treatment was comprised of blueberries situated on a coarse mesh platform 2.5 cm (1 inch) above the soil substrate. The final treatment involved the blueberries sitting directly on top of the substrate, but these arenas also had 28.3 g (1 ounce) by volume of blueberry duff over the soil. The tops of all arenas were covered with very fine mesh held in place with a rubber band to prevent SWD maggots from crawling out during the trial. There were a total of 20 replicates per treatment for a total of 60 arenas. Half of each treatment was placed in the lab under room temperature conditions and half were placed in a fruiting blueberry field at the blueberry research farm in Jonesboro. The replicates in the field had aluminum roofs (17 x 17 cm) propped over the tops of the arenas to prevent rain entering during the trial. The arenas were misted with water approximately every other day during the

trial. The infested berries were placed in the arenas on 17 Sep and the trial ended on 28 Sep. After the trial ended, the arenas were frozen to stop pupal development. The berries and substrate in the arenas were examined carefully for the presence of pupae. We selected ordinal logistic regression to detect differences in pupation site between treatments and experiment locations.

Fig. 1. SWD adult ovipositing in blueberries (left) and the three treatment arenas (right): soil substrate, elevated above soil and soil-duff substrate.



RESULTS: Most larvae pupated in the substrate. This was true for both the lab and field trial. The proportion of pupae in the fruit were significantly different between the lab and field trials ($\chi^2_{(1)} = 31.31$, $P < 0.0001$). The lab trial had higher proportions of pupae in the substrate with 4 to 15% of pupae in the substrate compared to the field trial which only had up to 2% of its pupae in the substrate. The lab trial had 0 to 9 pupae in the fruit and 0 to 49 pupae in the substrate with respective averages of 2.25 to 17.62 pupae. The field trial had 0 to 1 pupae in the fruit and 0 to 31 pupae in the substrate with respective averages of 0.033 to 10.34 pupae. The proportion of pupae in the fruit were significantly different among treatments ($\chi^2_{(2)} = 10.71$, $P = 0.005$). In both the lab and field trials, of the three treatments, the treatment with the fruit elevated above the substrate had the largest number of pupae in the fruit (Tables 1 & 2). In the field, only the elevated treatment had pupae in the fruit, all other treatments in the field had 100% of pupae in the substrate. In both locations, fewer larvae developed successfully to pupae when the fruit was elevated above the substrate compared to fruit that sat on the substrate.

Table 1. Pupation location lab study results.

Arena type	Pupae in substrate	Pupae in fruit	Total pupae	% pupae in substrate	% pupae in fruit
Fruit on soil	175	8	183	96	4
Fruit elevated above soil	63	11	74	85	15
Fruit on duff	273	44	317	86	14

Table 2. Pupation location field study results.

Arena type	Pupae in substrate	Pupae in fruit	Total pupae	% pupae in substrate	% pupae in fruit
Fruit on soil	114	0	114	100	0
Fruit elevated above soil	48	1	49	98	2
Fruit on duff	138	0	138	100	0

CONCLUSIONS: The 2015 results were similar to our 2014 study. In 2014 we found almost all pupae in the field trial in the soil substrate and in the lab trial there was more of a mix of pupae in the fruit and soil substrate. However, the 2015 study found considerably fewer pupae in the fruit compared to 2014. In 2014, the lab trial used highbush blueberries which may have influenced pupation site selection. It was interesting to note that there was again a considerable difference between the lab and field trials. This suggests that environmental factors such as temperature, light, and humidity might have an influence on pupation site selection. The 2015 summer was unusually hot and dry and most of the fruit in the experiment had shriveled and dried by the end. The substrate may have been a more favorable location because of moisture. Pupation site may potentially have a large impact on predation. Natural enemies are able to find and consume SWD pupae when they are in the substrate (see Study 4 of this report), but it is unknown how well they can locate the pupae when they are in the fruit. Further studies are needed to understand how environmental factors influence pupation site selection. It would also be beneficial to study how far a maggot travels before choosing a pupation site.

Study 4. Field predation of spotted wing drosophila pupae

OBJECTIVE: Spotted wing drosophila (SWD) is a relatively new pest in Maine, first detected in 2011. Repeated surveys over the last two years have not revealed any parasitoids attacking this fly. SWD spend most of their immature stages inside of fruit, which makes them hard to control and is difficult for predators to locate. Previous studies have revealed that a majority of SWD pupae pupate outside of their fruit host which makes this stage more vulnerable to predation than the egg and larval stages. We sought to quantify the number of SWD that were consumed by predators and to determine which types of predators may be responsible for consuming these individuals.

METHODS: Spotted wing drosophila pupae were collected from our lab colony and examined under a microscope to verify the presence of living pupae. To prevent emergence during the trial which would confound our results, we froze and killed all living pupae for 24 hours prior to using them in our experiments. The freshly killed pupae were attached to two, 7 cm strips of double sided tape on top of a 9 cm Petri dish. Each strip of tape had 10 pupae on them for a total of 20 pupae per plate. This was replicated 33 times per field location.
In the field, the dishes of pupae were set up in three transects 3 m apart from other dishes and each transect was set 3 m apart from the other transects. The dishes were set up under one of four different treatments (Fig. 1). The first treatment was a plate under a coarse diameter wire cage to exclude large predators (1 x 1 cm openings) such as birds and rodents. The second

treatment was set out in the open but covered with a layer of blueberry duff to simulate pupae that pupate in the substrate. The third was a dish set out in the open which allowed access from any predators. Our final treatment was a plate set inside a coarse cage covered with fine mesh to prevent access from any predators. This last treatment acted like a control to verify that wind and rain were not dislodging the pupae. The open, wire, and duff treatments were replicated ten times per field, and the control was replicated three times per field. A total of 660 pupae were inspected, set up, and monitored for predation at each field.

In addition to the plates with SWD pupae, we also set up a transect line of pitfalls traps. The pitfall traps were arranged in a single linear transect with 3 m between traps. The holes were dug using a post-hole digger and then a 20 oz. (6.35 x 11.43 cm) deli cup was set into the hole so that the lip was at ground level. The pitfalls were filled half way with soapy water and covered with a 17 x 17 cm tin rain cover propped up by nails.

This trial was replicated at two field sites. One site was located in a commercial organic blueberry field in Stockton Springs. The trial was set up on 16 Sep in a pruned field, but next to a rock wall with unharvested blueberries running alongside it. The second trial was set up on 15 Sep at the University of Maine's Blueberry Hill Farm in Jonesboro in a fruiting field.

The dishes were checked every other day for six days for a total of three checks. The dishes covered in a layer of blueberry duff were only checked once at the end of the experimental period since it was too difficult to examine them while they were covered in the duff layer. The soapy water in the pitfalls was replaced every other day and all collected insects were stored in alcohol. The pitfalls were left out for the same amount of time as the pupae dishes. Statistical analysis was not possible with this data as most of the treatments experienced 100% predation by the second day after deployment in the field. We analyzed the data using logistic regression in JMP12®.

Fig. 1. Four treatments, from left: a coarse screen cage around dish, open dish not enclosed by any cage, control with fine mesh surrounding the pupae, and an open dish covered in a layer of blueberry duff.

RESULTS: Pupae predation rates were high at both fields. Most pupae were eaten within the first 48 hours after being placed in the field. In Jonesboro, predation was much higher in the caged and open plates compared to the plates covered in duff. In this site, 90% of the pupae were eaten in the open and caged treatments but only 36% of pupae were missing from the duff plates. In Stockton Springs, all three treatments were nearly identical with over 95% predation for all treatments. No pupae were missing from the controls (Table 1). However, the mesh came unglued for two of our controls in Stockton Springs and predators were seen inside the mesh; therefore, those were excluded from the results. Overall, the three treatments were significantly different from one another ($\chi^2_{(2)} = 17.32$, $P = 0.0002$). Contrast analysis revealed that the caged and open treatments were significantly different from the duff treatments, but not from each other (Table 2). Predation between the two sites was also significantly different ($\chi^2_{(1)} = 16.96$, $P < 0.0001$). The most common predators observed were crickets, ants and harvestmen. Very few ground beetles were captured in pitfalls (Table 3). In 2014, a laboratory study showed that ground beetles were voracious predators on SWD pupae. However, it appears from this year's study that other members of the predator community in blueberry fields might also be eating SWD pupae.

Table 1. Percent of pupae missing across three dates for both trials.

Location	Treatment	Percent Missing Pupae Sample 1	Percent Missing Pupae Sample 2	Percent Missing Pupae Sample 3
Jonesboro	Caged	94.5	96	96
Jonesboro	Open	89	92.5	94
Jonesboro	Duff	ns*	ns	36
Jonesboro	Control	0	0	0
Stockton Springs	Caged	99	100	100
Stockton Springs	Open	99.5	100	100
Stockton Springs	Duff	ns	ns	95.5
Stockton Springs	Control	0	0	0

*ns = not sampled until last sample date

Table 2. P-values from contrast analysis.

	Caged	Duff	Open
Caged	------	0.0003*	1
Duff	------	------	0.0003*

* = indicates statistical significance

Table 3. Total numbers of predators and scavengers collected in all pitfall traps by field.

Predators/Scavengers	Stockton Springs	Jonesboro
Crickets	105	81
Beetles	1	2
Harvestmen	2	21
Ants	20	47

CONCLUSIONS: Pupae predation was very high in both trials and in all treatments in 2015. Predators or scavengers appeared to find the pupae even when the pupae were hidden under a layer of blueberry duff. This is very promising since it appears that many SWD pupae tend to pupate in the substrate (see Study 3 of this report). These results indicate that any predators in the field may still be able to locate and consume the pupae even when buried in the substrate. The duff treatment in Jonesboro was the only incident of less than 90% predation. The different mix of predators in Jonesboro may have contributed to the lower predation rate in the duff treatment. There were fewer crickets in Jonesboro which might scavenge on SWD pupae. We collected very few beetles during the 2015 study especially compared to our 2014 study. It was a very dry summer and may have contributed to a decline in natural enemies. We are encouraged that even with the hardships faced by predators this summer; predation on SWD pupae was still very high. However, SWD has a very high fecundity and even 95% predation may not prevent damaging levels in a wild blueberry field, but it is highly likely that these predation levels will slow the population increase at the end of the summer.

Study 5. <u>Exclusion netting as an alternative method for control of spotted wing drosophila</u>

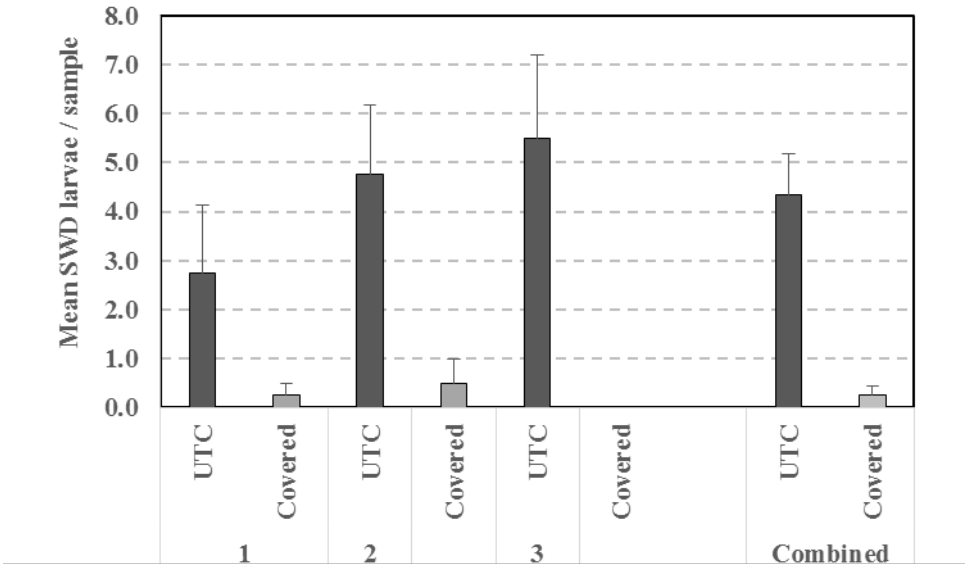
METHODS: Exclusion netting was evaluated for a second year to determine its effectiveness in preventing infestation of fruit by spotted wing drosophila (SWD). Anti-Insect Netting, (25 Mesh) was placed in three areas of a fruit-bearing wild blueberry field at Jonesboro, ME (Fig. 1). The nets were deployed on the fields prior to fruit ripening.

Fig. 1. Exclusion netting covering wild lowbush blueberry

Two replications (13' wide x 50' long) were set on 29 Jul and the third (13' wide x 100' long) was set on 30 Jul. Red cups with standard yeast/bait solution were placed in each trial area to monitor for the presence of SWD adults. Fruit samples were taken periodically to determine the beginning of the fruit infestation period. On 25 Sep the exclusion netting was removed and four fruit samples were taken from areas protected by each block of netting and paired areas not protected by any netting. Each sample was ca. 2 cups of ripe fruit. The fruit was processed to determine maggot infestation using the Salt Extraction Method described the new factsheet published by Drummond and Yarborough (http://umaine.edu/blueberries/factsheets/insects/210-spotted-wing-drosophila/).

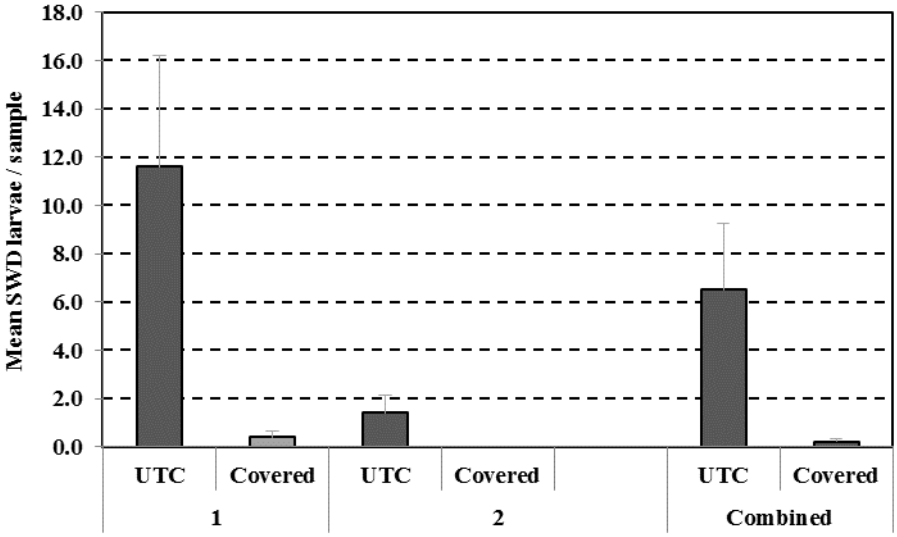
RESULTS: Logistic Regression was used to evaluate the results. In this study, exclusion netting appeared to be an effective means of suppressing fruit infestation in limited areas (Fig. 2). We did not find any evidence for a block effect (χ^2 = 1.1018, P = 0.5764) or any treatment by block interaction (χ^2 = 3.4187, P = 0.1810). However, we did find a significant treatment effect (χ^2 = 15.851, P < 0.0001). Significantly fewer maggots were found in fruit samples collected from the exclusion (net covered) areas compared with the unprotected check areas (UTC).

Fig. 2. Bar graph showing mean SWD larvae per treatment. Lines are standard errors of the mean. 2015 data.



These results support the results of our 2014 trial where netting was placed in two fruit-bearing wild blueberry fields at Jonesboro, ME. One replication in time was set on 9 Jul and the second on 18 July. We found that there was a time replication effect ($F_{(1,9)} = 3.877$, $P = 0.081$), suggesting that the first time replication (Block 1) had higher SWD infestation than the second time replication (Block 2). We did not find any evidence for a block effect ($F_{(4,9)} = 0.713$, $P = 0.604$) or a treatment by block interaction ($F_{(4,9)} = 0.858$, $P = 0.524$). However, we did find a significant treatment effect ($F_{(1,9)} = 5.478$, $P = 0.044$). Significantly fewer maggots were found in fruit samples collected from the treated (net covered) areas compared with the unprotected check areas (UTC). Where infestation was high (Block 1), < 1 maggot/sample was found; two of five samples had one maggot each. At lower infestation levels (Block 2), no maggots were found in any fruit sample from the protected area (Fig. 3).

Fig. 3. Bar graph showing mean SWD larvae per treatment. Lines are standard errors of the mean. 2014 data.



CONCLUSIONS: This was the second year in which we evaluated exclusion netting as a possible alternative control method against SWD. In both years, the netting has proven be highly effective deterrent to infestation by SWD. This control tactic appears to have promise for the small grower. An inexpensive netting will be trialed in 2016 with the hope of demonstrating a netting (Reemay® $700 / acre) with an estimated lifespan of six years.

Study 6. Survey of parasitoids of spotted wing drosophila

Spotted Wing Drosophila (SWD) is an invasive pest in Maine with no known native, natural enemies in this state. Other states have detected parasitoids that attack and can successfully complete their development in SWD. Surveys for parasitoids across Maine in 2014 did not yield any parasitoids. SWD spend their egg and larval stages inside of fruit but many pupate outside of their fruit host. We continued our efforts to locate parasitoids in 2015 by placing SWD-infested fruit in wild blueberry fields.

METHODS: We began sampling for parasitoids once SWD maggots were confirmed in fruiting fields. We collected weekly fruit samples across 14 wild blueberry fields and checked all samples for the presence of SWD maggots. We stopped fruit sampling as soon as the fields were harvested. SWD in 2015 came later than they did in 2014, so most blueberry fields were harvested before maggots were observed in the fruit. The only field that was not harvested before we detected maggots was the University of Maine's research farm in Jonesboro. Therefore, the only site we monitored for SWD parasitoids was the University farm.
As soon as maggots were confirmed in fruit samples at the Jonesboro farm, we set out dishes filled with SWD-infested fruit. The infested fruit dishes consisted of a 7 x 3 cm cup filled with sand and a mixture of strawberries, wild blueberries, and bananas. The fruit cups were placed into a sleeve cage with adult SWD. The SWD had access to the fruit for one week before the cups were placed in the field. The time period ensured there was a mixture of eggs, larvae and

pupae in each infested fruit cup. Each week, eight fruit cups were set out at two locations at the University farm and were left for one week in the field. Four cups were set along the edge of a fruiting and pruned field. The other four cups were set along a fruiting field edge next to a wooded edge. The cups had tin roofs propped above them to keep them from filling with rain. After removing them from the field, the fruit cups were set inside of Tupperware® and placed inside totes with damp paper towels. The cups were checked weekly for any parasitoid emergence. The first infested fruit traps were set outside on 10 Sep and the last set were set out on 6 Oct. A total of 48 infested fruit cups were deployed over a six-week period. All infested fruit cups were kept in the lab for at least one month after being retrieved from the field.

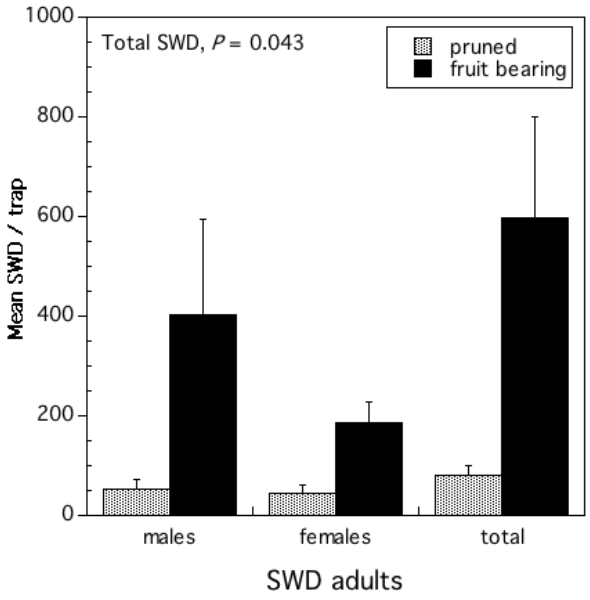
RESULTS: Similar to 2014, were unable to locate any parasitoids during the course of this study. All infested fruit cups had SWD emerge from them, so we assume there were plenty of prey available and that they were kept under conditions that would allow parasitoid development. 2015 was a particularly dry summer, which does not favor SWD development. This might have contributed to SWD's late arrival as SWD prefer conditions to be cool and damp. Blueberry growers in Maine also harvested their fields earlier to avoid SWD infestation so we were very limited with how many fields we were able to sample for parasitoids. Any parasitoid populations may be too small and fragmented for us to find using a single field. We may need much larger samples across multiple sites in order to locate SWD parasitoids.

CONCLUSIONS: The establishment of parasitoids in Maine could be highly beneficial to blueberry growers, especially in years where SWD build up in numbers earlier than they did in 2015. We will continue to survey for SWD parasitoids in 2016 as well as attempt to obtain parasitoids from the USDA for release in the state.

Study 7. Infestation of wild fruit by spotted wing drosophila

Spotted wing drosophila (SWD) is in invasive insect that attacks wild blueberries in Maine. In 2012, we found that SWD were common in pruned fields, although not as abundant as in fruit-bearing fields (Fig. 1). So, a question that arose is, how can SWD proliferate in prune fields. Use of alternative wild fruits was our first hypothesis. SWD has a very large host range which has helped make it a widespread and successful invader. Surveys from other states in the country have found dozens of different cultivated and wild hosts. Known wild hosts include blackberry, cherry, elderberry, and honeysuckle to name a few. We attempted to survey alternative wild SWD fruit hosts in Maine by focusing on wild fruit found adjacent to wild blueberry fields.

Fig. 1. Abundance of spotted wing drosophila flies in pruned and fruit-bearing fields in 2012.



METHODS: We collected non-blueberry wild fruit during a one-month period along 15 different blueberry fields. We tried to collect fruit at an additional 16th field, but no wild fruit could be found in the vicinity. We began sampling on 4 Aug which coincided with the beginning of our first SWD fly captures in blueberry fields. We collected wild fruit in each field until that field was harvested or until wild fruit could not be located. Our final wild fruit collection was 1 Sep. After a piece of wild fruit was collected, it was placed into a one ounce Solo® cup with a lid. Each cup was labeled with a unique identifier and placed into a tote filled with dampened paper towels to provide humidity. The fruit was checked every other week for SWD development and emergence. In addition to collecting wild fruit from each of these sites, we also deployed four SWD adult traps and took four, ½ cup blueberry samples from each field weekly to check for SWD maggots in blueberries.

RESULTS: A total of 2,257 pieces of fruit were collected during the one month collection period. Of those, 100 pieces of fruit had confirmed SWD infestation which gives us an overall infestation rate of 4.43%. We found the following fruit bordering wild blueberry fields: blackberry, bunchberry, catberry, cherry, chokeberry, dewberry, honeysuckle, juneberry, raspberry, rosehips, and wild raisin (Fig. 2). All of those fruit had confirmed SWD infestation except the chokeberry, juneberry, rosehips, and wild raisin. Raspberry had the highest overall SWD infestation rate with nearly 11% of raspberries collected infested and blackberry followed with just over 9% infestation (Table 1). The first recorded incidence of SWD infestation in wild fruit started on 11 Aug and increased during the course of this study (Fig. 3). We recalculated infestation rates by fruit type starting with the first date of SWD infestation and included all proceeding dates. These late infestation rates were much higher, with roughly 30% of raspberries and bunchberries infested. The first adult SWD were captured in these fields starting on 21 and 27 Jul for our southern and Downeast fields respectively. We found the first SWD maggot infested blueberry samples on September 1st.

Table 1. Infestation rates of wild fruit.

Fruit	Overall infestation rate	Late infestation rate	# sites where fruit collected	Percentage of sites with infestation
Blackberry	9.05	15.27	5	40
Bunchberry	5.05	30.16	7	28.57
Catberry	4.40	4.40	1	100
Cherry	5.74	8.17	8	25
Chokeberry	0	0	6	0
Dewberry	6.52	13.04	2	50
Honeysuckle	1.89	5.66	3	33.33
Juneberry	0	0	3	0
Raspberry	10.95	31.89	8	37.5
Rosehips	0	0	3	0
Wild raisin	0	0	1	0

Fig. 2. Distribution of wild fruit collected.

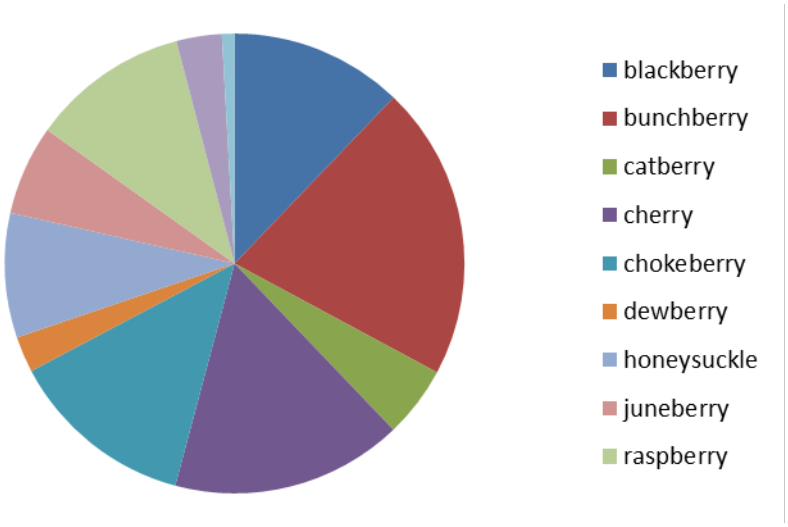
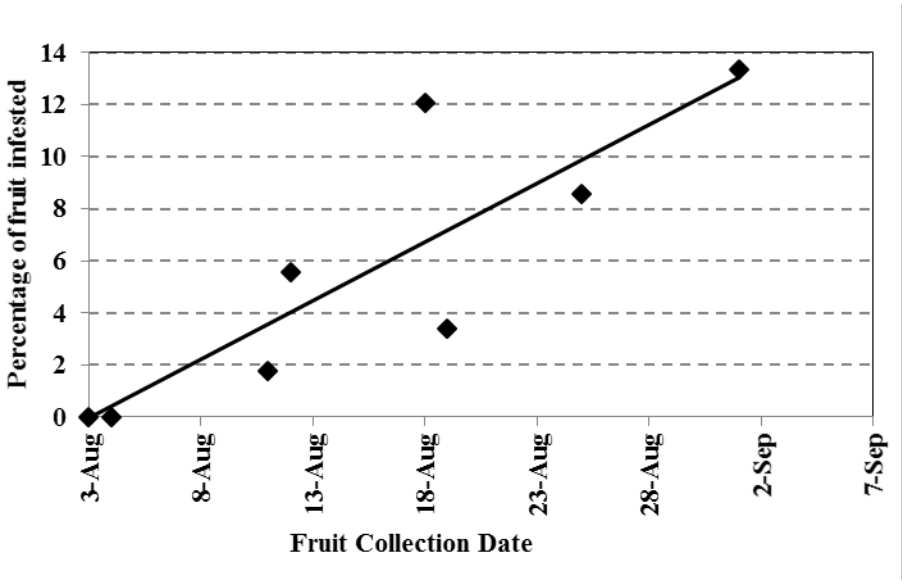


Fig. 3. Spotted wing drosophila infestation in wild fruit increased as the sampling period progressed.



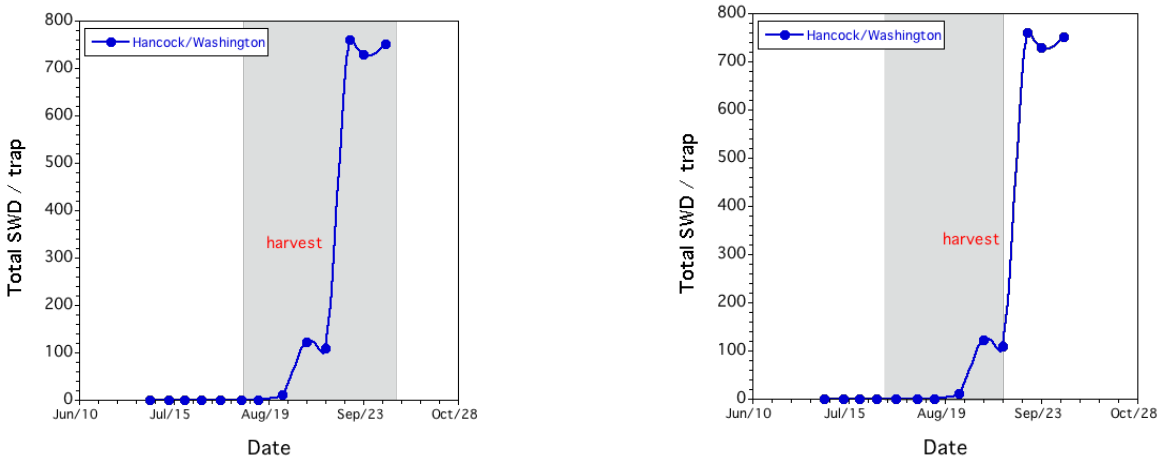
CONCLUSIONS: Blueberry field edges are commonly wooded edges that contain a variety of alternative host plants for SWD. These field edges provide not only egg laying sites, but also shade and moisture that SWD need. These field edges may act as sources of infestation that can allow SWD to move into blueberry fields. Infestation rates were high in several species, especially raspberries, blackberries, and surprisingly, bunchberries. Raspberries and blackberries are known popular hosts, but bunchberries were a previously unrecorded host. This is of particular interest because bunchberry is commonly distributed in blueberry fields. However, SWD did not use bunchberry as a host until roughly the same time as blueberries, so although it is a common plant, it may not help SWD build up in high numbers prior to blueberry infestation. Infestation in wild fruit began several weeks before infestation in blueberries, despite blueberries being ripe and vulnerable to SWD attack. This suggests that SWD prefer these wild fruit earlier in the season. This preference may help to increase SWD densities before they move into blueberries.

Study 8. <u>Spotted wing drosophila and climate change, a hypothetical analysis</u>

Spotted wing drosophila (SWD) is here to stay in Maine wild blueberry. Most often when a new invasive pest becomes established it is not possible to eradicate it. This will be the case with SWD due to its utilization of our abundant wild fruits in Maine. Currently, the blueberry industry is practicing fairly good management of SWD based upon an early harvest tactic along with adult monitoring with traps to determine if insecticide applications are warranted. One question might be…how stable is this management strategy and how likely is it to be perturbed with a change in climate toward milder winters and warmer summers? This question can only be answered by modeling the population dynamics of SWD.

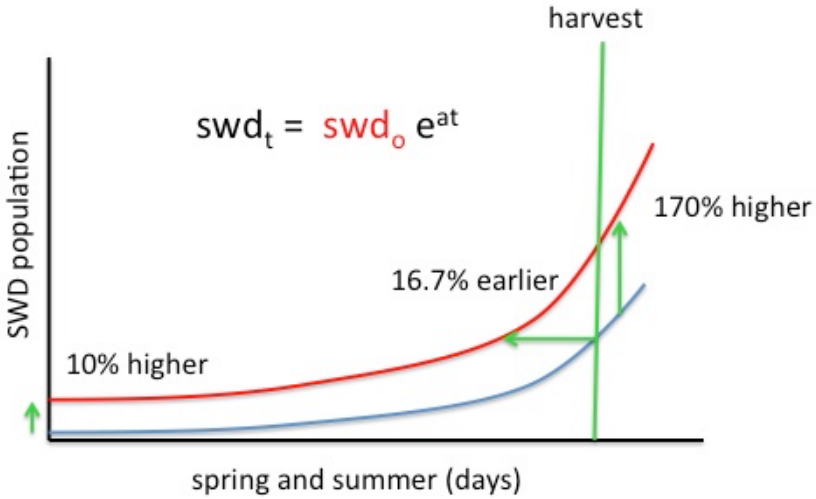
METHODS: We constructed a very simple computer simulation model of SWD population dynamics based upon what we have seen in Maine (Fig. 1). We used an exponential growth model to depict the within-growing season growth of SWD. The overwintering survival represented the early spring initial overwintering population of adult flies. To answer this question in general terms we investigated what might happen under two scenarios: 1) a 10% increase in overwintering survival due to milder winters, and 2) a 10% increase in SWD development rate under hotter spring and summer conditions.

Fig. 1. The population increase of SWD in 2012 as measured by adult fly capture in traps (left), and the effect of early harvest (right). Gray boxes are harvest window.



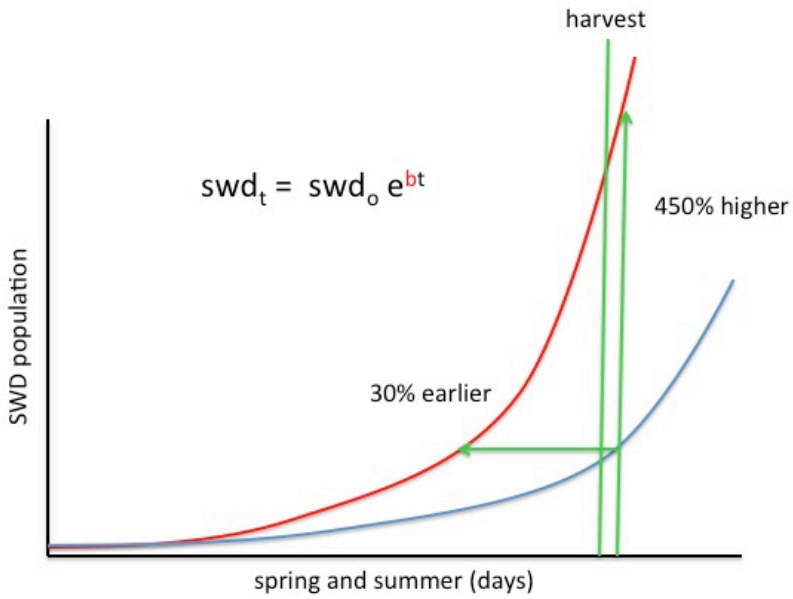
RESULTS: The computer simulation suggests that milder winters that result in 10% better survival increase the late season buildup of SWD that is 170% higher at harvest and 16.7% earlier, relative to the current buildup of SWD (Fig. 2).

Fig. 2. Simulation results for SWD with normal winters (blue) and milder winters (red). Green line depicts harvest time and green arrows depict change in population numbers.



When summer conditions become hotter such that spotted wing drosophila develops more quickly during the growing season, one can see that SWD populations take off and result in abundances 450% greater at harvest compared to typical current summer conditions (Fig. 3).

Fig. 3. Simulation results for SWD with normal spring and summers (blue) and hotter spring and summers (red). Green line depicts harvest time and green arrows depict change in population numbers.



CONCLUSIONS: The conclusions of these simulations have to be considered with extreme care. They do not represent other aspects of SWD population dynamics other than overwintering survival success and development rate as a function of temperature increases. For instance, extreme hot weather has been shown to be detrimental to SWD and reduced summer survival. In addition, if SWD does blow in from states further south of Maine in the early spring, then these populations could alter what we see independent of winter conditions. These simulations only suggest that current management tactics appear to be working for the Maine blueberry industry, but that constant vigilance in monitoring is important as climatic and near-term weather conditions can change and SWD populations might rapidly change. However, one last point. Increase in development due to increases in spring and summer conditions may result in greater population increases than increases in winter temperatures and overwintering success of this pest.

RECOMMENDATIONS: Our research in 2015 provides a growing amount of evidence that monitoring for male SWD can be used as an early warning system for decision making for protecting the crop and preventing damage OR not investing in protecting the crop if it is unnecessary. Essentially, after three years of monitoring and damage assessment we have preliminary action thresholds. The thresholds range from conservative (lowest level of risk) to liberal (moderate risk). The conservative threshold is the first male SWD trap capture in a field, this corresponds to a 99% chance that when this threshold is reached the field will NOT be infested until the following week; likewise, a 1% chance exists that the field is already infested

by the time the first male fly is captured. A threshold of three male SWD is a middle of the road threshold and suggests that there is a 90% chance that with this threshold the field will have no infestation for a week and only a 10% chance that the field is already infested. The most risky threshold is 12 male SWD per trap the week prior to first detection of maggots in the fruit. This corresponds to a 50% likelihood that there will be no infestation for a week, but on the other hand a 50% chance that the field is already infested.

Another piece of data that we collected in 2015 suggests that if your blueberry field is surrounded by other species of wild plants with fruits that you may want to sample more intensely and more frequently for SWD. This fly is using wild fruits along the edges of blueberry fields at a very high rate.

Predation of SWD by existing insect predators in wild blueberry fields is high. 90-100% of all SWD that pupate in blueberry fields are consumed. This is NOT to say that the insect predators will prevent a problem with fruit infestation by SWD, but rather if protection of insect predators can be practiced by spraying when other insect pests are at threshold levels, then the buildup of SWD may be retarded and allow many growers to harvest before damage is detected. This being said, our modeling studies with climate change also suggest that warmer springs and summers and milder winters might increase population buildup and cause more damage in these warmer years. Because of this annual monitoring for SWD is very important. Not having a problem with SWD one year does NOT mean that a problem will not occur the following year.

The last recommendation is that for a few growers, netting laid down on the crop to exclude SWD can work. The netting has to be staked down well to prevent wind from carrying it off the field. Whether netting can be obtained at a cost effective price needs to be investigated.

ENTOMOLOGY

INVESTIGATORS: F. A. Drummond, Professor of Insect Ecology/Entomology
J. A. Collins, Assistant Scientist of Insect Pest Management
E. Ballman, Research Associate in Invasive Species / Entomology

5. TITLE: IV. Biology of blueberry bees, and blueberry pollination.

Study 1. Pollination project

The fourth year of a five-year project assessed bee abundance and subsequent fruit-set and yield in sixteen wild blueberry fields in 2015. At the end of year five (2016) of this study, the relationship between bee abundance and fruit-set and yield will be quantified for more than 140 blueberry fields since 1993.

METHODS:

Blueberry flower-counts and subsequent fruit-set

In late May-early June (peak bloom), six blueberry clones were selected within each of twelve fruit-bearing blueberry fields. Nine of the fields were located in Downeast Maine and seven were located in the mid-coast area of Maine. For each clone, we counted the number of flowers on each of six stems. The stems were marked with numbered metal plant tags. In late June, three marked stems from each clone were cut, placed in individual zip-lock bags, and brought

into the laboratory where fruit-set was evaluated by counting the number of developing fruit on each stem**.** This was repeated in late July-early August with the remaining three marked stems. Sample dates are given in Table 1. The reason that both early and late fruit-set were measured is that we hope to assess soil fertility and leaf nutrient levels on physiological fruit drop (fruit drop between initial fruit formation [swelling of the calyx] and fruit ripening). We collected the same data in 2014. Abiotic conditions such as air temperature, rainfall, and soil moisture are also hypothesized to be responsible for fruit drop; however, it would be necessary to collect fruit-set data and monitor daily weather conditions at each site for many years.

Table 1. Sample dates for flower density and early and late fruit counts in wild blueberry fields located in two wild blueberry growing regions in 2015.

			Flower	**Fruit-set**	
Location	**Status**	**Site #**	**Counts**	**Early**	**Late**
Downeast	High	1	3-Jun	24-Jun	3-Aug
	Medium	2	3-Jun	24-Jun	28-Jul
	Organic	3	3-Jun	24-Jun	3-Aug
	High	4	3-Jun	24-Jun	28-Jul
	Low	5	22-May	24-Jun	21-Jul
	High	6	3-Jun	24-Jun	28-Jul
	Low	7	22-May	24-Jun	21-Jul
	High	8	3-Jun	24-Jun	28-Jul
	Medium	9	3-Jun	24-Jun	28-Jul
Mid-Coast	Medium	10	25-May	22-Jun	14-Jul
	Medium	11	27-May	22-Jun	14-Jul
	Organic	12	25-May	24-Jun	21-Jul
	Organic	13	22-May	24-Jun	21-Jul
	Low	14	27-May	22-Jun	14-Jul
	Low	15	25-May	22-Jun	14-Jul
	Organic	16	27-May	22-Jun	14-Jul

Bee abundance

Two different methods were utilized to study bee abundance, 1) colored bowl traps and 2) visual estimates. On sample dates indicated in Table 2; blue, yellow, and white plastic cups were placed in each plot. Bees were sampled using colored bowl traps on two dates; peak bloom and late bloom. For each sample date, there were three replications of each color in each of 16 fields. Cups were placed such that the top of the cup was even with the top of the blueberry canopy. Each cup was filled ¾ full with water. A drop of unscented dish-washing detergent was added to the water to break the surface tension. Traps were left in the field for 24 hrs. At collection, traps from each site of the same color were pooled and brought back to the laboratory where they were

placed in urine cups with 70% ethyl alcohol for sorting and identification (identification to species is not complete as of the time of writing this report).
Hand collections were also utilized to assess bee populations. A minimum of 20 bees were collected at each site over various sample dates. Collection times (percent effort in person per unit time) were recorded as a measure of bee abundance. To visually estimate bee density, the number of bees (honeybees, bumble bees, and other native bees were counted in each of 16, m^2 quadrats per site (8/block). For each sample we counted the number of bees observed in 1 minute. This was repeated on each of two or three dates as indicated in Table 2. The data from all dates was combined as "mean bees/m^2/min" for the analysis.

Table 2. Sample dates (x – denotes no sample taken).

	Colored bowls		**Visual Estimates**		
Site #	**Sample 1**	**Sample 2**	**Sample 1**	**Sample 2**	**Sample 3**
1	22-May	11-Jun	x	5-Jun	10-Jun
2	24-May	11-Jun	x	5-Jun	10-Jun
3	22-May	11-Jun	21 May	x	10-Jun
4	22-May	11-Jun	x	5-Jun	10-Jun
5	17-May	8-Jun	18-May	28-May	5-Jun
6	22-May	11-Jun	x	5 Jun	10-Jun
7	17-May	8-Jun	18-May	28-May	5-Jun
8	22-May	11-Jun	x	5-Jun	10-Jun
9	22-May	11-Jun	x	5-Jun	10-Jun
10	17-May	4-Jun	18-May	28-May	4-Jun
11	18-May	4-Jun	18-May	29-May	4-Jun
12	17-May	8-Jun	18-May	28-May	5-Jun
13	17-May	8-Jun	18-May	28-May	5-Jun
14	18-May	4-Jun	18-May	29-May	4-Jun
15	18-May	4-Jun	18-May	29-May	4-Jun
16	18-May	4-Jun	18-May	29-May	4-Jun

RESULTS: There was no evidence to suggest a significant difference in early fruit-set ($F_{(3,12)}$ = 1.60, P = 0.242) among the four production systems (organic, low input, medium input, and high input; Fig. 1). Although a trend can be seen that decreases from high input to organic production in Figure 1, but the variation within each production system is high. Early fruit-set is mostly due to bee foraging visitation to the blueberry flowers; although, mummy berry disease and frost can also affect early fruit-set.

Fig. 1. Comparison of percent early fruit-set among production systems, dashed line is the expected background pollination.

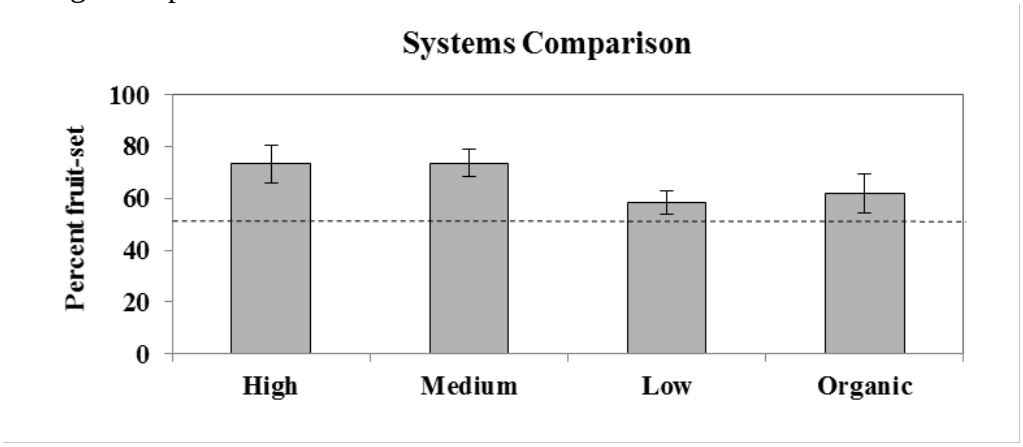


Figure 2 shows early fruit-set for each site within a production system. As expected, early fruit-set measured in late June (Fig. 3) was higher than late fruit-set measured in July just prior to harvest. The lower fruit-set in July might be attributed to a number of factors including insect and disease damage and physiological fruit drop.

Fig. 2. Comparison of percent early fruit-set within each production system, dashed line is the expected background pollination.

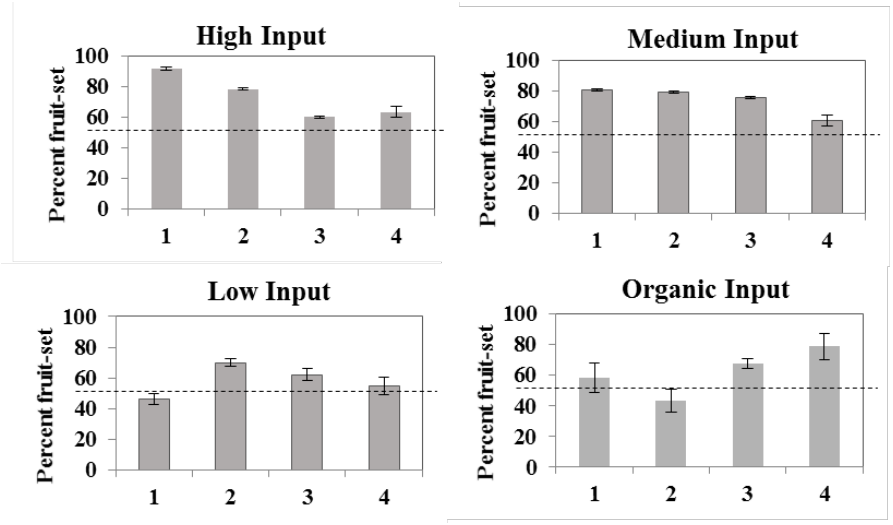
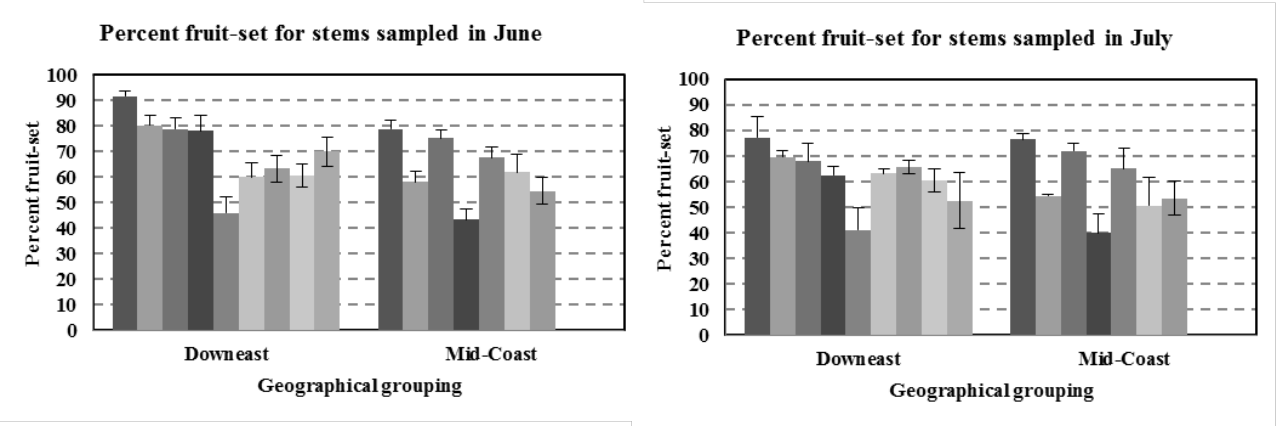
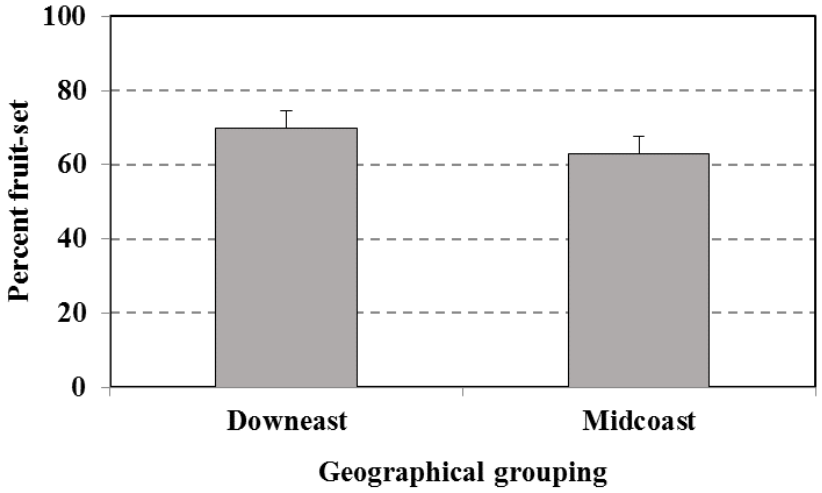


Fig. 3. Percent fruit-set for stems sampled in June or July.



There was no significant difference ($F_{(1,14)} = 1.12$, $P = 0.3083$) in early fruit-set (which is a measure of percent pollination) between the nine sites located in Downeast (69.9%) compared with the seven sites located in the mid-coast area (62.8%), (Fig. 4).

Fig. 4. Percent early fruit-set in fields sampled in Downeast compared with fields sampled in mid-coast, ME.



Figures 5 and 6 show the results of modeling yield based on fruit-set and production system. There was a relationship between yield and early fruit-set as a measure of percent pollination. We have observed this in some years in the past and not in others due to all of the pest and weather factors that often reduce yield from potential yield (fruit-set). Yield was a function of production system ($F_{(1,12)} = 1.12$, $P = 0.043$). Yields were significantly higher in the high and medium input systems compared to the low and organic system. Finally, there was no significant difference in berry weight among the systems (($F_{(1,12)} = 2.44$, $P = 0.115$) (Fig. 7).

Fig. 5. Relationship between yield and fruit-set (percent pollination).

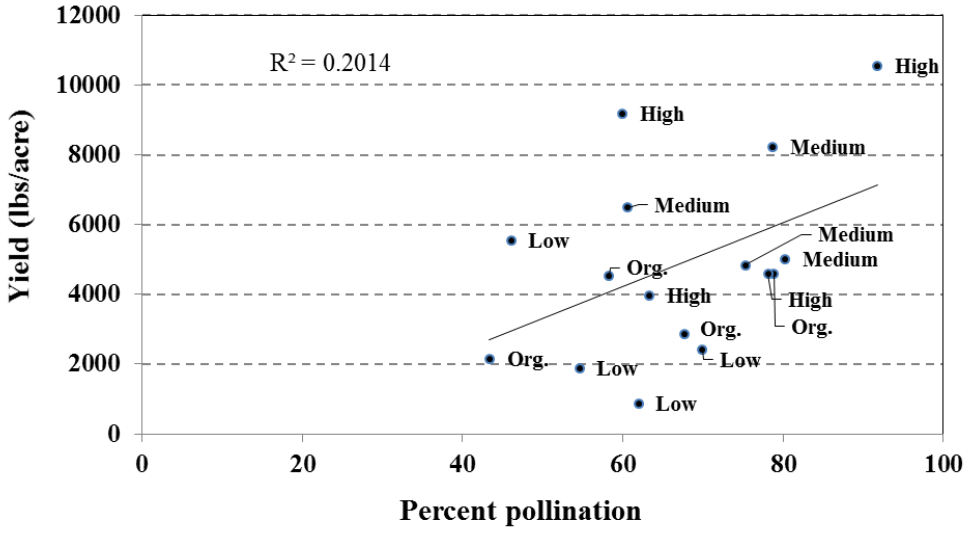


Fig. 6. Yield as a function of production system.

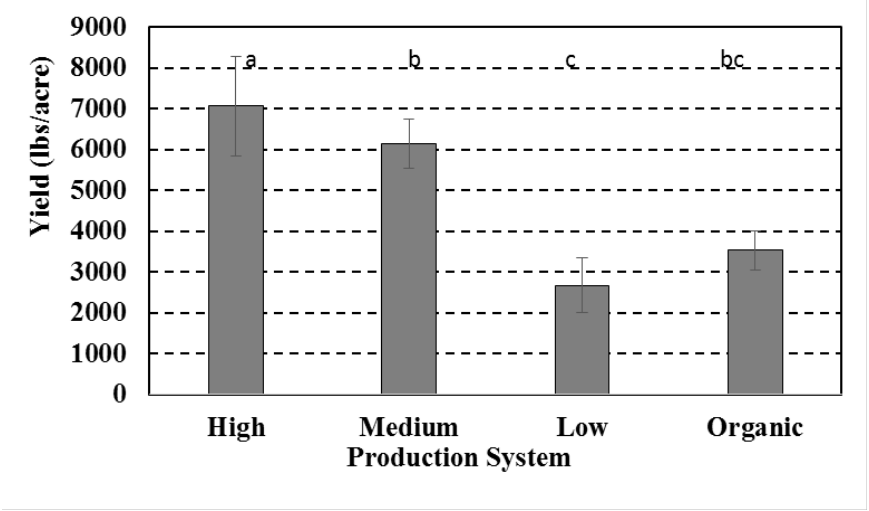
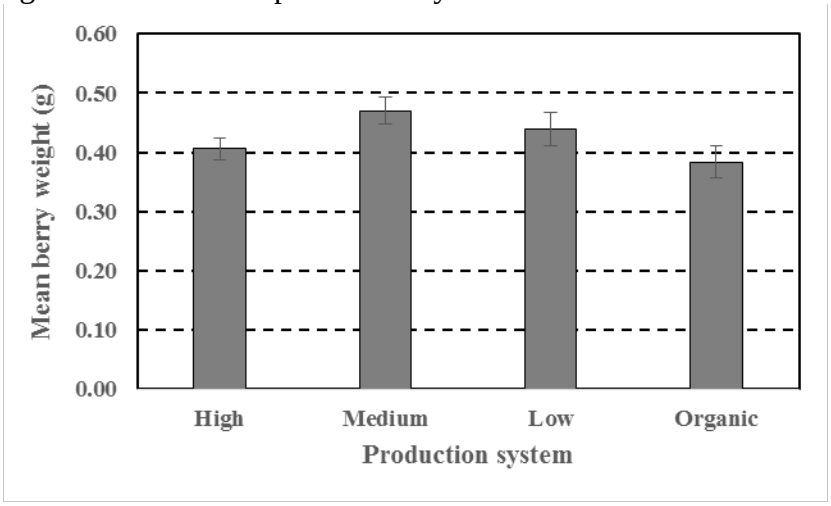


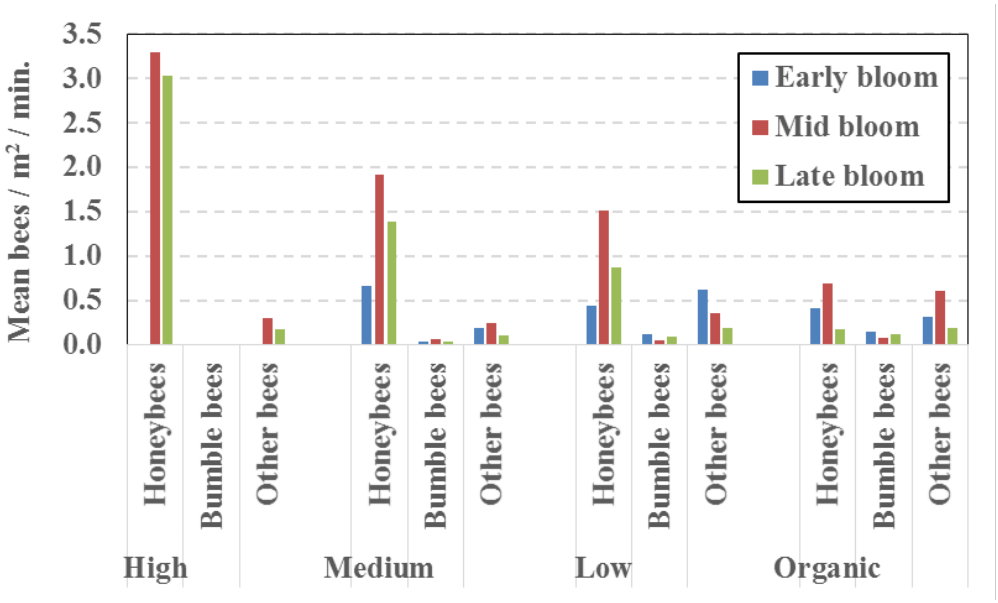
Fig. 7. Berry weight as a function of production system.



Bees - Visual estimates in quadrats

Bee density (bees/m^2/min) derived from the timed 1 minute quadrat counts during bloom are shown in figure 8. Analysis of variance suggests that bee densities were dependent upon the wild blueberry production system. Honeybees were at higher densities in the high input system (3x) compared to the organic system ($F_{(3,12)} = 4.983$, $P = 0.018$). The middle input and low input systems did not differ from either the high or organic systems. Bumble bee densities were significantly higher in organic and low input systems compared to the high input system ($F_{(3,12)} = 11.081$, $P = 0.0009$). The medium input system was not different from either the low and organic or the high input systems. There was no difference in the density of other native bees due to production system ($F_{(3,12)} = 1.426$, $P = 0.283$). The total bee density (all bee groups) was only marginally different among production systems ($F_{(3,12)} = 3.071$, $P = 0.069$). This was due to the inverse relationship between honeybees and bumble bees.

Fig. 8. Relative bee abundance across the four production systems. Visual counts of bees/m^2/min for honeybees, bumble bees (both wild and managed) and other native wild bees.



Bowl traps

Bowl trapping was also conducted in the 16 wild blueberry fields in 2015. Bowl trapping is conducted to collect bees for species identification so that we can estimate the diversity and richness of bee communities in wild blueberry. We use three colors of bowl traps in our sampling so that all bee species will be attracted to the traps. Figure 9 shows the response of both honeybees and native bees to the three bowl colors. Honeybees appear to prefer white or blue bowls to yellow bowls ($F_{(2,78)} = 7.932$, $P = 0.0007$). The native bees have a different color preference. They prefer white and yellow bowls with a slight less preference for blue bowls; however, the native bees did not show a statistical preference for any bowl color ($F_{(2,78)} = 3.142$, $P = 0.375$). The rankings of honeybees and native bees in the four production systems was similar to that observed in the quadrat counts. Native bees were trapped in higher numbers in organic compared to medium input fields and high and low input systems were not different from either organic or medium systems ($F_{(3,12)} = 4.336$, $P = 0.028$). Honeybees were more abundantly trapped in high and medium input systems compared to the organic system ($F_{(3,12)} = 5.924$, $P = 0.010$). Figure 10 illustrates these differences.

Fig. 9. Honeybee and native bee abundance relative to bowl color (both samples combined)**.**

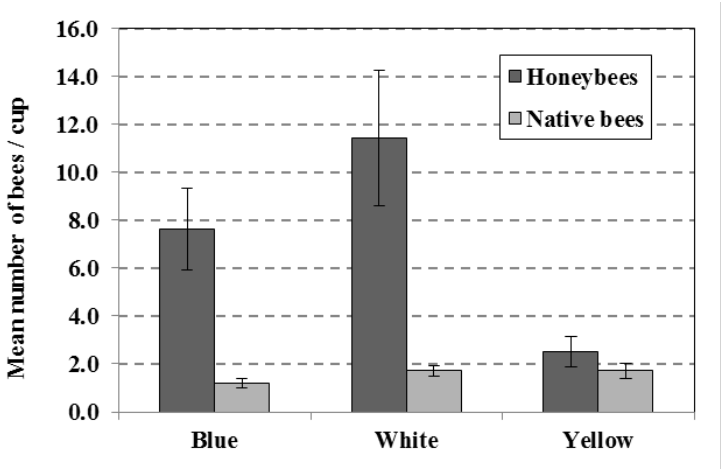
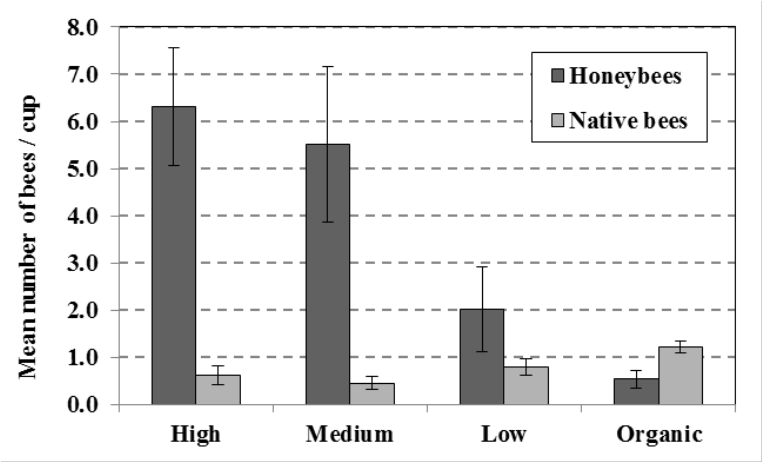
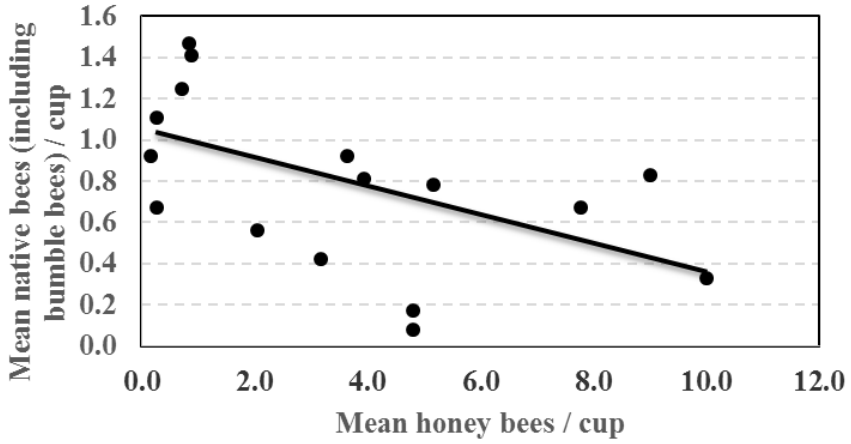


Fig. 10. Honeybee and native bee abundance relative to production system (both samples combined).



The number of honeybees trapped was observed to be in an inverse relationship to native bees trapped (Fig. 11). The negative correlation was significant (r = - 0.521, P = 0.036). This is either due to a detrimental effect on native bees by honeybees or due to the large importation of honeybees into the barrens that do not support large densities of native bees to begin with. This phenomenon has been demonstrated by previous years' data. This relationship may not be suggestive of a problem in native bee communities, but it should be looked at in the future.

Fig. 11. Relationship between numbers of honeybees and numbers of native bees captured in colored bowl traps (an average of sampling dates during bloom).



Hand-collections of bees

Hand collecting bees on flowers was conducted to assess the bee community species diversity associated with wild blueberry, but not necessarily foraging on wild blueberry flowers. The technique involves searching for bees on all flowers in a field and collecting all bees observed. The sampling period is timed as a measure of sampling effort and the bee population abundance is reported as bees/min * #people sampling. There was a significant difference in the number of total bees collected / min. (all bee species combined) among the production systems ($F_{(3,12)}$ = 8.17, P = 0.003). Bees were more easily collected on the high and medium input sites compared to the low and organic sites (Figs. 12 and 13). This is most likely due to the dominance of honeybees in these two systems and supports the findings of the quadrat and bowl sampling.

Fig. 12. The mean bees hand-collected per minute, by site, for each production system. Lines are standard error of the mean.

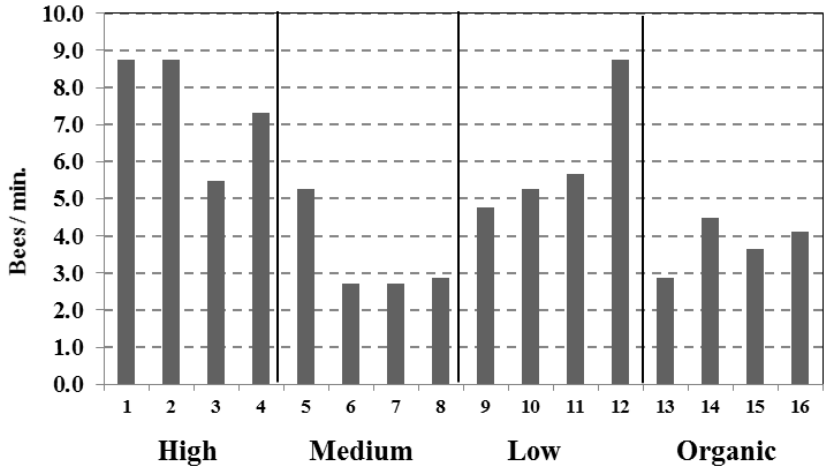
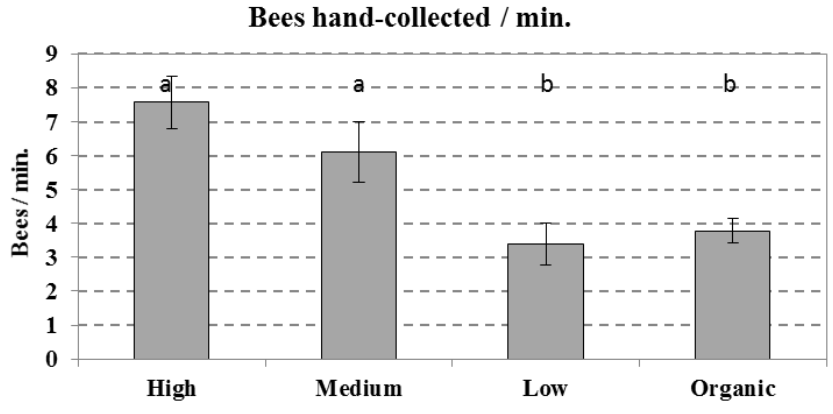


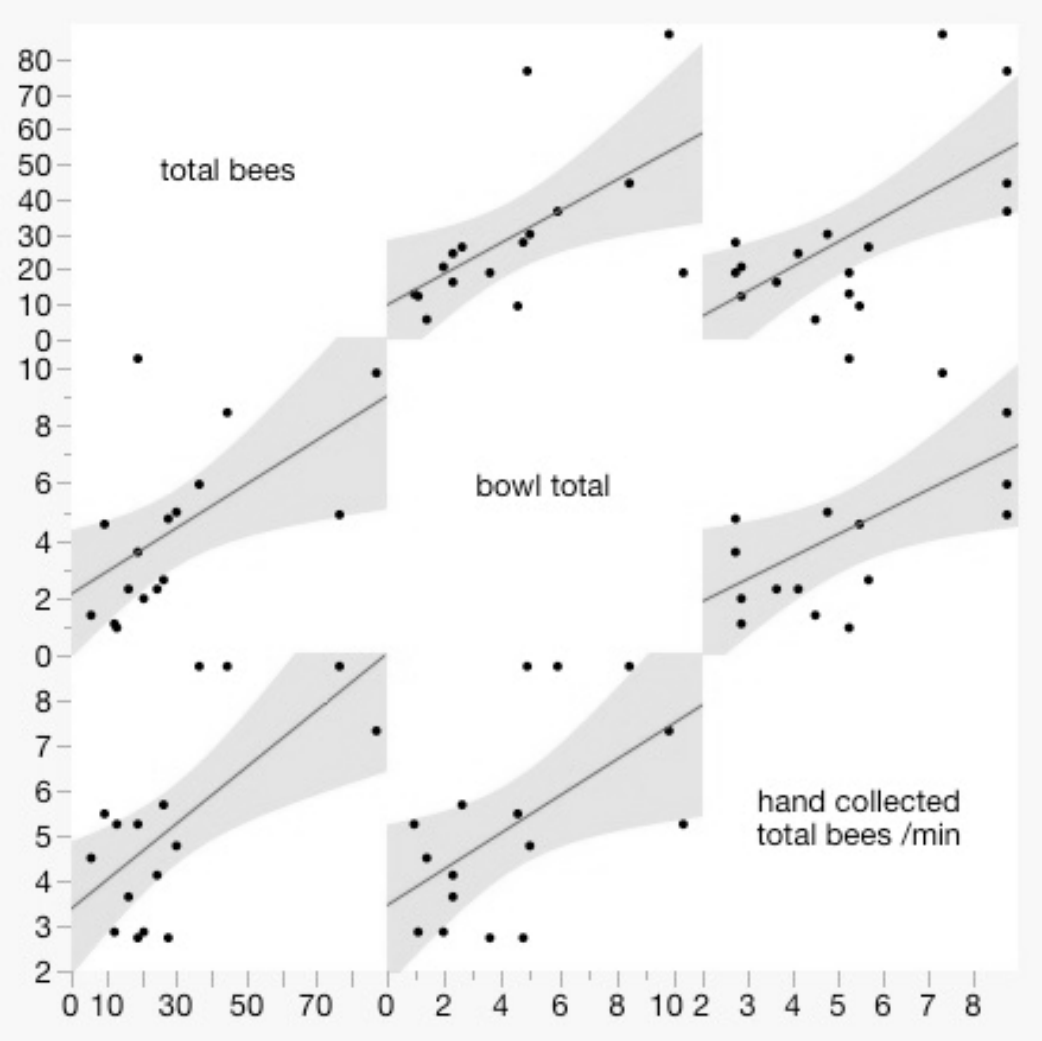
Fig. 13. The mean number of total bees collected in the different wild blueberry fields, by production system. Lines are standard error of the mean.



Comparison of bee capture methods

Since the hand collection technique has only yielded total bees (honeybees + bumble bees + other native bees) by the time of this writing we decided to compare all three bee monitoring methods using total bees. We assume that if each of the methods provides a relative measure of the actual bee population density in a wild blueberry field during bloom, then they should be correlated with one another. Linear correlations among all sampling techniques of total bees were significantly correlated (quadrat vs bowl: $r = 0.68$, $P = 0.004$; quadrat vs hand: $r = 0.492$, $P = 0.052$; hand vs bowl: $r = 0.529$, $P = 0.035$; see Fig. 14). Therefore, it can be stated that all three sampling methods provide estimates of bee population levels. This provides a robust measure of the bee community.

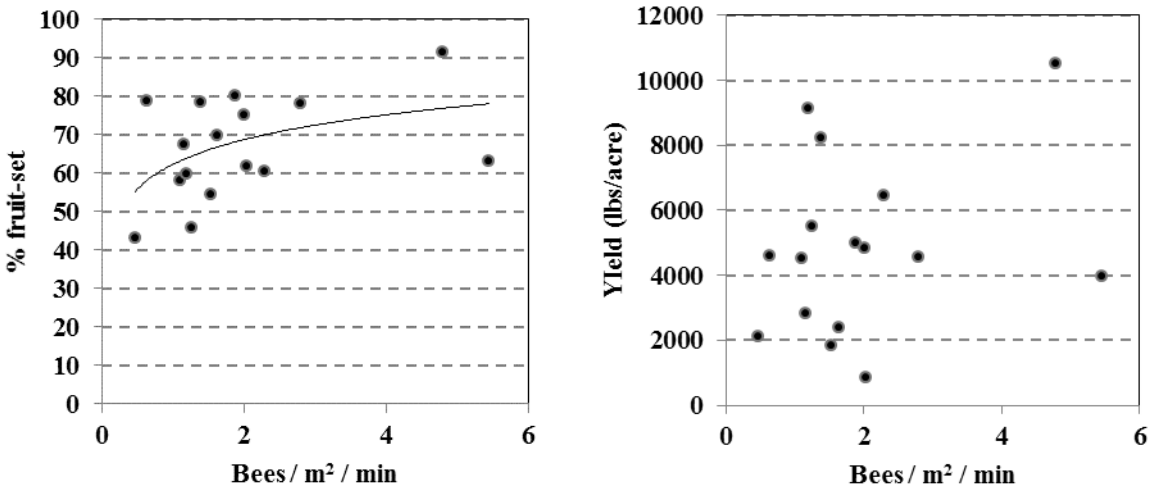
Fig. 14. Graphical representation of the relationship between total bees sampled in each of 16 wild blueberry fields by quadrat sampling, bowl trapping, and hand collecting.



Effect of bee abundance on fruit-set and yield

Bee abundance as a predictor of fruit-set and yield is one of the main tools that growers can use to assess the native bee community and the effectiveness of honeybee stocking. To determine if in 2015 bee density was a good predictor of fruit-set and yield we chose the quadrat measure of bee density as the predictor (bees/m^2/min). In 2015, total bees/m^2/ min were significant predictors of fruit-set ($F_{(1,14)} = 3.546$, $P = 0.081$, $r^2 = 0.202$). Figure 15 depicts the relationship between total bee density during bloom and percent fruit-set. Forty-four percent of the variation in fruit-set was explained by bee density. However, in 2015, there was no relationship between bee density and yield ($P > 0.05$). This is not highly unusual due to other factors that affect yield such as pest damage, drought stress, and fruit drop.

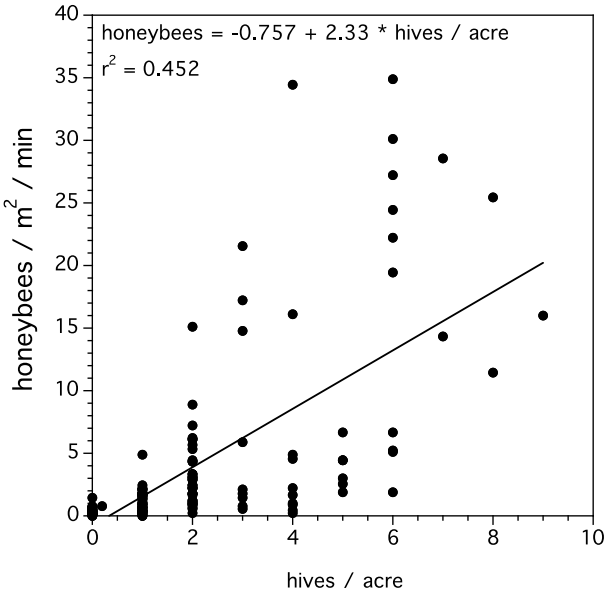
Fig. 15. Relationship between (A) percent fruit-set and (B) yield and the resulting bee densities in each field for visual counts of bees/m^2/min. Total bees = combined data for honeybees, bumble bees and other native bees.



The long-term pollination research that we have conducted by measuring fruit-set, yield and native bee and honeybee densities is now complete. We have data from 162 wild blueberry fields sampled over 11 years (1993-2015). The first result, as expected, we found a significant relation between honeybee hives/acre and the density of honeybees foraging in wild blueberry fields ($F_{(1,160)} = 132.084$, $P < 0.0001$, $r^2 = 0.452$, Fig. 16). The percent of variation in honeybee density explained by the measured number of hives is only 45.2%. This is not surprising since other sources of variation affect honeybee forager density in a fruit-bearing wild blueberry field in bloom. The strength of the colony in a hive, weather conditions during foraging, alternative blooming plants outside the wild blueberry field, all would affect this relationship. Therefore, a grower that rents 4 hives per acre may have varying numbers of bees field to field or year to year.

The main result of this study demonstrates that both native bee and honeybee densities (bees / m^2 / minute) have significant effects on fruit-set. In addition, the production system also has a significant effect on fruit-set. The best model predicting percent fruit-set used the predictor variables: 1) year, 2) production system (i.e. organic, low, medium, and high input production systems), 3) native bee density, and 4) honeybee density. The percent variation in percent fruit-set explained by these predictors was 42.6%. This model suggests that fruit-set is significantly affected by weather (a year effect), production system (most likely due to factors such as mummy berry infection, insect pests, weeds, and potentially soil fertility), and of course honeybee and native bee densities ($F_{(15,156)} = 7.209$, $P < 0.0001$, $r^2 = 0.426$). The bee density coefficients suggest that on a PER BEE basis native bees are roughly 13.1 times more efficient than honeybees. Although on average honeybees are 20.2 $\pm$ 6.7 (SE) times more abundant across all fields in the 11 years of sampling.

Fig. 16. The relationship between measured hives per acre and honeybee foraging strength (honeybees/ m^2/min, n=162).



A predictive model for yield did not include honeybee or native bee density as significant predictors. However, percent fruit-set does predict yield ($F_{(1,156)} = 54.832$, $P < 0.0001$, $r^2 = 0.255$), but fruit-set only explains 25.5% of the variation in yield. This is most likely due to factors such as weed, disease, and insect pest pressure between bloom and harvest, harvest date, soil fertility, and soil moisture. Previously we showed how percent fruit-set is predicted by honeybee and native bee density; however, the best predictors of yield are: 1) year, and 2) production system. This predictive model explains 50.8% of the variation in yield across all fields and years ($F_{(13,144)} = 13.494$, $P < 0.0001$, $r^2 = 0.508$), but only year and production system are significant predictors. However, this does NOT mean that bees are unimportant for good yields, they are. Bees drive fruit-set and then fruit-set drives yield, but a SIGNIFICANT part of yield is ALSO responding to events that occur after bloom such as insect pests, weeds, diseases, plant nutrition, weather, and irrigation.

CONCLUSIONS: In conclusion, bee densities are highly determined by the production system, either intentionally through the rental of honeybees or due to the surrounding landscape and management practices. We found that all three bee monitoring techniques yield an index of bee population levels. In 2015, bee density was a very good predictor of fruit-set, but not a predictor of yield. This should not be interpreted that bees are not important if they are not directly related to yield. Bees are highly indirectly related to yield by being directly related to fruit-set. In fact the relationship between fruit-set and yield was significant ($F_{(1,14)} = 3.529$, $P = 0.081$, $r^2 = 0.201$).

From our 11 years of pollination research that includes 2015 (1993-2015) we have developed predictive models of percent fruit-set (predictors: year, production system, honeybee density and native bee density). We have shown that native bees are much more efficient than honeybees, but honeybees are brought in at 20 times the density of the natural native bee density. Honeybees clearly supplement the pollination due to native bees. This supplementation is an additive one

and we found no evidence that honeybees cause a decline in native bees. We also found that honeybee density in blooming fields is linearly and directly related to hive stocking density in those fields. Fruit-set only explains about 25% of the variation in yield and the best predictors of yield are year and production system. The strong year effect demonstrates the importance of weather and the geographic location of the field. The production system effect on yield is strong evidence that management after bloom is key to enhancing yield.

Study 2. Alternative bee forage in wild blueberry fields

Wild blueberries are reliant on insects for pollination services to produce fruit. Many smaller farms rely exclusively on native pollinators, though farms that rent honeybees also benefit from additional pollination from native bees. Although wild blueberries provide a large food source for bees, the bloom period only lasts 3-4 weeks and bees need resources both before and after that period. Native pollinators rely on alternative floral hosts when blueberry flowers are not in bloom. These alternative sources of food are important before and especially after blueberry bloom. We recorded the types and amount of wild floral resources bordering blueberry fields in two of Maine's major wild blueberry growing regions.

METHODS: We surveyed flowering plant abundance and number of species within and along the edges of 16 blueberry fields in 2015. Seven fields were located in Downeast Maine and nine fields were located in Maine's mid-coast blueberry growing region. Six of the seven fields Downeast were conventionally managed and located on the blueberry barrens; whereas, the remaining Downeast site was a small organic farm. The farms in mid-coast Maine were a mix of organic and conventional fields. Each field was sampled three times during the summer: during early bloom, during early fruit development, and during fruit ripening (May, June, and July; respectively). The perimeter of each field was inspected and all flowering plants were recorded. We also quantified the percentage of landscape the flowing plants made up and were classified as zero, less than 1%, 1%, 2% 3-5%, 10%, or greater than 10%.
We also compared bee abundance between fields of varying bloom percentages. The bee abundances were collected in a different study and were measured by the use of bee bowls or quadrate sampling. We measured the association between bee abundance and floral bloom percentages by a general linear model in JMP12®. For this analysis, we calculated the average field perimeter and field interior that was in bloom and represented it as a percentage of the field edge and interior in bloom. This was performed over all sampling dates for each field. We used data from both 2014 and 2015 studies to determine if the response of bees to flowering plants in wild blueberry fields was consistent.

RESULTS: Downeast surveys had five flowering plants species in the early survey, 10 in the second sample, and 18 in the final sample. The mid-coast survey had six flowering plants species in the early survey, 25 in the second sample, and 33 in the final sample (Tables 1 & 2). The total number of flowering species in and around fields increased over time and was higher in the mid-coast fields compared to the Downeast fields (Fig. 1). The average percent blooming landcover rating was less than 1% for both sets of fields along and within the field. Landscape bloom percentages ranged from zero to five percent for our southern sites and zero to ten percent for our Downeast sites; although, the vast majority of sites Downeast were less than 1%. Eight

percent of mid-coast fields had no bloom in field edges; whereas, 30% of Downeast field edges had no bloom during the course of this survey (Figs. 2 & 3).
We compared the number of bees in bowl traps and quadrat samples among fields with different field bloom percentages over BOTH 2014 and 2015. In our bee bowl analysis, year-floral assessment interactions ($F_{(1,24)} = 6.49$, $P = 0.002$) was significant indicating that the response of bee abundance to floral assessments was different between years. Because of this interaction with year, we analyzed the number of bees per bowl by year. There was no relationship between floral resources and number of bees per bowl in 2014 ($F_{(1,10)} = 0.98$, $P = 0.34$). In 2015 there was a significant positive relationship between floral resources and bee abundance in bowl traps ($F_{(1,14)} = 4.74$, $P = 0.048$) (Fig. 4).

Fig. 1. Floral diversity increased over time and was higher in mid-coast wild blueberry fields compared to fields Downeast.

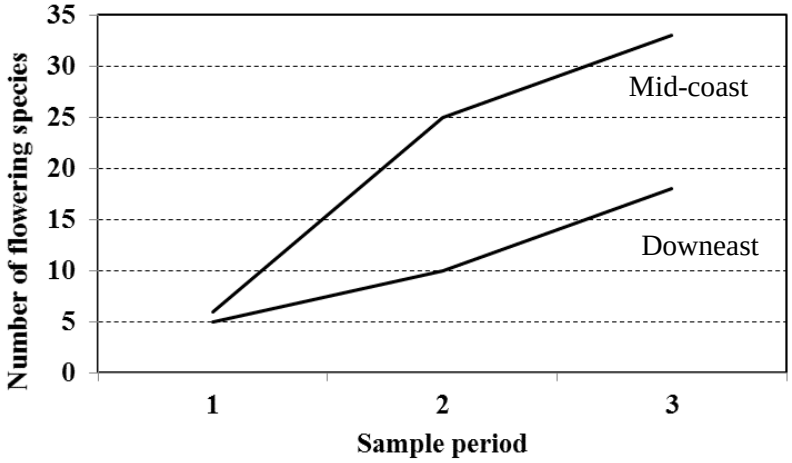


Table 1. Downeast flowering plants.

Location	Flowering Plants Sample 1	Flowering Plants Sample 2	Flowering Plants Sample 3
Flowering Plants	Bluets, bunchberry, cherry, honeysuckle, violets	Black chokeberry, blackberry, bunchberry, cinquefoil, dandelion, clover, false solomon's seal, hawkweed, sheep laurel, sorrel	Blackberry, blue-eyed grass, bristly sarsaparilla, bunchberry, cinquefoil, clover, cow wheat, dandelion, dewberry, dogbane, goldenrod, hawkweed, meadowsweet, rose, sheep laurel, st john's wort, winterberry holly, yellow loosestrife
Average % of Edge Landscape in Bloom	Less than 1% in bloom	Less than 1% in bloom	Less than 1% in bloom
Average % of Field Interior Landscape in Bloom	Less than 1% in bloom	Less than 1% in bloom	Less than 1% in bloom

Table 2. Mid-coast flowering plants.

	Flowering Plants Sample 1	Flowering Plants Sample 2	Flowering Plants Sample 3
Flowering Plants	Bluets, cherry, dandelion, strawberry, violets, willow	Autumn olive, bedstraw, black chokeberry, blackberry, blue-eyed grass, bluets, bunchberry, buttercup, Canada toadflax, chickweed, chives, cinquefoil, clover, dandelion, hawkweed, honeysuckle, iris, lilac, pea, pin cherry, raspberry, sorrel, strawberry, trillium, yarrow	Aster, bedstraw, blackberry, black-eyed susan, blue-eyed grass, bluets, bristly sarsaparilla, bunchberry, campion bladderwort, chickweed, cinquefoil, clover, daisy, dewberry, dogbane, goldenrod, goldmoss stonecrop, gray dogwood, hawkweed, meadowsweet, milkweed, northern bush honeysuckle, queen anne's lace, rabbit foot clover, rose, smartweed, st. john's wort, thistle, tigerlily, vetch, whorled loosestrife, wild rose, yarrow
Average % of Edge Landscape in Bloom	Less than 1% in bloom	Less than 1% in bloom	Less than 1% in bloom
Average % of Field Interior Landscape in Bloom	Less than 1% in bloom	Less than 1% in bloom	Less than 1% in bloom

Table 3. Most common flowering plant species near Mid-coast wild blueberry fields by month.

Flowering plant	Total number of fields present	Number of fields in May	Number of fields in June	Number of fields in July
St. john's wort	9	0	0	9
Vetch	9	0	0	9
Clover	8	0	2	8
Dandelion	8	7	3	0
Milkweed	8	0	6	1
Black-eyed susan	8	0	0	8

Table 4. Most common flowering plant species near Downeast wild blueberry fields by month.

Flowering plant	Total number of fields present	Number of fields in May	Number of fields in June	Number of fields in July
Blue-eyed grass	6	0	0	6
Cinquefoil	6	0	5	2
Sorrel	6	0	6	
Dogbane	5	0	0	5
Sheep laurel	5	0	3	4

Fig. 2. The distribution of field edge bloom percentages in coastal blueberry fields during all three sampling periods.

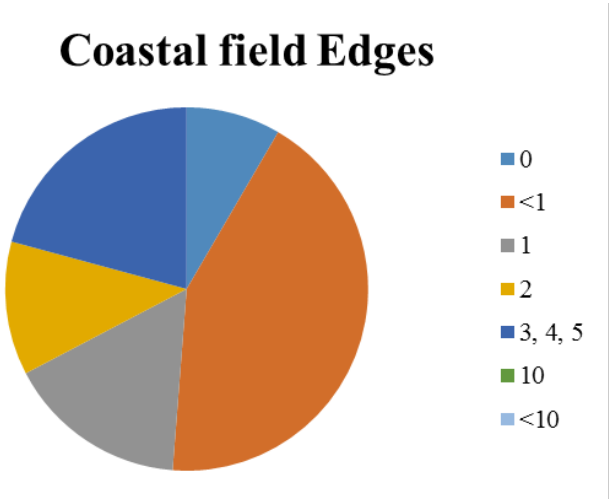


Fig. 3. The distribution of field edge bloom percentages in Downeast blueberry fields during all three sampling periods.

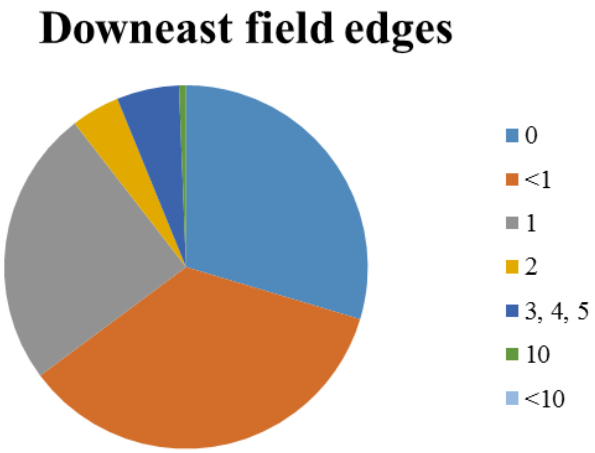
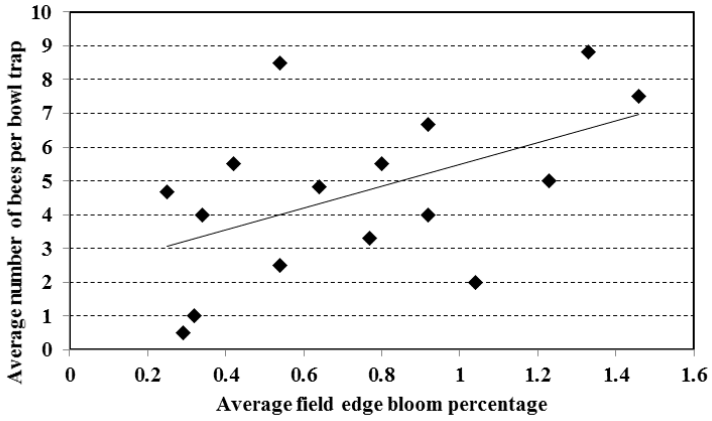


Fig. 4. In 2015 the average number of bees caught in bowl traps was higher in fields with larger bloom percentages.



CONCLUSIONS: Similar to our survey in 2014, we found that plant diversity increased as the season progressed with more species of flowering plants later in the season. Although we recorded a large number of flowering plants across all fields, most fields only had a small percentage of land area with these plants. This trend was especially prevalent in the fields Downeast that typically only had a few species of flowering plants, and many field edges had no flowering plants at all. The most common plants in the mid-coast blueberry fields were different from the most common plants found in the Downeast fields. There are no blueberry flowers during these later sampling dates and so it is an important time of year for native pollinators to locate alternative forage. In 2015, bee abundance in bowl traps was higher in fields with larger bloom percentages. This indicates that the presence of these floral resources before and after blueberry bloom can help sustain a large and healthy native bee population. This trend was not seen in 2014. This highlights the complex changing nature of bee populations from year to year and site to site, as the 2014 fields were different from the 2015 fields. Additional data is needed to get a clearer sense of how floral resources impact native pollinators. A previous three-year study from 2010-2012 representing 40 wild blueberry fields did not find an effect of within field and edge flowering plant abundance on the number of wild bees in a field during pollination. In addition, another study conducted by a recent graduate student, Ms. Shannon Goff, suggests that the landscape surrounding blueberry fields may be the most significant factor explaining the abundance of the native bee community in a wild blueberry field during pollination. However, based upon these studies and our 2014 and 2015 study, it may be that the relationship between within-field flowering plant abundance and bee abundance in highly context dependent and that if the surrounding landscape is factored into the "equation" that a stronger relationship between flowers in a field and the bee community will be seen.

Study 3. Bee preferences for alternative forage resource, update on the bee module experiment
Report from Dr. Alison C. Dibble, Dr. Lois Berg Stack, and Dr. Frank Drummond

Bees require flowers for their food; they use nectar and pollen to provision their brood. To improve pollinator security, growers can either plant for pollinators, or manage vegetation near blueberry fields to encourage plants that bees visit. The effectiveness of pollinator plantings and habitat improvements depends on the plants selected for inclusion. To support native wild and managed bees associated with wild blueberry agriculture, we observed bee visitation rate upon flowers of native and introduced plants in a replicated experiment.

METHODS: In a 5-year study funded by the USDA and University of Maine, from 2012-2014 at four sites (Blueberry Hill Farm in Jonesboro, Rogers Farm in Old Town, and two wild blueberry farms in Blue Hill), we quantified bee visitation rate on 70 plant species or cultivars. Data consist of one-minute observations on flowers and nest habitats.

RESULTS: Data for 2012-2014 include 11,448 one-minute observations, with 3293 honey bee sightings, 1396 orange-banded bumble bees, 4166 other bumble bees, 1541 sweat bees, and 1157 other bees (Table 1).
For bees observed on a given plant, we adjusted data as a percent of the total bees observed on all plants in flower at that site and day (average 54.8, min. 3, max. 140). Bee groups differed in

their visitation rates. A few plants attracted bees in all groups. In general, bees tended to visit fancy cultivars such as double-flowered marigolds less often than related wildtypes.

Table 1. Visitation rate (as percent of bees seen on a given day/site) for the most-visited plants tested in the Bee Module experiment 2012-2014.

Bee group	**Plants, percent of bees at date/site**	**Bee group**	**Plants, percent of bees at date/site**
Honey bees	Summersweet 10% Borage, especially white flowers 6% Butterfly milkweed 4.9% Yellow sweetclover 4.2% California poppy 4.2% Anise hyssop 4% Sunflower, wild 3.9% Blanketflower, wild 3.6%	Bumble bees other than orange-banded	Foxglove beardtongue 'Mystica' 33.2% Bee's friend 14.5% Butterfly milkweed 12.9% Blue cowslip 9.6% Anise hyssop 6.6% Yellow sweetclover 6.2% Borage, especially white flowers 5%
Orange-banded bumble bees	Butterfly milkweed 6.4% Bee's friend 4.3% Greek oregano 3% Purple coneflower 2.4% Yellow sweetclover 2.3% Anise hyssop 2.2%	Sweat bees	Catmint 8.8% Foxglove beardtongue 'Mystica' 4.8% Bebb's willow 4.4% Greek oregano 2.9% Black chokeberry 2.5% Buckwheat 2.3%
Other bees (includes miner bees, leaf-cutter bees, plasterer bees, etc.)	Bebb's willow 37% Foxglove beardtongue 'Mystica' 4.8% Shadbush 'Regent' 4% Black chokeberry 3.5% Mustard Pacific Gold 2.8%	All/Any	Anise hyssop Bee's friend Blanket flower Borage Buckwheat Butterfly milkweed Meadowsweet Oregano, Greek and red Pasture rose Purple coneflower Yellow sweetclover Willow (percent varies with each bee group)

CONCLUSIONS: Our observations indicate that valuable bee forage habitat is associated with minimally managed areas around the farm, such as field edges, wet spots, roadsides or rock piles that contain such plants as willow, shadbush, black chokeberry, meadowsweet, roses, goldenrods, milkweed, and asters. Strips planted in clovers, buckwheat, and bee's friend are likely to benefit bees on the farm. Data from 2015 will be reported in 2016, and will include two native goldenrods, a hawkweed and additional asters.

ADDITIONAL RESOURCE: See "A Pocket Guide to IPM Scouting in Wild Blueberries, 2nd Edition", compiled and edited by D. Yarborough, F. Drummond, S. Annis, J. Cote, and A. Dibble (2014), UMaine Cooperative Extension, for more about bees and plants related to lowbush blueberry.

Study 4. <u>Determination of floral characteristics that attract bees to flowers</u>
<u>Report from Megan Leach (Master's student), and Dr. Frank Drummond</u>

Pollinator plantings are suggested to wild blueberry growers and home gardeners as way to increase the native bee community. These plantings provide bees with more forage outside of blueberry flowering, when there is little else in the landscape. Some flower species have been shown to have higher visitation by bees than others. It has also been shown that flower preference can be different among bee species. The reason behind different visitation for flower species is not currently known. It has been suggested that higher nutritional quality of nectar and pollen is a reason for the differences in visitation and that higher nutrition flowers have higher visitation by bees. It has also been shown that floral traits can drive higher visitation. The first objective this summer was to measure floral characteristics as well as pollen and nectar nutrition to determine which are attracting more bees to flowers. The second objective was to conduct a controlled experiment to determine if visitation to flowers can be altered through soil fertility.

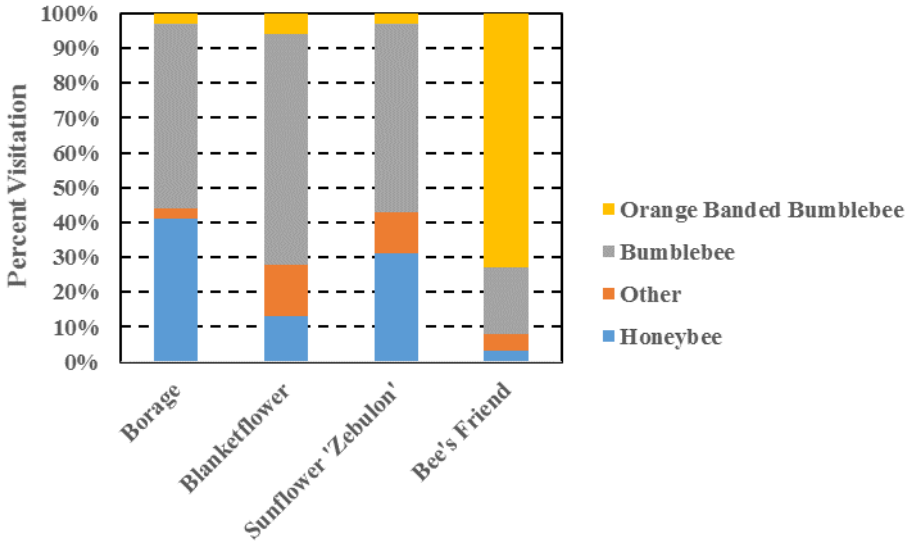
METHODS: This past summer we planted four different flower species at four different sites that were established in 2012, called the "Bee Module" (Study 3 of this report). One site was located in Stillwater, two were located in Blue Hill, and one was located in Jonesboro. These sites have 36 different plant species and nest habitats planted in m^2 plots. The plots are observed for visitation to determine which plant species and nest habitats have higher visitation by bees. Plants were placed in four plots at each site. The plant species chosen were from two families, Asteraceae and Boraginaceae. In the family Asteraceae we chose the species *Gaillardia aristata* (blanketflower) and *Helianthus annuus* 'Zebulon'. In the family Boraginaceae we chose the plant species *Borago officianalis* (Borage), and *Phacelia tanacetifolia* (Bee's friend). All four plant species had been in the bee module in previous years, so visitation to these flowers was already known. Flowering times were also very similar for all four plant species. For each sample there were three, one-minute observation periods and any bees visiting the flowers were then recorded. We also recorded floral traits by measuring diameter, density, corolla depth, and foraging area for each species.

To collect pollen and nectar without disturbing the visitation plots, a flower strip of each species was planted at the Stillwater field site. In these strips, we collected pollen and nectar, pollen grains per flower, and volume of nectar per flower. We will be analyzing pollen for amino acid content using High Pressure Liquid Chromatography (HPLC). To analyze nectar we will be using a handheld refractometer to get sugar concentration and Ultra Performance Liquid Chromatography (UPLC) to determine what sugars are present in the nectar.

For the second objective, we wanted to determine if visitation could be manipulated for one flowers species. We planted three treatments of *Impatiens capensis* (Jewelweed); one treatment with high fertilizer (27g Osmocote), one with low fertilizer (7g Osmocote) and one with no fertilizer. We then recorded visitation to the flowers, recorded floral characteristics, and collected pollen and nectar for chemical analysis. The pollen and nectar will be analyzed using the same methods that were used for pollen and nectar analysis described for the first objective.

RESULTS: The preliminary results for objective one show that each flower species has different visitation profiles (Fig. 1). Bee's Friend in particular has much higher percent visitation for the orange-banded bumble bee than the rest of the flowers species. We are currently analyzing the results to determine if the differences are statistically significant. We are also creating an analysis method to determine the amino acid profiles for the pollen of each species as well as the sugar profiles for the nectar of each plant species. Once all of the data is collected, we can run further analysis to determine which floral characteristic or characteristics explain the visitation data.

Fig. 1. Percent visitation by each bee group. Preliminary results for each bee group observed on the flowers for the 2015 field season.



Preliminary results for objective two show that visitation was much higher in the high fertilizer treatments (Fig. 2). The zero fertilizer treatment did not produce any flowers, so no observations or measurements were taken on those plants. The high fertilizer treatments produced more flowers than the low fertilizer treatment (Fig. 3). We will be analyzing the pollen and nectar this spring to determine if the nitrogen availability in the soil affects the pollen and nectar nutrition in the plant. Once analysis of pollen and nectar is complete, we will statistically analyze the data to determine which factors contribute to higher or lower visitation.

Fig. 2. Percent visitation for high and low treatments at each observation date for objective two.

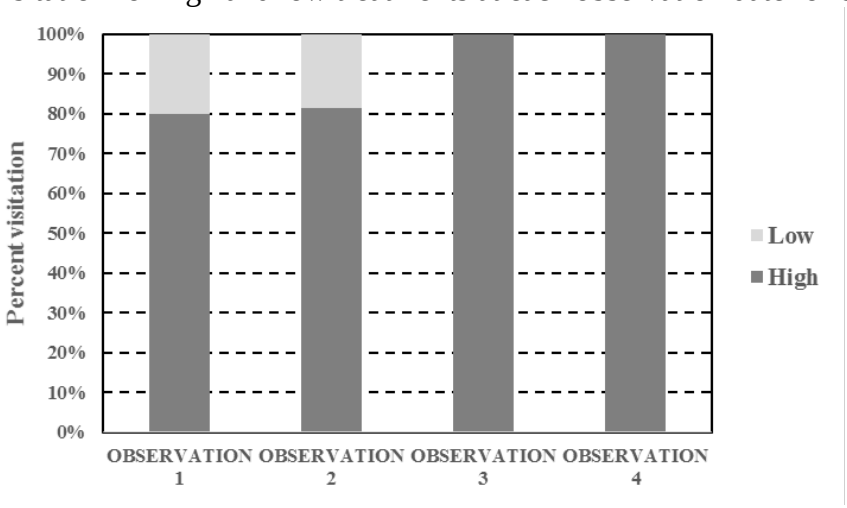
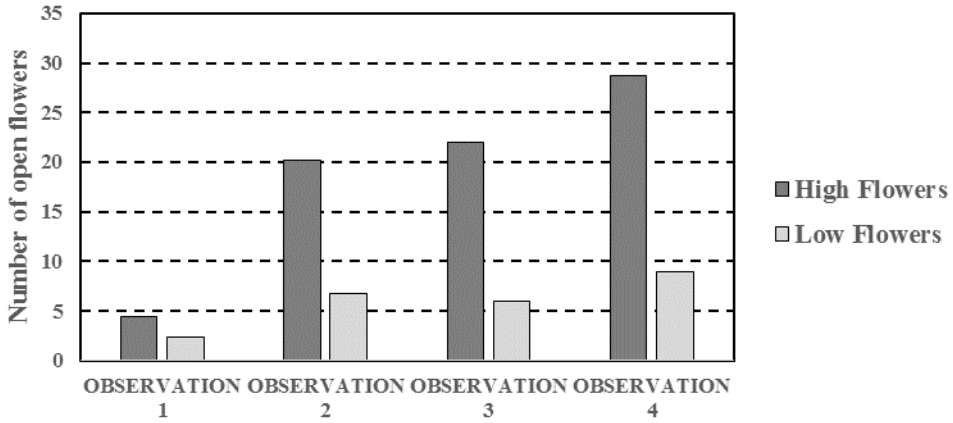


Fig. 3. Number of open flowers at each observation date for objective two.



CONCLUSIONS: This study will determine which characteristics draw bees to flowers for collection of nectar and pollen. Pollen and nectar nutrition is important to bee growth and development and by selecting to visit higher nutrition flowers brood would benefit as well as population growth. This study will contribute which factors are influencing visitation and if there are any characteristics that we can select for pollinator plantings that would contribute to pollinator conservation. We will also be able to make suggestions for pollinator plantings to increase attraction to pollinators.

Study 5. <u>Establishment of floral plantings in wild blueberry fields</u>
<u>Report from Eric Venturini, Dr. Lois Stack, Dr. Alison Dibble, and Dr. Frank Drummond</u>

In June 2012 an experiment was conducted to assess several techniques for establishing a pollinator planting in wild blueberry. In 2013, sampling of the treatment plots was conducted to determine the initial effect of these techniques on the establishment of the planting mix (Maine Wild Blueberry Mix, Applewood Seed Company). Then in 2015, the plots were revisited to assess the outcomes of the planting techniques in the fourth year of the planting.

METHODS: The wildflower mix establishment site was located at the University of Maine's wild blueberry research facility, Blueberry Hill Farm in Jonesboro, ME. To achieve a soil pH of 6.0 from a starting pH of 4.7 at the establishment site, on 18 May 2012, we applied 7846 kg/ha of lime using a Gandy® T36 drop spreader and incorporated with tillage to a depth of 7.5 cm. Shallow tillage was repeated on 31 May. On 8 July we raked the seedbed and broadcasted seed using an Earthway® hand crank seeder. Seeds were bulked with vermiculite to ensure an even distribution of seeds. Immediately after broadcasting, all plots were rolled with a 1.5 m wide 1,180 kg compacter. Plots were irrigated to receive 2.5 cm of rain per week. If rain fell equal to or in excess of 2.5 cm in a given week, no additional irrigation was applied.

Each of four replicates (statistical blocks) contained five treatment plots (4x8 m). Treatment 1 was an unsown rototilled control, all other treatments were seeded to the same mix of wildflowers. In addition to wildflower seed, treatment 2 was mowed once annually in the fall, treatment 3 was sown with a low density of sheep fescue (*Festuca ovina*), treatment 4 included both mowing and sheep fescue, and treatment 5 was seeded to the wildflower mix alone. Each treatment plot was split into two sub-plots. One sub-plot in each treatment plot was also sown with oats (*Avena sativa*). See Table 1 for a list of seed and seeding rates.

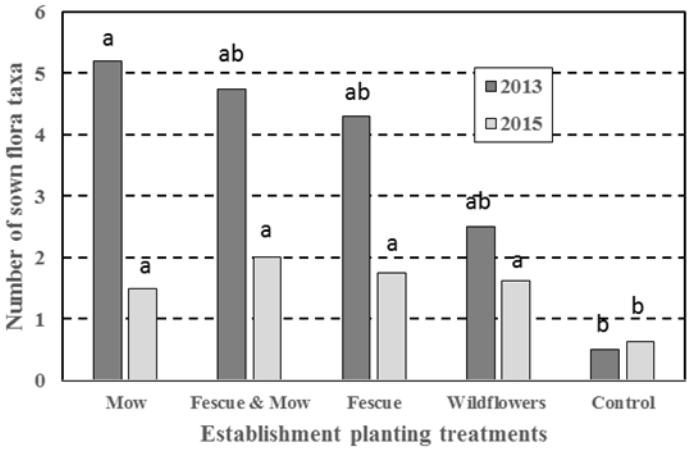
In September 2013, we randomly selected 0.5 m^2 from each sub-plot for cutting to assess plant biomass. All above-ground plant material in each sub-plot was cut, bagged by species, and dried for two weeks in a drying room (40-45 ºC) at the University of Maine Analytical Laboratory and Maine Soil Testing Service in Orono, ME. All plant material was identified, labeled as sown or unsown, and weighed. Opened, unopened, and past inflorescences were identified to species and recorded. In September 2015, we randomly selected 0.5 m^2 from each subplot and estimated floral species richness, flower density and biomass of flowers. Split-plot analysis of variance was used to test the establishment techniques with respect to sown plant species richness, inflorescence density and biomass. In 2015, Split-plot ANOVA was used to assess establishment techniques on total sown plant inflorescence density and dry weight and non-sown (weed) plant inflorescence density and dry weight. Poisson regression was used for the analysis of floral richness and individual inflorescence densities and dry weight per m^2 due to the prevalence of plots with no flowering plants in the mix (0 counts in plots).

Table 1. Seeding rates for wildflower. No. live seeds/ft^2 unavailable for clovers. Not shown, nurse crop of *Avena sativa*. Wildflower statistics shown are supplied by Diane Wilson at Applewood Seed Company.

Common Name	Species	Habit	No. live seeds/sq foot	No. live seeds per acre
Wildflower Treatment				
Plains Coreopsis	*Coreopsis tinctoria*	*Annual*	9.55	416,000
Indian Blanket	*Gaillardia pulchella*	*Annual*	7.66	333,600
Sunflower	*Helianthus annuus*	*Annual*	2.75	120,000
Lavender Hyssop	*Agastache foeniculum*	*Perennial*	5.29	230,400
Lance-Lvd. Coreoopsis	*Coreopsis lanceolata*	*Perennial*	5.79	252,000
Canada Tick Trefoil	*Desmodium canadense*	*Perennial*	1.82	79,200
Purple Coneflower	*Echinacea purpurea*	*Perennial*	4.69	204,400
Common Boneset	*Eupatorium perfoliatum*	*Perennial*	3.53	153,600
Bergamot	*Monarda fistulosa*	*Perennial*	4.01	174,720
New-England Aster	*Symphyotrichum novae-angliae*	*Perennial*	4.52	196,800
		Total Wildflower -->	**49.60**	**2,160,720**

RESULTS: In September of 2013, establishment technique significantly influenced the species richness of sown species ($F_{(4,12)} = 3.689$, $P = 0.035$), with mowing having the greatest effect on sown species richness (Fig. 1). However, mowing was only significantly different from control plots. All treatments except wildflower seed alone had a significantly greater richness of sown species than the control. In 2015, only the non-planted control was different in the sown wildflowers species richness from all of the planted plots (Fig. 1, $\chi_{(4)}^2 = 14.890$, $P = 0.005$). There was no effect on plant species richness from the seed mix of sowing oats in plots ($\chi_{(4)}^2 = 0.612$, $P = 0.962$).

Fig. 1. The richness (# species) of sown species associated with the techniques for establishment, 2013 & 2015.

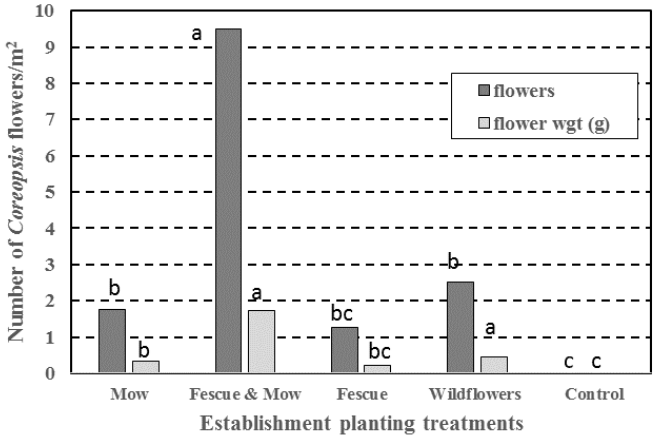


Coreopsis lanceolata, *C. tinctoria*, *M. fistulosa*, and *S. novae-angliae* were the four most common plants in samples in 2013. There were no significant treatment effects on *C. lanceolata* dry matter weight, open flower or total plant inflorescence density, and dry weight number. *Coreopsis tinctoria* finished flowering by the time of our sample, explaining why there was no treatment or split-plot effects on the number of open flowers. However, both *C. tinctoria* dry matter weight ($F_{(4,12)} = 4.762$, $P = 0.016$) and total flower heads ($F_{(4,12)} = 4.426$, $P = 0.020$) were affected by treatments. Means comparison (Tukey HSD) revealed that when wildflower was seeded alone, *C. tinctoria* produced significantly more flowers (443.50 flowers/m^2) and plant material (31.52 g/m^2) than any other treatments except when seeded with fescue (94.75 flowers/m^2 and 5.91 g/m^2, respectively). Inflorescence density and dry weights of *Monarda fistulosa* were also not affected by establishment technique treatment or oat seeding. However, when wildflowers were sown alone and not mowed, *M. fistulosa* produced more dry matter (LS mean = 70.91 g/m^2, $F_{(4,12)} = 2.443$, $P = 0.104$). *Symphiotrichum novae-angliae* was in bloom during our sampling. Although neither treatments nor split-plot (oat seeding) effects influenced *S. novae-angliae* at the α=0.05 level, the interaction term treatment x split plot significantly influenced the number of open flowers produced at the $\alpha = 0.10$ level ($F_{(4,12)} = 2.452$, $P = 0.091$). Although not highly significant, treatments involving fescue and mowing consistently resulted in greater numbers of open *S. novae-angliae* flowers than wildflower seed alone.

In 2015, in rank order, the most common plants were *Desmodium canadense*. *Monarda fistulosa, and Coreopsis lanceolata*. Although by 2015, 26% of the total floral resource (#Inflorescences/m^2) in all plots was weedy non-planted species, the majority of which were goldenrod species. In addition, sown floral plant richness had decreased almost 70% by 2015. The total sown inflorescence density no longer appeared to be influenced by establishment techniques when sampled in 2015 ($F_{(4,12)} = 2.081$, $P = 0.141$) nor by the sowing of oats ($P = 0.567$). This was also the case when inflorescence dry wgt/m^2 was analyzed ($P > 0.05$). However, when investigating the non-sown plant response (mostly goldenrod species) we found that the sowing of oats significantly reduced goldenrod inflorescence density ($F_{(1,12)} = 8.036$, $P = 0.012$) and inflorescence dry weight ($F_{(1,12)} = 7.798$, $P = 0.011$) by 2015. There was also a non-significant trend toward higher goldenrod inflorescence densities and inflorescence dry weights in the non-sown treatment plots ($P > 0.05$).

When investigating individual sown plant species responses to the establishment techniques we found no significant treatment or oat seeding effects on *Desmodium* inflorescence density or dry weight ($P > 0.05$) and no significant treatment or oat seeding effects on *Monarda* flower density or dry weight ($P > 0.05$). Although the effect of the split-plot treatment on *Monarda*, the seeding of oats, was marginally significant for flower density ($\chi^2_{(1)} = 3.304$, $P = 0.069$) and for flower wgt/m^2 ($\chi^2_{(1)} = 3.342$, $P = 0.065$). The mean flower number/m^2 was 2.19 for plots not sowed with oats compared to 5.92 for plots sowed to oats. The mean number of dry flower wgt/m^2 was 0.89 for plots not sowed with oats compared to 2.39 for plots sowed to oats. There were treatment effects on both *Coreopsis* flower density ($\chi^2_{(4)} = 23.867$, $P < 0.0001$) and flower dry weight ($\chi^2_{(4)} = 25.899$, $P < 0.0001$). Figure 2 shows that the control treatment had the lowest *Coreopsis* flower number and dry wgt/m^2; whereas, the mowing in combination with the *Fescue* treatment had the highest *Coreopsis* flower density and dry wgt/m^2.

Fig. 2. *Coreopsis lanceolata* flower density and dry weight per m^2, 2015.



CONCLUSIONS: General conclusions from this study suggest a practical longevity of a pollinator flower planting in wild blueberry to be four years under the low intensity management that we implemented. The initial establishment of the planting appeared to be favored by mowing; although, this was strictly only significantly better than the non-sown control plots. Four years after the planting was sown, the establishment techniques were not strong determinants of sown floral density or biomass, except for *Coreopsis lanceolata* that appeared to be positively influenced by an establishment regime of both mowing and fescue cropping. *Monarda fistulosa* was positively influenced by over sowing oats on the wildflower mix. Goldenrod was observed to be favored in plots that were not over sown with oats. This study is finished, but a demonstration pollinator planting will be replanted in 2016 and maintained for growers to visit at the University of Maine, Blueberry Hill Research Farm.

Study 6. The effect of pollinator plantings in enhancing native bee populations and fruit-set in wild blueberry

Report from Eric Venturini and Dr. Frank Drummond

Pollinator plantings have been pursued around the world as a means of increasing native pollinator community abundance in many bee pollinated high value crops. In mid-May and early June 2012, Mr. Eric Venturini planted four pollinator plantings (4,950 ft^2) designed for wild blueberry. Three of these plantings were adjacent to wild blueberry fields. Each planting consisted of a strip of wild flower mix (10 wild flower species), a clover strip (three species of clover), and a non-planted seed bank regeneration strip. An additional plot adjacent to the pollinator strips was planted to 36 different bee plant species (400 ft^2). The effect of these plantings (5,350 ft^2) on native bee abundance and fruit-set and yield were assessed in 2013 during the second year of the planting. A second assessment was completed in 2015.

METHODS: In 2013, three blueberry fields (Jonesboro and Blue Hill) with adjacent pollinator plantings were sampled for native bee and honeybee abundance (bees/ m^2/min), fruit-set, and yield. Neighboring fields (controls, at least 5 miles away) were also sampled for native and honeybees and fruit-set and yield. In 2015, the same fields were sampled for native and honeybees (bees/m^2/min) and fruit-set, but not yield. ANOVA (RCB) was used to determine if the presence of adjacent pollinator plantings had any effect on native bee density and fruit-set.

RESULTS: In 2012 and 2013 there was no significant difference ($F_{(1, 1)} = 0.028$, $P = 0.868$) between native bees in wild blueberry fields with adjacent pollinator plantings and those without pollinator plantings (controls). In 2015, there were significantly higher densities ($F_{(1,2)} = 48.396$, $P = 0.020$) of native bees in the fields associated with pollinator plantings than in the control fields (Fig. 1). Fruit-set was also not significantly different in 2013 between fields with pollinator strips and fields without strips (Fig. 2). However, in 2015 we observed a significant difference at the $\alpha = 010$ level. This suggests a significant affect, but also a difference that might be explained by chance with a 10% probability. Therefore, this is not a strong effect, but this is not unexpected considering the size of the pollinator planting which was just a bit larger than 0.25 acres in fields that were in the 5-10 acre size.

Fig. 1. Abundance of native bees in fields with pollinator strips compared to bee abundance in fields without pollinator strips

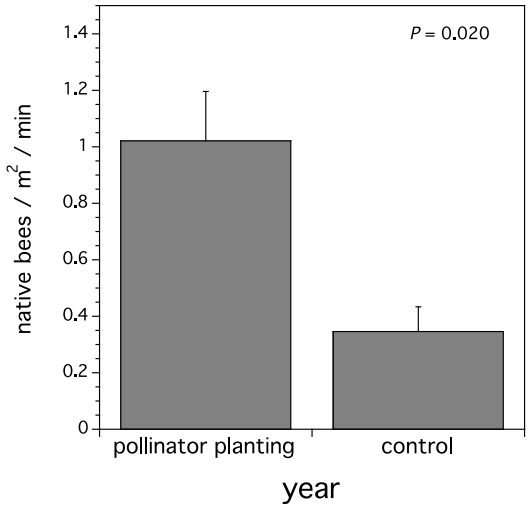
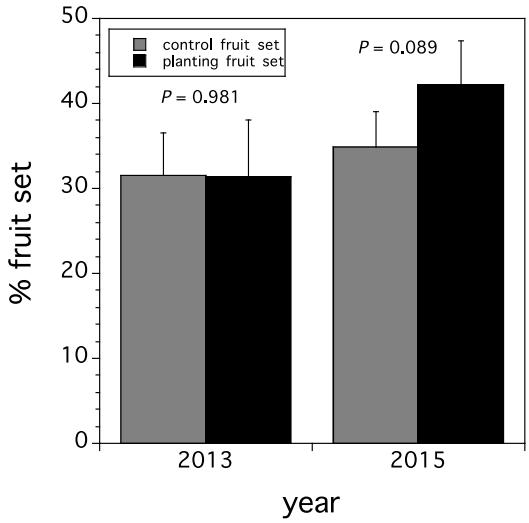


Fig. 2. Fruit-set among fields associated with pollinator plantings compared to fields without plantings in 2013 and 2015.



CONCLUSIONS: We conclude that pollinator plantings can result in enhanced pollination, but that this may take several years to be manifest. This is most likely due to the fact that bee populations increase at a relatively slow rate. In addition, pollinator plantings of a larger land area may have resulted in a stronger effect. Our pollinator strip to blueberry field size ratio was approximately 1-5%. Some researchers suggest a ratio of 10-20%. However, this will usually not be possible or economically feasible in most blueberry landscapes. In the future, interested growers will have to experiment with plantings in order to determine the correct ratio for their needs. We have shown that it can work, but we cannot determine the optimal size of a planting necessary for enhancing the pollinator community and pollination in commercial production.

Study 7. Influence of land cover type on wild bee abundance and diversity, 2015

Report from Brianne Du Clos (Ph.D. candidate), Dr. Cyndy Loftin, and Dr. Frank Drummond

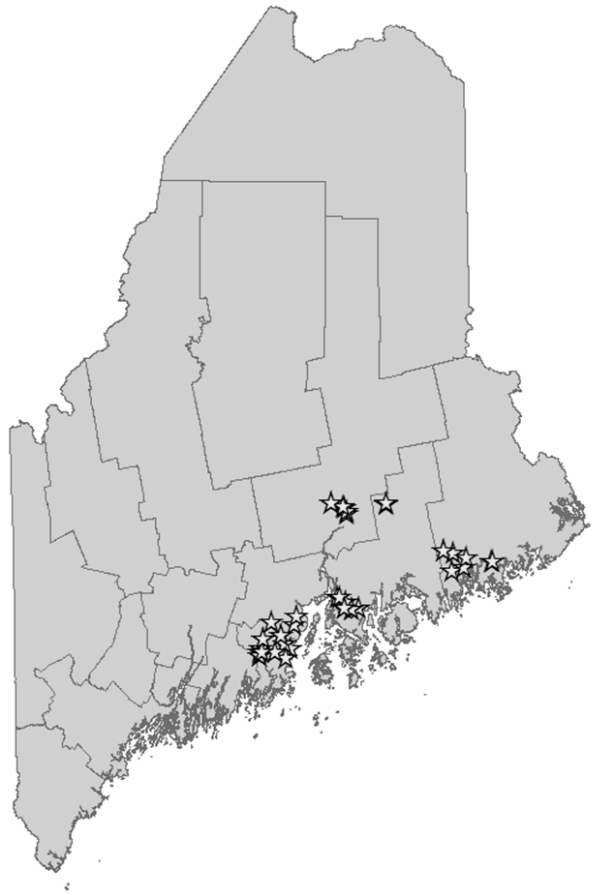
Little is known about wild bee communities outside of wild blueberry fields in Maine. We will quantify bee community richness and abundance in eight land cover types throughout Maine's wild blueberry growing region (Table 1). This study is an expansion of a similar project completed last summer. The purpose of this project is to provide more accurate parameter values for the InVEST Crop Pollination model applied in the wild blueberry crop system. Field-collected information about bee communities in non-crop land cover types is not available, yet this information will greatly improve accuracy of model predictions of wild bee abundance across the landscape. This information also will be incorporated into a computer based landscape visualization tool to show growers estimated abundance of wild bees in landscapes surrounding their crop fields based on the InVEST Crop Pollination model predictions. The value of this tool is dependent on prediction accuracy, which will be improved with bee survey data collected in non-crop land cover types.

Table 1. Land cover types surveyed for wild bees.

Agriculture/field
Developed
Emergent wetlands/scrub
Wetlands/water
Commercial blueberry field
Coniferous forest
Deciduous/mixed forest edge
Deciduous/mixed forest interior

METHODS: We established five blocks of survey sites throughout Maine's wild blueberry production landscape: two in Knox/Waldo counties, one in Penobscot County, one in Hancock County, and one in Washington County (Fig. 1). Each block consisted of eight sites, one of each land cover type, for a total of 40 survey sites (Table 1).

Fig. 1. Study sites for land cover survey.



We assessed nesting resources at each site by looking for standing dead wood, fallen dead wood, woody shrubs with hollow twigs, and open, sandy soils. We surveyed bee communities at each site in early, mid-, and late summer 2015. Each survey consisted of 30 minutes of live-netting bees and a 24-hour deployment of bowl traps. We also recorded blooming flowers and collected samples of flowers for identification. There was no live-netting of bees at sites that did not have blooming flowers. All bees were prepared for identification to genus in the lab.

RESULTS: All bees have been identified to genus and most have been verified by a bee taxonomy expert. Here we present preliminary results, as thorough statistical analysis has yet to be completed. We collected 1,723 bee specimens, many of which were found in blueberry, deciduous/mixed forest edge, and developed/other land cover types (Fig. 2). Emergent wetland and deciduous/mixed forest edge had the highest number of wild bee genera; bees in 18 genera (of 24 total genera found) were collected in these land cover types (Fig. 3).

Fig. 2. Wild bee abundance by land cover type.

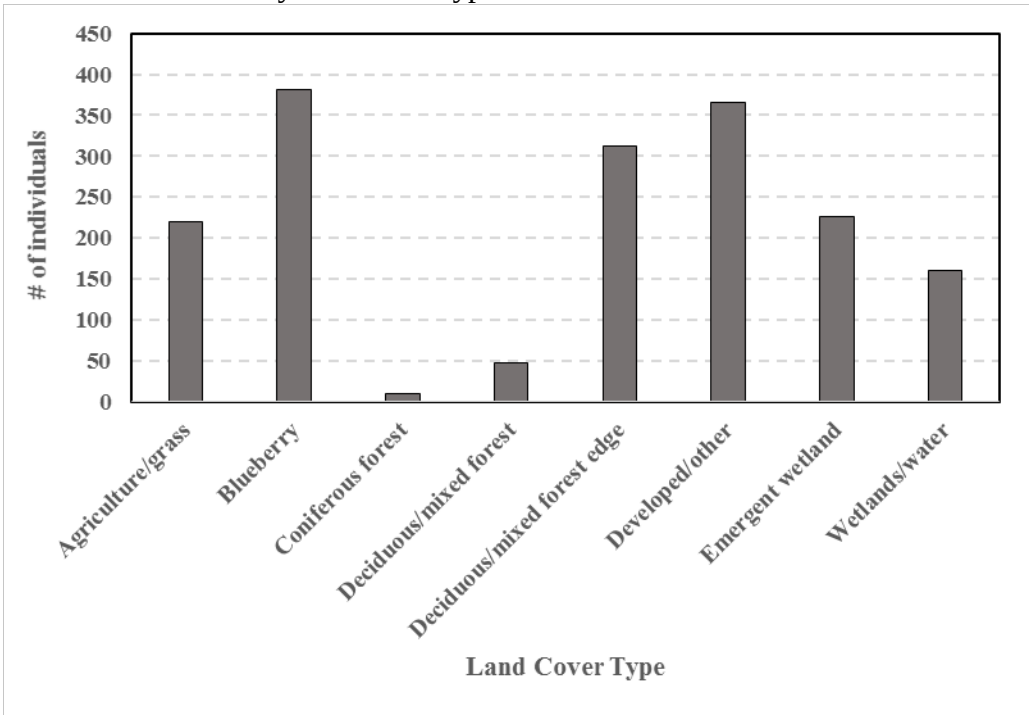
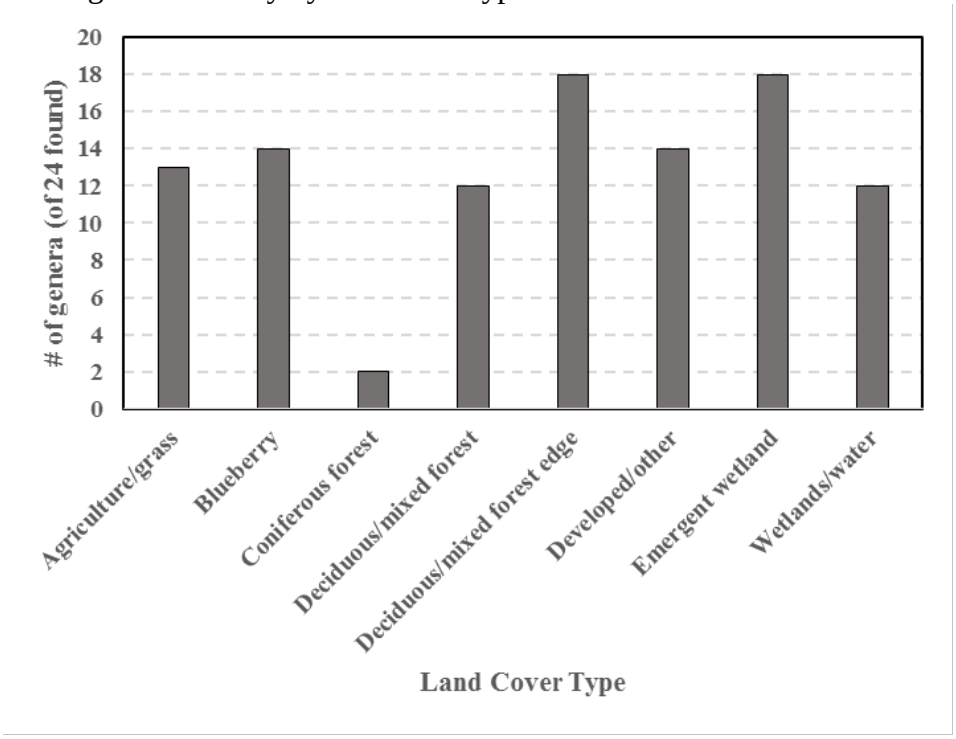


Fig. 3. Wild bee genus diversity by land cover type.



Our statistical analysis will include quantifying the bee communities in each land cover type and comparing our field collected data to current expert opinion. We will use our data to create new InVEST model parameters to improve the performance of InVEST to predict wild bee abundance in Maine's wild blueberry landscape.

CONCLUSIONS: Our wild bee surveys will contribute to a robust data set of wild bees in Maine. The surveys of non-blueberry land cover types are especially important as they address a crucial knowledge gap of wild bee natural history in Maine. This work will be the first comparison of expert opinion to bee community survey data for parameterization of the InVEST Crop Pollination model. The comparison will determine the best data for use as model parameters, resulting in robust predictions of wild bee abundance at the crop field level.

Study 8. Development of a web-based tool for grower assessment of wild bee abundance in the wild blueberry production landscape

Report from Brianne Du Clos (Ph.D. candidate), Dr. Sam Hanes, Dr. Cyndy Loftin, and Dr. Frank Drummond

We are developing a novel web-based tool for wild blueberry to visualize estimated bee abundance associated with land cover in the landscape around focal crops. The tool will show growers where their conservation efforts are best focused at the landscape scale. Wild blueberry growers can use the information presented in BeeMapper to determine placement of honeybee hives during blueberry pollination, establish a pollinator conservation plan for particular crop fields, and understand wild bee communities in different types of land

METHODS: BeeMapper is currently available for much of the Downeast wild blueberry growing region in Washington and Hancock counties. BeeMapper displays two maps over a Google Maps-style interface: one is a custom land cover type map created to determine potential wild bee habitat; the other is output of the InVEST Crop Pollination model and shows predicted wild bee abundance across the Downeast wild blueberry growing region. We conducted in-depth one-on-one sessions with six growers located in this region in March 2015. These growers encompassed a variety of crop management practices and are influential in the wild blueberry grower community. After the interviews, we updated the tool to accommodate grower requests.

RESULTS: BeeMapper is still in development and is not yet available to the public. Interviews with growers indicated the need for a simple, concise summary of data in the tool and a strong desire for a printable output. We have worked in a simple data summary into the tool (Fig. 1); work is ongoing for the printable output. We continue to develop the web site that will be associated with the tool (www.umaine.edu/beemapper; Fig. 2), and we are compiling documentation on how to use the tool. This documentation will be provided to wild blueberry growers for review in 2016.

Fig. 1. Concise summary of data displayed in BeeMapper web tool.

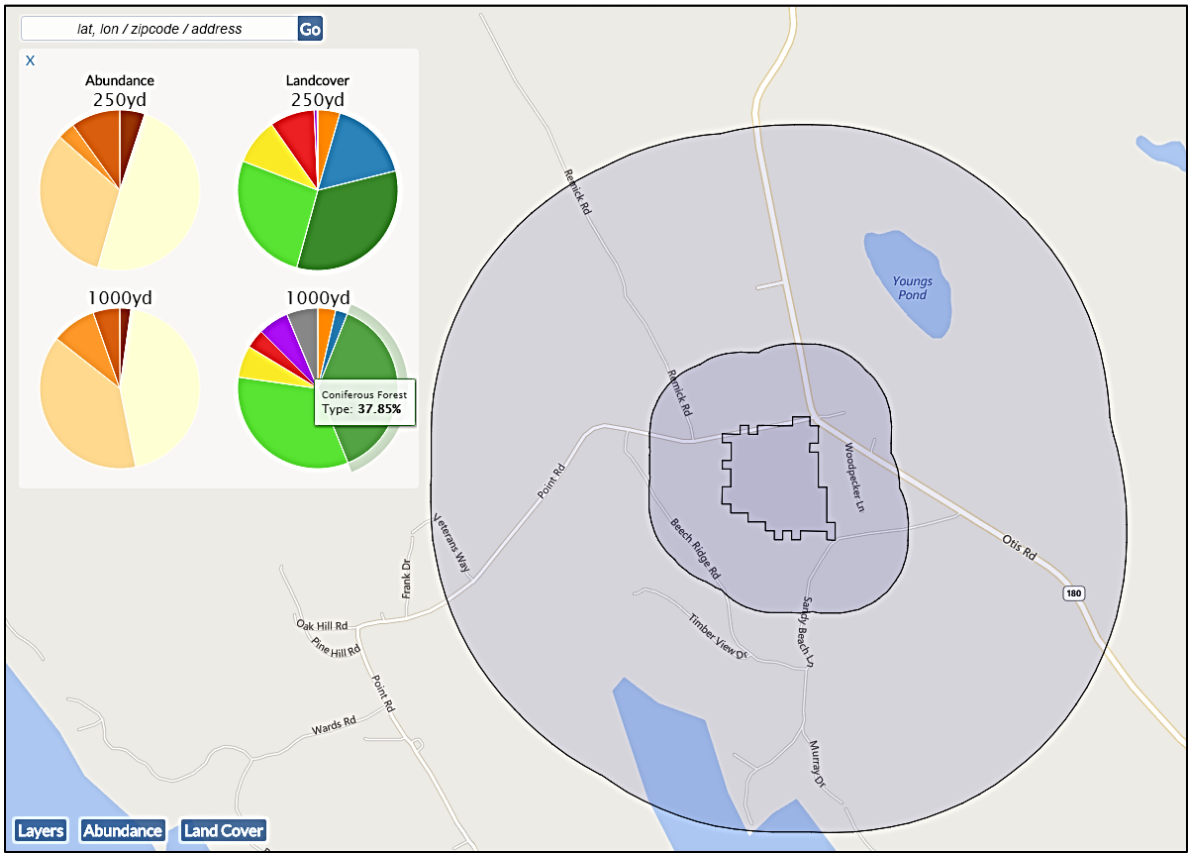
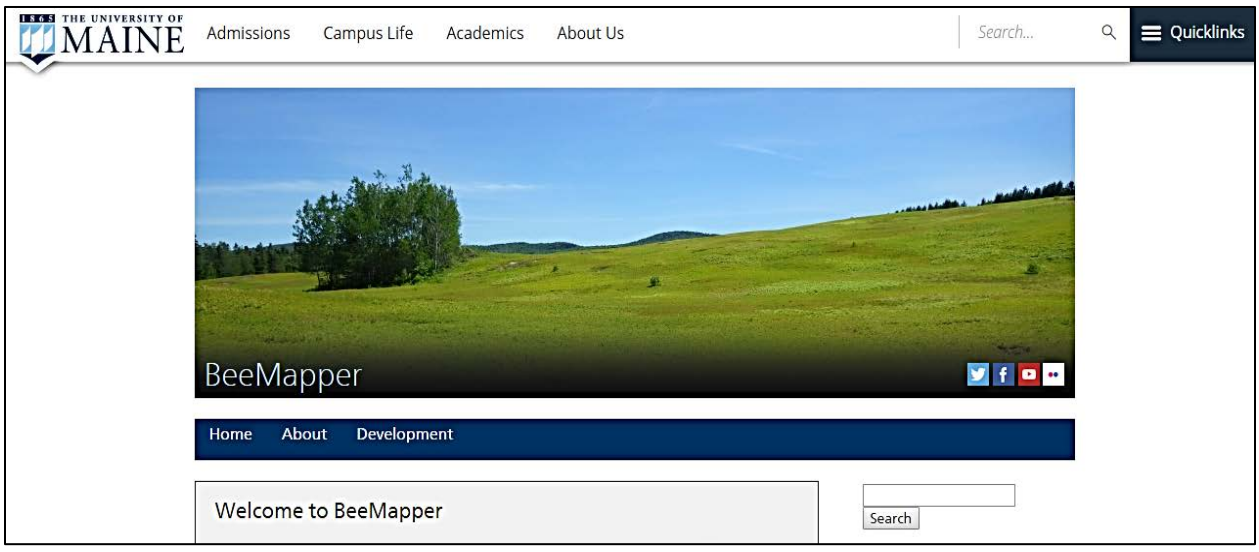


Fig. 2. BeeMapper web site.



We are working to improve the data displayed in BeeMapper on two fronts: we have conducted an extensive field survey to improve InVEST model parameters (see Study 7 of this report), and we are expanding coverage of BeeMapper to the mid-coast growing region of Knox and Waldo

counties. Completion of these projects is expected to take some time, but the delay in making the tool accessible to growers is necessary to make the tool accurate and effective.

CONCLUSIONS: We will continue to develop the web tool and seek feedback from wild blueberry growers throughout the tool development process. The final version of the tool will help growers visualize the contribution of the landscape surrounding their fields as wild bee habitat and inform their decisions about land management to enhance crop pollination as well as wild bee conservation.

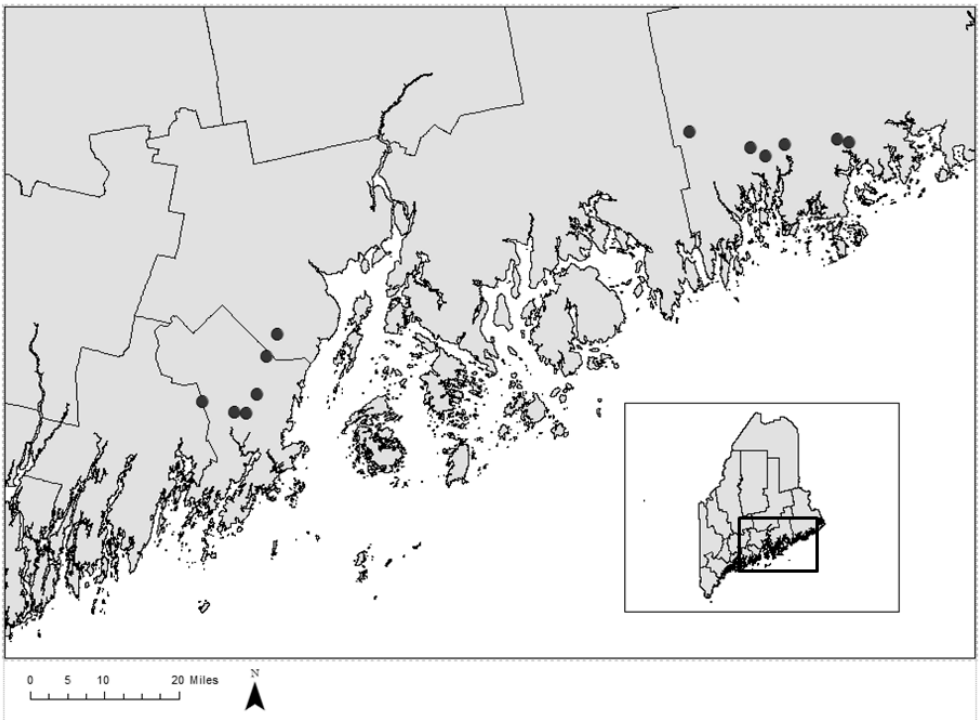
Study 9. Characterizing wild bee communities in power line rights of way in Maine's wild blueberry production landscape

Report from Brianne Du Clos (Ph.D. candidate), Dr. Cyndy Loftin, and Dr. Frank Drummond

Wild bees are a critical resource for pollinating Maine wild blueberry; however, Maine's wild blueberry production landscape is heavily forested. Wild bee communities decrease in diversity and abundance with an increasing proportion of forest cover in the landscape surrounding crop fields, though this relationship is not universal among all bees. Power line rights of way (ROW) occur throughout the blueberry landscape and may provide a consistent source of habitat for wild bees. Management guidelines for ROW often call for the removal of tall vegetation for safety purposes, creating paths of early-successional, shrub-scrub habitat. The dense, thick shrub composition that develops from the use of selective herbicides is typically complex enough to support bee communities that are more stable and diverse than those in mowed or unmanaged habitat and should remain stable over time. Pollinator communities in power line ROW are not well-studied. We surveyed wild bee and flower communities in power line ROW over two field seasons to assess their capacity as wild bee habitat.

METHODS: We established 12 survey sites in large power line ROW; six in Washington County and six in Knox and Waldo counties (Fig. 1). These two blocks of sites allow us to assess the effect of landscape complexity on wild bee communities in power line ROW. Within each block, three sites were near blueberry fields and three sites were isolated in forest to assess the effect of a mass flowering crop on bee communities. The sites near blueberry fields were different in 2014 and 2015 due to the blueberry cropping cycle, but the sites isolated in forest remained the same throughout the study period. Pollinator community surveys were conducted in early, mid-, and late summer of 2014 and 2015. Each survey consisted of one hour of live-netting bees and a 24-hour deployment of bowl traps. We surveyed blooming flowers along two, 25 meter transects during each sampling occurrence after live netting bees. All bees were collected for identification to genus in the lab, and pressed samples of all flowers were made to identify them to species.

Fig. 1. Map of power line ROW survey sites, 2015.



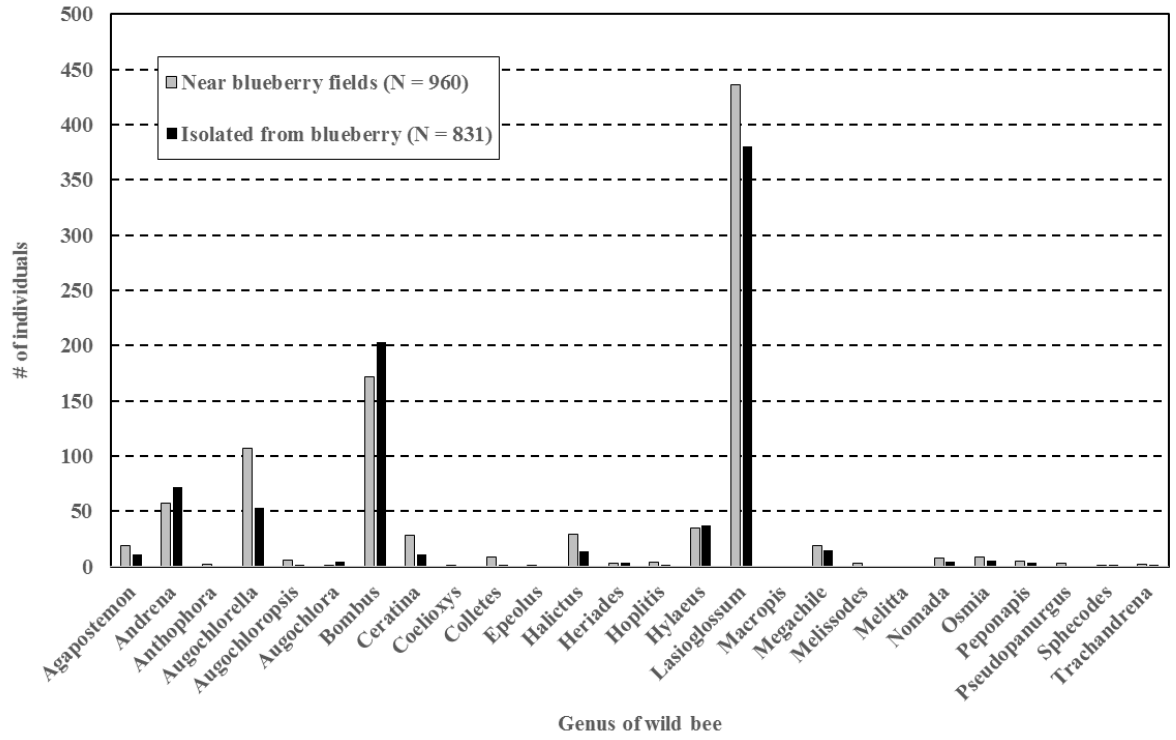
In the Knox/Waldo block of sites, we surveyed wild bees in the wild blueberry fields near to the power line ROW throughout 2014 and 2015.

RESULTS: All bees have been identified to genus, and expert verification of the identifications is pending. Here we present preliminary results for wild bee genera found in power line ROW near to and isolated from wild blueberry fields (Fig. 2). We collected 960 bees from sites near blueberry and 831 bees from sites isolate from blueberry.

Once verified, we will use statistical methods to compare bee communities between power line ROW sites near to and isolated from wild blueberry fields and between the two survey blocks. Preliminary results from a 2013 pilot study in Washington County indicate that there is a significant difference in Shannon diversity of live-netted bees between sites near blueberry fields (H=1.84) and sites isolated in forest (H=1.77) (t-test, $P = 0.01$). We expect to see this trend for both study regions in our subsequent study. We also expect that wild bee communities will be more diverse and abundant in Knox/Waldo counties than in Washington County.

We will use the surveys in nearby wild blueberry fields in Knox/Waldo counties to assess wild bee communities in nearby power line ROW during and after wild blueberry bloom. We expect that bee communities in power line rights-of-way and blueberry fields will have similar species diversity, but blueberry fields will have a greater abundance of wild bees during crop bloom. Bee abundance will increase in the power line rights-of-way post-bloom as bees shift foraging to floral resources there.

Fig. 2. Wild bee genera by power line ROW site type.



CONCLUSIONS: This study will contribute to growing knowledge of power line ROW as wild bee habitat. Power line ROW may serve as a source of forage for wild bees after the wild blueberry bloom, and they provide nesting habitat in open soils and woody debris. Natural habitat such as that found in ROW has been repeatedly demonstrated to enhance wild bee abundance and diversity in nearby crop fields.

Study 10. Monitoring honeybee health in wild blueberry during bloom. 2014-2015. Report from Jenn Lund, Rebecca Riverminder, and Dr. Frank Drummond

Honeybee decline is a serious threat to wild blueberry production. Since 2006 honeybees have been experiencing high levels of colony collapse. Parasitic mites, pathogens, narrow genetic diversity of honeybee stocks, and continuous pesticide exposure are some of the factors that can cause ill health to honeybees.

METHODS: A two-year study aimed at assessing colony health of honeybee colonies on wild blueberries during pollination (mid-May to mid-June) was initiated in 2014 and continued in 2015. Several hive locations in wild blueberry were monitored each year. We monitored nine hive locations (3 hives/location) in 2014 and 10 locations (5 hives/location) in 2015. Colony health and potential factors that might affect health were measured during bloom. Colonies were sampled three times during the 25-day (approximate) bloom period in May and June. Colony health was measured by sampling queen status (whether queen was present and laying eggs), preparations for queen supercedure (presence of supercedure cells), sealed brood population (the % area of wax comb on varying sized hive bodies with sealed brood and then converted to

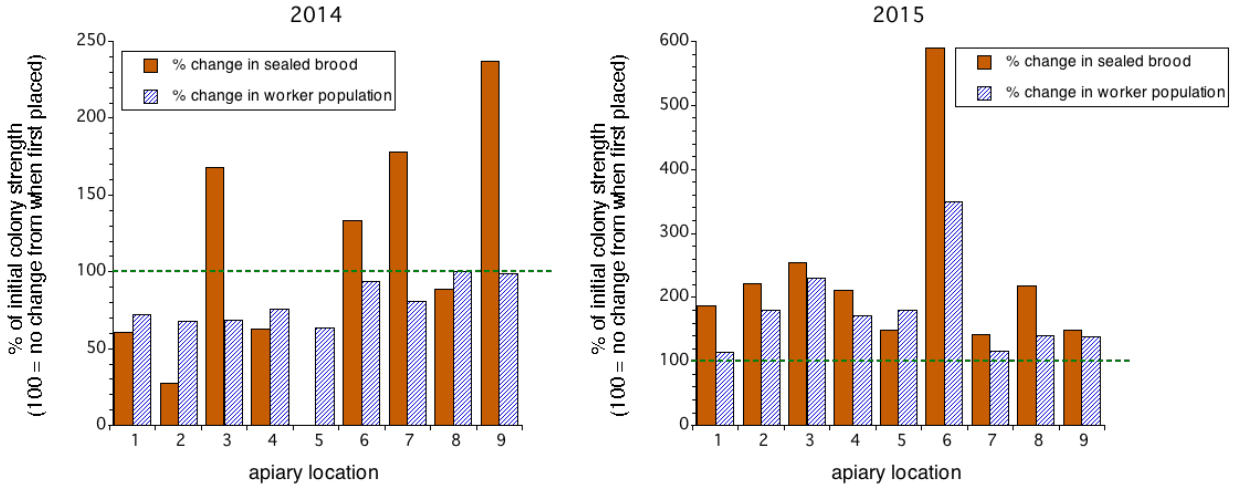
numbers of brood using published formulae), and worker population size (the % area of wax comb on varying sized hive bodies with workers and then converted to numbers of workers using published formulae). The health status sampling was performed soon after hives were deployed in the blueberry fields and then again just before bloom ended. A measure of population growth was calculated by determining the % growth from the first sample to the second sample of both sealed brood and workers.

Pesticide analysis was conducted on trapped pollen (sampled during peak bloom on 3 colonies/location, wax comb taken from the brood area of five hives (late bloom) and bee bread extracted from wax comb (only 2015 in late bloom). *Varroa* mite and tracheal mite infestation were sampled by collecting ca. 300 workers from the brood area in each of three (2014) or five (2015) hives at each location and then ETOH washes for *Varroa* or dissection in the lab was performed for tracheal mite. Samples were sent to the Connecticut Agriculture Experiment Station (2014) or the Gaston USDA lab (2015) for chemical analysis.

During late bloom a sample of 200 workers from each hive was taken to assess virus (ABPV, BQCV, CBPV, DWV-A, DWV-B (*Varroa destructor* virus), IAPV, LSV, and SBV), *Crithidia* or *Trypanosome*, and *Nosema* pathogens by submitting the honeybee samples that had been stored at -80ºC to a molecular lab in Beltsville (2014) or North Carolina (2015). Quantitative PCR was used to assess not only presence of the pathogens, but also the intensity of infection (estimated number of viral copies). ANOVA, linear regression, and linear correlation were used to assess factors that may have contributed to ill colony health.

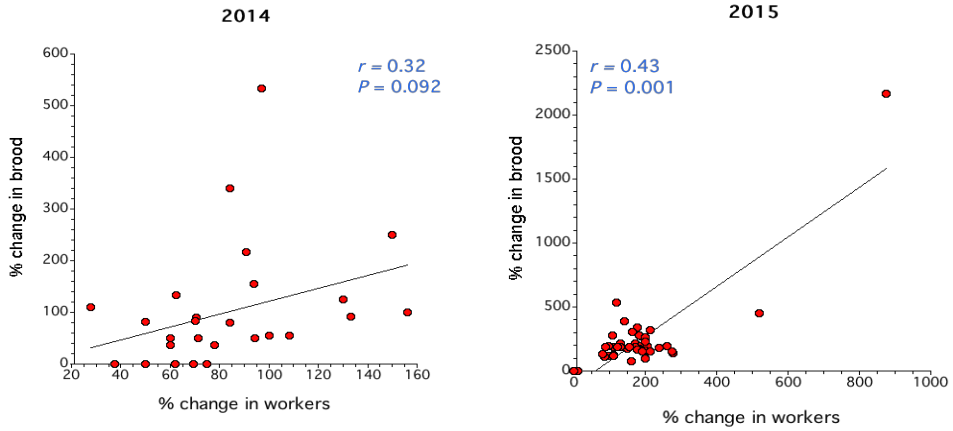
RESULTS: We found that 2014 was very different from 2015 in terms of colony health as we measured it (Fig. 1). We used a measure of change in colony strength, both workers and larvae, from the beginning of bloom to the end of bloom. In general, many colonies became weaker (more abundant population at the start of bloom compared to the end of bloom) in 2014 compared to 2015. The locations were not the same among years; although, some sites were in adjacent fields. The green dashed line on both graphs shows the line of no population increase from the beginning of bloom to the end of bloom. Any bars below the dashed line had populations that declined over bloom and bars above the dashed line increased during bloom. Overall, it can be seen that the change in both brood and worker populations in 2014 suggests that many apiaries did not prosper during bloom; whereas, in 2015 all apiary locations had either neutral or positive population increase. In 2014, just after bloom finished, many hives (an estimated 25%) that left wild blueberry collapsed on the subsequent floral crop or bee pasture that they were deployed on. We have not heard that this was the case in 2015. The sampling of colonies in 2014 indicates that many colonies were in decline (4 of the 9 locations) over the bloom period. The central question is…are colonies coming to Maine already stressed and unhealthy, or are the conditions during bloom in wild blueberry causing colonies to become unhealthy and then collapse or dwindle after they leave blueberry. The sampling of pesticide exposure and parasites and pathogens cannot definitively determine the cause of the health status of the sampled colonies, but it can shed light onto what may be happening.

Fig. 1. Change in adult worker bee and larval bee (brood) abundance from the beginning of bloom to the end of bloom (green line denotes no change line), 2014 & 2015.



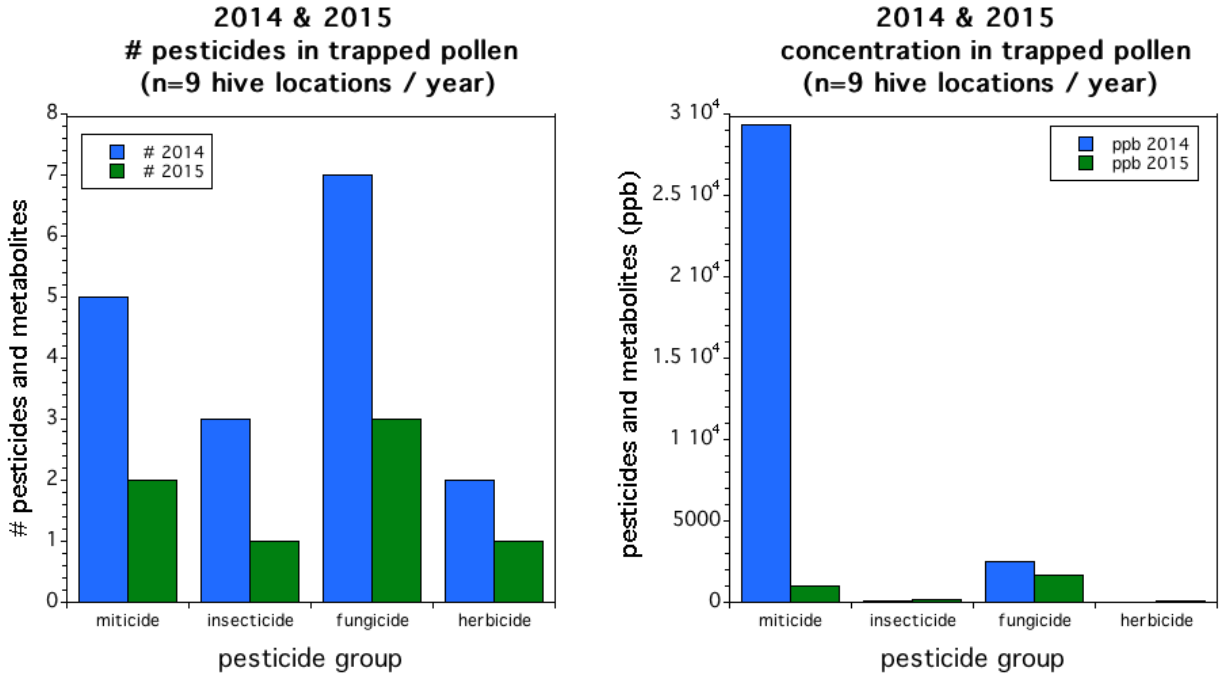
Although the correlations between worker and brood populations are not strong, they are significant in both years (Fig. 2), although highly leveraged in 2015 by one extreme colony. The rate of change in brood relative to a change in workers is greater in 2014 (slope = 1.24) than in 2015 (slope = 0.54), suggesting more brood production for a given level of worker population. Although it cannot be assumed that the rate of buildup is related to prior health conditions of the colonies or conditions during blueberry bloom that affected colony health.

Fig. 2. Relationship between % change in workers and brood, 2014 & 2015.



Pesticide exposure (both parent chemical compounds and their metabolites) in trapped incoming pollen was greater in 2014 than in 2015 (Fig. 3). Both the number of pesticides for each group and the concentration were greater in 2014. Trapped pollen, in MOST cases is thought to represent exposure from the flowers that honeybees are foraging on, but because there is a high level of miticides in the pollen, this has to be questioned.

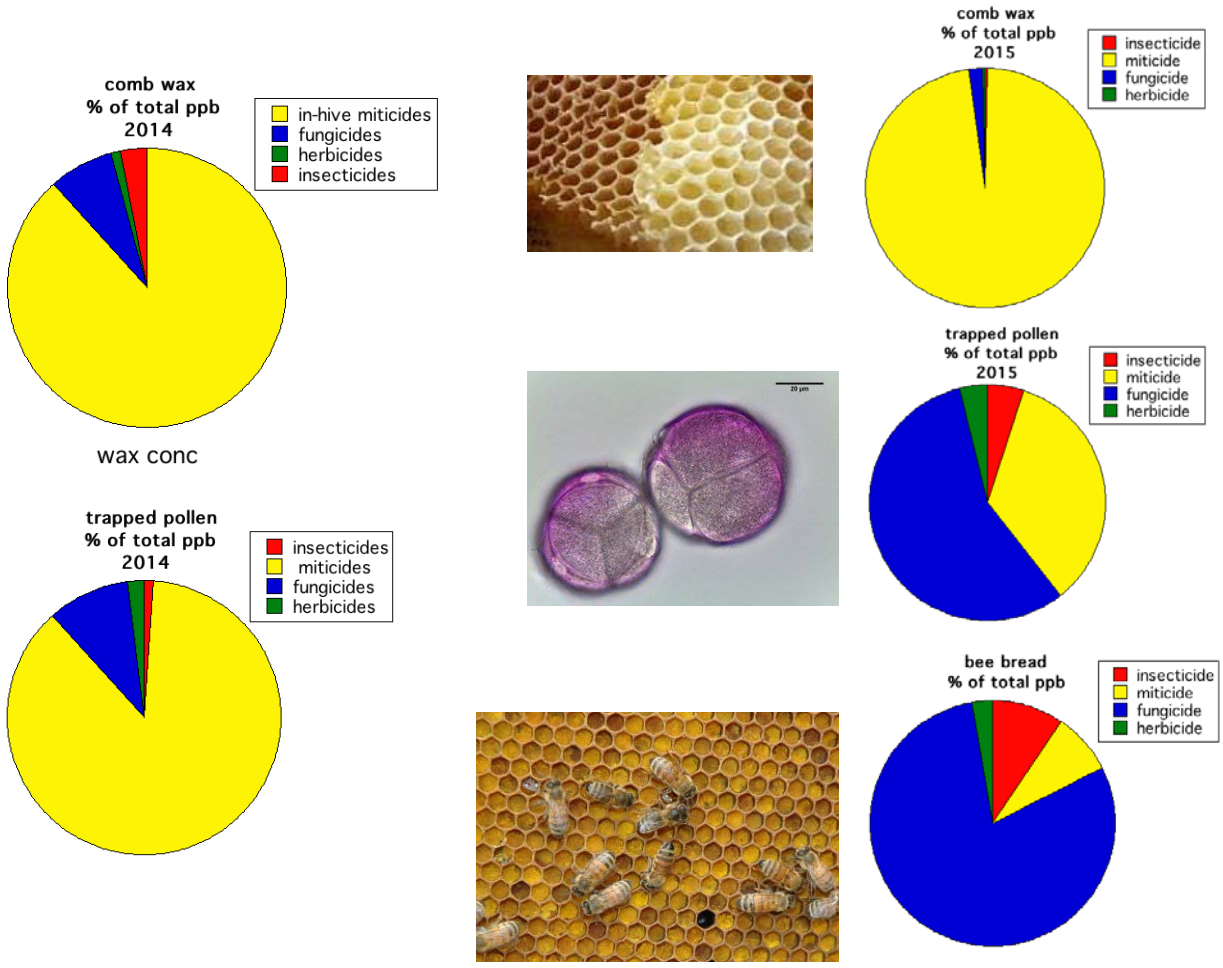
Fig. 3. Number of pesticides and pesticide concentration (ppb) in trapped pollen, 2014 & 2015.



We also sampled wax comb and bee bread (a mixture of pollen and nectar that is fed to the larvae). Looking at the relative exposure (% of total concentration by each pesticide group) one can see that in 2014 (pie charts on the left) the greatest concentration of pesticide exposure is in-hive miticides (yellow) both in trapped pollen and wax comb (Fig. 4). Fungicides (blue) make up the next greatest proportion; although, it is very small (about 10-15%), followed by insecticides (red) and herbicides (green).

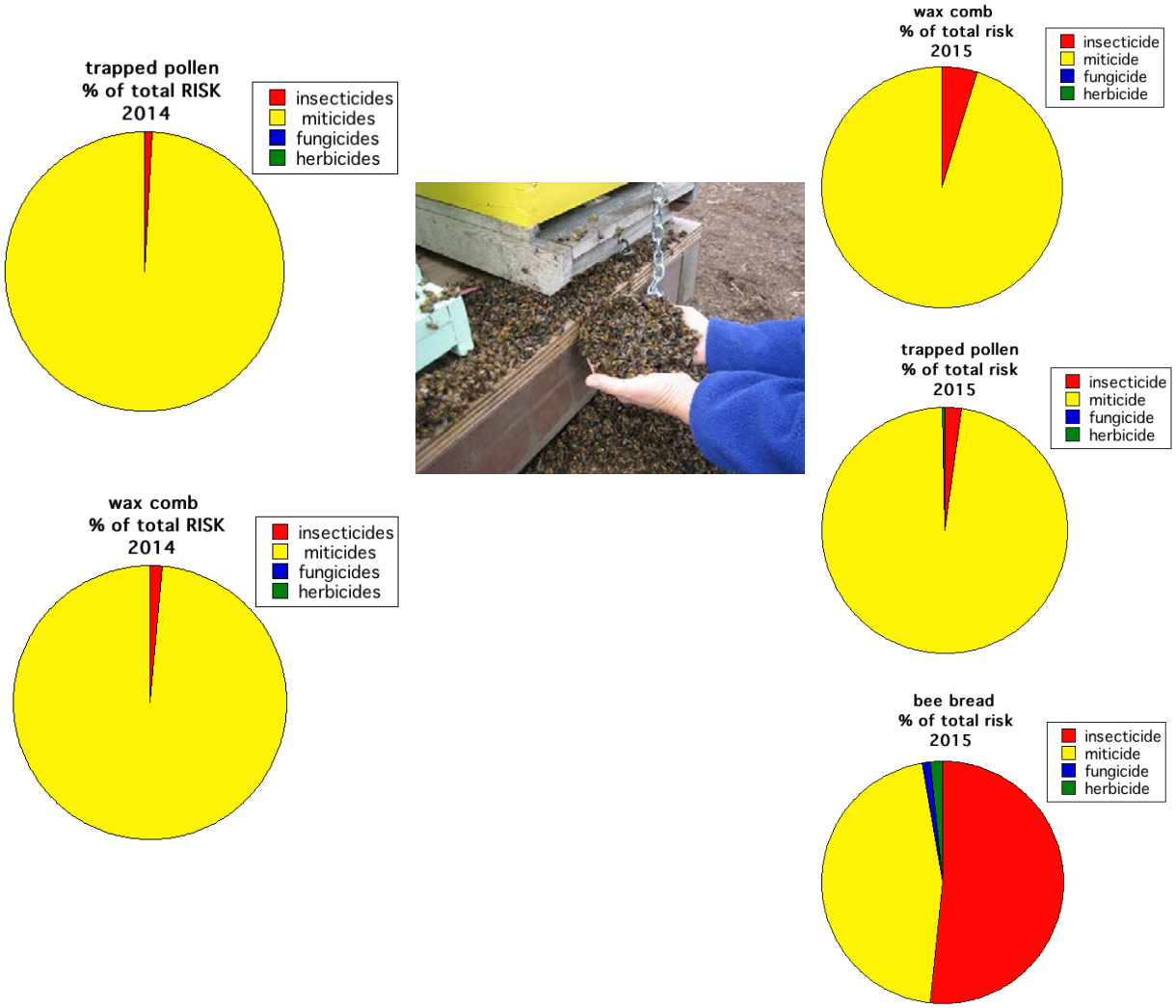
In 2015, again wax comb mostly contaminated by in-hive miticides (yellow), followed by fungicides (blue), and a much lower level of insecticides (red). Trapped pollen the future larval bee food has the most contamination from fungicides (blue) followed by in-hive miticides (yellow). The bee bread (more near-term bee larval food) is predominantly contaminated with fungicides(blue), then insecticides (red), followed by in-hive miticides (yellow) and herbicides (green). The concentrations are informative because they suggest what bees are coming in contact with, but it does NOT tell us what might be the potential threat to honeybees in wild blueberry.

Fig. 4. Estimated relative exposure to honeybees of each pesticide group in pollen, wax comb, and bee bread, 2014 & 2015.



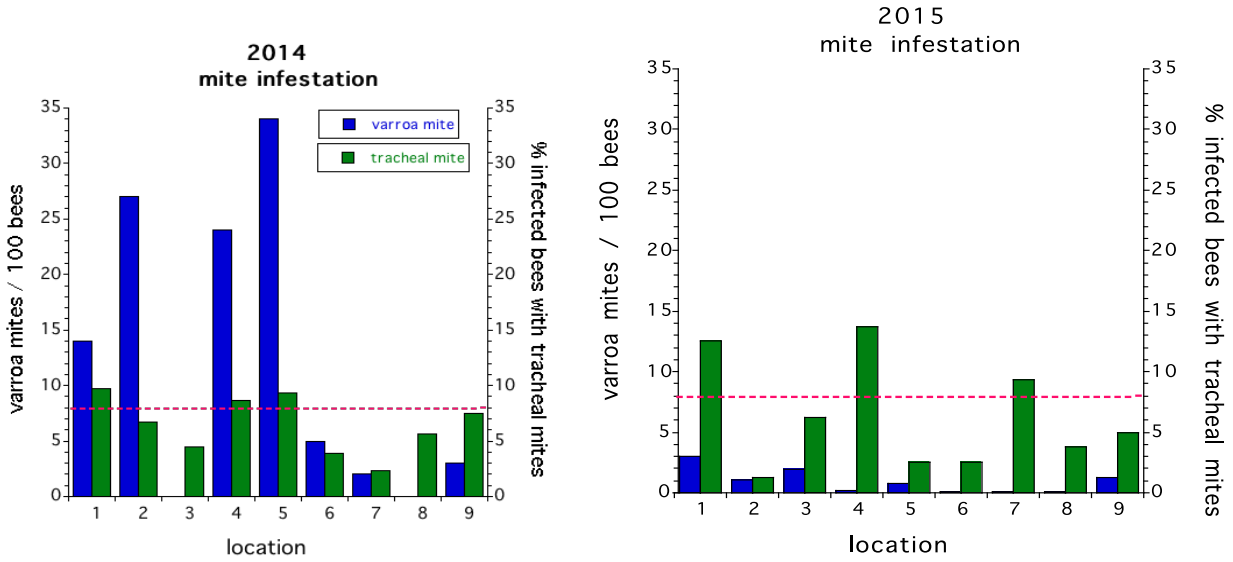
When the relative exposure is viewed by measuring the "RISK" to honeybees in the colony (RISK = ppb concentration / the amount needed to kill 50% of the colony…i.e. LD_{50}), a different picture emerges (Fig. 5). In 2014 (left side of panel), in both trapped pollen and wax comb, it can be seen that by far the greatest risk is from miticides (yellow). In 2015 (right side of panel), wax comb and trapped pollen show the greatest risk to mimic 2014, being miticides (yellow). However, the risk in bee bread was observed to be mostly insecticides (phosmet among others) followed by miticides (yellow). It is interesting that bee bread was not similar to trapped pollen or wax comb. It is difficult to know exactly what is going on in the colonies, in terms of risk to exposure, since wax comb and bee bread represent a combination of exposures picked up before the colonies arrived in blueberry as well as when they arrived in blueberry. Even the trapped pollen does not solely represent exposure routes from the outside environment, as can be seen by the large exposure to chemicals that are for the most part in-hive miticides. Also, there are no miticides used in wild blueberry for pest management.

Fig. 5. The calculated Risk Quotient calculated for honeybees in 2014 and 2015.



In 2014 and 2015 (Fig. 6) tracheal mite infestations (green bars) were high for the spring of these years, but not unusually so. *Varroa* mite infestations (blue bars) were extremely high in 2014 and were at levels that threatened colony collapse (see red dashed line). In 2015 (Fig. 6, on right) *Varroa* mite infestations were moderate to low; although, location one was climbing toward a level of concern with 3 *Varroa*/100 worker bees.

Fig. 6. The tracheal and *Varroa* mite infestations for 2014 and 2015.



Also during honeybee dissections for tracheal mites we discovered what we believe is parasitism of honeybees in 2015 by the phorid fly, *Apocephalus borealis*. Further research in 2016 by R. Riverminder will focus on confirmation of phorid fly parasitism. Figure 7 shows the percent parasitism observed at each location. This fly was reported in California in 2012 as a potential cause of colony collapse and is also a natural parasite of bumblebees in the Northeast. We were surprised to see it in 2015 since we did not find it in 2014.

Figure 8 from quantitative PCR shows the level of viruses (black, red, blue), protozoans (*Trypanosome*, mostly *Crythidia*, in gray), and fungi (*Nosema* in purple). Black queen cell virus was present at all sites, but at low to moderate levels in 2014. Deformed wing virus and sac brood virus were at moderate to high levels as was *Nosema ceranae*. *Trypanosome* levels were extremely high at three locations. It is difficult to predict what the ramifications of the infections are, but the ubiquitous nature of infection is probably related to high *Varroa* mite levels and overall, average ill health of the colonies during bloom.

Fig. 7. Phorid fly percent parasitism of honeybees in 2015.

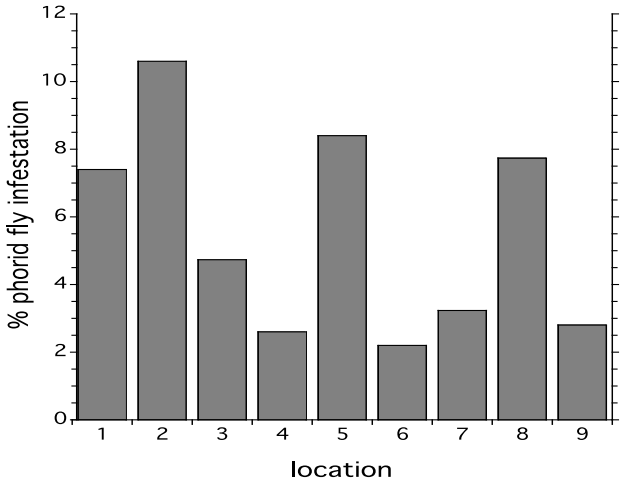


Fig. 8. Pathogen levels in 2014 colonies (standardized to vary between 0 and 25).

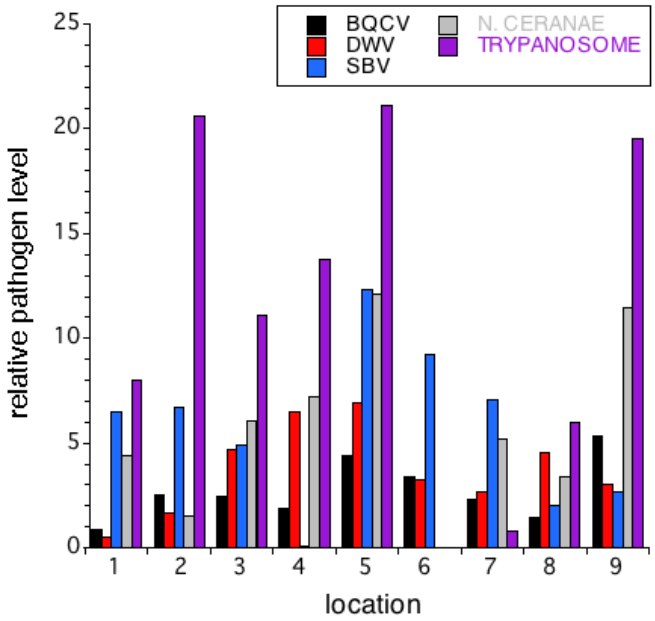


Figure 9 depicts the relative abundance of pathogens in honeybee colonies in 2015. Several more pathogens were screened for in 2015. The most abundant pathogens were *Nosema*, Deformed wing virus (DWV-A), *Varroa* destructor virus (DWV-B) and Lake Sinai virus (LSV). Many of the pathogens were correlated in their levels across the 10 locations. Table 1 shows the pairs of pathogens that co-varied (Spearman's rank correlation). In addition, *Varroa* mite levels were correlated with the levels of deformed wing virus (Fig. 10).

Fig. 9. Pathogen levels in 2015 colonies (standardized to vary between 0 and 25).

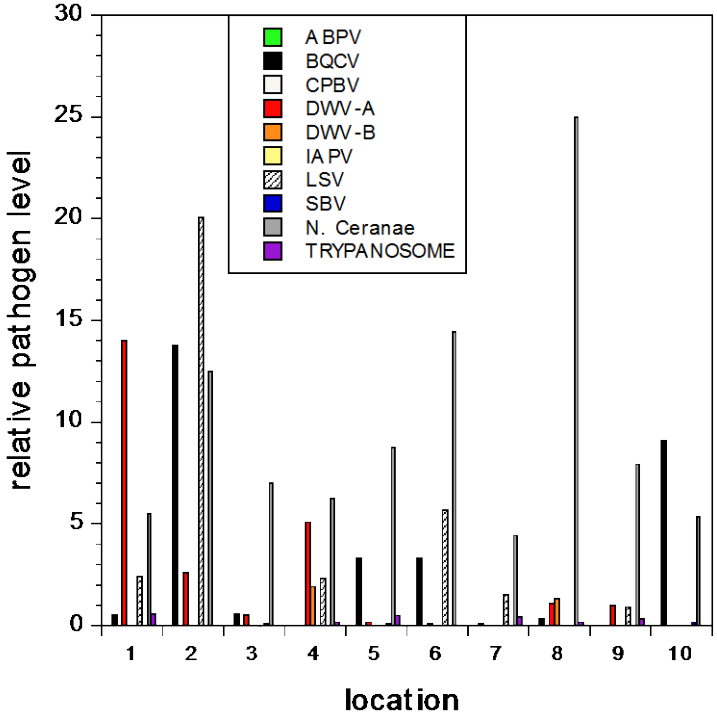
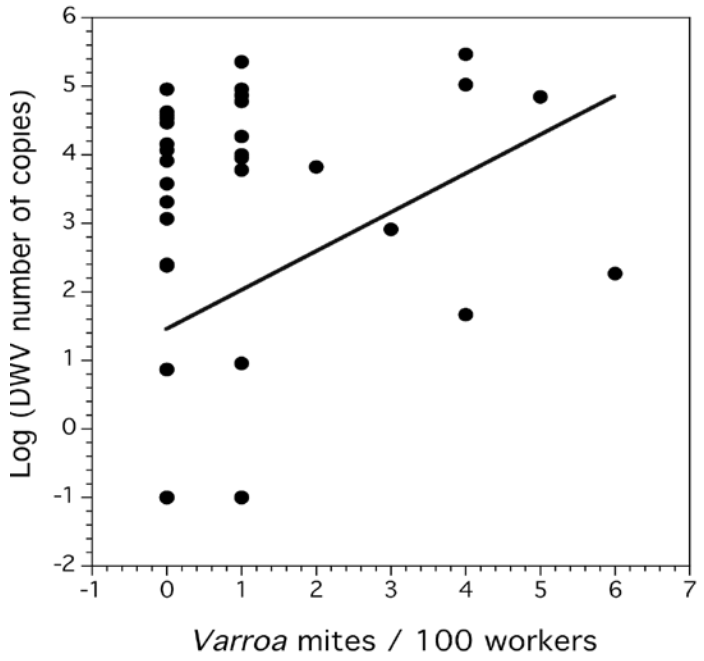


Table 1. Associations among viruses in 2015 across the apiary locations.

Pathogen association	Correlation coefficient	P-value
Deformed wing virus vs Acute bee paralysis virus	0.381	0.007
Varroa destructor virus vs Acute bee paralysis virus	0.385	0.006
Deformed wing virus vs *Varroa destructor* virus	0.311	0.028
Israeli acute paralysis virus vs Black queen cell virus	0.357	0.011
Sac brood virus vs Black queen cell virus	0.332	0.019
Nosema vs Black queen cell virus	0.284	0.046
Trypanosome vs Acute bee paralysis virus	0.415	0.003
Trypanosome vs Deformed wing virus	0.358	0.011

Fig. 10. The relationship between *Varroa* mite levels and Deformed wing virus levels, 2015.



A principal component analysis (PCA) of all colonies at the 10 apiary sites monitored in 2015 was conducted. All the pathogen, parasitic mite, and colony health measures were included in the ordination. Figure 11 shows the ordination as a plot in the first two dimensions (32% of the total variance explained by the first two axes). This ordination shows that there is no obvious separation of the apiary sites due to pathogens, parasites or colony health measures. Multiple analysis of variance confirms this finding that apiary sites did not form separate populations ($F_{(9,39)} = 1.635$, $P = 0.139$). As can be seen in the ordination there was much variability between colonies within apiary sites (hive drops). However, there was an apiary x pathogen diversity interaction, suggesting that the pathogen complex did vary from apiary to apiary ($F_{(81,209)} = 1.516$, $P = 0.009$). Table 2 shows examples of these differences among apiaries.

Fig. 11. An ordination (principal components analysis) of the colonies monitored in 2015. Symbols of the same color and shape are colonies from the same apiary (hive drop).

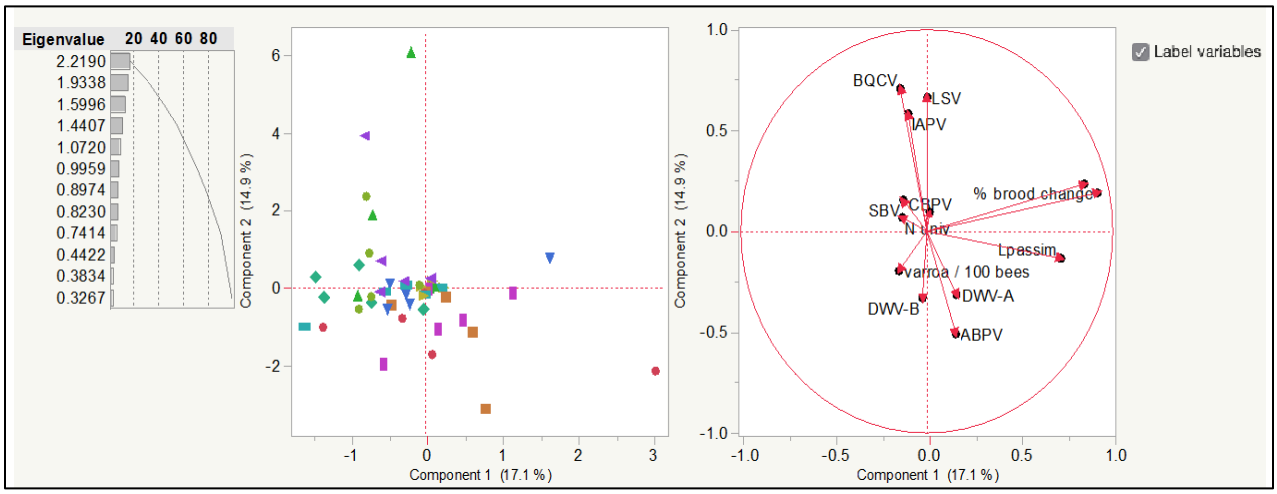


Table 2. Individual pathogen differences among apiaries (Wilcoxon rank test).

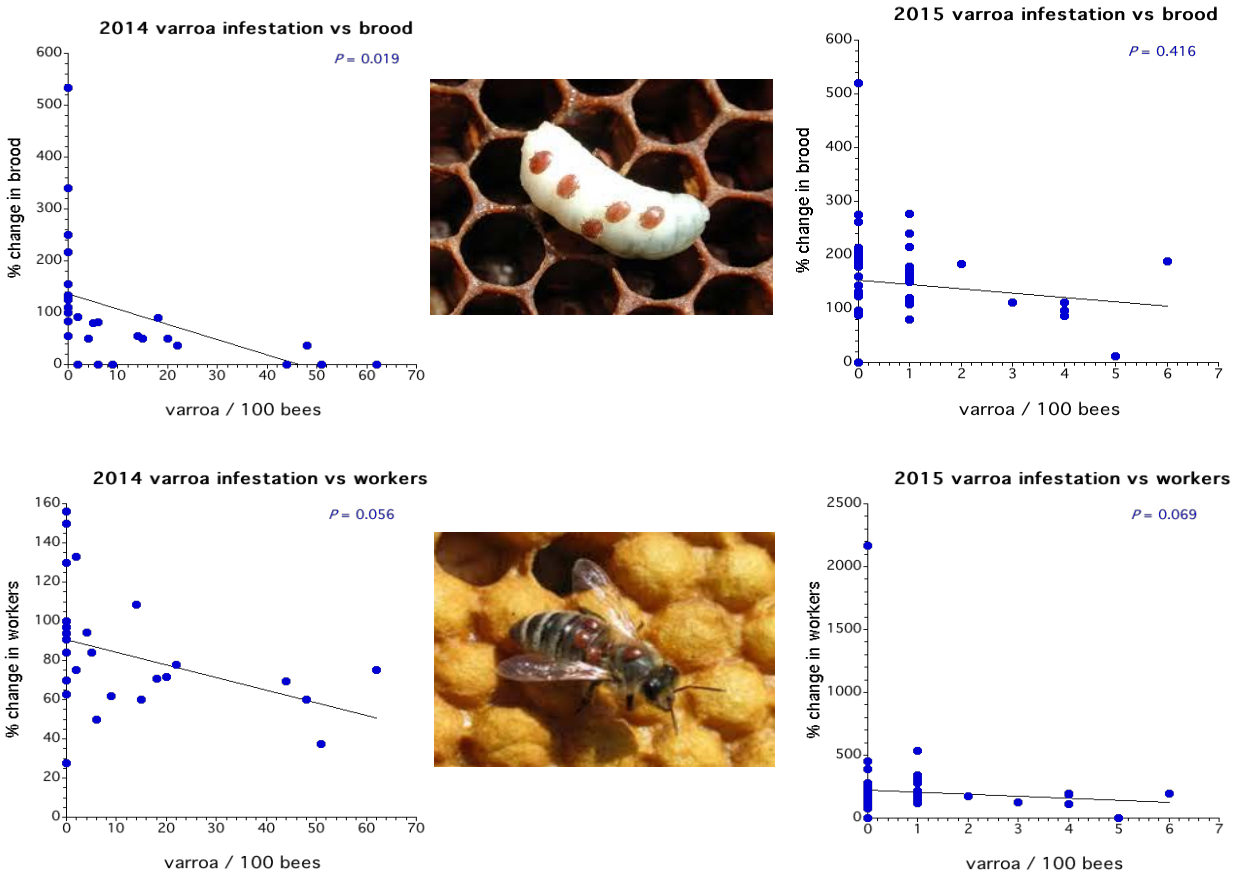
Pathogen	Wilcoxon statistic	P-value	Apiary differences
ABPV	31.367	0.0003*	Wilson > Aurora & Wreath Burnt Camp > Aurora & Wreath
BQCV	26.859	0.0015*	Aurora > Burnt Camp & Wilson EO Morse > Wilson
CBPV	7.298	0.6061	
DWV-A	28.541	0.0008*	Airport > Long Pond & Columbia Burnt Camp > Long Pond
DWV-B	13.938	0.125	
IAPV	10.987	0.156	
LSV	9.334	0.223	
L. passim	19.044	0.025*	Wilson > BBHILL Airport > Aurora & BBHill & EO Morse & Wreath Tibbets & Burnt camp > Wreath
Nosema	7.176	0.6188	
SBV	21.179	0.0119*	EO Morse > Tibbets

The main point of Table 2 is not that specific apiaries have higher or lower levels of specific pathogens than other apiaries, but that the variation in pathogen levels does differ among apiaries, but there is NO discernable pattern.

Factors related to colony health: Linear statistical models were used to assess factors that were related to the variation in health/population growth status of the colonies in both 2014 and 2015. Pesticide exposure did not correlate with either brood or worker changes during bloom. We

looked at total concentration, concentration by pesticide type, number of pesticides, risk quotients of both total and by type of pesticide and no evidence of significant effects were found. These analyses were conducted for trapped pollen, wax comb, bee bread, and by pooling over all substrates. However, it should be noted that our analyses of pesticide contamination and risk could only be conducted for each hive site and not for each hive because of the expense involved in the quantitative chemical analysis. There was no relationship to colony health and pathogen levels or tracheal mite infestation in 2014, nor any relationship between colony health and pathogen levels, tracheal mite infestations, or phorid fly eggs and larval infestations in 2015. However, there was a relationship between colony health and *Varroa* mite infestation levels in 2014 and 2015 (Fig. 12). *Varroa* mites were sampled from every colony and in 2014 significant decreases in brood ($P < 0.05$) and worker growth rates ($P < 0.10$) were associated with *Varroa* mite infestation levels that were exceedingly high. In 2015, *Varroa* mite levels were quite a bit lower and colony health at each site was better, in terms of population growth. However, there was a relationship between *Varroa* levels and brood growth ($P < 0.10$).

Fig. 12. Relationship between *Varroa* mite and change in brood and workers, 2014 & 2015.



CONCLUSIONS: A two-year honeybee health-monitoring program was conducted in the Maine blueberry barrens. The two years represented highly different patterns in colony health as measured by brood and worker population growth. The 2014 colonies experienced a high level of collapse after leaving Maine. We do not know if this was the case in 2015. The only correlates to honeybee colony health in the two year study was *Varroa* mite infestation which

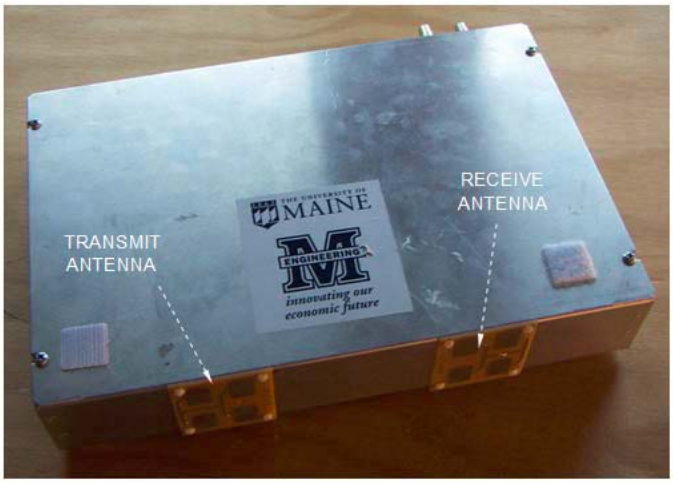
was extremely high in 2014. *Varroa* infestation could be the causal factor for colony ill health, but one should not ignore the potential effects of pesticides, especially in-hive miticides. A cautionary note is that pesticide exposure is never a good thing for honeybee colonies and the blueberry industry by practicing sound pest management should minimize exposure whenever possible.

Study 11. Development of a Doppler radar microphone for honeybee colony health assessment
Report from Dr. N. Emanetoglu, Dr. H. Aumann, and Dr. F. Drummond

Honeybee health monitoring requires intensive and sometimes invasive hive inspections. The hive inspections are very labor intensive and preclude assessment of a large number of hives in a small amount of time. RADAR techniques were investigated to monitor honeybee hives, with the aim to assess colony strength in a minimal amount of time.

METHODS: A portable Doppler radar microphone was built and tested in the field. The modulation of the radar signal due to the mechanical bee vibrations emitted by bees living in the hive was measured and correlated with honeybee activity. A second unit was developed based on measuring the signal transmitted through the hive instead of the signal reflected from its surface. This Bee Hive Transmissivity (B-HiT) unit provides the capability to determine how full a beehive box is, while discriminating between honey and honeybees.
Aumann and Emanetoglu developed a 5.8 GHz prototype Doppler radar microphone (DRM) in 2014. A second, field-portable Doppler radar microphone unit was built using funds from this grant. The two units will be referred to as Doppler1 and Doppler2, respectively. By operating in the unlicensed 5.8 GHz Industrial, Scientific and Medial (ISM) radio frequency band, the instrument can be implemented safely and inexpensively with readily available wireless components. Both DRM units transmit a maximum power of 10 mW, significantly below the 1 W limit imposed by FCC regulations. The power radiated from the radar unit is below even Wi-Fi router signal power levels, and is safe to operate.

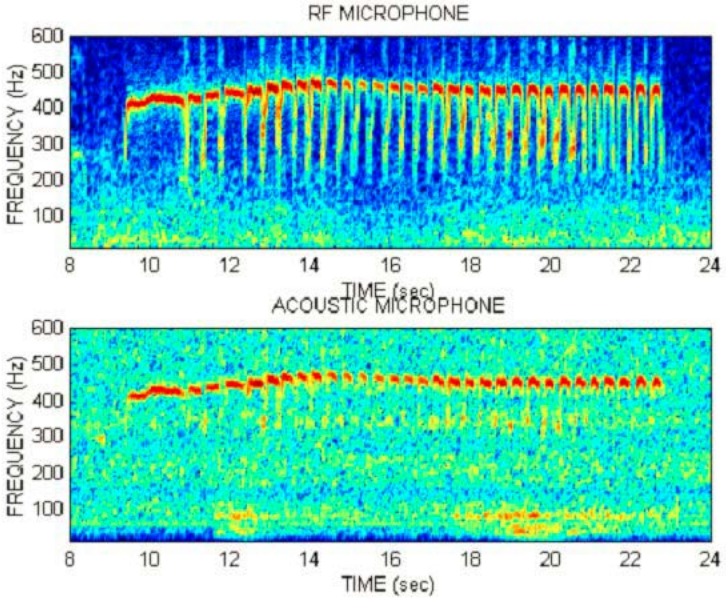
Fig. 1. Doppler Radar Microphone developed at the University of Maine for honeybee research.



The original prototype (Doppler1) was mounted on a fixture near an observation hive at the Aumann Farm site (AFS). The DRM was used to take measurements of the observation hive daily, from June through August, multiple times a day. The DRM output was recorded using a laptop, in the WAV file format. Simultaneous recordings were also made using acoustic microphone, and these recordings were also save in WAV file format.
The second unit (Doppler2) was used in experiments at the University of Maine Apiary at the end of Grove Street Extension on campus. On 29 July eight hives ranging in size from 2-4 hive boxes were sampled early in the morning (7 AM) for honeybee worker strength (number of workers per frame per box, estimated by percent coverage of each frame surface) and the number of frames containing honey. In the afternoon, transmission of radio frequencies were measured through each hive box the colonies. Regression analysis was used to relate the received strength of radio frequencies and hive box worker numbers and the amount of honey. Radio transmission measurements and colony assessments were made again on 19 Aug and 20 Sep.

RESULTS: The data from both sets of measurements was analyzed using Matlab. Data analysis was performed by Drs. Aumann and Emanetoglu, as well as Ms. Catherine Ahola, a Bangor High School (BHS) student who was funded by BHS. The Doppler radar microphone data was correlated with visual observations and the acoustic microphone data. The captured data showed audio signatures between 200 Hz and 250 Hz when the bees were active, and 150 Hz to 180 Hz when the bees were resting. Figure 2 shows a comparison of the data collected with the Doppler radar microphone and an acoustic microphone when the queen bee was active.

Fig. 2. Comparison of DRM (RF microphone) and acoustic microphone recordings.



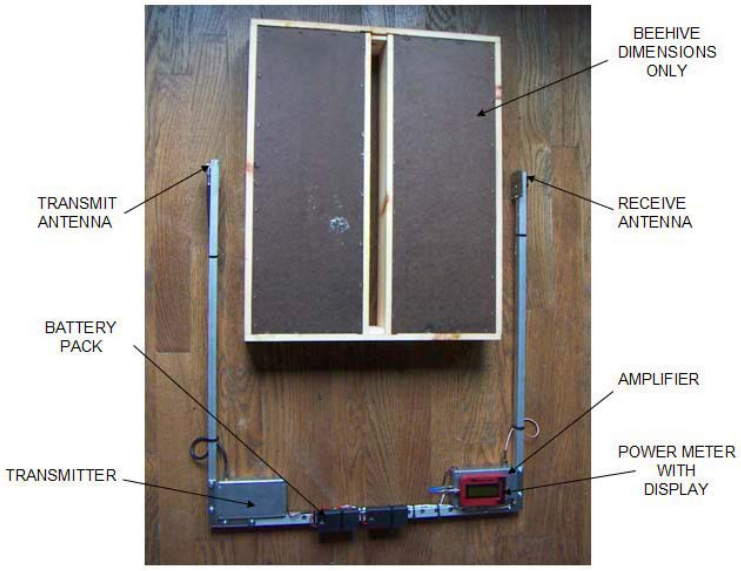
We used the DRM design for the first set of measurements in June. This experiment conducted also revealed that the original DRM design was very sensitive to the distance between the radar and beehive. The received signal strength was not only dependent on the bee colony strength, but also on the distance to the beehive in a complicated manner. It was expected that the signal strength would rapidly decrease due to the fact that the return signal is inversely proportional to

the fourth power of the distance, i.e. $1/R^4$ where R is the distance between the radar and beehive. However, it was discovered than even a quarter of an inch change in the distance could cause the signal strength to increase or decrease. It was determined that this was due to the phase difference between the modulated reflected signal and the static reflected signal (i.e. the background reflection). The DRM transmit frequency could be tuned to obtain the best signal-to-noise ratio. However, this would require either a trained technician to operate the DRM or an auto-tune and distance calibration function to be added to the DRM, significantly increasing its complexity and therefore cost. Two alternative designs were investigated, with promising results.

The first alternative design took advantage of the static reflection to reduce the complexity of the DRM's receiver stage. The traditional direct downconverter design uses a local oscillator signal as a reference, at 5.8 GHz for the DRM, and mixes it with the received signal to obtain the audio frequency signal. In the modified version, the received signal was fed directly to a power detector circuit instead. The static background signal acted as the reference signal, and an audio frequency output was obtained from the power detector that was identical to the traditional design. This alternative Doppler radar design reduces the receiver component count by 80% and thus reduces the cost of the DRM's receiver stage. This alternative design was developed at the end of the performance period (August 2015) and was not thoroughly tested with the observation beehive. However, initial measurements were nearly identical to those obtained from the original DRM. More experiments with beehives will be carried out with the alternative design.

A second design, the bee hive transmissivity unit (B-HiT), was developed to address the problem with signal strength being a function of distance as well as honeybee colony size by fixing the distance between transmit and receive antennas, as shown in figure 3. As this distance is fixed, the signal attenuation is due only to material present in the beehive: honey and honeybees.

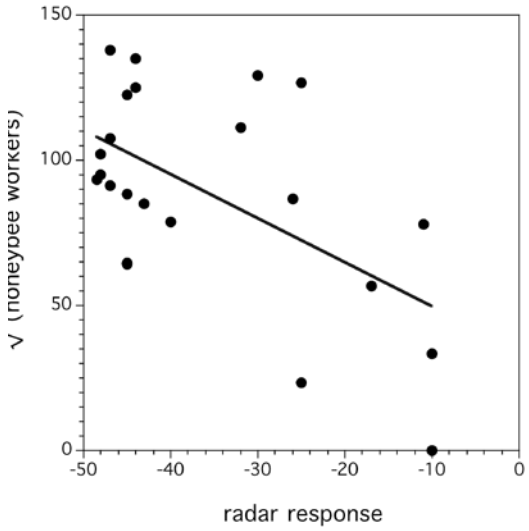
Fig. 3. The second alternative design**,** B-HiT unit.



We found that the Doppler microphone was able to predict worker number in each box fairly well (explaining 32% of the variance in worker bee numbers, $P = 0.005$, Fig. 4). Including the amount of honey in the hive, it was found that the Doppler microphone can predict hive strength

much better when measured as BOTH worker bee numbers and the amount of honey. The overall percent variation in predicting colony strength increases to 51% of the variation explained (P = 0.0003). This is because both honey and honeybee workers are comprised of high levels of water and water is a good absorber of radio waves.
Both honey and honeybees absorb and attenuate the RF signal transmitted by the B-HiT unit. However, the honeybees also modulate the signal at acoustic frequencies, allowing for the bees to be detected similar to the DRM. The strength of the modulation signal detected by the B-HiT can be unambiguously correlated with bee colony strength, as the transmission distance is fixed. In principle, the B-HiT unit is superior to the DRM as: (1) it doesn't suffer from ambiguities caused by distance variations, and (2) it can detect the presence of honey, which the DRM cannot.

Fig. 4. Relationship between Doppler microphone readings and worker population per hive box.



Two B-HiT units were constructed using components available in Dr. Aumann and Dr. Emanetoglu's labs, as well as components purchased using funds from this grant. The two units operated at 5.8 GHz and 2.4 GHz, both of which are unlicensed ISM bands. Both units transmitted at a signal strength of 10 mW. The 2.4 GHz version proved to be more reliable due to two reasons: (1) less attenuation at 2.4 GHz vs. 5.8 GHz, (2) larger antennas which resulted in a larger sampling volume. The larger sampling volume made the 2.4 GHz B-HiT unit less sensitive to honey or bee colony distribution in the hive.

CONCLUSIONS: The B-HiT units were constructed and tested in July, August and September. While the experiments were sufficient to determine that the 2.4 GHz B-HiT unit performed more consistently, more data needs to be collected to form a reliable analysis base to correlate the signal attenuation and modulating signal strength to the quantity of honey and/or honeybees in the hive.

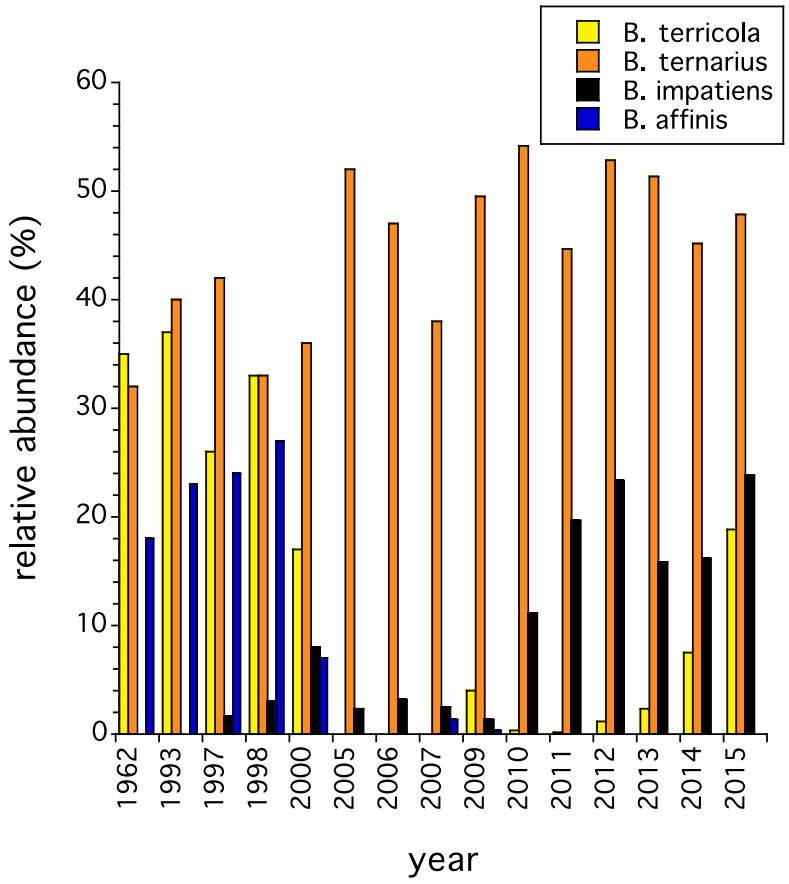
Study 12. <u>The apparent comeback of a once common bumblebee that went into decline. Report from Kalyn Bickerman-Martens (Ph.D. candidate), and Dr. Frank Drummond</u>

The yellow-banded bumble bee (*Bombus terricola*) was once one of the more common bumble bees in wild blueberry fields. In the late 1990s populations of the bee began to decline across the state. We have continued to look for it over the past 20 years and starting in 2012, we saw an increase in its populations and so we started a survey in 2013 to assess its well-being.

METHODS: Locations throughout Maine and the Maritimes were surveyed for the yellow-banded bumble bee. The survey sites were both producing blueberry fields and non-agricultural landscapes. At each site 30 minutes of searching for bumble bees on flowers and recording their numbers provided a measure of the abundance of the yellow-banded bumble bee relative to other species of bumble bees. Therefore, relative abundance was calculated on a per site basis, but only for sites where it was found. At a subset of sites, the flower species that the bumble bees were collected on was also recorded. In addition, in 2014, at a site in South Thomaston, Maine, ten sequential visits of 30 minutes each over a three day period were made to assess the likelihood of detecting the yellow-banded bumble bee on a per visit and multiple visit basis. Logistic regression was used to determine factors that might influence the yellow-banded bumble bee abundance and prevalence (percent of locations where it was detected).

RESULTS: Figure 1 shows a historical perspective based upon our data (1962 is data collected by Boulanger and Osgood) on the relative abundance of several key bumble bee species in Maine. *Bombus ternarius* (the red belted bumble bee) has remained common in Maine for the last 6 decades (orange bar). *Bombus affinis*, the rusty patch bumble bee (blue bar, *B. affinis*) was common until the 1990s when it declined rapidly as is the case with the yellow-banded bumble bee (yellow bar, *B. terricola*). However, one can see that the yellow-banded bumble bee was found again in 2009 in low abundance, but then increased in abundance from 2012 until present. The impatient bumble bee, *B. impatiens*, was not common in Maine in the 1960s, being a more southern bumble bee. However, in the late 1990s it started to be common, although in low densities, and presently it comprises 20-25% of the bumble bee abundance in Maine. There is no known cause of the declines of the rusty patch bumble bee and the yellow-banded bumble bee, but some scientists have conjectured that it is due to diseases brought into the Northeast by the commercial bumble bee for pollination. The rise of the impatient bumble bee is thought to be either due to climate change and warmer winters or due to introducing it into Maine with the growth of the commercial bumble bee industry.

Fig. 1. Relative density of key bumble bee species in Maine between 1962 and 2015.



Intensive bee survey since 2012 shows the increase of the yellow-banded bumble bee across Maine (Fig. 2 A-D). Glancing at the four panels it becomes apparent how fast the yellow-banded bumble bee has spread across Maine since its decline. The percentage of sites that it is found in has increased from 1-2 % in 2009-2012 to 27.3% in 2012, 9.4% in 2013, 49.5 % in 2014, and 65.9 % in 2015. It appears that this bee is in a resurgence; although, sampling in western and northern Maine has been sparse. Figure 3 shows that the yellow-banded bumble bee uses common summer flower species. This suggests that the cause of its decline was most likely not floral resources.

Fig. 2. Survey locations and presence (red dots) and absence (black dots) of the yellow-banded bumble bee, 2012 (top left), 2013 (top right), 2014 (bottom left), and 2015 (bottom right).

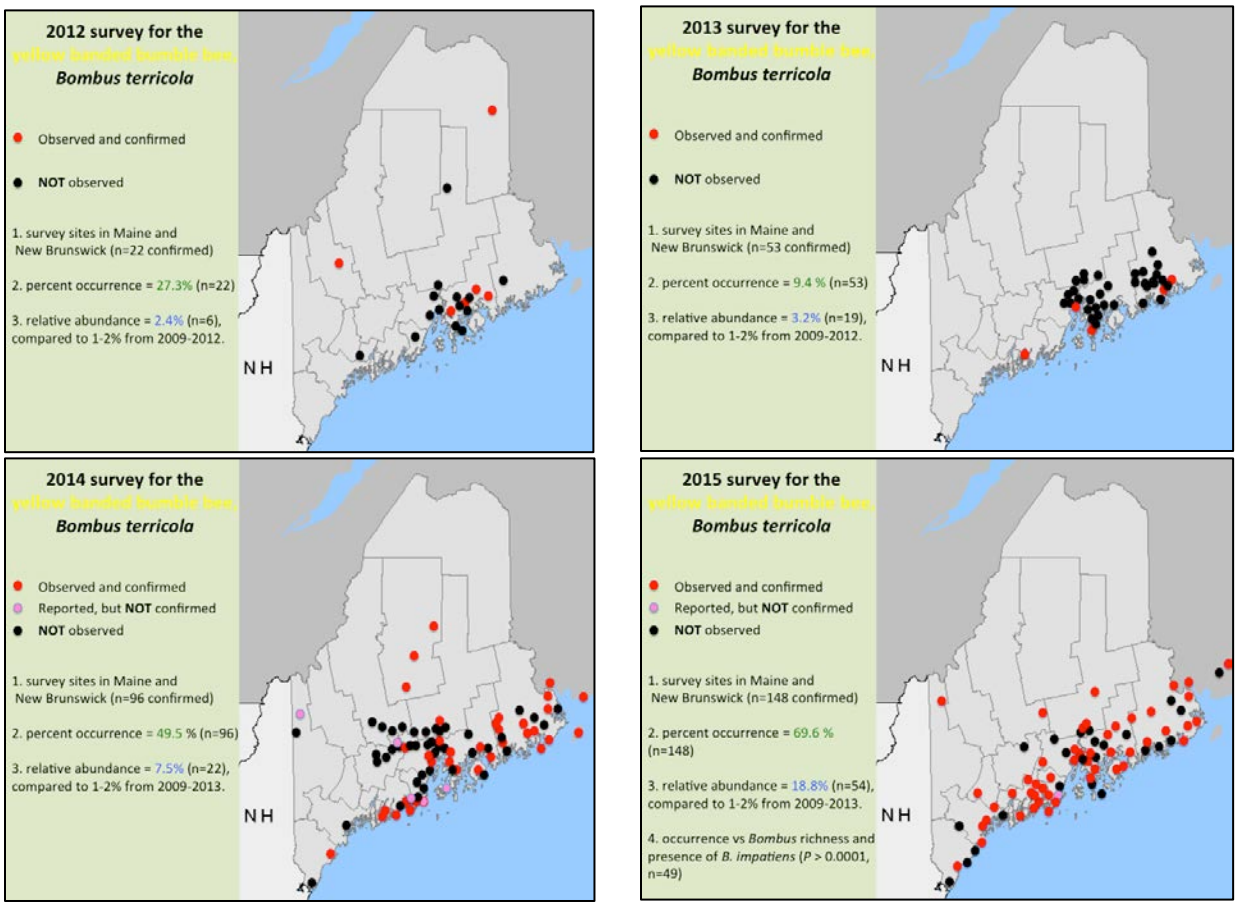
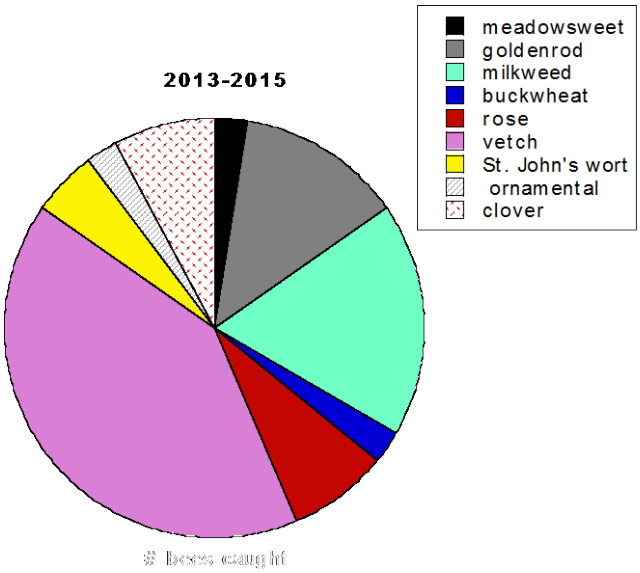


Fig. 3. Flower species on which the yellow-banded bumble bee was captured.



We conducted an analysis to determine what factors might be responsible for their presence in some locations and not others. We found that year (2014 compared to 2015) did not explain their prevalence (% of sites occupied) ($P = 0.649$) and agriculture (wild blueberry production) also did not affect their occupation of a site ($P = 0.874$). It is just as likely to find the yellow-banded bumble bee in a wild blueberry field as any other landscape type. However, we found that a site is more likely to be occupied if there is a high bumble bee richness (other species of bumble bees at the site) ($\chi^2_{(1)} = 26.268$, $P < 0.0001$) and the site is less likely to be occupied if the impatient bumble bee is present at the site ($\chi^2_{(1)} = 17.556$, $P < 0.0001$). Figure 4 shows how bumble bee richness and the presence of the impatient bumble bee affect the presence of the yellow-banded bumble bee at a site.

Fig. 4. The effect of increasing bumble bee richness and the presence of the impatient bumble bee on the likelihood that the yellow-banded bumble bee will be present at a site. Blue line is in the absence of the impatient bumble bee and the red line is in the presence of the impatient bumble bee.

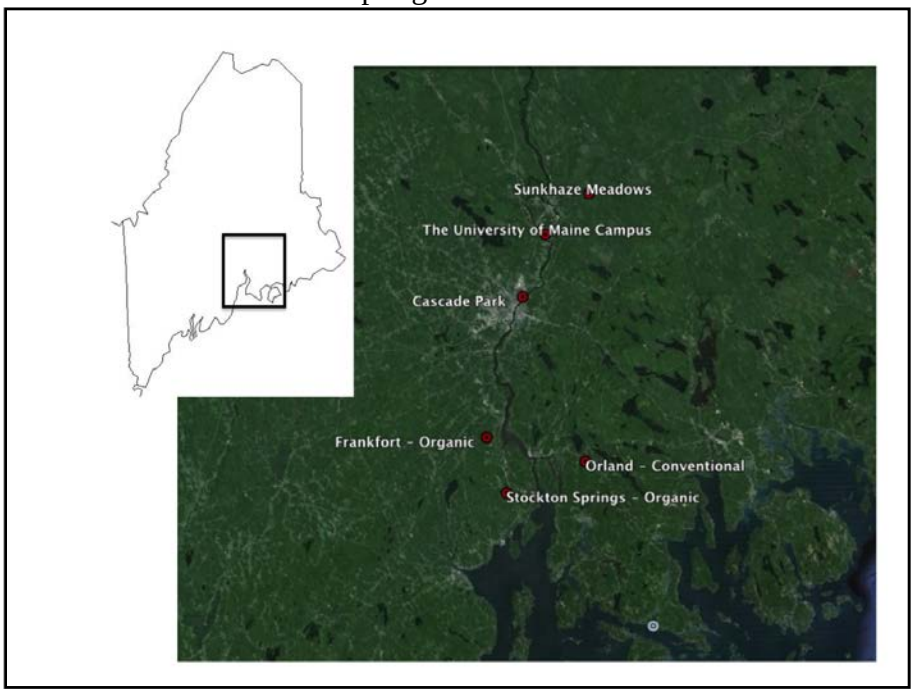
CONCLUSIONS: It is nice to see a bumble bee that was in decline to be increasing throughout coastal Maine. The increase of this species may be greater than we have presented with our data. This is because the chance of detecting the yellow-banded bumble bee in a single site visit is actually quite low (30%, detection in only 3 of 10 visits). It is also reassuring that its presence does not appear to be affected by wild blueberry production. However, all may not be perfect as we did identify that the impatient bumble bee is increasing in abundance and is associated (not

necessarily causing its absence from a site) with the absence of the yellow-banded bumble bee from a site. Only time will tell how the dynamics play out.

Study 13. Bumble bee species distributions and phenology in mid-coast and Downeast Maine Report from Kalyn Bickerman-Martens (Ph.D. candidate) and Dr. Frank Drummond

METHODS: We completed an intensive bumble bee survey of various sites around mid-coast and Downeast Maine between 8 Jul and 7 Oct 2015. During this time, 18 sites were sampled at least once and of those sites, six were sampled 8 or 9 times on a biweekly basis (Fig. 1). The six sites were comprised of two organic wild blueberry fields, one conventional wild blueberry field, the University of Maine Campus, a park in the urban center of Bangor, and a national wildlife refuge. Total time spent collecting at each site varied depended on the number of field technicians but was always a combined effort of 1.5 hours during which as many bees were captured as possible. Along with the date and site, the flower (if known) that the bee was foraging on when captured was documented.
Samples were taken back to the lab, frozen at -20°C, and then identified to species. Unknown and difficult to identify specimens were mailed to a collaborator at the University of Vermont for species identification. The abdomens of the specimens will be dissected to look for macro parasites (conopid fly larvae, phorid fly larvae, mermithid nematodes) and the gut tissue removed to visually examine for *Nosema bombi* spores on a phase contrast microscope at 400x magnification. Any macro parasites found will be preserved in 70% ETOH and all dissected bee specimens will be preserved at -80°C.
Data collected in the summer of 2015 will be compared to those data collected in the summers of 2012, 2013, and 2014.

Fig. 1. Sites of intensive bumblebee sampling in 2015.



RESULTS: A total of 1,876 bumblebees were caught in 2015. Of those, 1,514 were captured at the six sites that were sampled intensively. The breakdown of bumblebee species relative abundances differed quite a bit by site (Fig. 2).
Bombus ternarius (the orange-belted bumble bee) was found at all six sites (BRIGHT RED), but had the highest relative abundance (89%) in the heavily forested Sunkhaze Meadows in Milford. *B. ternarius* made up approximately half of all species caught in the organic blueberry fields in Stockton Springs and Frankfort as well as the conventional field in Orland. The more urban centers of Bangor and Orono; however, were dominated by *B. impatiens* (BRIGHT BLUE - the common eastern bumble bee), making up around half of all bees caught on the University of Maine Campus and 72% of all bees caught in Bangor. *B. vagans* (GREEN – the half black bumble bee) and *B. bimaculatus* (MINT – the two spotted bumble bee) were present at all sites. The species that is thought to be in decline throughout its range, *B. terricola* (PURPLE – the yellow-banded bumble bee), was found at nearly all sites sampled except for the organic field in Frankfort and Sunkhaze Meadows. *Bombus rufocinctus* (ORANGE - the red-belted bumble bee) was only collected at one site, Cascade Park in Bangor. Notably, this is also the first recorded collection of *B. rufocinctus* from 2012 through this summer for the project. Also, *B. fernaldae* (Fernald's cuckoo bumble bee) was only captured in the organic field in Stockton Springs.
We were also able to observe a phenological difference in each species, with some being more common earlier in the summer (and in some cases are the dominant species in one day's collections) and disappearing by August (*B. bimaculatus, B. perplexus, B. terricola*, and *B. rufocinctus*). Other species appear to build up in numbers through time and are present until first hard frost (*B. impatiens, B. ternarius*, and *B. vagans*). The number of unique confirmed species captured per collection day appears to decrease as the summer progresses (Fig. 3). This suggests that there is a higher diversity of bumble bee species earlier in the summer than later. There was a significant effect of date on the total number of species captured at all six fields ($P < 0.004$).
Bumble bee species diversity for each field was calculated as Shannon's Diversity Index (Table 1). The Shannon's Diversity Index (H) is a method that is used to characterize species diversity in a community and is based on the species richness (the number of species present) and species abundance (the number of individuals of each species). The higher the number, the higher the species diversity. We also calculated the species evenness of each field (E), which is a measure of how similar the abundances of each species are. The closer the number is to one, the more even the species abundance at that site. We see that the most diverse site the organic field in Stockton Springs with the highest H (1.39) and the least diverse site was Sunkhaze Meadows in Milford (0.49). Sunkhaze Meadows is also the least even site with a value of 0.27, which can be compared to the pie-chart in figure 2 where we see the site is heavily dominated by *B. ternarius*.

Fig. 2. Relative species abundance at each site and all sites in the summer of 2015.

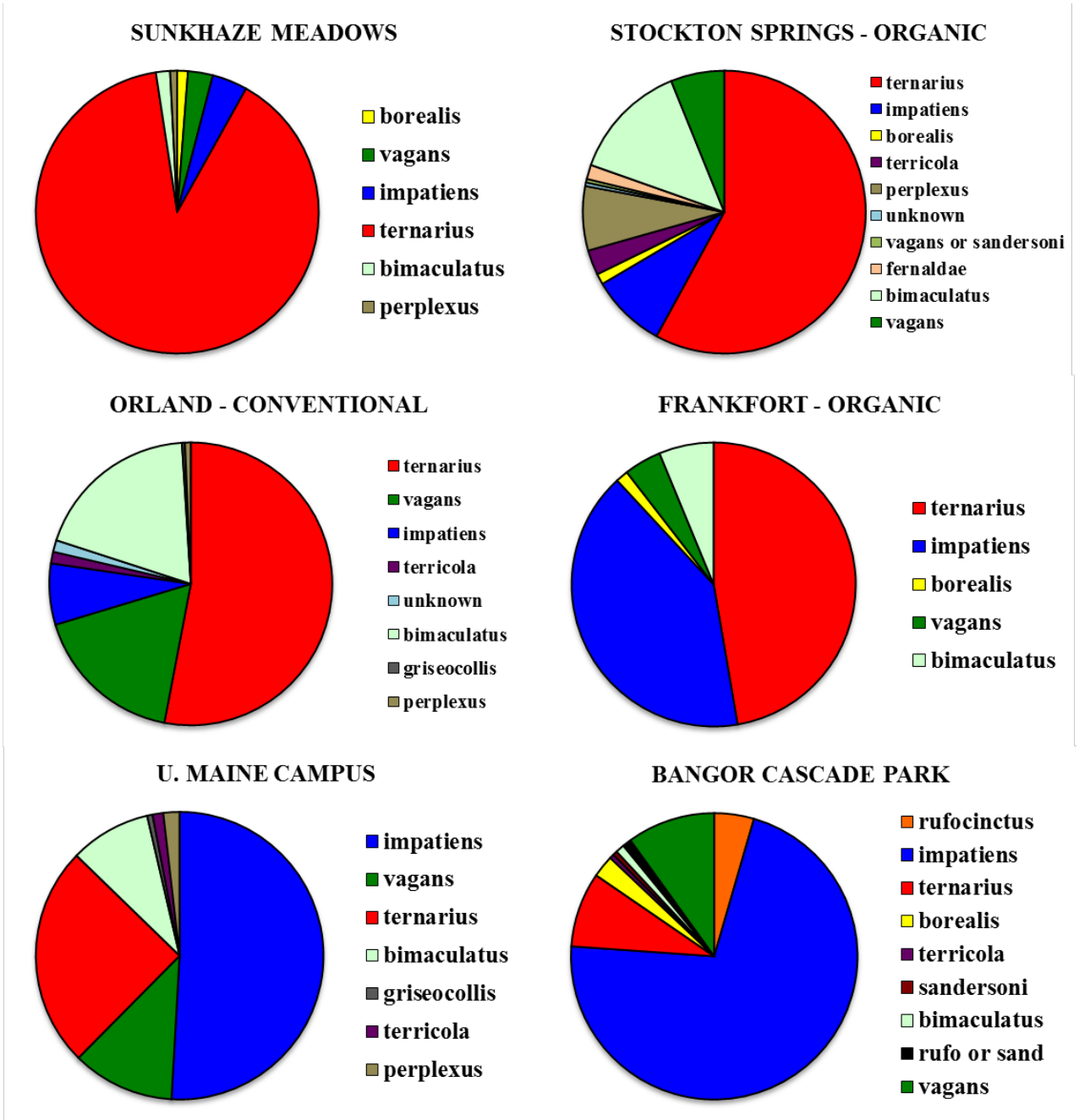


Fig. 3. The number of distinct species captured per day. There is a significant effect of date on the number of species caught ($P < 0.004$).

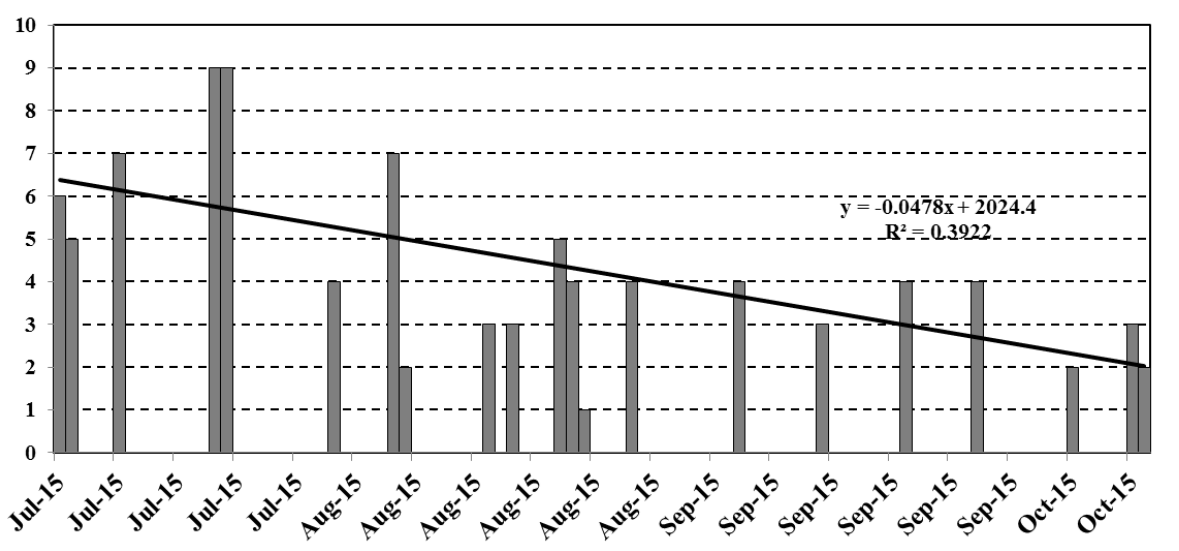


Table 1. Diversity of bumble bees at the six sampled sites in 2015.

Site	Shannon's Diversity Index (H)	Evenness
Bangor	1.00	0.48
UMaine Campus	1.31	0.68
Frankfort - Organic	1.09	0.68
Orland - Conventional	1.26	0.65
Stockton Springs - Organic	1.39	0.67
Sunkhaze Meadows	0.49	0.27

CONCLUSIONS: The six sites that were sampled intensively varied greatly in species diversity as well as species evenness. The heavily forested site in Milford, Sunkhaze Meadows National Wildlife Sanctuary, had the lowest species diversity and evenness and was predominately populated by *B. ternarius*. In contrast, the most diverse site was the organic wild blueberry field in Stockton Springs. We hope that by evaluating the percent land cover data with a 1 kilometer buffer around each site (the average maximum flight distance of a bumble bee), we can identify habitat characteristics that may predict the presence of a bumble bee species. The University of Maine campus and Bangor Cascade parks may both be considered "urban" and floral resources in these sites contain many ornamentals. In both sites the dominant species is *B. impatiens*.

In addition, many sites seemed to have fairly low relative abundances of species that had earlier emergence (*B. bimaculatus* and *B. perplexus*), but in many cases by the time we were collecting in mid-July, males of these species had already emerged. These indicate that the colonies were already in their reproductive part of their cycle while others were still building up their workers. The loss of these earlier emerging bees in later collections can be seen in the loss of diversity in our collections: fewer unique species were collected per day as the season progressed.

Study 14. <u>Andrenid bee nesting and abundance in wild blueberry</u>

Native bees often emerge just prior to wild blueberry bloom. Bumble bees and sand bees (Andrenidae) are among the most numerous pollinating field force present in blueberry fields during early bloom. In 2015 we assessed the density of sand bees in three fields in coastal Maine just at the onset of bloom. We also measured in 16 wild blueberry fields sand bee nesting presence and relative density in both pruned and burned fields. We took field ground temperatures to determine if burned fields had higher spring ground temperatures and thus might be more likely to harbor nesting sand bees that are in need of warm conditions in the spring for rearing young.

METHODS: Starting on 6 May, four wild blueberry fields located in Union, Waldoboro, Winterport and Cherryfield, ME were surveyed for sand bee foraging activity on the earliest clones. Bee foraging activity was measured by counting the number of adult female sand bees visiting blueberry flowers in a 1.0 m^2 plot within a clone for a 1 minute period. Seven to 10 plots were setup in any of the few clones that were flowering. During this period, less than 1% of the entire field was in bloom. The estimated fruit-set due to the foraging activity in the early clones was estimated using the formula: expected fruit-set = 14.5 + (17.7 * (# andrenids / m^2 / min)) (as reported in: Factsheet 629, Honeybees and Blueberry Pollination. Starting on 15 May, field surveys were initiated in 16 pruned wild blueberry fields (8 mowed and 8 burned) to estimate the prevalence and relative abundance of nesting sand bees. In each field, three, 100m long transects were walked in search of ground nesting sand bee aggregations. If aggregations were found they were ranked low or high depending upon nest density. Less than 100 nests in all three transects were ranked low relative abundance and greater than 100 nests were ranked high. In twenty-one fields (10 mowed and 11 burned) ground temperatures were taken using an infra-red soil temperature scanning gun. Mowed and burned fields were paired so that measurements were taken at the same time of day (within 10-15 minutes of each other). Five replicated temperature measures were taken in each field. Ordinal logistic regression was used to compare prevalence and abundance of sand bee nesting in relation to the pruning type. ANOVA, RCB was used to compare the ground temperatures among mowed and burned fields.

RESULTS: In 2015, sand bees were fairly abundant throughout much of Maine's growing regions. In fact, several growers called in to report the presence of these bees nesting in their fields. Early season foraging on the first blooming clones and the expected resulting fruit-set is shown in Figure 1. Bee foraging activity ranged from 2.6 sand bees/m^2/min down to 0.2 sand bees/m^2/min. Expected fruit-set reflected the sand bee foraging abundance and ranged from 56.4% to 18.0%. Soil ground surface temperatures were slightly warmer ($F_{(1,19)} = 2.812$, $P = 0.109$) in burned fields, 94.4 ºF, versus 84.1ºF in mowed fields (Fig. 2). One of the reasons, I believe, that burned field ground surface temperatures were not much greater in warmth than mowed fields is that bare spots, as well as blueberry covered ground, may have both been included in the replicated measurements as it is difficult to always know the target area that the infra-red gun uses for measurement. It appears that the warmer soil temperatures might be a potential factor, in addition to soil composition, to the likelihood of sand bees nesting in a wild blueberry field. Figure 3 shows that greater prevalence of sand bee aggregations are in burned compared to mowed fields (mowed = 12.5% versus 100% of burned fields having aggregations). There was also a significant difference in abundance of sand bee nests between mowed fields

and burned fields ($\chi^2_{(1)}$ = 15.724, $P < 0.0001$). The burned fields also contained higher densities of sand bee nest aggregations.

Fig. 1. Sand bee (Andrenidae) foraging activity and calculated expected fruit-set in four wild blueberry fields in 2015.

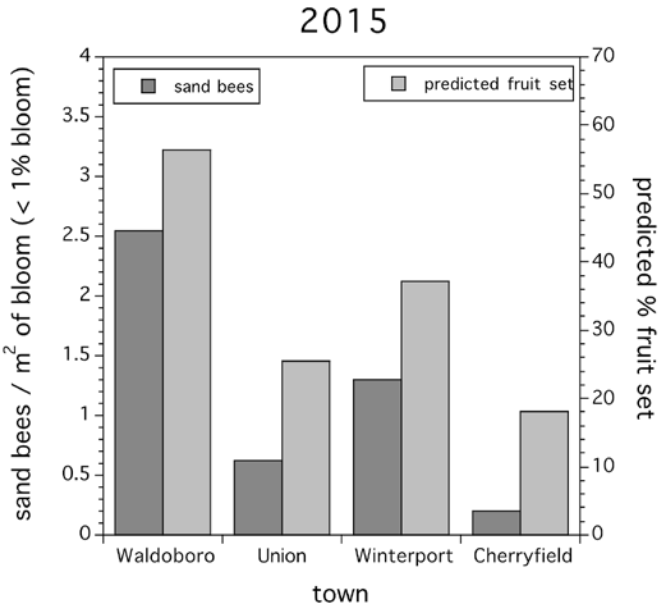


Fig. 2. Average ground surface temperatures in mowed versus burned wild blueberry fields.

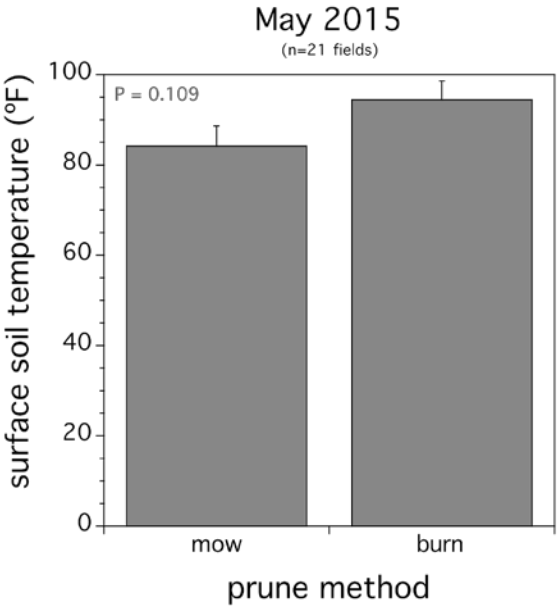
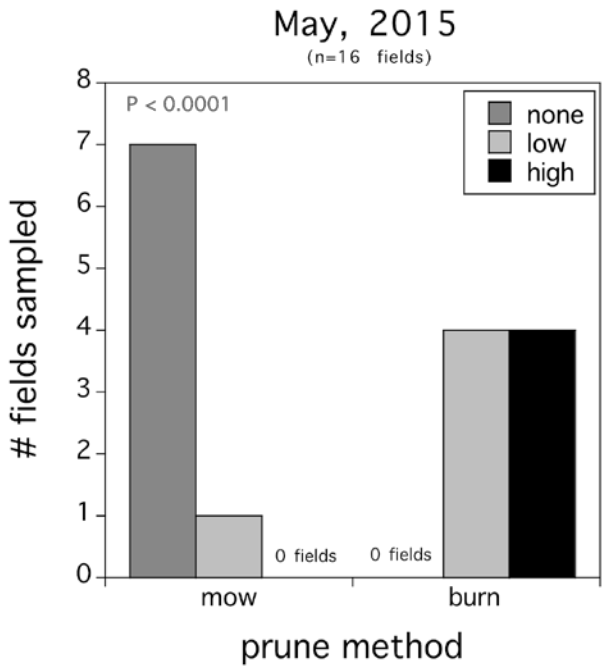


Fig. 3. Sand bee relative nesting density (ranked categories) in mowed versus burned fields.



CONCLUSIONS: Sand bees are important pollinators. Previous studies of mine have shown that they are highly efficient pollinators and that they are well synchronized with wild blueberry bloom. It was shown in 2015 that they forage heavily on the first blooming clones, well before honeybees are placed in most fields and that they tend to nest predominantly in burned fields. Soil ground temperature may be a factor that is correlated to their prevalence in burned fields. Other factors that affect their nesting locations would most likely floral resources that bloom prior to blueberry bloom and a sandy well-drained soil. It has also been observed that sand bees like bare ground areas to nest in and so the decrease in burning fields compared to mowing as a technique of pruning should not lead to the demise of these important bees.

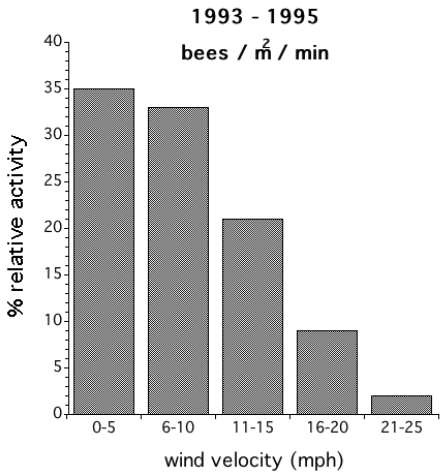
Study 15. Potential effects of climate change on pollination in wild blueberry

Climate change is not only predicted to result in a rise in sea level, increase in air temperatures, lengthening of the growing season, increased extreme weather events, but also increase precipitation, especially along the coast. We used a wild blueberry plant growth model to estimate the potential effects of climate change on bee foraging activity. Bee foraging activity has already been shown by us to be related directly to fruit-set.

METHODS: A degree-day model developed and validated in Nova Scotia was used to estimate the beginning of bloom and the end of bloom, between which is the bloom "window". This bloom window was estimated using historical maximum and minimum air temperatures to calculate cumulative degree-days. In this study, we used air temperatures from Blue Hill, Maine from 1960 to 2015 (55 yrs). However, bees are not actively foraging on all days. In order to predict the number of pollination days during the bloom window, we subtracted days that were

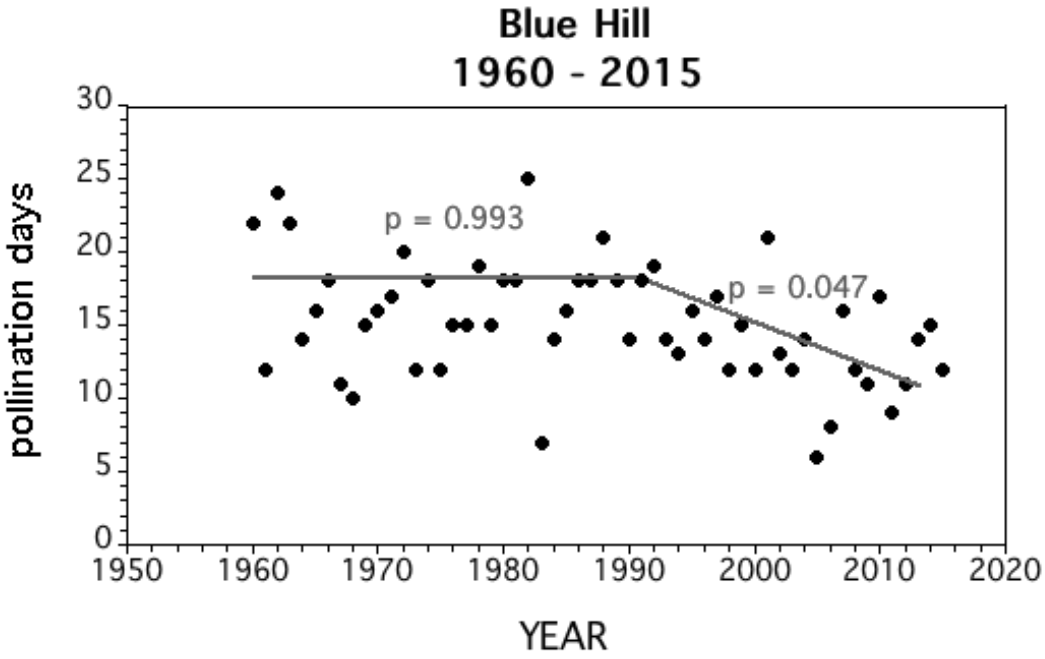
below 40ºF, or had 1 inch or more rain, or had winds of > 20 mph (Fig. 1.). This yields the number of days during bloom that bees pollinated flowers or "pollination days".

Fig. 1. Data collected on bee activity in wild blueberry during varying wind conditions between 1993 and 1995.



RESULTS: Our estimate of pollination days for Blue Hill between 1960 and 2015 shows that there was no significant change ($P = 0.993$) in the average number of pollination days from 1960 until 1990 (30 years). On any given year the number of pollination days is estimated from the models to range from 7 to 25 days. Starting in the early 1990s average pollination days appear to have declined at a linear rate ($P = 0.047$) through to 2015. The major cause of this trend is the increase in rainy springs during bloom. Therefore, it can be stated that climate change is already happening and wild blueberry growers are facing fewer days for pollinators to visit flowers in their fields then 30 years ago. This has great implications on the bee populations necessary during bloom for adequate levels of pollination. However, even during the decline period there have been years with very good pollination windows.

Fig. 2. The number of estimated pollination days in the Blue Hill Region from 1960 to 2015.



CONCLUSIONS: This study is a modeling exercise and only presents EXPECTED or PREDICTED modeled pollination days. The degree-day model is not entirely accurate in Maine and so there is error in the predictions. In addition, our estimation of days that are not suitable for bees are currently calculated from daily estimates of weather conditions. An estimate based upon hourly conditions should be much more accurate. In the near future we will be improving these estimates with more accurate models that will be validated over the next few years.

Study 16. <u>Long-term annual fluctuations of a wild bee community in Maine wild blueberry</u>

The wild bee community provides either total pollination services to some wild blueberry fields in Maine or contributes to the pollination that honeybees provide. We have investigated how abundances of wild bees vary from location to location and we are now in the process of making a web-based tool for growers to use that will make predictions of bee abundance around their field. BUT, the question remains how bee communities might fluctuate from year to year. We have an answer to this question by conducting annual bee sampling in a wild blueberry field in Winterport, Maine.

METHODS: Starting in 1989, the bee community was sampled annually in a wild blueberry field in Winterport, Maine. At peak bloom, twenty m^2 quadrats were observed for 1 minute. The quadrats were approached slowly and the observer (Dr. Drummond) waited 1 minute without moving and then proceeded to count the number of bees in and entering the quadrats for 1 minute. This sampling was conducted 2-3 times during peak bloom and only performed on sunny days between the times of 10 AM and 2 PM when air temperatures were > 60ºF and winds were < 15 mph. Bees were recorded as belonging to the following groups: *Bombus* (bumble

bees), Megachilidae (leafcutting or mason bees), Andrenidae (sand or digger bees), and Halictidae and others (sweat bees, cellophane bees, yellow face bees, and others). Bee density was converted to a per acre basis for graphical analysis. The estimated fruit-set contribution by the bee community was calculated using the formula we have derived for estimating fruit-set in wild blueberry based upon native wild bees per m^2 per minute. Several years are not represented by samples. The years 1998, 2001, and 2005 were missed because of conflicts with other research projects. In 2009 commercial management of the field was commenced after its cessation in 1990. Because the entire field was put on the same cycle, 2010, 2012, and 2014 were not sampled because they were prune years.

RESULTS: The time series of bee community fluctuations are shown in Figure 1. Inspection of the figure shows that bee communities fluctuate greatly from year to year. This is important when considering relying solely on the native bee community for pollination needs. Some years will result in much better pollination than others (Fig. 2). An attempt at modeling the fluctuations in total bee numbers resulted in a significant predictor model, but with poor overall predictability (Fig. 3). The model only explains 7.3% of the variation in bee numbers over time. The significant factors are the numbers of bees the previous year ($P = 0.11$) and a random stochastic random walk factor ($P = 0.0009$). Modeling the individual groups of bees did not result in any better predictions. What the statistical model suggests is that the dynamics of native bees are driven by density dependence factors such as disease or competition for nesting sites or flowers, but also fluctuations from year to year are a result of random events such as weather, the stronger effect. This is what we would have assumed to begin with, but it is important to show that these factors are in fact both driving bee numbers from year to year.

Fig. 1. Bee group annual fluctuations in a Winterport wild blueberry field, 1989-2015 († denotes beginning of commercial management).

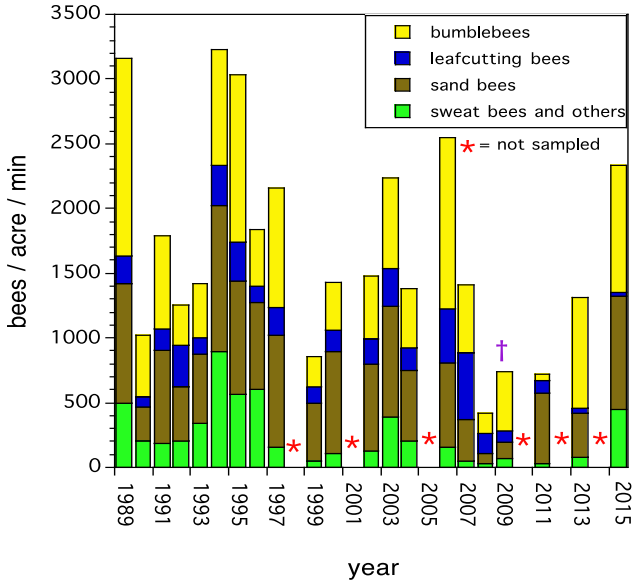


Fig. 2. Predicted fruit-set in the Winterport wild blueberry field as a function of total native bee community over time.

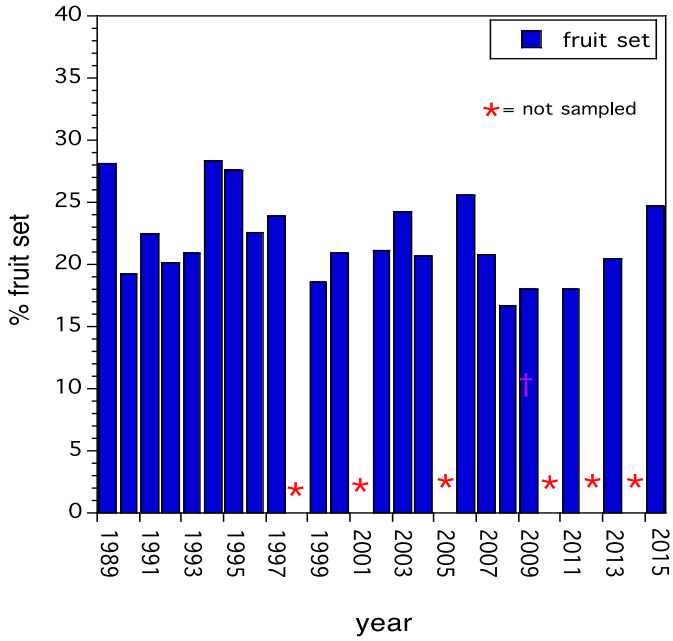
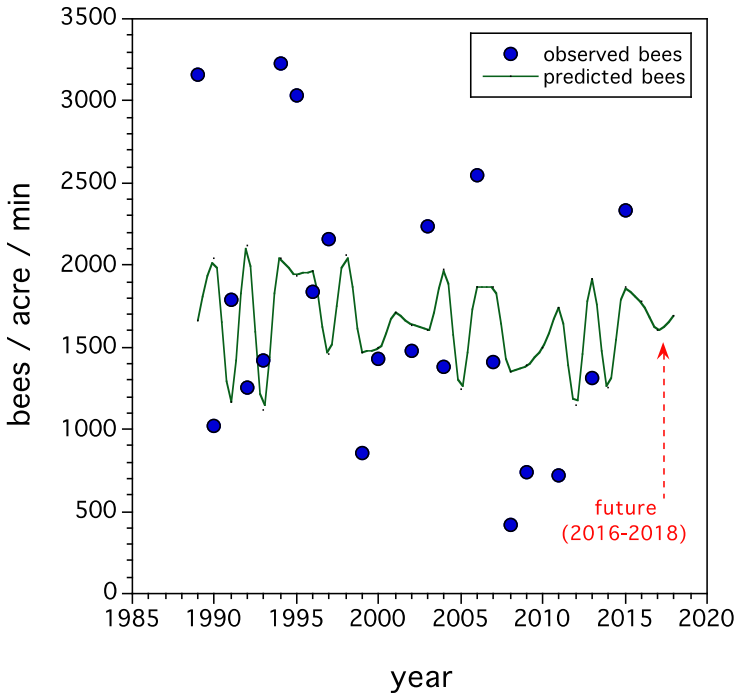


Fig. 3. Observed total bee densities and predicted total bee densities derived from an ARMA (autoregressive moving average) time series model.



CONCLUSIONS: Native bees are an important resource for wild blueberry growers. Some growers depend upon them entirely for pollination and other growers benefit from their supplemental pollination to the service that honeybees provide. Some wild blueberry fields will

have higher numbers of wild bees in their fields year to year based upon the surrounding landscape of a wild blueberry field. This study shows that although a given field may have high potential for native bee communities in their fields, the number of bees fluctuates from year to year and that this fluctuation might affect annual pollination levels and appears to be driven by density-dependent factors such as disease and competition, but also driven by random events such as weather.

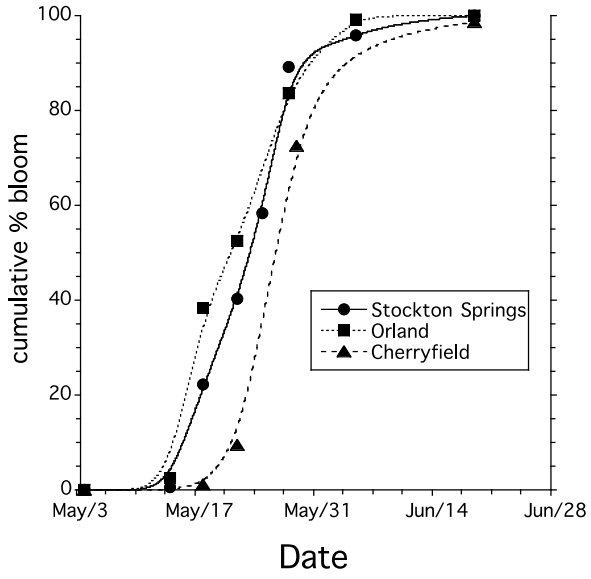
Study 17. Predicting the bloom period in Maine wild blueberry

An accurate bloom model for wild blueberry in Maine will be helpful for the development of a simulation model to assess pollination strategies and also investigate changes in the climate. Currently, a degree-day model exists for Nova Scotia, but it is not dependable because it assumes that the soil does not freeze between January and March. We initiated an extensive data collection process in 2015 to develop a more appropriate model for Maine under conditions where plants do not begin to develop until at least March.

METHODS: In 2015 we visited 18 wild blueberry fields in the mid-coast (11 fields) and Downeast (7 fields) growing regions of Maine. At each visit we collected 10 stems and counted the number of open flower buds and calculated the proportion of flowers in the field in bloom. To develop a degree-day model for Maine we collected daily maximum and minimum air temperatures from local weather stations in the vicinity of each field. The weather data collected started in January 2015 and ended in July 2015. Using this data the number of degree-days was calculated for threshold base temperatures from 32-50ºF (every 2 degrees) and for 55 and 60ºF using the formula: degree-days = (average daily air temperature – threshold base temperature), where average temperature is: [(maximum air temperature + minimum air temperature) / 2]. Using this information we determined the precision of each degree-day (DD) model base for estimating three endpoints: initial bloom (1%), mid-bloom (50%), and full bloom (100%). The measure of precision used was the standard error to mean DD ratio.

RESULTS: Figure 1 shows the bloom progression in three of the 18 fields that we sampled as an example of the temporal pattern that we are attempting to model. It can be seen that the Downeast field in Cherryfield is behind the Orland and Stockton Springs field in the progression of bloom.

Fig. 1. The progression of bloom in three wild blueberry fields in Maine, 2015.



Weather stations were not deployed at every field and so some locations such as the blueberry barrens could only be modeled by using two weather stations to predict bloom in several fields. When the degree-days were calculated for each day at each field they were summed from March 1 and the accumulations at dates where 1%, 50%, and 100% bloom were observed were recorded. The precision of each prediction for each base temperature was calculated and is shown in Figure 2. This graph shows that the base temperature most likely to provide the best prediction is no larger than 45ºF. Figure 3 displays the degree-day model (degree-days necessary) for 1%, 50%, and 100% bloom when a base of 45ºF is used.

Fig. 2. Precision, measured as the standard error as a proportion of the mean DD for 1%, 50%, and 100% bloom. The vertical dashed line demarcates a point of inflection where the precision declines rapidly and therefore suggests the base temperature for the degree day model (45ºF).

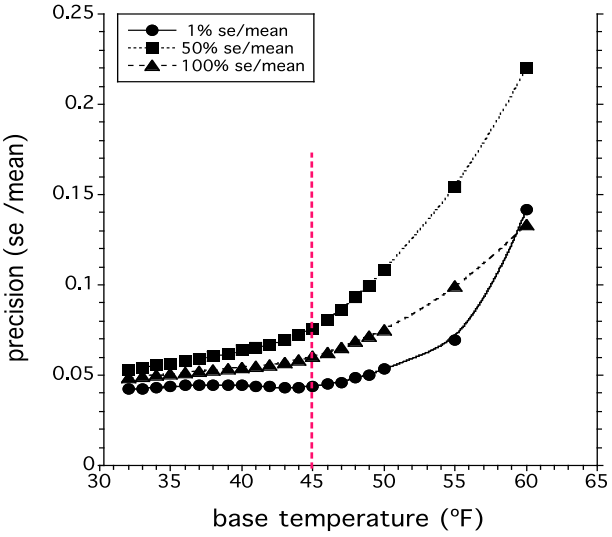
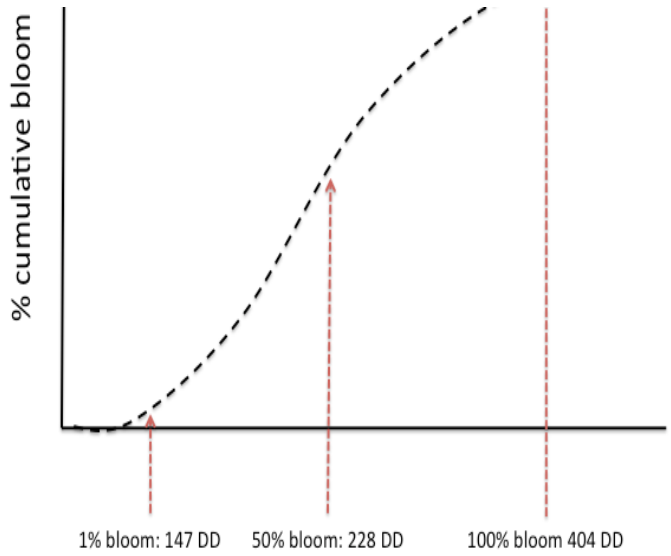


Fig. 3. The degree-day model for 1%, 50%, and 100% bloom depicts (vertical arrows) the degree-day accumulations (base = 45ºF) for the three phases of bloom.



CONCLUSIONS: A degree-day model for wild blueberry bloom in Maine is suggested based upon field sampling across the two major growing areas in Maine. This model is different than a previous model developed in Nova Scotia since air temperatures are not accumulated until March. In addition, a 45ºF base is used instead of the 50ºF base used for Nova Scotia. Validation of this model will be conducted in 2016 by sampling another set of wild blueberry fields to estimate the progression of bloom.

Study 18. Reproductive biology of wild blueberry

Wild blueberry plants are tetraploid (i.e. four sets of chromosomes). This results in several very unique characteristics regarding their reproductive biology. Studies in 2015 on blueberry pollination involved bee foraging and fruit-set relationships that have now resulted in a database of 140 fields over 8 years (reported on elsewhere). This report describes several experiments conducted in 2015 that pertain to other aspects of wild blueberry pollination not directly pertaining to bee visitation. We repeated a previously conducted experiment designed to ascertain the length of time that stigmas are receptive to viable pollen in the field. Length of bloom period was documented for three clones in a specific field in Winterport, ME. Another study measured the phenology of bloom across 16 fields in both Mid-coastal and Downeast, Maine (See Bio. Study 17 of this report). Three previous years of experiments have documented that self-incompatibility and outcrossing compatibility are expressed as gradients across the range of genotypes (clones) in fields. The ratio of plants that are self-compatible compared to those that are obligate out-crossers changes through the season with early season blooming genotypes being more likely to be self-compatible. This has important implications for pollination. The focus of the study in 2015 was to repeat experiments determining the phenology or self-compatibility to determine if this is a general phenomenon across many blueberry fields. The last experiment conducted in 2015 was a “chase” experiment. This experiment involved

four clones and either self-pollinating or cross-pollinating flowers in each clone sequentially over time to provide evidence if self-incompatibility is pre- or post-zygotic and if the phenomenon of priming (initial exposure to self-pollen breaking down the incompatibility system with subsequent self-pollen deposition) occurs in wild blueberry.

METHODS: Four studies were conducted in 2015. The first study involved determination of the length of stigma receptivity. A wild blueberry field in Winterport, ME was the experimental site. We selected five clones (genotypes) on 5 May and starting on 10 May we covered at least 10 stems with no open flowers with mesh bags to prevent pollination when flowers first opened. Starting on 15 May bags were checked daily and newly opened flowers were marked with a Sharpie® of a specific color on the calyx. Pollen was collected from five adjacent clones (not the selected target clones). Pollen was collected by cutting stems with open flowers and bringing them into the lab for 24 hrs. This allowed the flowers to dry overnight. During the next morning the flowers were vibrated with a tuning fork (440 Hz) over a petri dish and the pollen was collected. Pollen from all five clones was mixed and taken out to the field for hand pollination. At least five flowers from each clone of each age 1 to 10 days old were hand pollinated with a paintbrush with approximately 50-100 pollen tetrads. The mesh bags were left on the stems for 10 days after the last pollination (11 June). On 2 July all marked flowers were examined for fruit development indicated by swollen a calyx. The calyx ends were remarked and then examined again on 21 July for well-developed fruits and proportion of fruit-set from the initial number of hand pollinated flowers and the proportion of mature fruit from the initial number of flowers were calculated.

The second study measured cumulative bloom in three clones in the Winterport wild blueberry field in 2015. Starting on 8 May and continuing every few days throughout bloom the number of flowers per stem on 10 stems in each of three flagged clones were counted. The number of stems/m^2 was also measured for each clone. A cumulative curve was constructed from the data and the number of flowers available for bee visitation was calculated during peak bloom (40-60% cumulative bloom) by using the formula: Peak flowers / day / m^2 = (% * flowers / ha / 100) / (flower receptivity days) / 10^4, and integrating the value between 40 and 60% bloom.

The third study was designed to assess the proportion of self-compatible genotypes during early bloom (<10% or < 100 DD), mid or peak bloom (ca. 50% bloom), and then at the end of bloom (ca. 80% or > 700 DD). This study was conducted at the University of Maine Blueberry Hill Research Farm in Jonesboro, ME. During each of the three bloom periods, six clones were selected that had <50% of the flower buds opened. These clones were self-pollinated by hand with a paintbrush by selecting 3-5 stems each with several flower bud clusters in loose cluster. The stems were bagged and then 1-3 days later, the bag was removed and five newly opened flowers on each stem were marked on the calyx end with a Sharpie® ink marker and hand pollinated with a paintbrush. Fruit-set was assessed on 1-3 July. An analysis of variance (RCBD, year*field as a block) of fruit-set relative to clone bloom phenology was conducted on data collected in the same manner in 2009, 2010, 2011, and 2015. At this point only fruit-set has been analyzed. Fruit weight and seeds/berry data will be collected this winter and subsequently analyzed.

The fourth study conducted in 2015 was a "chase" study. The purpose of this study was to determine if the first deposition of pollen on a stigma, assuming a heavy deposition, will determine the fate of fruit-set (either poor fruit-set and abortion) or a high level of fruit-set and viable seeds; OR IF, the first pollen deposition "primes" or stimulates the plant to be less

discerning about pollen quality and result in high fruit-set. This hypothesis is based upon the unknown mechanism of whether self-incompatibility is a pre-zygotic messaging system or a post-zygotic physiological disruption system such as an abortive process. This study was conducted in a commercial wild blueberry field in Winterport, ME. Four clones were selected when the field was exhibiting 25% bloom. Stems with non-open flowers were bagged and monitored every 2-3 days for the presence of bloom. Bags were removed at bloom (flower age was two days) and hand pollinated with ca. 100 pollen tetrads. Sets of marked flowers (5-10) on each of 12 stems in each of the four clones were selfed OR outcrossed and then: 1) rebagged and not pollinated again, or 2) rebagged and then 1 or 4 days later were pollinated with either self or outcross pollen collected from random neighboring clones and then again rebagged. Two weeks after petal fall, on 25 June, fruit-set was estimated and bags were removed. On 10 September fruits were harvested so that this winter viable aborted seeds per berry, and berry size can be estimated.

RESULTS: The time series of proportion initial fruit-set and mature fruit from increasing aged flowers is shown in Figure 1 for both 2007 and 2015. The patterns were similar; although, in 2015 almost no fruit drop occurred after initial fruit-set from flowers five days old or older. The average viability of the stigma is about four days in both experiments. After a flower is four days old the stigma viability decreases rapidly with age. The mature fruit reflects berry load in relation to fruit drop and not stigma viability. Fruit drop is probably both a function of genotype and environmental stress.

Cumulative bloom in the Winterport field for three clones can be seen in Figure 2. The number of flowers per m^2 during bloom that can be visited (although not all stigmas have optimal viability) is estimated at 1,676. This means that if 50% fruit-set is to be obtained, at least 838 flowers.

Self-compatible clones are significantly more predominant during early bloom than in mid or late bloom (Fig. 3). There is much variation in clone (genotype) response. This is due to the fact that clone self-compatibility is not a yes or no characteristic, but is actually on a continuum and so overlap not only occurs among bloom periods but also close to 100% compatible clones are observed in early bloom but also during late bloom.

Fig. 1. Relationship between flower age (days after opening) and the proportion of flowers that set fruit and then retained fruit, 2007 (left) and 2015 (right).

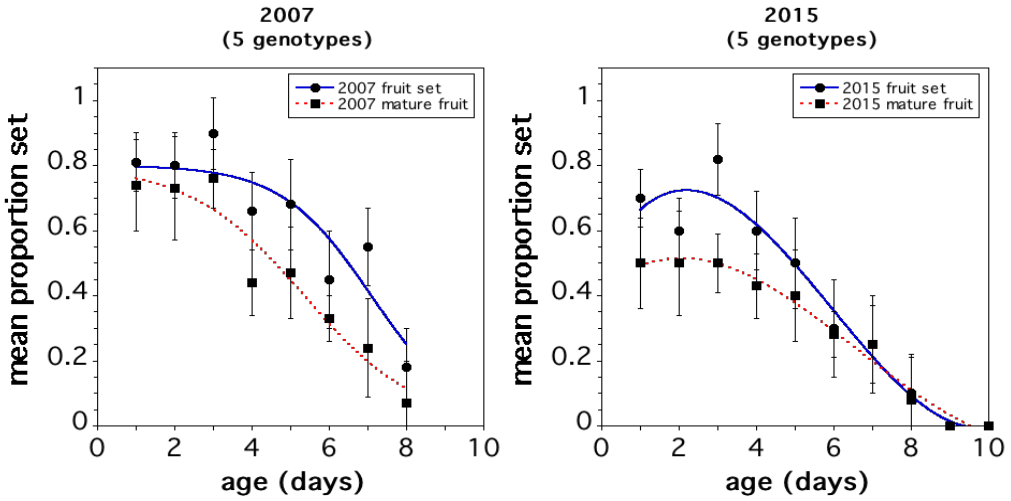
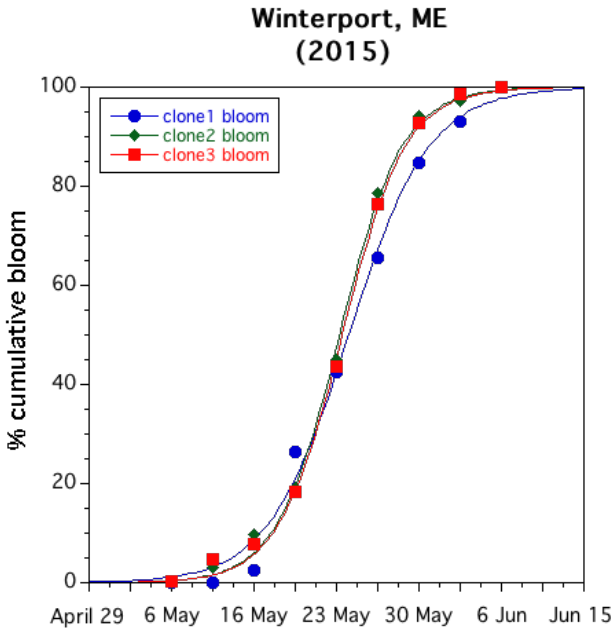
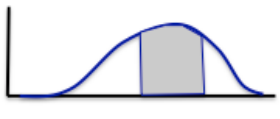


Fig. 2. The cumulative bloom of three clones in Winterport, ME and the number of flowers available for bee visitation during peak bloom. The number of flowers during peak bloom (between 40-60% cumulative bloom) was calculated using the formula below and integrating the area under the transformed curve depicted.





Peak flowers / m^2 = (% * flowers / ha / 100) / (flower receptivity days) / 10^4 = 1,676

Fig. 3. Results of four years of experiments assessing clone bloom phenology and the likelihood of the clone being self-compatible.

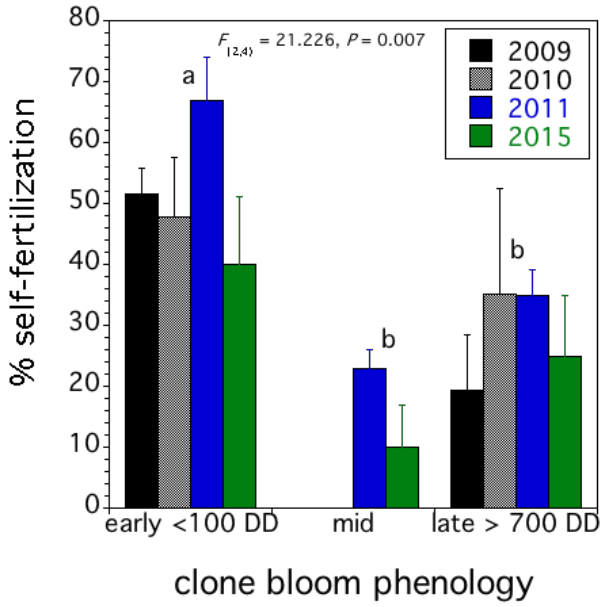
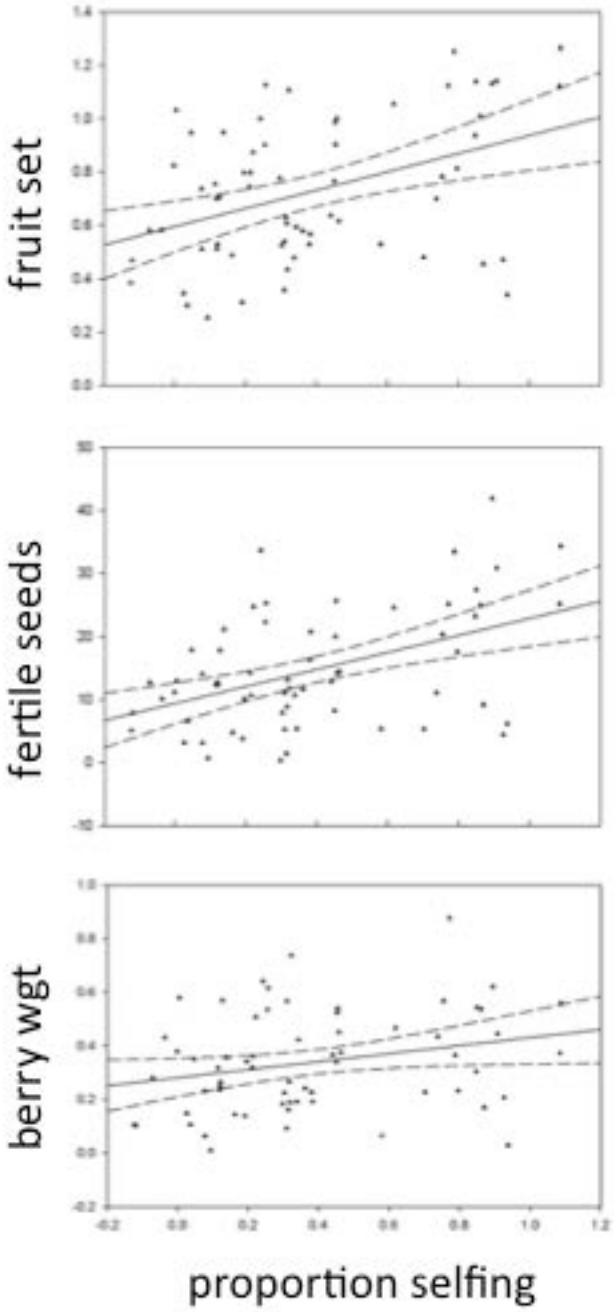


Figure 4 shows the results of a previous study of Dr. Dan Bell (Bell et al. 2010) that illustrates the greater fruit-set and yield of self-compatible clones. This relationship is not only because "selfers" can utilize their own pollen, but also because they are universal "mothers" and are compatible with many genotypes when outcrossed. This might be desirable for a grower, but it might not be beneficial for plant fitness if the progeny (seeds) resulting from self-fertilization are less viable.

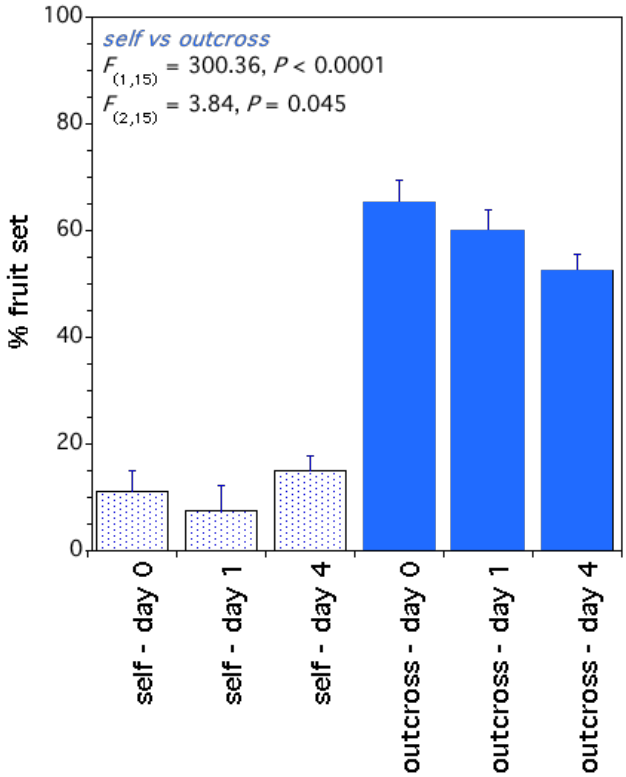
Fig. 4. The relationship between cross and self-compatibility represented as a gradient and fruit-set, fertile viable seeds per berry, and berry weight.



The results of the 2015 “chase” experiment show that clones randomly selected had low levels fruit-set when 2-day-old flowers were initially “selfed”, but significantly higher levels of fruit-set when initially cross pollinated (Fig. 5). Not seen in the averages plotted, but one clone was fairly self-compatible (38% fruit-set), but the average of all four resulted in a low average fruit-set. Subsequent pollination events on post-day 1 and 4 did not improve fruit-set in either the

continually selfed or outcrossed flowers (Fig 5). There also does not appear to be a priming effect that might be seen as an increase in fruit-set over time. It appears, although not significant, to be a negative sequential pollen effect on the outcrossed flowers. We are not sure why this is the case, but it might reflect a drop in stigma viability over time as the flowers aged. It is possible that we damaged the stigma surface with repeated hand pollinations and thus negatively affected germ tube growth (2-4 day process in wild blueberry). However, this may also not be a real trend, as we did not observe a similar pattern in the selfed flowers.

Fig. 5. Mean fruit-set in four clones either "selfed" or outcrossed sequentially in Winterport, 2015.



CONCLUSIONS: We conducted four experiments in 2015 about blueberry plant reproduction. The first experiment demonstrated that wild blueberry flower stigmas in two different years last up to 10 days, but that receptivity or viability declines quickly after four days. Using this stigma receptivity duration we also estimated from three clones that peak bloom in a typical blueberry field can produce 1,676 flowers for bee visitation. This of course will vary greatly from year to year due to a temperature dependent rate of bloom and the stem and bud/stem density per m^2 in a given field. We have completed four years of selfing experiments. We have not found significantly more selfing in early bloom when each year is analyzed, but when all four years' data is analyzed together, significantly more self-compatible clones appear to have an early phenology than later during the bloom period. The four years of experiments represented three different fields. So this does appear to be a general phenomenon. We have shown in other years that in the beginning of bloom bees aggregate to the few clones in bloom and so much higher levels of selfing must occur than later when the opportunity to move among clones is greater.

Therefore, one might suggest that a positive selection force might exist to bloom early if one is self-compatible so that all the bees can be "selfishly" attracted at least until other clones begin to bloom. However, this would be outweighed by the cost of suffering frost damage. The "chase" experiments are not really very informative because in reality lots of floral visits only result in few tetrads being deposited upon the stigma by bees (Drummond, unpublished data) and so the experiment discussed here is the extreme case and may not represent reality. It would be interesting to repeat this experiment with smaller amounts of pollen deposition and different combinations of sequential pollen depositions in regard to self and outcrossed pollen.

Study 19. Assessment of the mechanisms behind growth and development changes in Maine's wild blueberry induced by flower removal.

Report from Alex Bajcz (Ph.D. candidate) and Dr. Frank Drummond

This study has two components. The first is designed to assess if previously demonstrated changes in Maine wild blueberry growth and development brought about by a flower removal treatment can be altered by three subsequent treatments, each reflecting a possible underlying mechanism for why flower removal induces these changes. The second assesses the degree to which these changes are altered by variation in the level of flower removal.

METHODS: During the first weeks of May, 2015, 31 wild blueberry clones were selected at Blueberry Hill Farm in Jonesboro, ME. In 11 of these clones, ten 1/8m^2 plots were delineated, and in the other 20 clones, four plots of the same size were delineated. In all plots, all inflorescence buds on all stems were hand-counted. In six of the ten plots in the first set of clones, approximately 70% of the buds were removed from each stem via hand-pinching (hereafter "X" plots) while, in the other four plots (hereafter "C" plots), no buds were removed. In the other set of clones, 0% ("N" plots), 20% ("T" plots), 40% ("F plots"), and 60% ("S" plots) were removed, respectively, in the four plots in each clone. Because of inter- and intra-clonal differences in average numbers of inflorescence clusters per stem, actual removal rates per plot varied. In four of the ten plots in the first set of clones (two X plots and two C plots), no further manipulations were performed; these plots served as internal controls. The remaining six plots in each of these clones received one (and only one) additional manipulation: inflorescence removal that was biased by position on the stem (two X plots), foliar fertilization (one C plot and one X plot), or near-total defoliation (one X plot and one C plot). Each of these treatments was designed to interact with the effects of flower thinning by affecting thinned plots more or less than unthinned ones.

Several measures of plant performance will be compared across all plots; using mixed-effects regressions, we will identify treatments and/or treatment combinations that most strongly impact performance. Furthermore, in extent to which the effects of flower removal are linear or non-linear will be accessing using the second set of plots. The measures of plant performance surveyed will include:

Total vegetative, reproductive, and stem mass per plot, within and across collections.
Rates of reproductive failure at the stem, inflorescence, and individual flower levels.
Mass of reproductive structures per inflorescence.
Individual fruit size, shape, and mass.
Fruit ripening rate.

Total marketable fruit yield per unit area.
Leaf length and area, within and across collections.
Individual leaf and fruit wet and dry masses, as well as the wet:dry ratio thereof.
Canopy photosynthetic rate.
Canopy development rate as measured by leaf area index.
Ripe fruit titratable acidity and fruit pulp pH.
Fruit and leaf total soluble solids content.
Ripe fruit anthocyanin pigment content.
Leaf chlorophyll and carotenoid content.
Seed set and number of mature seeds per ripe fruit.
Leaf tissue nutrient concentrations.

RESULTS: In assessing the linearity of impacts stimulated by flower removal, several early trends in the data are apparent. For the most part, where flower removal's impacts were significant, the effects were decidedly non-linear with the level of flower removal. Instead, in many cases, there appeared to be a clear threshold of removal that stimulates significant growth and development changes, and above and below this threshold the exact level of removal is immaterial. For example, the total soluble solid content of ripe fruits at harvest (a proxy for sugar content) was substantially increased by any level of flower removal over 20% but was not increased any further by additional removal (Fig. 1). Similarly, the proportion of fruits that were ripe at harvest time (first week of August, Fig. 2) and the mass of vegetative tissue per stem at harvest (Fig. 3) were both significantly increased by removal levels of 40% or more. However, in both cases, 20% removal did not impact either measure, nor did 60% removal impact either measure more than 40% removal did.

Fig. 1. Average total soluble solids content, measured in % Brix as a proxy for sugar content, of random ripe wild blueberry fruits from plots subjected to a range of flower removal levels (0%, 20%, 40%, and 60%), line segments on bars are standard errors.

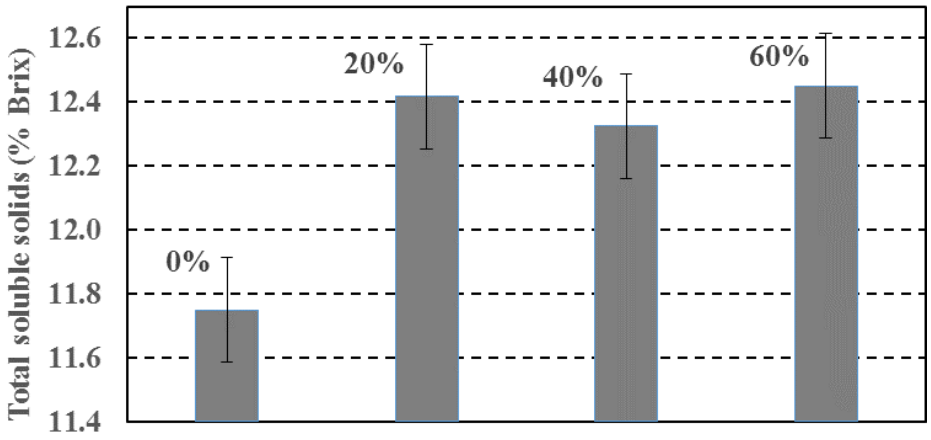


Fig. 2. Average proportion of fruits that were fully ripe at harvest from plots subjected to a range of flower removal levels (0%, 20%, 40%, and 60%), line segments on bars are standard errors.

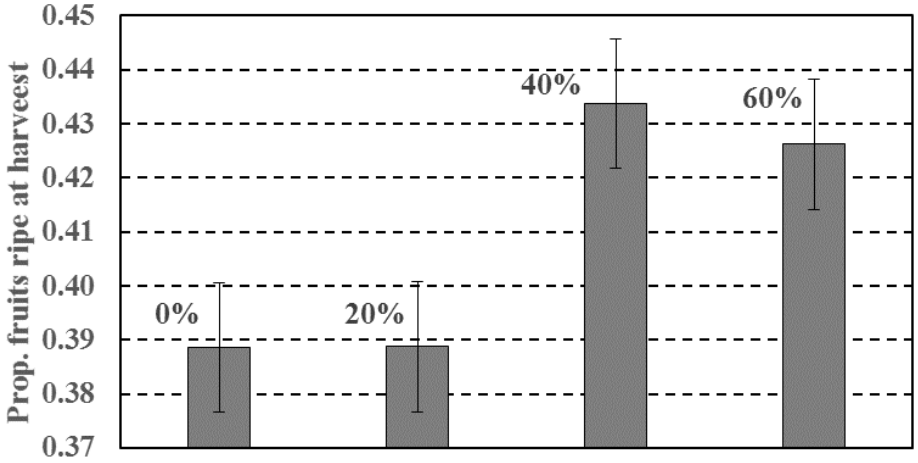
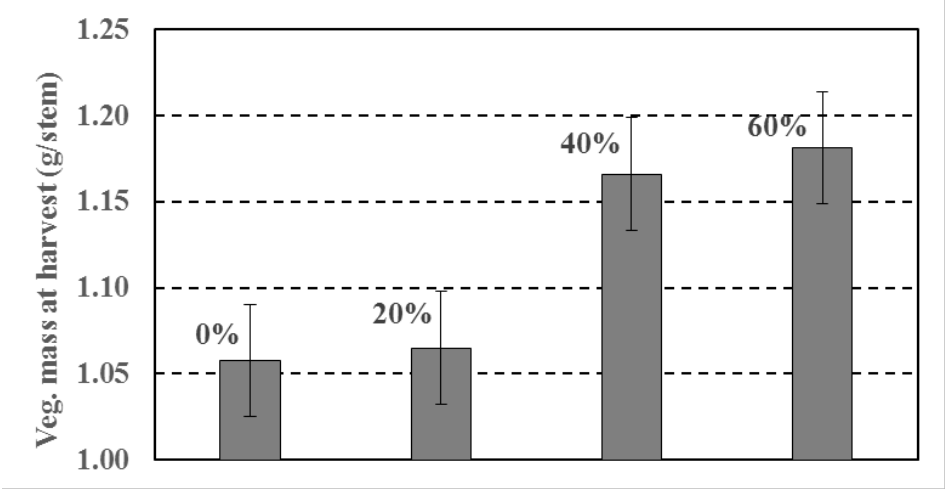


Fig. 3. Average vegetative mass (new leaves and new stems) per stem at harvest time from plots subjected to a range of flower removal levels (0%, 20%, 40%, and 60%).



CONCLUSIONS: Despite the popular conception that flowers are cheap to produce and maintain and that removal of flowers should thus prove inconsequential, flower removal has consistently been found to induce significant growth and development changes. However, it is highly debated why this might be so. Currently, there are nine popular hypotheses in the literature. Of these, three are thought to be the most comprehensive, *i.e.* able to explain both vegetative and reproductive changes. These are: 1) the (short-term) resource limitation hypothesis; 2) the spatio-temporal dominance hypothesis; and 3) the compound interest effect hypothesis. Each hypothesis basically suggests that flower thinning acts by "correcting" some underlying biological "problem" that limits growth and development because of implicit and unavoidable trade-offs related to resource allocation.

The short-term resource limitation hypothesis contends that nutrient mobilization within plants is slow and that stunting and abortion of structures can occur even when the resource deficiencies responsible are transient and localized rather than systemic. By removing some proportion of

structures, flower removal may make these resource deficiencies less likely to occur in remaining structures. Because nitrogen is the most crucial plant nutrient for growth and development, foliar fertilization with nitrogen should also reduce nitrogen deficiencies, mimicking the action of flower removal and making it less "beneficial." This hypothesis is supported when fertilized plots, both thinned and non-thinned, perform better than their unfertilized counterparts but with a greater margin of improvement for non-thinned plots than for thinned ones.

The spatio-temporal dominance hypothesis argues that stunting and abortion of structures can occur when resources are distributed based on a dominance/access hierarchy rather than based on need. Hierarchies arise when structures in certain locations on the plant (e.g. those nearer to the stem or those insulated from frosts) have better access to resources and thus command a greater share of them largely for that reason or when structures that happen to develop first or more quickly take a greater share of total resources simply because they have first access.

Flower removal may reduce the level of antagonism between structures for resource access to a point at which even subordinate structures can get sufficient resource access. If dominance is related to location on the stem (top-most or bottom-most reproductive clusters are dominant), removal of mostly subordinate inflorescences will reduce antagonism much less than removal of dominant inflorescences would, and in which case one direction of biased thinning would outperform the other.

The compound interest effect hypothesis stipulates that all structures, vegetative and reproductive, initially develop each season from the same stored resource pool, and that these two types of structures compete for these resources antagonistically. That is, if reproductive structures take more resources and develop more quickly, vegetative structures will take fewer resources and develop more slowly, and vice versa. However, eventually, reproductive structures will become dependent on vegetative ones for their resource supply as stored resources dwindle. As a result, rapid early reproductive development can stunt later reproductive maturation by impeding early vegetative development. Flower removal can help prevent this by shifting early development back in favor of vegetative structures. If a large fraction of vegetative structures are removed around the time point when reproductive structures become dependent on them, this action should hurt both thinned and non-thinned plots, but it should hurt non-thinned plots to a significantly greater degree.

Lastly, it is unclear whether growth and development changes stimulated by flower removal are linear or based on thresholds. Is a certain level of flower removal necessary before significant changes begin or can be detected? Or is the extent of stimulated change commensurate with the amount flower removal?

While results from this year's work are still pending, results from last year suggest that there is very little evidence to suggest that the short-term nutrient limitation hypothesis operates in wild blueberry, at least in terms of nitrogen. Results from last year also indicated that if the compound interest effect hypothesis has merit for blueberry, its impact is limited to affecting the developmental characteristics of individual fruits rather than larger, more systemic characteristics. Lastly, the spatio-temporal dominance hypothesis appears to have significant merit; plots that underwent flower removal in a biased fashion consistently performed at the highest levels of any plot types under study. However, the hypothesis may need refinement because it is unable to explain why both top- and bottom-focused inflorescence removal would be equally beneficial for plants.

It is clear that the changes induced by flower removal are not linear, and that any beneficial growth and development changes induced by flower removal materialize only if a critical

number of flowers are removed. If any less than this amount is removed, no effect will be observed. Furthermore, if more flowers than this critical number are removed, the results suggest no additional benefit would be generated.
While these results are preliminary, they indicate trends with substantial management implications. First, they suggest that blueberry plants do not respond to flower loss in a progressive or continuous manner. Rather, their responses might fairly be described as "either/or," with either a dramatic response or no response, depending on the extent of removal. This might mean that, for some traits, almost any level of flower loss or damage is likely to stimulate changes, whether they are desirable or not, while for other traits, small levels of flower loss may result in no discernable plant response at all. It may also mean that, for some traits, the level of desirable change to growth or development may be unattainable with any level of flower removal because the level of change does not apparently increase beyond a critical level of removal. Lastly, and more positively, it may also mean that the amount of removal needed to stimulate a desirable change could be fairly accurately pinpointed through more targeted research and be broadly applicable across genotypes, assuming environmental factors are appropriately considered.

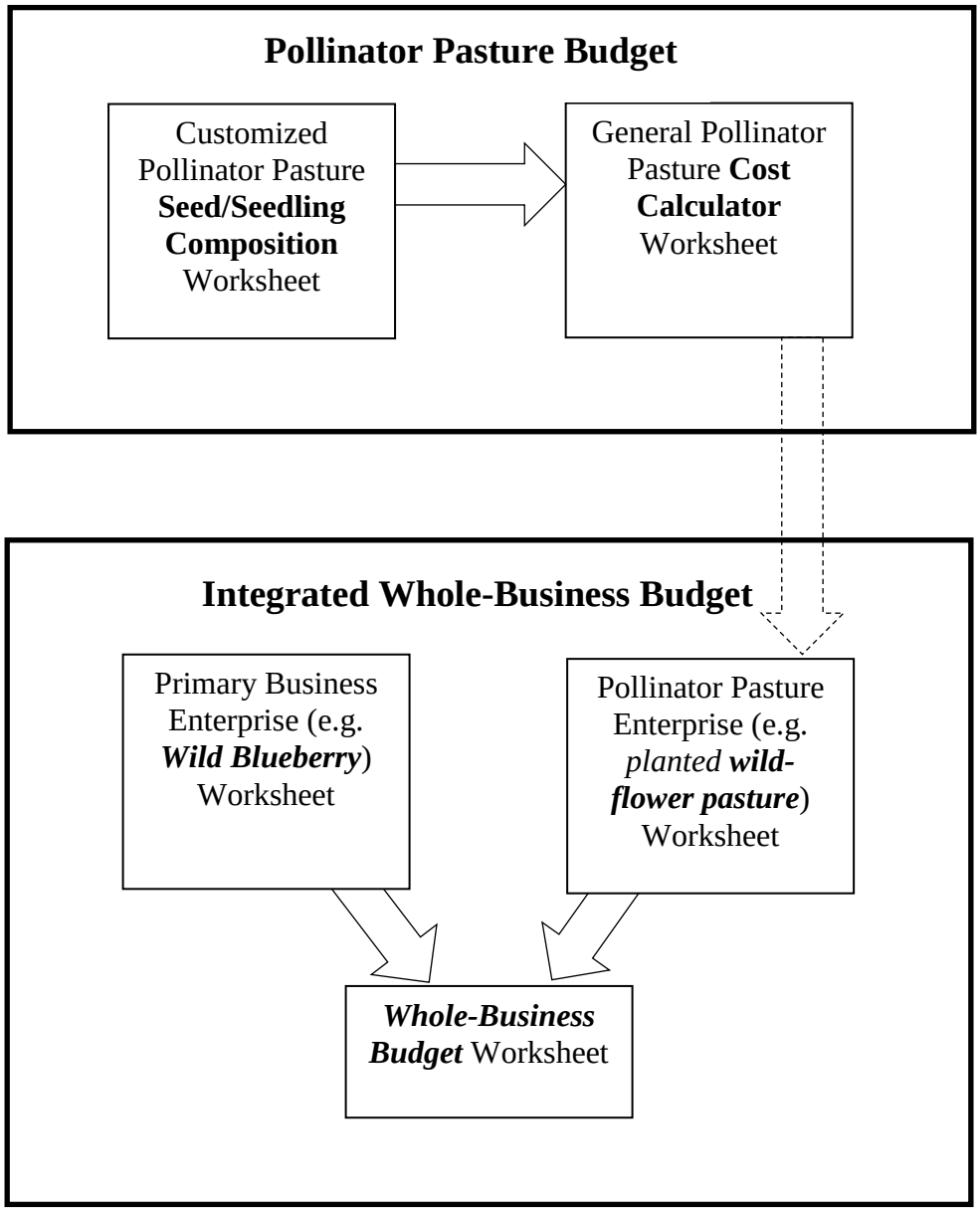
Study 20. Pollinator pasture budget calculator
Report from Dr. Aaron Hoshide, Dr. Cathy Neal (Univ. of New Hampshire) and Dr. Sam Hanes

As rented honey bee hives may become more expensive due to Colony Collapse Disorder (CCD) and other causes of over-winter mortality and as some wild pollinator species have declined, agricultural producers managing pollinator dependent crops have implemented pollination alternatives to hive rental. These have included other rented pollinators such as *Bombus* quads, but have recently focused on improving floral and nesting resource availability for native pollinators. In the Northeast, early agricultural adopters of alternatives to renting honey bees have included commercial wild blueberry, cranberry, apple, squash and pumpkin producers as well as other diversified farmers managing orchard and cane crops, highbush blueberries, squash and pumpkin, cucurbits, tomatoes and peppers, and pollinator-dependent vegetable seed production. In addition to farmers, citizens, businesses, as well as government and other entities have also been improving pollinator habitat for invertebrate conservation. Improving floral resources for native pollinators have generally involved two approaches. The first more common approach is to allow natural re-generation of perennial flowers such as asters, goldenrod, and milkweed by restricting mowing to the late fall. This approach can also involve rotational mowing to extend bloom of pasture clovers. This approach ($200 per acre per year) is about one-third to one-sixth the cost of actively planting pollinator pastures ($555 to $1,235 per acre per year). However, natural re-generation is dependent on the density and distribution of existing native floral species visited by native pollinators. This may require more extensive land base to support the same populations of native pollinators compared to an actively planted pollinator pasture. It may also be less suited for early-season pollinator species such as *Andrenid* bees which have completed their life cycle well before re-generational floral bloom. The second approach to improving floral resource availability for native pollinators is to actively plant perennial and annual species visited by pollinators. This includes planting perennial woody species such as male pussy willows as direct transplants, perennial herbaceous species such as asters, milkweed, and clovers, as well as annual species such as borage and poppies. While this

may provide floral resources that are denser and more suitable for specific pollinators, the additional expense ($355 to $1,035 per acre per year) of active bee pasture establishment may be more cost-prohibitive for farmers, homeowners, and businesses with extensive land base suitable for re-generation compared to those that are more land limited. In order to determine if actively planted pollinator pastures are feasible, it is important to calculate the costs of pollinator pastures with different species composition and establishment methods as well as to evaluate whole-business profitability when including such pastures (Fig. 1).

METHODS: A detailed budget for pollinator pasture planting was developed in collaboration with Dr. Cathy Neal at the University of New Hampshire (UNH) and is based on UNH research over the past 6 years. Model construction spanned September 2014 to October 2015. The budget model was beta tested with six entities at the New Hampshire Pollinator Summit held on November 2, 2015. The pollinator pasture model integrates two components: 1) a customized floral species composition and density worksheet and 2) a general pollinator pasture budget cost calculator. The floral species composition and density worksheet allows individual species specification, planting density, and seed or seedling cost. Any area of customized pollinator pasture can be modeled by specifying length and width dimensions of the planted area. The worksheet also allows any possible combination of planted areas established from either seed or seedling, as well as areas that include combinations of both seed and seedling establishment.

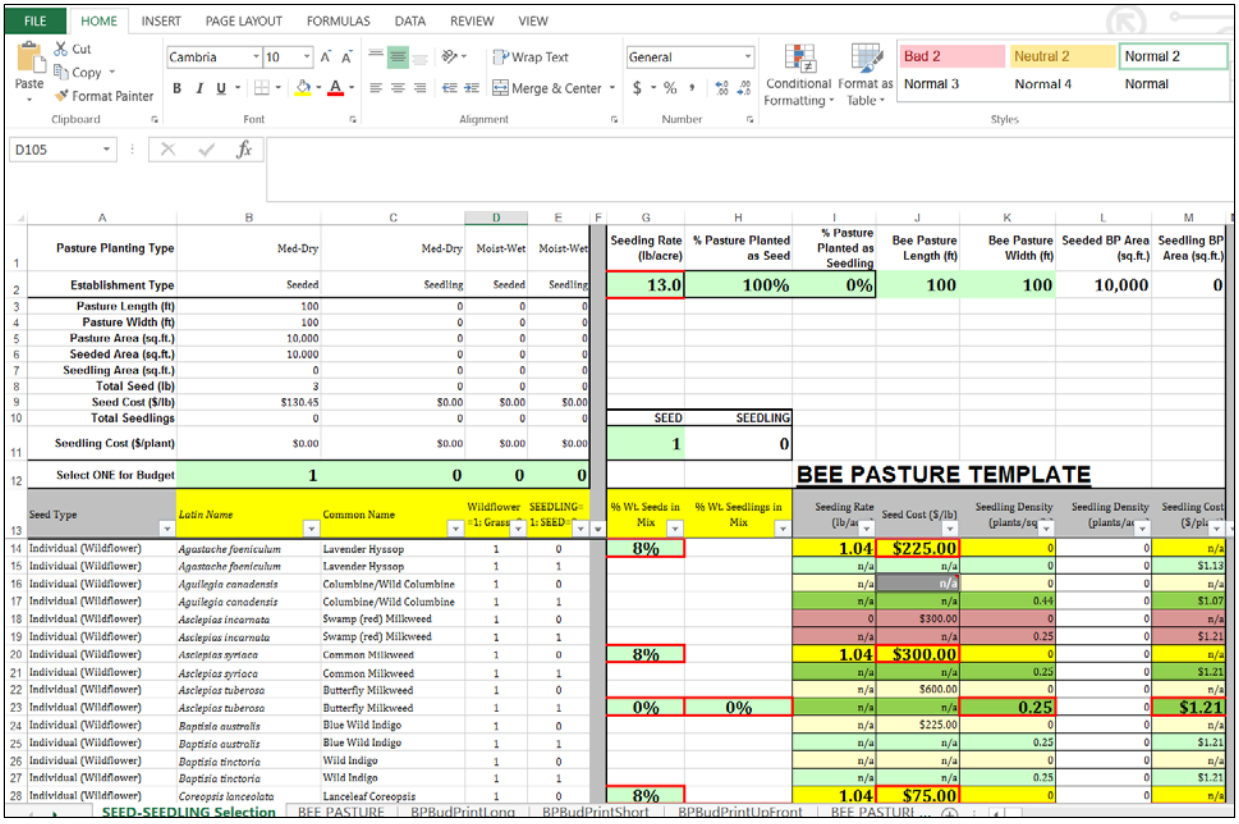
Fig. 1. Pollinator pasture budget and integrated whole-business models.



RESULTS: The customized pollinator pasture worksheet (Fig. 2) or default assumptions can be used by the general pasture budget (Fig. 3) by switching selection on or off between these two seed/seedling input data. The general pollinator pasture budget also uses a series of switches to specify the percent of the pre-defined pasture planted area established through smothering with plastic, tillage, or herbicides. The budget model also allows the user to model pollinator pasture establishment at different scales. For example, the percent of planted area using different sized equipment for tillage (roto-tiller, ATV tiller, tractor tiller) and mowing (push, riding, tractor rotary, tractor flail) can be specified. The model also allows herbicide control with Poast® using a backpack sprayer during the establishment year as well as establishment using a sod cutter. The general budget model calculates both variable (seed, labor, fuel, etc.) and fixed (building and

equipment depreciation, taxes, insurance) costs. Although this is a cost calculator, offsetting revenue sources can be specified from NRCS cost-share or other business enterprises. Cost estimates for both annual and start-up costs for pollinator pastures can be printed in standard budget formats. Unlike an annual budget, start-up costs include the total (rather than depreciated) cost of capital one can expect to invest during the establishment year. Smaller, more diversified perennial and annual crop producers may be interested in using this tool to estimate costs for installation of smaller, intensive pollinator pasture plots. Dr. Neal's research attributes successful pollinator pasture establishment and potentially greater pasture stand life to 1) fall seeding at higher seeding rates, 2) using transplants if spring planting without chemical herbicides, 3) post-emergence herbicide use for crab grass the year after establishment, and 4) regular hand weeding of invasive species. However, successful adoption of such actively planted pastures beyond the currently observed experimental phase by a few entities in the Northeast may also be dependent on better education on NRCS cost-sharing and alternative income streams from other pollinator-friendly enterprises such as cut flowers and herbs. Budget development in 2015 also involved collaborating with Dr. Sam Hanes to integrate a baseline wild blueberry enterprise budget with an enterprise budget for establishing pollinator floral pastures. This whole-farm budget combines the budget for 200 total acres of wild blueberries (100 acres fruiting in any given year) with the budget for any specified area of pollinator pasture. These budgets were also designed to print in short and long versions, with the long version specifying specific labor steps and any fuel use and costs associated with those steps, rather than just singular aggregated labor and fuel budget line items (Fig. 4). This representative budget model was then customized for three cooperating wild blueberry producers (small- and large-sized conventional and medium-sized organic) for beta testing the model during sit-down interviews scheduled with producers during June 2015. While the existing model uses a less complex enterprise budget for pollinator pastures than the one developed with UNH, it can utilize the cost estimates from this more complex pollinator pasture model.

Fig. 2. Customized pollinator pasture seed/seedling composition worksheet.



Pasture Planting Type	Med-Dry	Med-Dry	Moist-Wet	Moist-Wet
Establishment Type	Seeded	Seedling	Seeded	Seedling
Pasture Length (ft)	100	0	0	0
Pasture Width (ft)	100	0	0	0
Pasture Area (sq.ft.)	10,000	0	0	0
Seeded Area (sq.ft.)	10,000	0	0	0
Seedling Area (sq.ft.)	0	0	0	0
Total Seed (lb)	3	0	0	0
Seed Cost ($/lb)	$130.45	$0.00	$0.00	$0.00
Total Seedlings	0	0	0	0
Seedling Cost ($/plant)	$0.00	$0.00	$0.00	$0.00
Select ONE for Budget	1	0	0	0

Seeding Rate (lb/acre)	% Pasture Planted as Seed	% Pasture Planted as Seedling	Bee Pasture Length (ft)	Bee Pasture Width (ft)	Seeded BP Area (sq.ft.)	Seedling BP Area (sq.ft.)
13.0	100%	0%	100	100	10,000	0

SEED	SEEDLING
1	0

BEE PASTURE TEMPLATE

Seed Type	Latin Name	Common Name	Wildflower =1: Grass	SEEDLING= 1: SEED	% WL Seeds in Mix	% WL Seedlings in Mix	Seeding Rate (lb/ac	Seed Cost ($/lb)	Seedling Density (plants/sq	Seedling Density (plants/ac	Seedling Cost ($/pl
Individual (Wildflower)	*Agastache foeniculum*	Lavender Hyssop	1	0	8%		1.04	$225.00	0	0	n/a
Individual (Wildflower)	*Agastache foeniculum*	Lavender Hyssop	1	1			n/a	n/a	0	0	$1.13
Individual (Wildflower)	*Aquilegia canadensis*	Columbine/Wild Columbine	1	0			n/a	n/a	0	0	n/a
Individual (Wildflower)	*Aquilegia canadensis*	Columbine/Wild Columbine	1	1			n/a	n/a	0.44	0	$1.07
Individual (Wildflower)	*Asclepias incarnata*	Swamp (red) Milkweed	1	0			0	$300.00	0	0	n/a
Individual (Wildflower)	*Asclepias incarnata*	Swamp (red) Milkweed	1	1			n/a	n/a	0.25	0	$1.21
Individual (Wildflower)	*Asclepias syriaca*	Common Milkweed	1	0	8%		1.04	$300.00	0	0	n/a
Individual (Wildflower)	*Asclepias syriaca*	Common Milkweed	1	1			n/a	n/a	0.25	0	$1.21
Individual (Wildflower)	*Asclepias tuberosa*	Butterfly Milkweed	1	0			n/a	$600.00	0	0	n/a
Individual (Wildflower)	*Asclepias tuberosa*	Butterfly Milkweed	1	1	0%	0%	n/a	n/a	0.25	0	$1.21
Individual (Wildflower)	*Baptisia australis*	Blue Wild Indigo	1	0			n/a	$225.00	0	0	n/a
Individual (Wildflower)	*Baptisia australis*	Blue Wild Indigo	1	1			n/a	n/a	0.25	0	$1.21
Individual (Wildflower)	*Baptisia tinctoria*	Wild Indigo	1	0			n/a	n/a	0	0	n/a
Individual (Wildflower)	*Baptisia tinctoria*	Wild Indigo	1	1			n/a	n/a	0.25	0	$1.21
Individual (Wildflower)	*Coreopsis lanceolata*	Lanceleaf Coreopsis	1	0	8%		1.04	$75.00	0	0	n/a

Fig. 3. General pollinator pasture cost calculator worksheet.

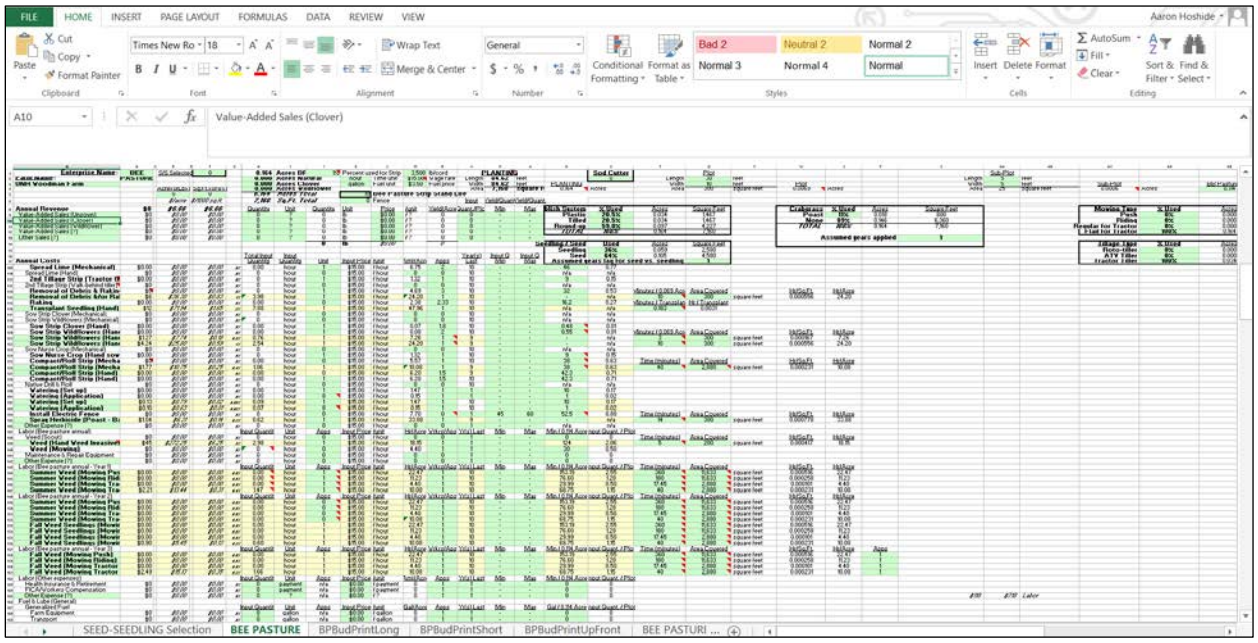


Fig. 4. Integrated whole-business budget for wild blueberry (100 acres) with planted wildflower pollinator pasture (3 acres) cost highlighted in light yellow.

	Total	Per Acre	Per Pound
Number of Acres	103	-	-
Wild Blueberry Yield (lbs)	340,415	3,305	-
Price ($/lb)	$0.73	-	-
Annual Revenue	**$240,935**	**$2,339.17**	**$0.73**
Annual Operating Expenses			
Fertilizer			
Granular	$7,954	$77.23	$0.02
Chemicals			
Fungicides	$1,650	$16.02	$0.005
Herbicides (Pre-Merge)	$11,295	$109.66	$0.03
Herbicides (Post-Merge)	$2,481	$24.09	$0.01
Herbicides (Wiping)	$154	$1.50	$0.0005
Herbicides (Bee Pasture)	$0	$0	$0
Insecticides	$942	$9.14	$0.003
Insect Traps	$73	$0.71	$0.0002
Pollination	$41,680	$404.66	$0.13
Sulfur & Other	$1,192	$11.57	$0.004
Lime	$135	$1.31	$0.0004
Seedlings	$0	$0	$0
Seed	$61	$0.60	$0.0002
Labor	$15,379	$149.31	$0.05
Labor (Bee Pasture)	$414	$4.02	$0.001
Diesel Fuel and Oil (Itemized)	$4,981	$48.36	$0.02
Diesel Fuel and Oil (Itemized - Bee Pasture)	$78	$0.76	$0.0002
Burning for Pruning	$7,000	$67.96	$0.02
Heating	$1,750	$16.99	$0.01
Heating (Bee Pasture)	$0	$0	$0
Maintenance and Upkeep	$8,329	$80.87	$0.03
Maintenance and Upkeep (Bee Pasture)	$3	$0.03	$0.00001
Managed Pollinator	$0	$0	$0
Miscellaneous			
Fire Permit	$20	$0.19	$0.0001
Rent or Lease (Electric Fence)	$2,000	$19.42	$0.01
Rent or Lease (Equipment)	$0	$0	$0
Rent or Lease (Tiller)	$0	$0	$0
Rent or Lease (Lime Spreader Push)	$0	$0	$0
Rent or Lease (Hand Roller)	$0	$0	$0
Rent or Lease (Land)	$0	$0	$0
Rent or Lease (Land - Bee Pasture)	$0	$0	$0
Replacement Teeth for Flail Mower	$167	$1.62	$0.001

	Total	Per Acre	Per Pound
Supplies	$500	$4.85	$0.002
Supplies (Bee Pasture)	$0	$0	$0
Soil & Tissue Testing	$0	$0	$0
Tax for Wild Blueberries	$2,479	$24.07	$0.01
Utilities	$600	$5.83	$0.002
Utilities (Bee Pasture)	$0	$0	$0
Interest	$963	$9.35	$0.003
Interest (Bee Pasture)	$0	$0	$0
Total Operating Expenses	**$112,281**	**$1,090.11**	**$0.34**
Total Overhead Expenses	**$4,595**	**$45.95**	**$0.01**
Annual Ownership Expenses			
Depreciation and Interest			
Buildings & Structures	$1,399	$13.59	$0.004
Buildings & Structures (Bee Pasture)	$17	$0.17	$0.00005
Excavation Equipment	$9,000	$87.38	$0.03
Drainage Tiles	$0	$0	$0
Burning Equipment	$540	$5.24	$0.002
Mowing Equipment	$1,075	$10.44	$0.003
Fertilization Equipment	$300	$2.91	$0.001
Spraying Equipment	$1,867	$18.12	$0.01
Wiping Equipment	$353	$3.43	$0.001
Irrigation	$0	$0	$0
Harvesting Equipment (Hand)	$0	$0	$0
Harvesting Equipment (Mechanical)	$2,700	$26.21	$0.01
Field Boxes	$800	$7.77	$0.002
Managed Pollinator	$0	$0	$0
Bee Pasture Equipment (Bee Pasture)	$602	$5.85	$0.002
Tools	$1,360	$13.20	$0.004
Tools (Bee Pasture)	$3	$0.03	$0.00001
Tractors & Vehicles	$10,608	$102.99	$0.03
Tractors & Vehicles (Bee Pasture)	$801	$7.78	$0.0024
Land	$23,884	$231.89	$0.07
Land (Bee Pasture)	$150	$1.46	$0.0005
Interest on Loans	$791	$7.68	$0.002
Interest on Loans (Bee Pasture)	$0	$0	$0
Insurance	$4,576	$44.42	$0.01
Insurance (Bee Pasture)	$36	$0.35	$0.0001
Taxes	$2,234	$21.69	$0.01
Taxes (Bee Pasture)	$45	$0.44	$0.0001
Total Ownership Expenses	**$63,142**	**$613.03**	**$0.19**
Total Annual Cost	**$180,018**	**$1,749.09**	**$0.54**

	Total	Per Acre	Per Pound
Net Farm Income (NFI)	**$60,916**	**$590.08**	**$0.18**
Return over Variable Cost (ROVC)	**$128,654**	**$1,249.06**	**$0.39**
Performance Measures			
Breakeven Revenue	$/hr	$/acre	$/lb
Long-run to Cover All Costs	-	$1,747.75	$0.54
Short-run to Cover Operating Costs	-	$1,090.11	$0.34
Return to Owner Labor			
Long-run Return to Owner Labor	$67.74	-	-
Short-run Return to Owner Labor	$143.07	-	-
Return to Total (Owner/Hired) Labor			
Long-run Return to Total Labor	$31.85	-	-
Short-run Return to Total Labor	$67.27	-	-

CONCLUSIONS: Both budget models can be improved in the future. The pollinator pasture cost calculator developed in collaboration with UNH can be improved in the following areas. A diversified organic mixed vegetable farmer in Connecticut has successfully established pollinator pastures on their farm using an Earthway® seeder to plant wildflower rows mulched once with grass clippings between rows to reduce annual weed competition. The current model does not allow for this establishment method. Herbicide application uses a back-pack sprayer but can be upgraded in the future to include tractor mounted or self-propelled sprayers of varying size. No-till planters have been used for establishing pollinator pastures over larger areas in other parts of the U.S. and this could be included in the calculator. The whole-farm budget model is currently being improved to include four different wild blueberry systems (organic, low input, medium input, and high input) and three scales of operation within each system for organic (5, 25, and 50 total acres) and conventional (25, 150, and 1,000 total acres). In the future, this integrated budget model could also be engineered to use the more complex pollinator pasture calculator developed with UNH rather than the simpler version currently used. Integrated budget models could also be developed for pollinator pastures coupled with other pollinator-dependent crops or other non-agricultural business enterprises.

RECOMMENDATIONS: Recommendations based upon our research findings will not be explicitly presented until many of our studies are complete. However, in the interim there are several specific observations that should be presented.

1.) Both native bee densities and honeybee densities are significant factors that determine fruit-set when assessed over a two-decade time frame. Because of this, native bees and honeybees should be protected and conserved in wild blueberry growing regions (see below). The levels of fruit-set that occur in wild blueberry fields are certainly predominantly due to bee densities, but are also dependent upon management of early season insect pests and diseases such as mummy berry. Other factors that explain variation in fruit-set are also factors not controlled by growers such as frost, rain events during bloom, and flowering plants located around a field periphery.

Yield is determined, on the other hand, by management of maturing fruit (set by bees). The production system (organic vs low, medium, and high input systems) affects soil fertility, prevalence of insect pests, weeds, and disease. Growers are certainly aware of these relationships, but with the quantitative measures that we now have, we will be assessing the economics of how the production system can alter these dynamics.

2.) The second observation is based upon the fact that both native bees and honeybees play a significant role in fruit-set. Therefore, an integrated strategy for pollination is recommended. The foundation of this strategy should be an evaluation of the native bee densities that are associated with a wild blueberry field. This can be assessed by sampling the native bee community during bloom as described by our video tutorial or when it is completed (in 2016) use of the internet tool, BeeMapper, for predicting the abundance of the native bee community in a blueberry field. Once the potential of native bee contribution toward fruit-set can be estimated, then decisions can be made to determine the number of honeybee colonies or commercial bumble bee colonies that need to be brought in to fields to provide desired levels of fruit-set. Another decision that can be made is to enhance native bee populations above the current carrying capacity of the surrounding landscape of a wild blueberry field. This can be performed through planting floral resources for bees. We do not have specific planting size requirements at this point, but we do know that even small plantings, the size of 0.1 acre can increase the native bee community and resulting fruit-set in wild blueberry fields. However, we are developing spreadsheets that can allow growers to estimate how large of a planting they can afford relative to their production system and budget. We have also shown that bees may respond to flowering plants along wild blueberry field edges (1 out of 2 years). Therefore, there is a benefit to having weedy flowering plants not in the field, but around the outside edge of blueberry fields.

3.) Protection of both native bee communities and imported honeybees and bumblebees in blueberry growing landscapes should be a priority. This can be achieved by minimizing exposure of bees to highly toxic insecticides. The wild blueberry Insect Control Guide factsheet (#209) can aid in selection of least toxic alternatives and use of sound IPM tactics and monitoring will lead to application of insecticides only when necessary. Our preliminary results suggest that simultaneous exposure to Assail and Orbit (for control of spring insect pests and mummy berry disease) may not be as harmful as we suspect. A detailed follow up study to be performed in 2016 on both honeybees and bumble bees should answer this question.

4.) Honeybees are the major pollinator (numerically) for the majority of current wild blueberry production (ca. 20 honeybees for every native bee). However, honeybee health is still a concern in the U.S. The wild blueberry industry in Maine imports almost 80,000 honeybee colonies during bloom. Our two-year study in Maine wild blueberry fields suggests that poor honeybee colony growth rates can be explained by *Varroa* mite parasitism and the disease agents that these parasites vector. However, we did observe pesticide residues in pollen from pesticides that are used in wild blueberry production. These residues were not correlated to low colony growth rates, but each additional stressor that honeybees are exposed to may result in additive effects toward decline of colonies. Therefore, growers should make an effort to minimize any factors that might be negative to honeybee colony survival. This includes not applying pesticides in fields in bloom when possible, selecting the least bee toxic pesticide for spring insect, disease, and weed management in BOTH crop and prune fields and when possible reducing the stocking density of honeybee colonies in fields by relying more upon commercial bumble bees or native bees. This last point is one that has not been researched, but a point made by many beekeepers that feel that their bees are starving in wild blueberry fields.

DISEASE MANAGEMENT

INVESTIGATORS: Seanna Annis, Associate Professor/Associate Extension Professor, School of Biology and Ecology
Rachael Martin, Research Assistant, School of Biology and Ecology
Tyler Case, MS Graduate Student, School of Biology and Ecology

6. TITLE: Research and control of mummy berry disease.

OBJECTIVE: Improve control of mummy berry and Botrytis blight through research and the deployment and operation of a disease forecasting system using weather stations.

METHODS: From April 23 to May 13, 2015, fourteen weather stations connected to the internet were deployed in blueberry growers' fields around Maine from Dresden Mills, Knox County to Wesley, northern Washington County (Fig. 1). Ten locations also had mummy berry plots that growers monitored through May. We contracted with Skybit to get virtual data for 10 locations where we also had real weather stations.
Data from the stations were used for the mummy berry forecasts starting in April and extending through May and then for Botrytis reports in May and June. The forecast reports were delivered in three ways: 1. in email messages sent out to an email list, 2. posted on the Wild Blueberry extension blog (http://umaine.edu/blueberries/blog/), and 3. recorded as answering machine messages. In September and October 2015, we put out new mummy berry plots for the next season in some grower fields and retrieved the weather stations for winter storage. Fields with weather stations were rated in May and September for disease.
In November 2015, we set up an incubator experiment to look at chilling hour requirements for pseudosclerotia germination. This experiment is being repeated from last year with a larger range of chill hours, moisture conditions and incubating temperatures post chill.

RESULTS: We had some hardware difficulties with the weather stations in the spring and so we were not able to get all of the weather stations working at the start of mummy berry season. We had numerous growers and members of the Blueberry Hill Research Farm who monitored mummy berry plots twice a week during the disease period. Throughout the disease risk season from early to late May, we were able to provide multiple forecast reports on mummy berry, as well as, the occurrence of frost for most of the blueberry growing areas. In May and June, we were able to provide some information on Botrytis blight risk to the growers. Ten out of the 14 stations had mummy berry plots, but at one site we did not get germination and at another we were not able to get the weather station out during the mummy berry season.
We ran into difficulty comparing the real and virtual data when we found the virtual data was estimated at 6ft of the ground and the real weather stations were taking measurements approximately 4 inches off of the ground. This produced a large discrepancy in temperature and length of leaf wetness. The virtual data is being recalculated and we hope to compare the two sets of data in the spring of 2016. We will compare virtual and real data for another year in 2016.

We had cooler conditions in April this year which delayed the start of mummy berry season until early May (for an example see Figure 2). The apothecia (cups) started to develop in late April, but most fields did not have susceptible plants until early May. The season was about two weeks in most areas with the last possible infection periods near to bloom about May 22nd. It was difficult to determine when the apothecia were gone this year since by May 16th most sites appeared to only have dried up apothecia but then on May 17th and 19th growers found more mature cups in some plots. Most growers reported using at least two applications of fungicides to control disease this year. Control was good in most fields with less than 5% disease, but some fields with inadequate control had up to 33% of stems with disease.
The incubator experiment on pseudosclerotia germination revealed that pseudosclerotia require at least 900 chill hours before germination and that longer chill hours require less post-chill hours for germination. Apothecia can persist from 2 to 4 days and many pseudosclerotia start germination but do not produce apothecia. This experiment is currently being repeated.

RECOMMENDATIONS: We recommend continuing to monitor conditions for mummy berry infection with the weather stations at this time. Weather stations will be set up at 15 locations next year with mummy berry plots at as many sites as possible with growers willing to monitor them. We will continue with the disease forecast and will improve the website for the weather stations by including a current prediction of infection for each monitored field. We will also continue our study comparing virtual and real weather station data to see if the virtual data is suitable as a substitute in the future.

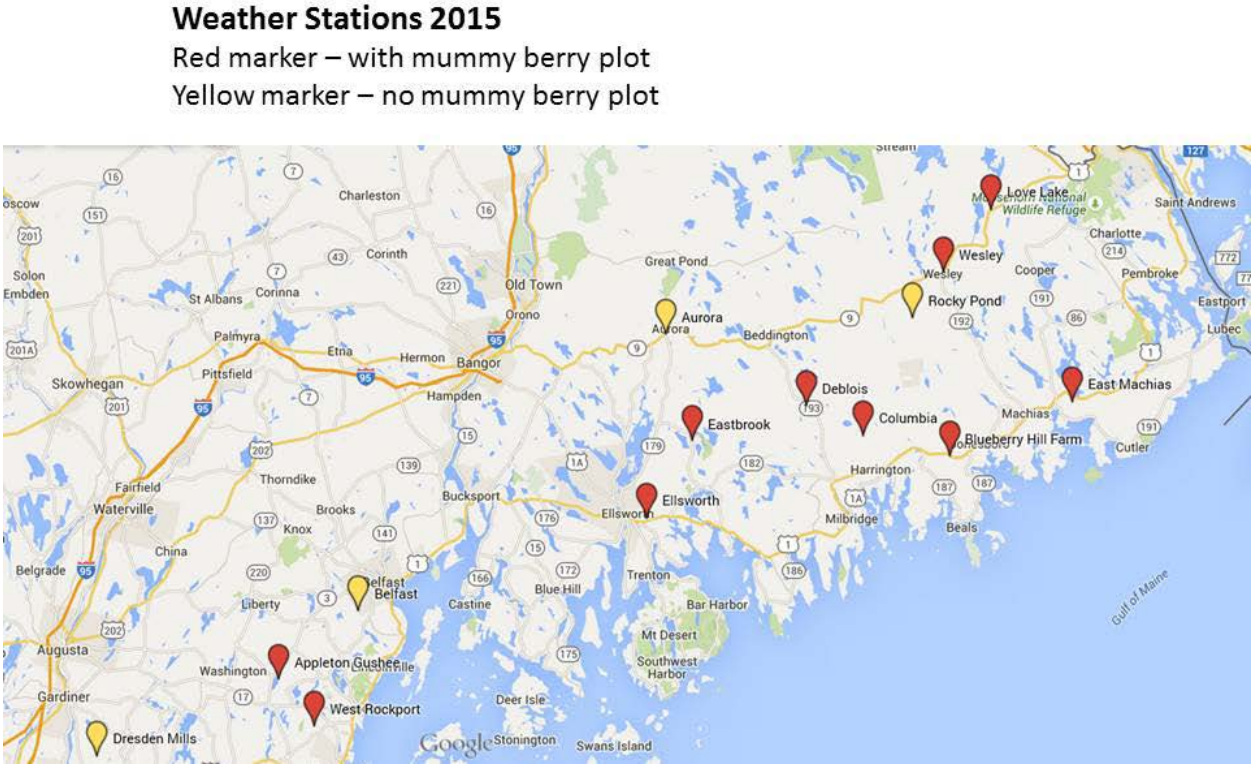


Figure 1. Locations of mummy berry forecast stations and mummy berry plots for 2015.

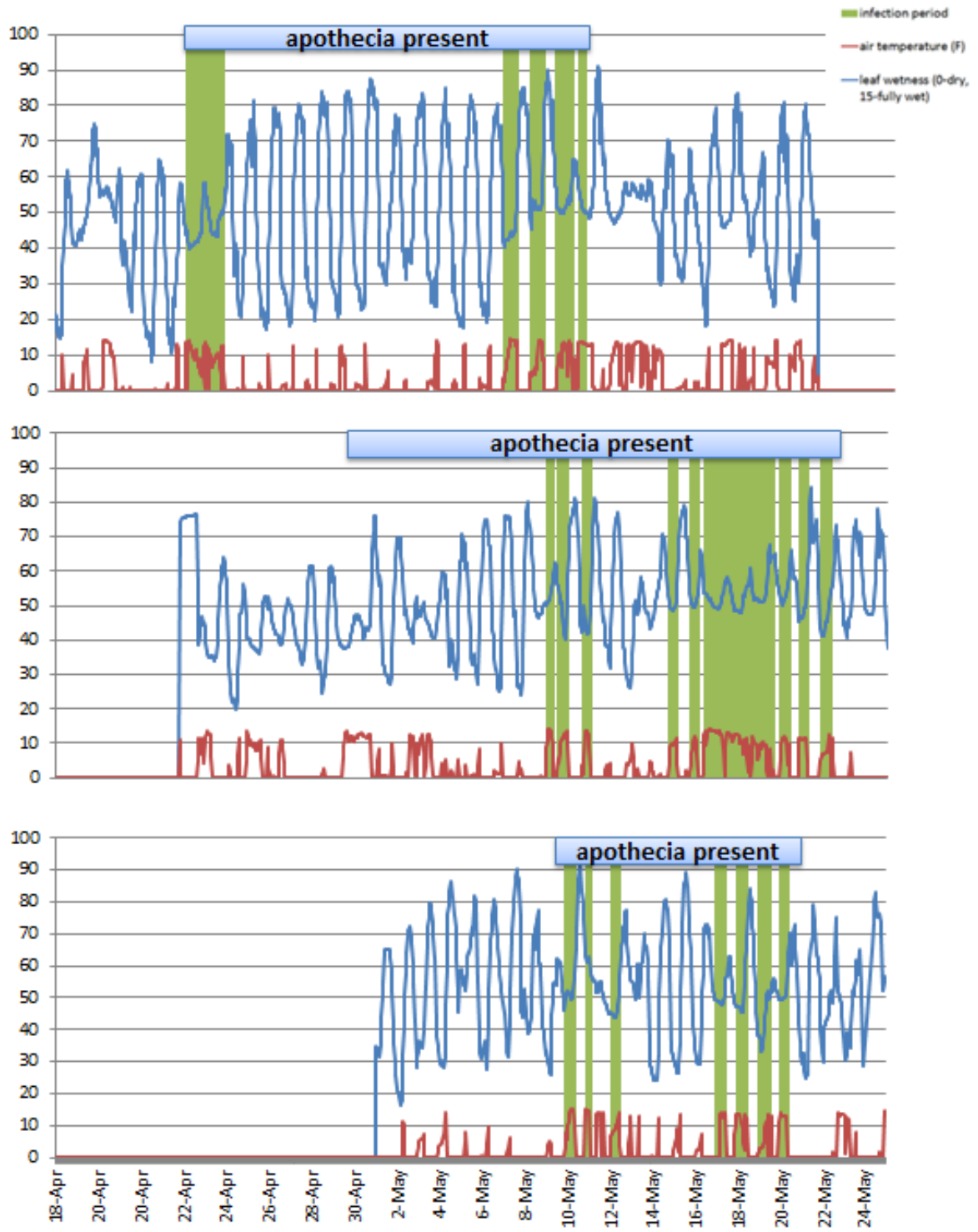


Figure 2. Comparison of infection periods from 2013 to 2015 at the Deblois site. Air temperature and leaf wetness were used to determine infection periods (green bars) for *Monilinia vaccinii-corymbosi*. Blue bars indicate when apothecia were present in fields.

DISEASE MANAGEMENT

INVESTIGATORS: Dr. Seanna Annis, Associate Professor/Associate Extension Professor, School of Biology and Ecology
Rachael Martin, Research Assistant, School of Biology and Ecology
Jennifer Cote, Assistant Scientist, School of Food and Agriculture

7. TITLE: Evaluation of fungicides for control of mummy berry on lowbush blueberry (2015).

OBJECTIVE: To evaluate control of the primary infection stage of mummy berry on lowbush blueberry (*Vaccinium angustifolium*) in Maine.
Primary disease target: Mummy berry, causal agent: *Monilinia vaccinii-corymbosi*

METHODS: Complete randomized block experiments were established in two lowbush blueberry fields with histories of mummy berry disease. One field was near Deblois and the other in Township 19, Maine. Fungicides (Table 1) were randomly assigned to 6' x 30' plots with a 3' buffer lane between each plot and replicated in 8 blocks per field. Fungicide applications were timed using the Mummy Berry disease forecast[1] according to locally monitored conditions of fungal and plant development and weather conditions favoring disease development. Fungicides were applied on May 8 and May 15 in the Deblois and Township 19 fields. Fungicides were applied at volumes equivalent to 20 gallons per acre at 35 psi with a CO_2 backpack sprayer equipped with a 4 nozzle boom, 8002VS Tee Jet tips and 50 mesh screens applied. Appropriate surfactants were added as recommended by the manufacturer (Table 1) and the negative control (check) plots received no spray applications. Due to applicator error Plot 24 at Township 19 (Serenade Optimum and Dyne-Amic) was not sprayed during the May 15 application.
Disease assessments in both fields occurred on June 3 and consisted of presence/absence of the disease symptoms on 40 blueberry stems along a transect through the middle of each plot. A rope with evenly spaced markings was stretched along the transect and the stem closest to each marking was inspected for disease symptoms on flowers or leaves. In addition, the number of markings at bare places (missing data) and frost damaged stems was recorded. The percentage of infected stems was the number of counted infected stems divided by the total number of rated stems (40 minus the number of bare locations) for each plot. Phytotoxicity was also rated at the same time disease assessments were made.
Blueberries were harvested on August 5, 2015. Harvesting occurred in a 2 foot strip down each plot center with a mechanical harvester and fresh weight was measured.

RESULTS: *Monilinia* apothecia appeared in Deblois and Township 19 fields during the first week of May, later than 2013 and 2014. However, mummy berry season started only a day earlier than in 2014 (May 9 in 2015, May 10 in 2014). The first infection period occurred the evening of May 9. Fungicides were preemptively applied on May 8. A second application was applied May 15 before the next set of infection periods. These applications should have covered all the infection periods. The season was more condensed than in 2013 or 2014, and there were fewer infection periods than in 2014. Most growers were able to obtain coverage with one or two applications.

[1]More information about the Mummy Berry forecast method can be found in UMaine Cooperative Extension Bulletin #217 (http://umaine.edu/blueberries/factsheets/disease) and the forecasts for last year are available at http://umaine.edu/blueberries/blog/.

Table 1. Fungicides tested in 2015 for control of mummy berry.

Treatment (Trade Names)	Application Rate (per acre)	Material	Manufacturer	FRAC group	EPA Reg. Number	Registered on Blueberries
Fontelis - Low	12 oz	penthiopyrad	DuPont	7	352-835	Yes
Fontelis - High	24 oz	penthiopyrad	DuPont	7	352-834	Yes
Proline	5.7 oz	prothioconazole	Bayer Crop Science	3	264-825	Yes
Serenade Optimum and Dyne-Amic (surfactant)	20 oz 0.25 % v/v	*Bacillus subtilis*, bacteria	Bayer Crop Science (Helena Chemical Company)	none	264-1160 (5905-50071-AA)	Yes
Positive Control - Bumper	6 oz	propiconazole	Adama	3	66222-42	Yes

Levels of mummy berry were moderate overall with untreated controls having 20% of stems with mummy berry in the Deblois field and 26% in the Township 19 field (Figure 1). The data was analyzed with all plots and omitting plot 24 (Serenade Optimum and Dyne-Amic) from the Township 19 trial which did not change the results.

At the Township 19 site, Serenade Optimum treatment did have significantly lower disease than the controls, but this was not seen in the Deblois field. In both fields, both high and low rates of Fontelis treatments and the Proline treatments had significantly lower levels of mummy berry and had similar levels to Bumper (propiconazole) which is currently the predominant fungicide used for mummy berry control in Maine. There was no significant difference between the two rates of Fontelis in either field.

There were no significant differences in yield among the treatments in either field (Figure 2). Frost damage or top kill was not observed at the Deblois site, and was present at levels below 2% at the Township 19 site and was not significantly different among treatments (data not shown). Phytotoxicity was not seen in any of the plants.

CONCLUSIONS/RECOMMENDATIONS: Proline is recommended for control of mummy berry disease based on the results of this trial and a 2014 trial. Proline without added surfactant (tested in 2014) is an effective product. Fontelis, without a surfactant, was also effective, and there was no significant difference between application rates, so the lower rate is recommended. Serenade Optimum showed some effectiveness at one site, so it may be an option for organic growers.

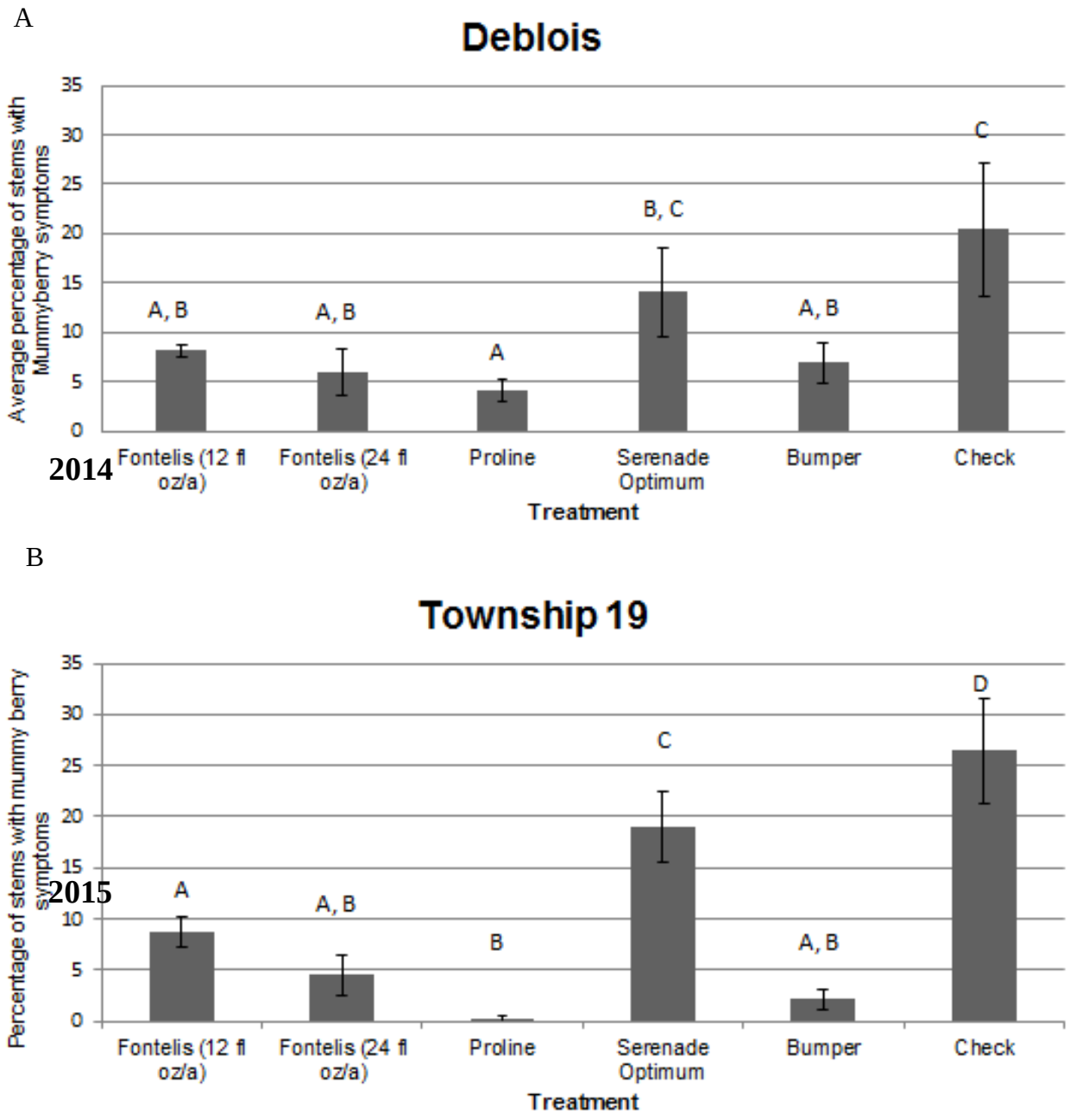
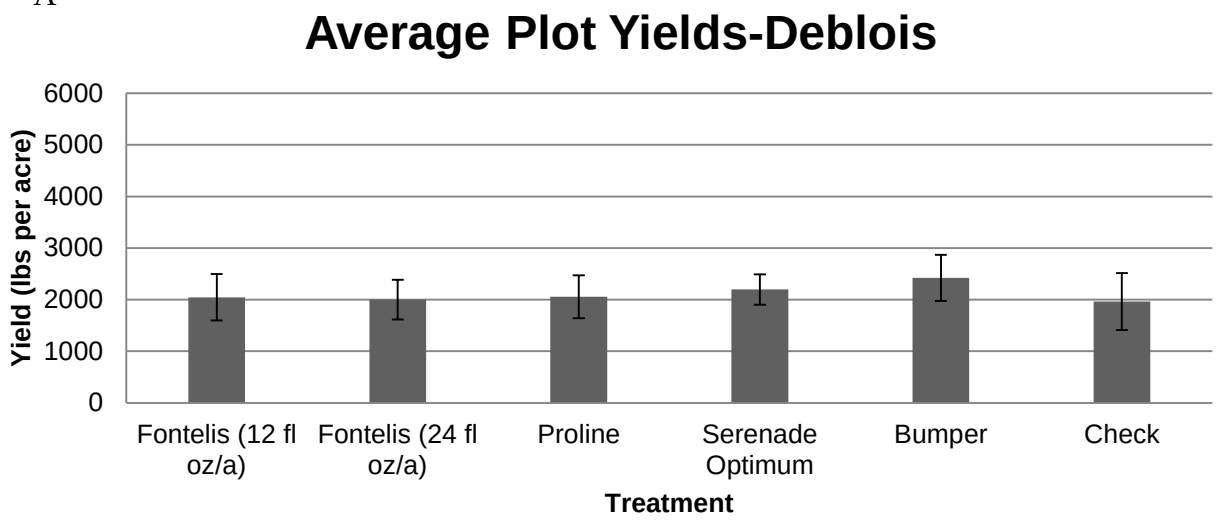


Figure 1. Average percentage of stems with symptoms of mummy berry disease in fungicide trials at A) Deblois and B) Township 19 fields. Error bars represent standard error of the mean of 8 replicates. Bars with different letters were significantly different at $p<0.05$ within the fields.

A



B

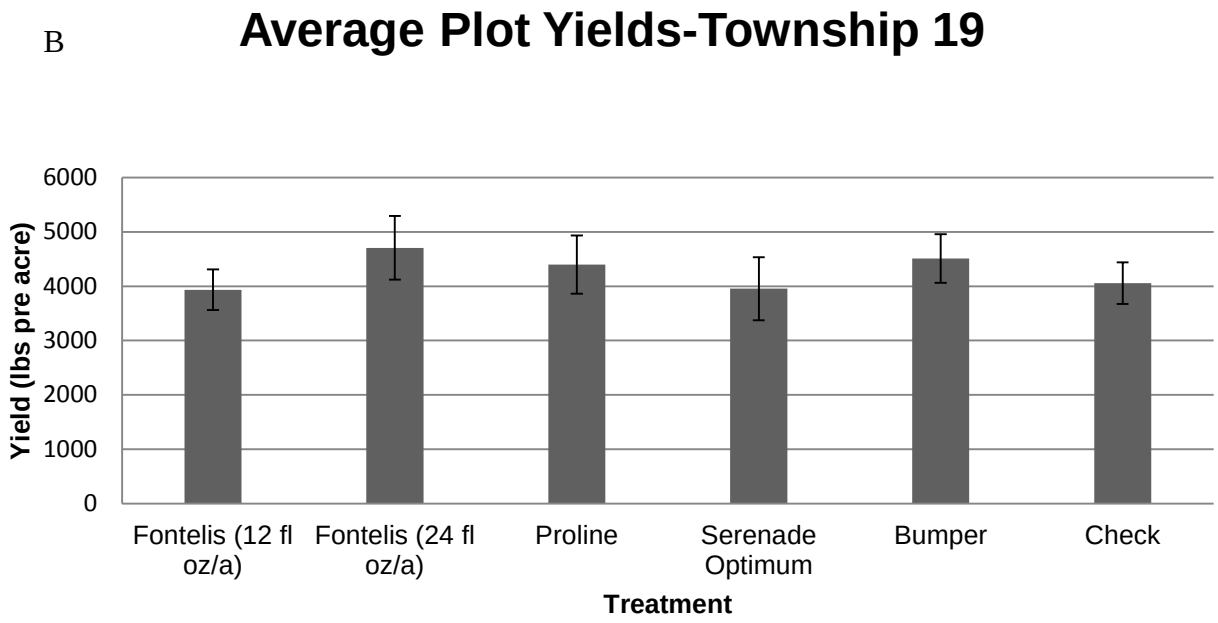


Figure 2. Average blueberry yield in pounds per acre for treatments in fungicide trials at A) Deblois and B) Township 19. Error bars represent standard error of the mean of 8 replicates. There were no significant differences among the treatments within the Deblois and Township 19 fields.

DISEASE MANAGEMENT

INVESTIGATORS: Dr. Seanna Annis, Associate Professor/Associate Extension Professor, School of Biology & Ecology
Rachael Martin, Research Assistant, School of Biology and Ecology
Jennifer Cote, Assistant Scientist, School of Food and Agriculture

8. TITLE: Evaluation of fungicides for control of leafspot on lowbush blueberry (2015).

OBJECTIVE: To evaluate control of leaf spot causing fungal pathogens on lowbush blueberry (*Vaccinium angustifolium*) in Maine.
Primary disease target: leafspot diseases (Septoria, Powdery Mildew, Rust)

METHODS: Complete randomized block experiments were established in a lowbush blueberry field in Jonesboro, Maine. Fungicides (Table 1) were randomly assigned to 6' x 15' plots with a 3' buffer lane between each plot and replicated in 6 blocks. Fungicides were applied on June 30 at volumes equivalent to 20 gallons per acre at 35 psi with a CO_2 backpack sprayer equipped with a 4 nozzle boom, 8002VS T Jet tips and 50 mesh screens applied. Appropriate surfactants were added as recommended by the manufacturer (Table 1) and the control plots received no spray applications.
A rope with 20 evenly spaced markings was stretched along a transect through each plot and the stem closest to each marking was cut and bagged. The next day stems were inspected for disease symptoms on flowers and leaves. The total number of leaves, nodes lacking leaves (leaves fallen) and the estimated percent coverage of each disease on remaining leaves was noted per stem. 120 stems per treatment (20 stems per plot) were rated. Phytotoxicity was also rated at the same time disease assessments were made. The first stems were collected August 10 and rated August 12. Stems were again collected on September 15, and rated on September 16. Stems were collected a third time on November 17 and were rated on November 18 and 19. At the time of the third rating, no leaves were left on the stems. Length of the main stem, number of leaf buds and flower buds was recorded.
Data was analyzed by plot averages in SAS (Statistical Analysis Software - SAS Cary, NC) using mixed model procedures (PROC GLIMMIX). Least Square means were used to determine specific differences among treatments ($\alpha = 0.05$). Correlation between treatments was assessed using Spearman's rank correlation in SAS (PROC CORR).

RESULTS: There was no effect of the treatments on leaf loss or disease levels in August (Fig. 1). Leaf loss ranged from 17 to 22% in August and was consistent among all treatments and the control. Leaf spot diseases were at low levels when measured in August. Septoria leaf spot affected from 1.5 to 5% of leaf area (Fig. 2). Powdery mildew as found on less than 4 % of leaf area (Fig. 3), and rust was negligible (Fig. 4). There was a significant correlation between levels of Septoria leaf spot and leaf loss in August ($R = 0.73657$ $p<.0001$, Fig. 5).
Levels of leaf loss were higher (25 to 45%) and were significantly different between treatments in September (Fig. 1). Leaf loss in September in the Proline, Pristine and Bravo Ultrex treatments was significantly lower than the control, but Serenade Optimum, Regalia and Procidic treatments did not differ significantly from the control.

Levels of Septoria and Powdery Mildew were very low (<1%) in September and not significantly different between treatments (Fig. 2 and 3). Rust levels in September were higher ranging from 1 to 8% than in August and were significantly lower in the Proline, Pristine and Bravo Ultrex treatments than in the control (Fig. 4). The levels of leaf rust in the Serenade Optimum, Regalia and Procidic treatments did not differ significantly from the control.
There was a strong correlation between levels of Rust and leaf loss in September (Fig. 6). Rust in September was significantly correlated with leaf loss in September (Figure 6).
From measurements in November, there was no significant effect of the treatments on stem length, number of leaf buds, or number of flower buds (data not shown). There was a significant but weak correlation of higher leaf loss in August correlated with lower levels of leaf buds. There were no significant correlations between leaf loss in September and disease levels or leaf loss in August.

Table 1. Fungicides tested in 2015 for control of leafspot.

Treatment (Trade Names)	**Material**	**Application Rate (per acre)**	**Manufacturer**	**FRAC group**	**EPA Reg. Number**	**Regis-tered on Blue-berries**
Proline	prothiocona-zole	5.7 fl oz	Bayer Crop Science	3	264-825	yes
Serenade Optimum and Dyne-Amic (surfactant)	*Bacillus subtilis*, Bacteria	20 oz 0.25 % v/v	Bayer Crop Science (Helena Chemical Company)	none	264-1160 (5905-50071-AA)	yes
Procidic	Citric extract	2.5 fl oz/10 gallons	Greenspire	none	n/a	yes
Regalia	*Reynoutria* spp. extract	2 quarts	Marrone Bio Innovations	P5	84059-3	yes
Pristine	pyraclostrobin and boscalid	18.5 oz	BASF Corporation	117	7969-199	yes
Bravo Ultrex	Chlorothalonil	3.6 lbs	Syngenta	M5	50534-201-100	yes

CONCLUSIONS/RECOMMENDATIONS: The conventional fungicides did decrease levels of leaf loss and rust in September but it is not clear what effects this would have on yield. The Serenade Optimum, Regalia and Procidic treatments did not significantly reduce Rust or leaf loss levels in September and may require an earlier application to control disease. The levels of leaf loss and disease in this trial are unlikely to affect yield in the following year considering the large number of factors that affect yield in the crop year. A new trial will be set up in 2016 in a prune year field to test earlier timings of fungicide applications to look at their efficacy for controlling leaf spots.

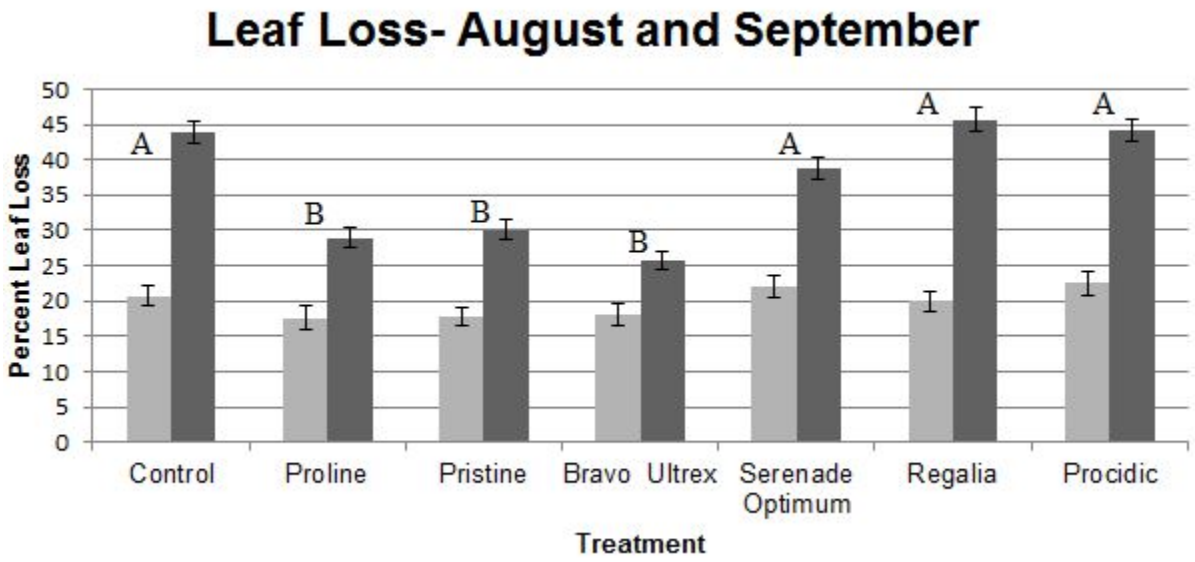


Figure 1- Levels of leaf loss in August (light grey bars) and September (dark grey bars). There were no significant differences among the treatments in August. Bars with different letters indicate statistically significant differences at $\alpha = 0.05$.

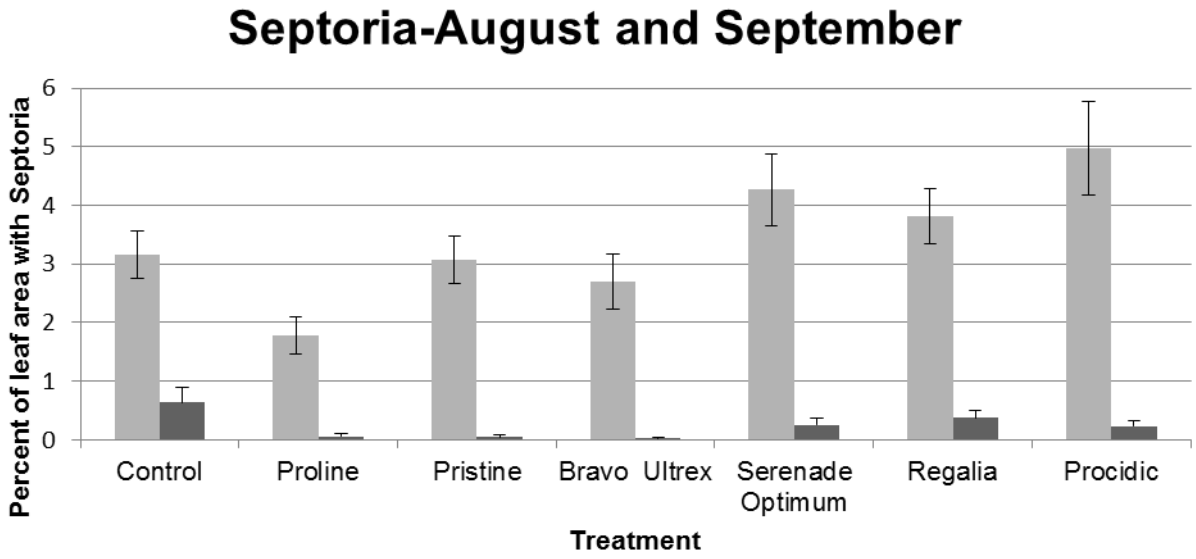


Figure 2- Levels of Septoria in August (light grey bars) and September (dark grey bars) for the treatments. There were no significant differences among treatments.

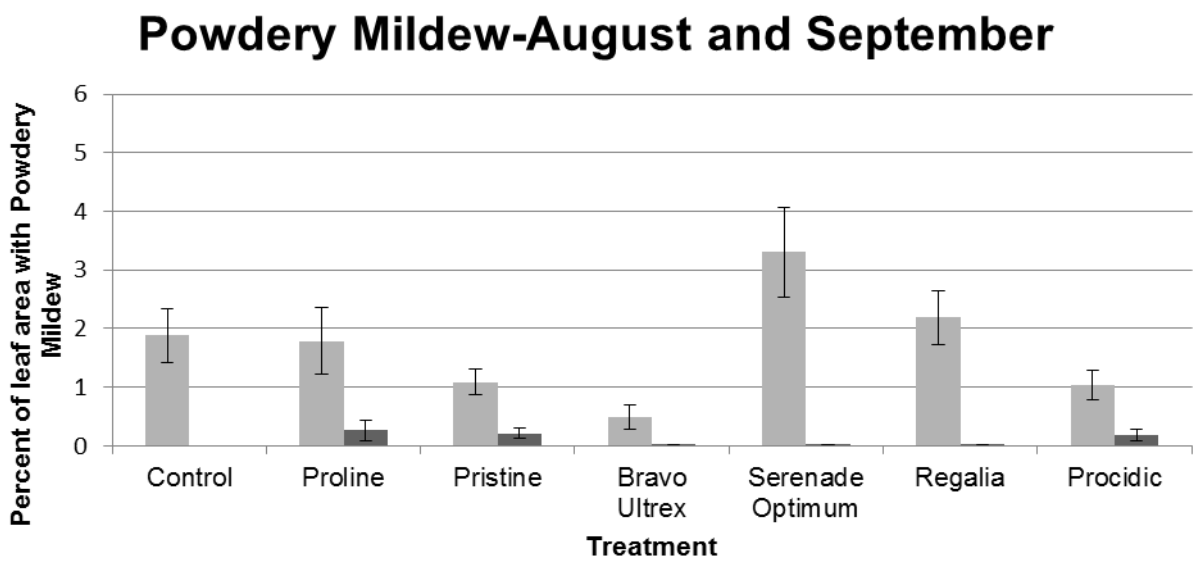


Figure 3- Percent of leaf area with Powdery Mildew by treatment in August (light grey bars) and September (dark grey bars). There were no significant differences among treatments.

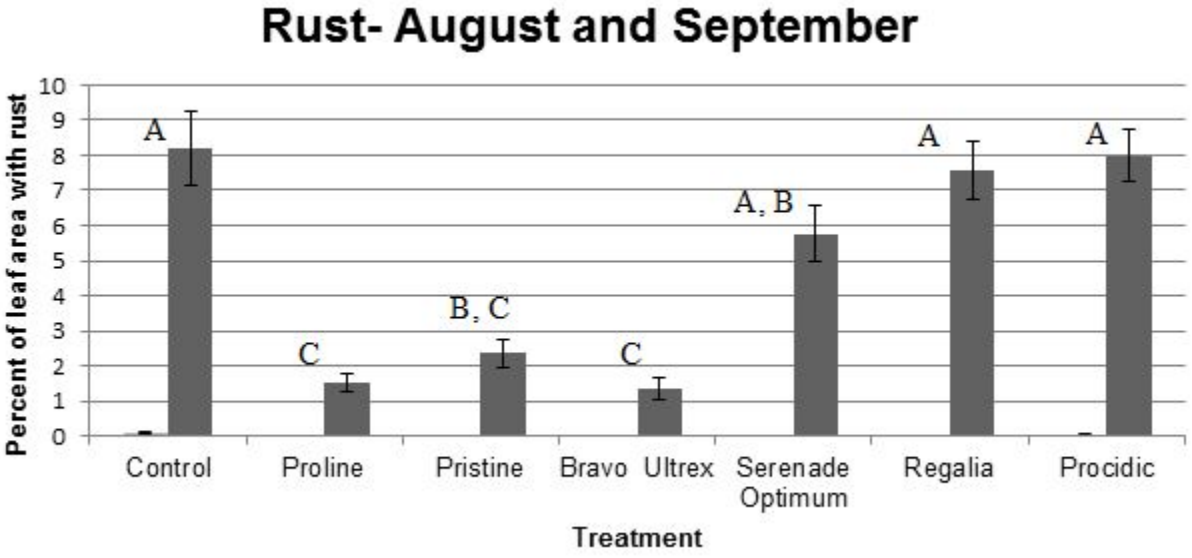


Figure 4- Percent of leaf area with Leaf Rust by treatment in August (light grey bars) and September (dark grey bars).). There were no significant differences among the treatments in August. Bars with different letters indicate significant differences at $\alpha = 0.05$ in the September rating.

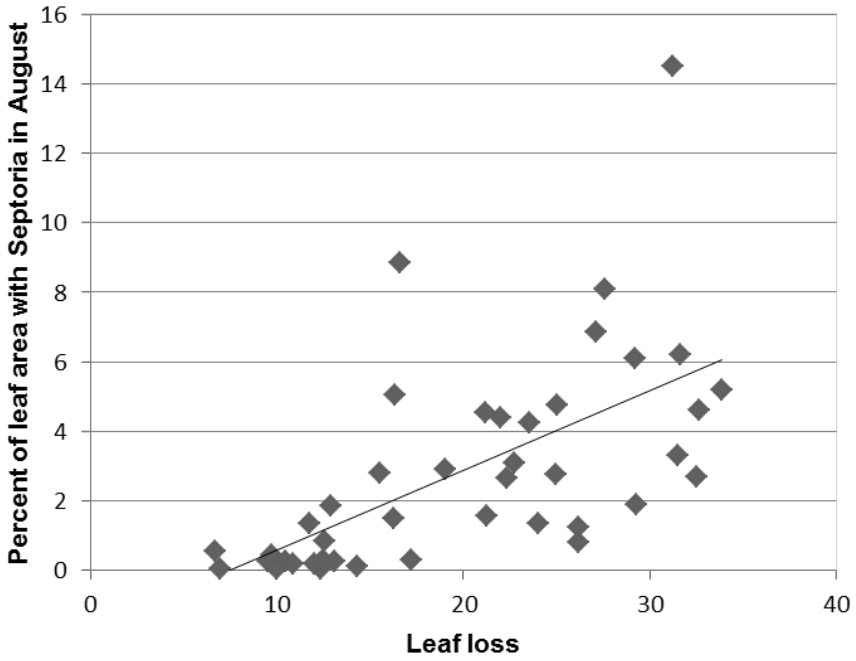


Figure 5- Average percentage of leaf area affected with Septoria versus leaf loss in August. The Spearman correlation was significant (R = 0.73657 p<.0001). The solid line represents a best fit linear regression line for illustrating the trend.

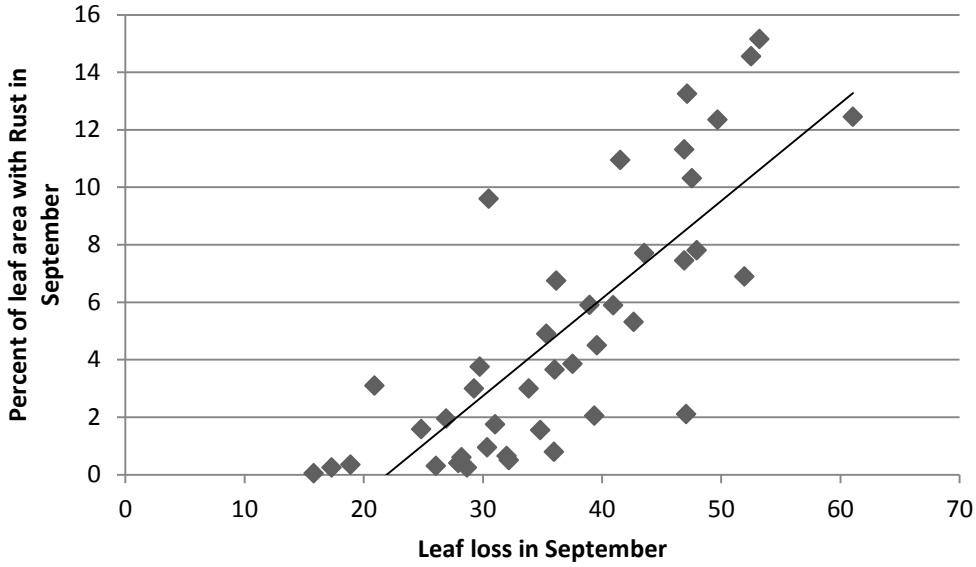


Figure 6- Average percentage of leaf area affected with Rust versus leaf loss in September. The Spearman correlation was significant (R= 0.83614, p <.0001). The solid line represents a best fit linear regression line for illustrating the trend.

WEED MANAGEMENT

INVESTIGATORS: David E. Yarborough, Professor of Horticulture
Jennifer L.D. Cote, Assistant Scientist

9. TITLE: Single vs split applications of post-emergent herbicides for spreading dogbane (*Apocynum androsaemifolium*) control in wild blueberry fields.

METHODS: Spreading dogbane (*Apocynum androsaemifolium*) is a major weed pest in wild blueberry fields, and is difficult to control with many of the industry's currently registered herbicides. In spring 2015 we initiated a trial at the University of Maine's Blueberry Hill Experiment Station Farm to examine the effect of Callisto and Matrix, on dogbane control. Dogbane was sprayed post-emergence to four 1 x 2 m plots in the following combinations, all with COC 1% v/v: Callisto 6 oz/a, Matrix 4 oz/a, Callisto 6 oz/a + Matrix 4 oz/a, Callisto 3 oz/a (2x), Matrix 2 oz/a (2x) and Callisto 3 oz/a + Matrix 2 oz/a (2x), on 3 June, and the split treatments were sprayed a second time on 17 June. In June and July, wild blueberry and dogbane cover and phytotoxicity were compared among treatments, to an untreated check and to the Blueberry Hill Farm's 5/13 pre-emergence application of Velpar 2 lb/a, Sinbar 2 lb/a and diuron 1.6 qt/a with Grounded additive at 2 pt/a combined with a 5/27 post-emergence application of Callisto 6 oz/a and Select 6 oz/a. Control of other broadleaf weeds and grasses were also assessed. It should be noted that at the June assessment, the split application treatments were assessed just prior to the second herbicide application. Cover data were determined by using the Daubenmire Cover Class system converted to percent, and phytotoxicity using a scale of 0-10 (0=no damage, 10=100% damaged/dead) which was converted to percent. The treatments were compared using Tukey's tests (α=0.05) to determine significant differences.

RESULTS: Initially in June, all treatments except Matrix at 4 oz/a and the split Matrix at 2 oz/a had significantly more dogbane injury than the check, but no treatment significantly reduced dogbane cover (Figure 1). By July, dogbane injury approached 90% in the Callisto split 3 oz/a and Callisto 3 oz/a + rimsulfuron 2 oz/a split applications, and dogbane cover was reduced to less than 10%. Dogbane cover was significantly lower in all treatments compared to Blueberry Hill Farm's treatments, but only the split Callisto treatment and the split Callisto + Matrix treatment were significantly lower than the check (Figure 1). The Matrix 4 oz/a and Callisto + Matrix split treatments resulted in slightly lower blueberry cover, but phytotoxicity (chlorosis) was 20% or lower at both evaluations and so was considered acceptable (Figure 2). The split Matrix and split Callisto + Matrix treatments resulted in the highest dogbane injury by July. Other weed cover in the plots was very low, below 12% overall, and there were no significant differences among treatments at either evaluation (Figure 3).

Figure 1. Dogbane cover and phytotoxicity following post-emergence applications of Callisto and Matrix (different letters denote significance at α=0.05).

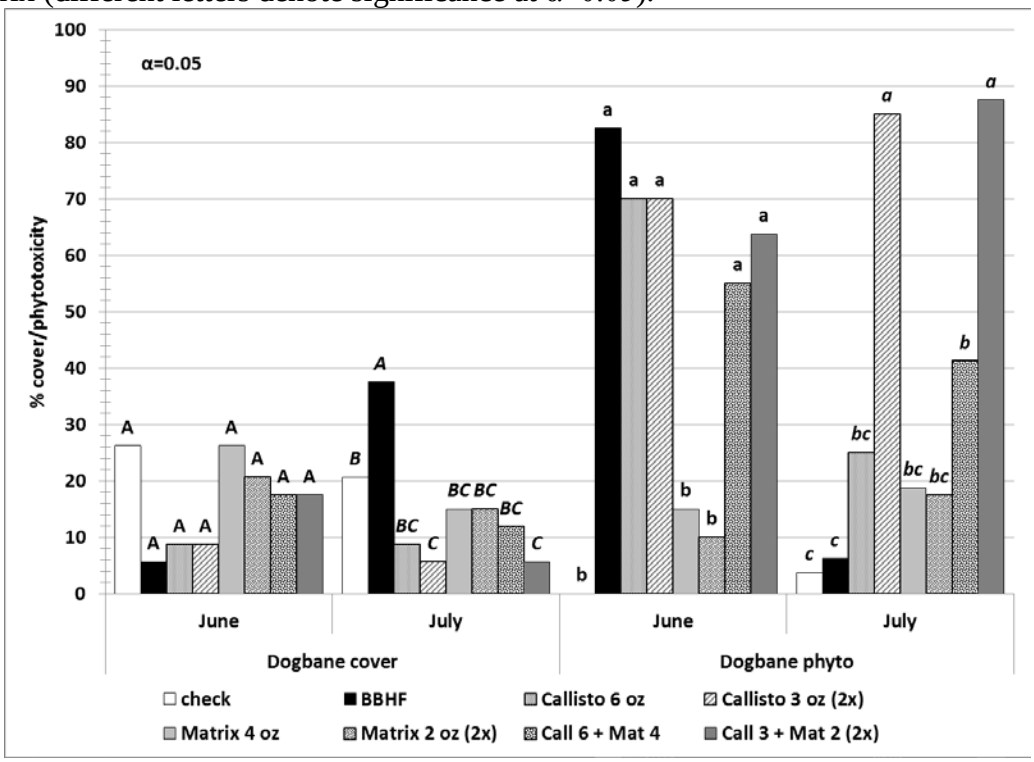


Figure 2. Wild blueberry cover and phytotoxicity following post-emergence applications of Callisto and Matrix (different letters denote significance at α=0.05).

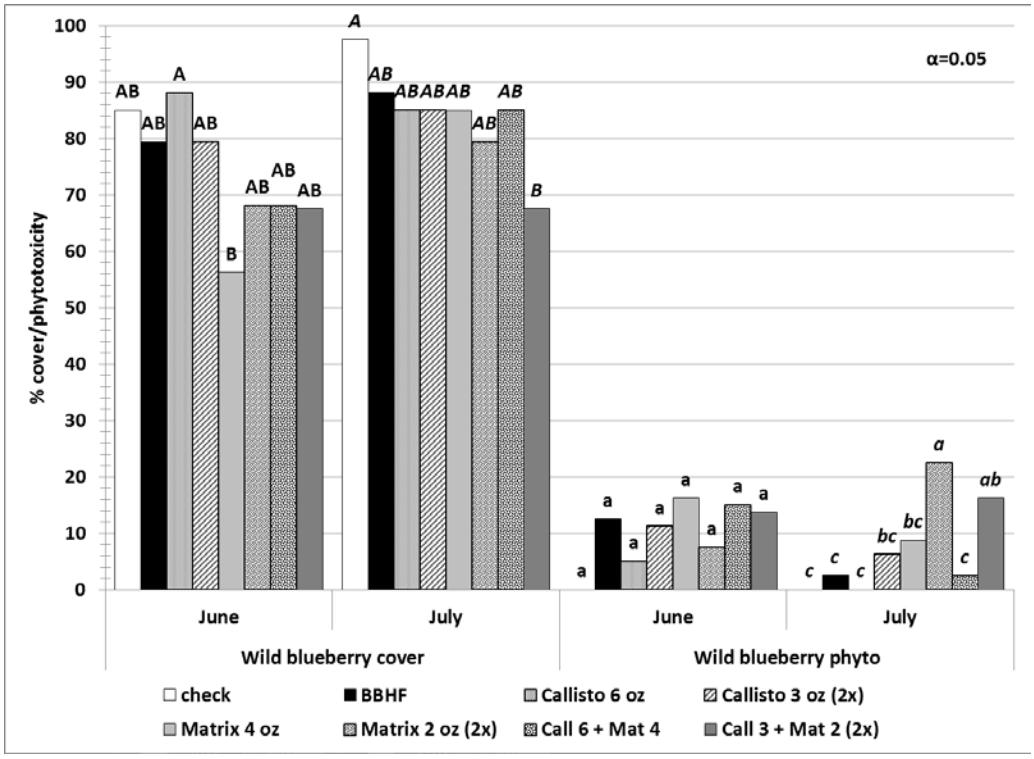
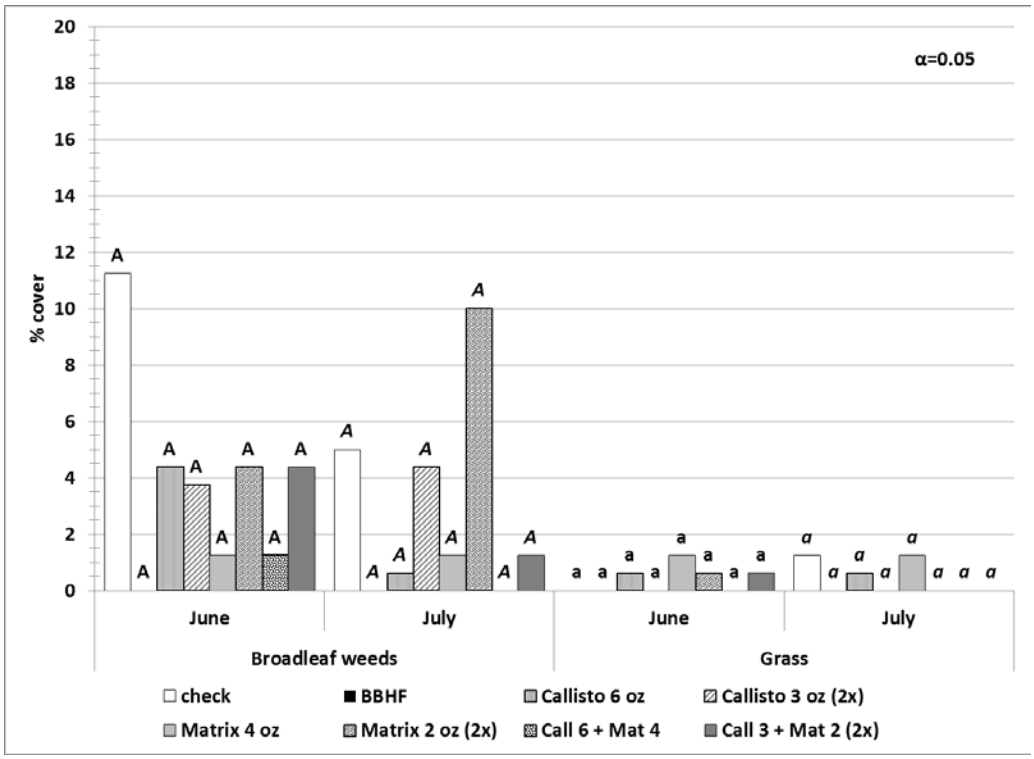
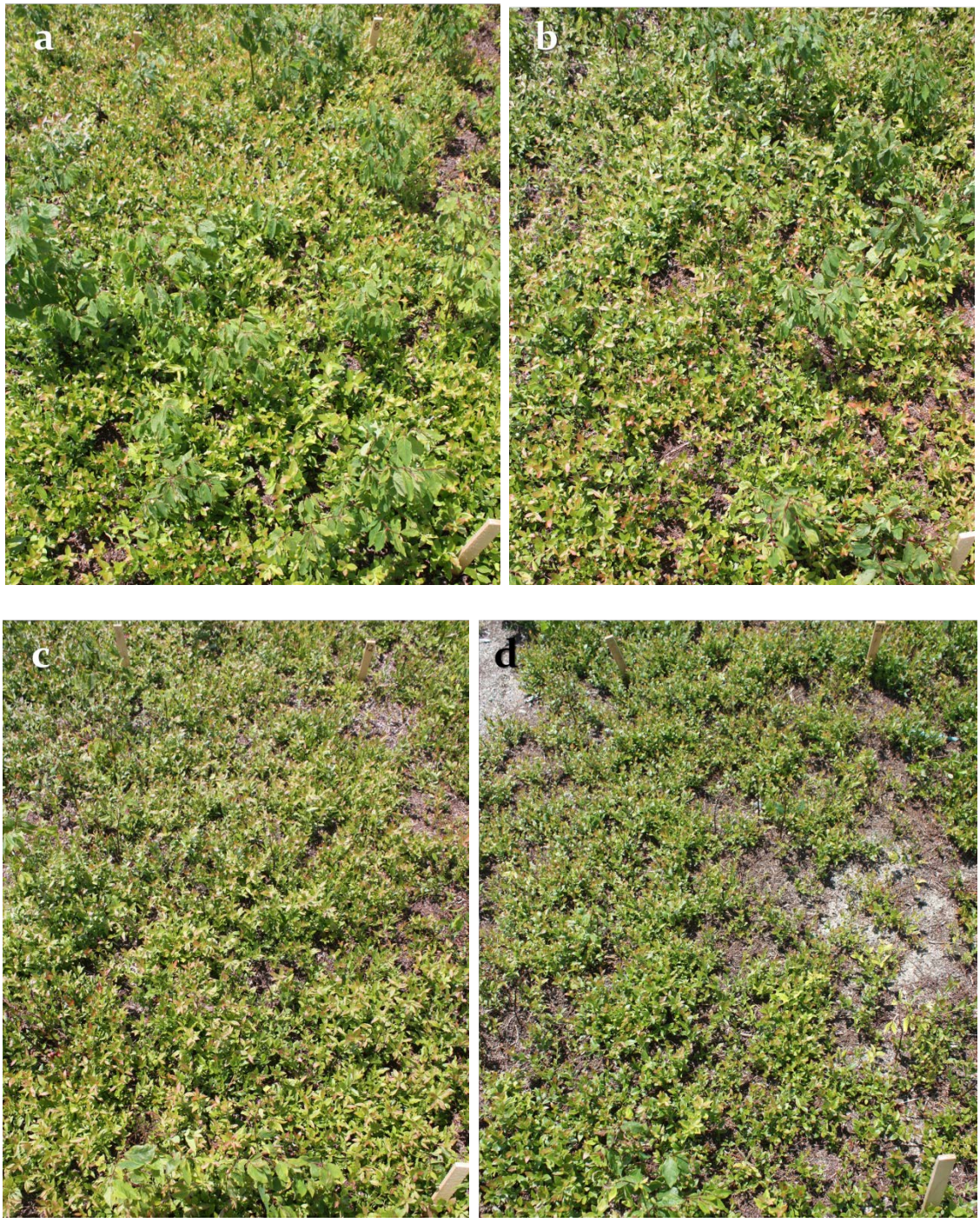


Figure 3. Broadleaf weed and grass cover following post-emergence applications of Callisto and Matrix (different letters denote significance at α=0.05).



CONCLUSIONS: The Callisto and Callisto + Matrix tank mixes resulted in higher dogbane injury and lower cover overall, indicating that Callisto was the most effective treatment for controlling this weed. The lack of total control was due to new dogbane emerging after the herbicide applications; new dogbane shoots were observed both at the second application of the split treatments and at the July assessment (Photo 1A-D). Although Blueberry Hill Farm also sprayed Callisto, the lack of long-term control in their treatment appeared to be because their post-emergence spray not only occurred a week earlier than ours, allowing more time for additional dogbane to emerge, but they only sprayed once post-emergence. In conclusion, the split Callisto treatment was the most effective in controlling spreading dogbane; it resulted in the lowest dogbane cover and greatest dogbane injury along with the split Callisto + Matrix treatment, but considering the two were equal in cover and within 2% of each other in phytotoxicity, the addition of Matrix did not improve the control and therefore is unnecessary. Additional applications of Callisto later in the season would be needed to get complete control of this weed.

Photo 1. Dogbane in **A**) the untreated check, **B**) Callisto 6 oz/a, **C**) split Callisto 3 oz/a and **D**) split Callisto 3 oz/a + Matrix 2 oz/a treatments. Note emerging dogbane seedlings.



RECOMMENDATIONS: Pursue a 24-C label change to increase the number of applications of Callisto.

WEED MANAGEMENT

INVESTIGATORS: David E. Yarborough, Professor of Horticulture
Jennifer L.D. Cote, Assistant Scientist

10. TITLE: Evaluation of fall applications of herbicides targeting horseweed in wild blueberry fields.

METHODS: Several herbicides, both registered and unregistered, are currently under review for use on wild blueberry. Alion (6.5 oz/a), Chateau (12 oz/a), Sandea (1 oz/a) and Trellis (1.33 lb/a) are pre-emergence herbicides, while Matrix (4 oz/a) may be used pre- or post-emergence. In this trial, we treated an emerging problem weed, horseweed (*Conyza canadensis*). Horseweed germinates in the fall and overwinters as a basal rosette, which can escape the notice and therefore control by growers. The following year, the plant bolts and flowers typically just prior to harvest, and the seeds can be spread by harvesting equipment.
The treatments were applied to horseweed on ten 1-m^2 plots on 11 November 2014, except Chateau which was applied on 11/26; five plots were also treated by the grower with Velossa (6.6 pt/a) and diuron (1.6 qt/a, 5/12/15), and Callisto (3 oz/a, 6/16/15). Effects on wild blueberry cover and phytotoxicity, horseweed and broadleaf weed and grass cover were evaluated in June and July. Cover data were determined by using the Daubenmire Cover Class system converted to percent; horseweed phytotoxicity was not rated as the plants were either dead or unaffected. Data were analyzed using Tukey's tests to determine significant differences (α=0.05).

RESULTS: Wild blueberry cover in the horseweed trial was low because the horseweed occurred in bare spots at this site (Photo 1). There were no significant differences in blueberry cover and no phytotoxicity for the treatments alone in June, but phytotoxicity was observed in the Chateau treatment in July (Figure 1, Photo 2). The grower-treatments resulted in minor phytotoxicity in June, of which the Matrix treatment was only significantly higher than Chateau treatment. However, there was significant phytotoxicity in the grower-treatments in July from the grower's Callisto post-emergence treatment (Photo 3). At the June evaluation, horseweed cover was significantly lower in the Matrix treatment compared to the other treatments alone (except Sandea); by July, Matrix remained lowest but was no longer significantly different (Figure 2, Photo 4). There was no horseweed in the grower-treatment plots at either evaluation. Broadleaf weed and grass cover were low in this trial as well, with no differences in broadleaf weed cover at either evaluation (Figure 3). Grass cover was highest in June on the Sandea treatment and was significantly higher than Chateau and all grower-treatments. In July, grass cover in the Sandea treatment almost doubled and was significantly higher than the grower-treatments, plus Alion and Chateau alone.

Figure 1. Wild blueberry cover and phytotoxicity in 2015 following fall 2014 herbicide treatments (different letters denote significance at α=0.05).

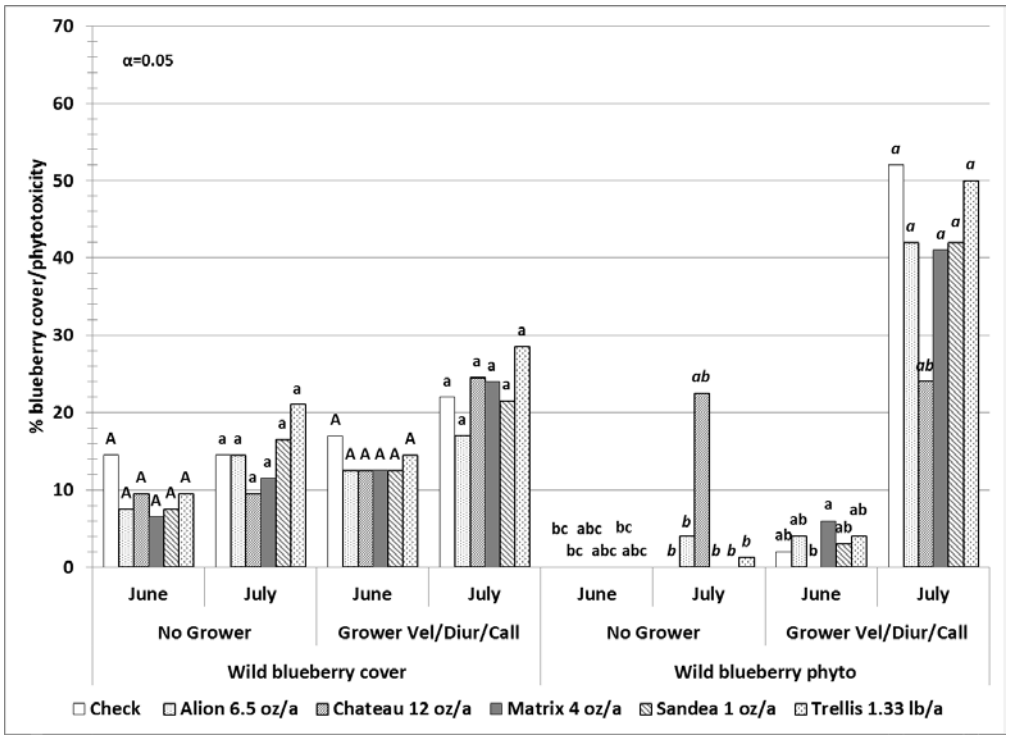


Figure 2. Horseweed cover in 2015 following fall 2014 herbicide applications (different letters denote significance at α=0.05).

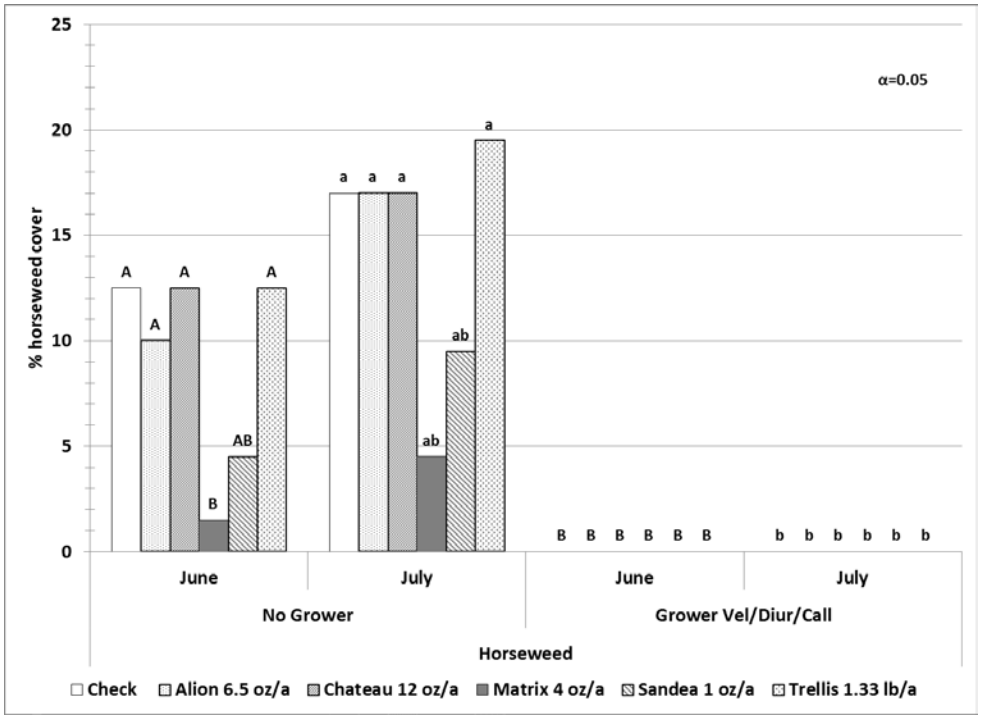


Figure 3. Broadleaf weed and grass cover in 2015 following fall 2014 herbicide applications (different letters denote significance at α=0.05).

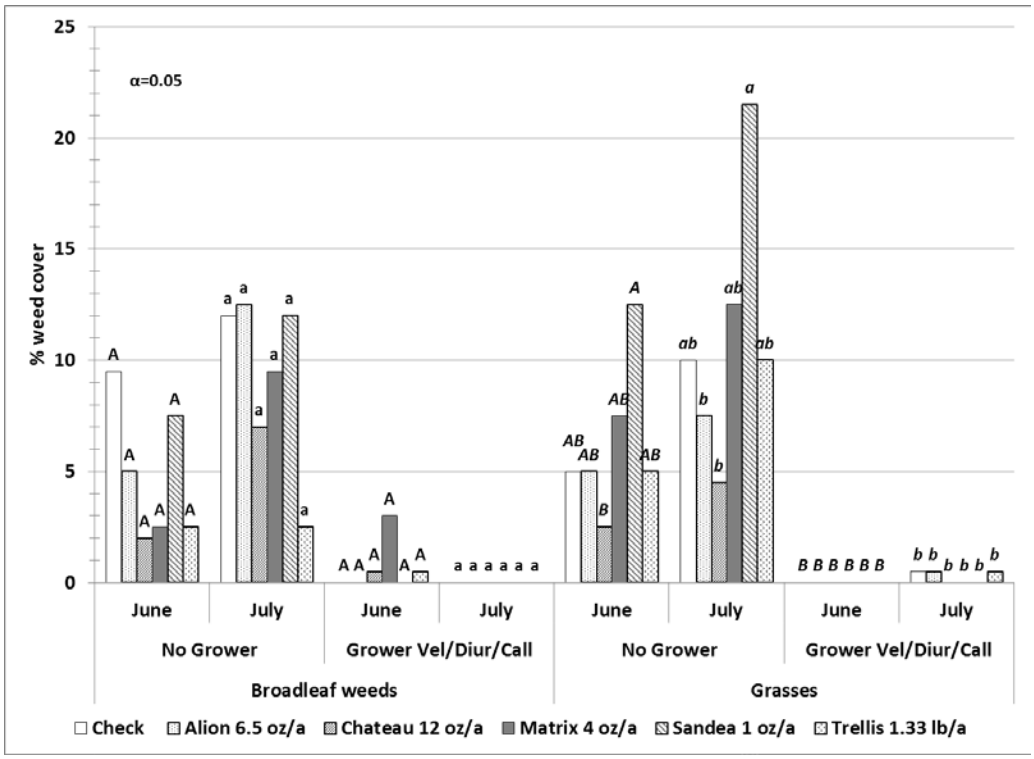


Photo 1. Untreated check plot in July; horseweed occurred in bare spots at this trial site.

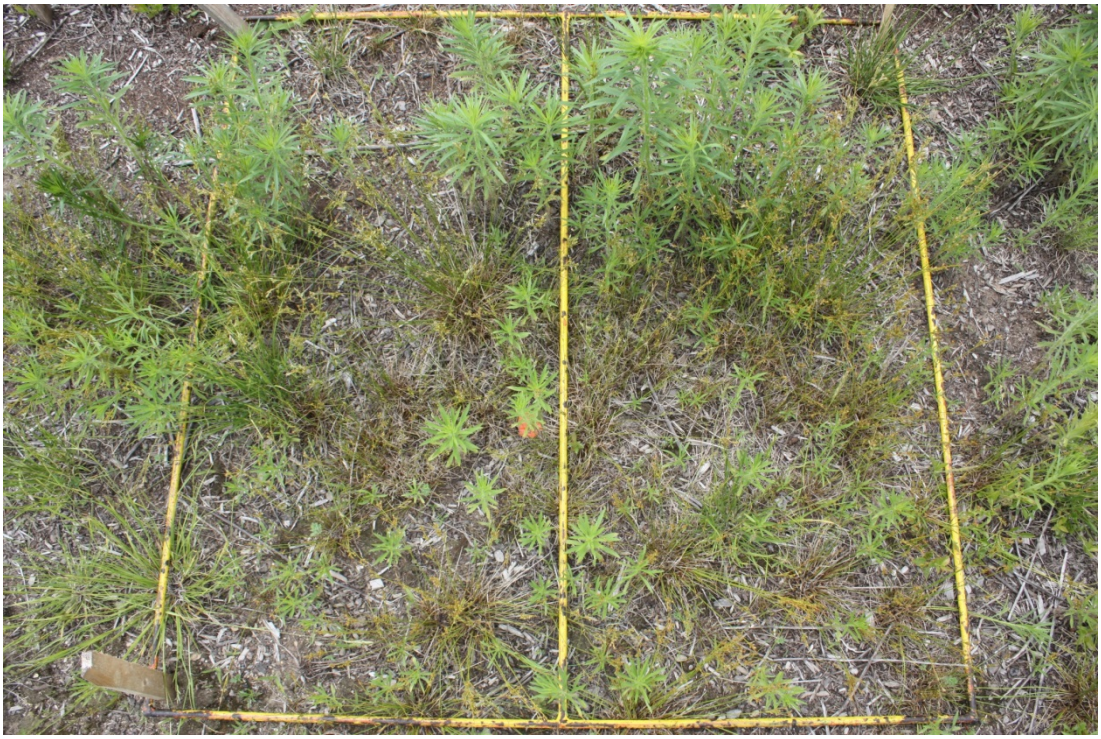


Photo 2. The Chateau treatment in July, showing lack of horseweed control and phytotoxicity to wild blueberry.

Photo 3. Wild blueberry phytotoxicity in July, resulting from the grower's Callisto application on a hot day.

Photo 4. The Matrix treatment in July; note horseweed control in plot and horseweed outside plot.

CONCLUSIONS/RECOMMENDATIONS: Horseweed was not resistant to the mixture of labeled herbicides used by the grower, so it is best controlled with registered herbicides as spring pre-emergence applications. At this time, there are no future studies planned for horseweed.

WEED MANAGEMENT

INVESTIGATORS: David E. Yarborough, Professor of Horticulture
Jennifer L.D. Cote, Assistant Scientist

11. TITLE: Herbicide combinations with Sinbar and Grounded to assess efficacy on weed control in wild blueberry.

METHODS: In spring 2015 we set up a trial in a prune field at Blueberry Hill Farm, to assess a new formulation of Sinbar. The product was held up at Canadian customs, so we applied the treatments in combination with Sinbar WDG 2 lb/a to examine efficacy on weeds when combined with other chemistries with and without Grounded. Grounded is an adjuvant "designed to enhance the deposition and absorption of ground applied pesticides" and some Maine growers have claimed it increased weed control in their fields. A Randomized Complete Block Design was replicated six times with 6' x 60' plots; all treatments except the check contained Sinbar and were tank mixed with: Nothing (Sinbar alone); Alion 5 oz/a; Callisto 3 oz/a; Matrix 2 oz/a; and Velpar 1 lb/a. Each 60' plot length was split in half, with 30' receiving

Grounded 0.25% v/v as part of the tank mix. The plots were sprayed late pre-emergence on 22 May 2015, and were to be evaluated early and late in the growing season. Cover data were determined by using the Daubenmire Cover Class system converted to percent; phytotoxicity data were gathered using a scale of 0-10 (0=no damage, 10=100% damaged/dead) converted to percent.

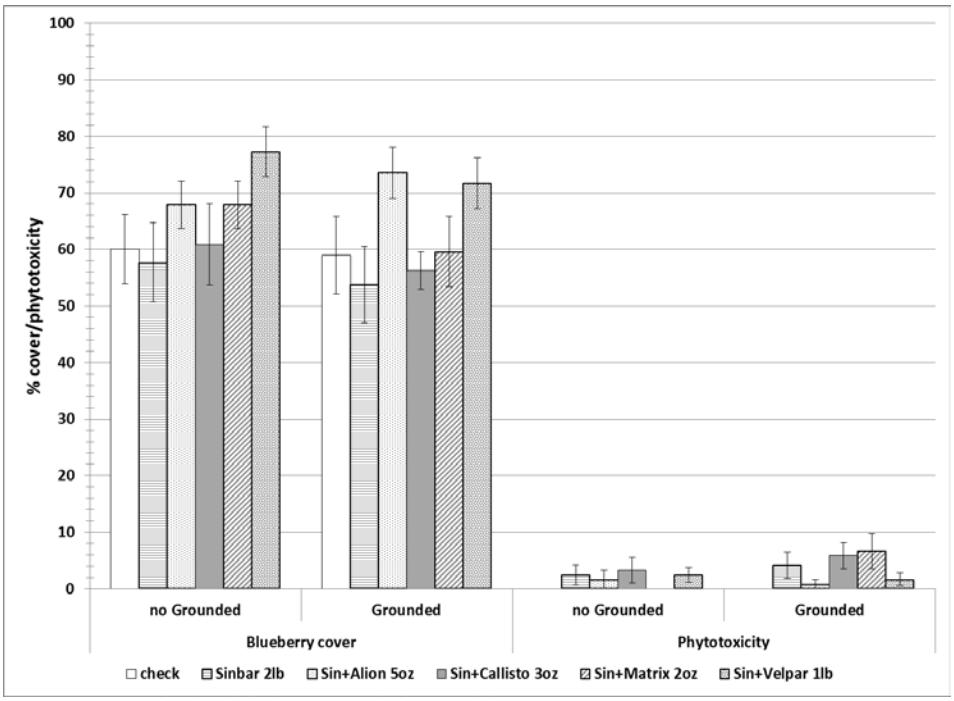
RESULTS/CONCLUSIONS: The first evaluation was completed on 7 July. However, the weeds in the trial area were so large and/or aggressive that treatment differences or control of weeds could not be observed, and the blueberry plants were shaded which would compromise yield differences in the crop year due to a lack of flowers/berries (Photos 1-2). The only visible trend observed onsite was that Grounded resulted in more weeds in some areas.

Photo 1. Blueberry was displaced by aggressive low-growing weeds such as bunchberry.

Photo 2. Blueberry was also shaded by large weeds such as brackenfern.

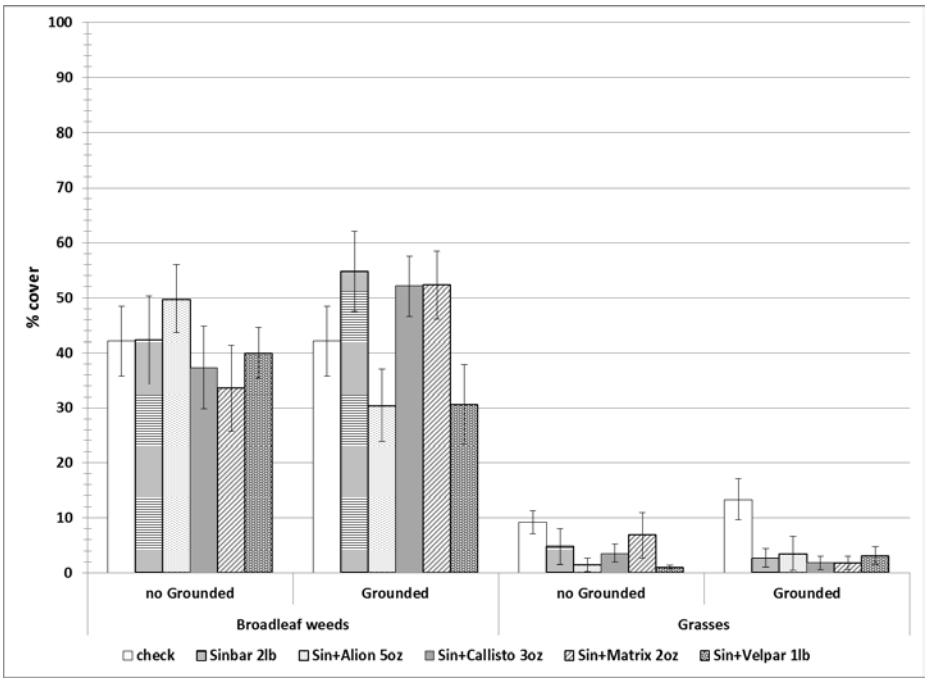
When the results from the first evaluation were graphed using the Standard Error of the Mean, blueberry cover without Grounded was similar with the exception of Sinbar+Velpar, which had slightly higher percent cover (Figure 1). Wild blueberry cover in the Grounded plots was higher in the Sinbar+Alion and Sinbar+Velpar plots; in both cases it is unclear whether the differences in blueberry cover are due to the herbicides, the amount of available sunlight or the amount of available area not already occupied by weeds. Phytotoxicity was very low for all treatments, and was similar among treatments. These results are more dependable because the herbicides were sprayed pre-emergence and therefore were taken up from the soil as the blueberry plants emerged.

Figure 1. Wild blueberry cover and phytotoxicity in July, following pre-emergence applications of Sinbar-herbicide combinations with and without Grounded (Std. Error of Mean).



Sinbar+Alion and Sinbar+Velpar with Grounded appeared to control broadleaf weeds more so than without Grounded, and more so than the other Grounded treatments (Figure 2). However, Grounded did not appear to consistently improve control, as shown in the Sinbar alone, Sinbar+Callisto and Sinbar+Matrix treatments. All treatments controlled grasses better than the check and Grounded alone, which is to be expected as Sinbar is the industry standard for grass control, but at <10% overall, treatment differences are minimal. In conclusion, the trial was discontinued as the treatment differences later in the year were not evident. If higher rates of the combinations were used then perhaps better weed control could be obtained. The higher rates of these materials are necessary to get lasting control when there is high weed pressure. The trial area was turned back over to Blueberry Hill Farm.

Figure 2. Wild blueberry cover and phytotoxicity in July, following pre-emergence applications of Sinbar-herbicide combinations with and without Grounded (Std. Error of Mean).



RECOMMENDATIONS: No recommendations; this trial has been discontinued.

EXTENSION

INVESTIGATOR: David E. Yarborough, Extension Blueberry Specialist

12. TITLE: Wild Blueberry Extension Education Program in 2015.

OBJECTIVE: To provide educational programming to bring research based knowledge to wild blueberry growers in Maine. To collaborate with Canadian researchers and provide relevant information to Maine growers.

METHODS: Conduct an educational program that will stress the use of best management practices in an integrated crop management program, which will improve the efficiency of culture and minimize the use of unnecessary pesticides and fertilizers. Conduct spring grower meetings and field days to introduce and reinforce the use of best management practices, integrated crop management and sound business management principles. Provide management information through the blueberry newsletters, fact sheets in the wild blueberry grower's guide both in print form and on the web at http://extension.umaine.edu/blueberries/, through telephone and correspondence, and conduct field visits as appropriate. Cooperate with County Educators and provide support for blueberry initiatives requested by the County office. Cooperate with the Wild Blueberry Advisory Committee Research, the Wild Blueberry Commission of Maine and

the Wild Blueberry Association of North America on blueberry related matters. Cooperate with county (Soil and Water Conservation Districts), state (Department of Agriculture, Board of Pesticides Control) and federal agencies (USDA, IR-4) on blueberry related matters. Needs are determined from the Wild Blueberry Advisory Committee Research and Extension priorities, Wild Blueberry Newsletter survey, and from individual client contacts. The advisory committee gave priority to grower outreach, IPM, pesticide recommendations for weeds, insects and diseases, food safety and groundwater. Needs identified by the survey include weed management, economics/marketing, pest management, general information and fertilization. Needs identified by individual grower contact reinforce those previously identified, but also added the need for blueberry quality and groundwater concerns.

RESULTS:

***Educational Activities*:**

This year the Blueberry Integrated Crop Management (ICM) program consisted of a presentation at the Agricultural Trade Show, at an expanded one day Spring Grower meeting in Bangor, field demonstration sessions conducted three times in three counties, an organic field day and the annual field day at Blueberry Hill Farm where we had a discussion on the effects of climate change on wild blueberry management. I also participated in a Barrens tour and educational sessions for Bloggers that Ethos had brought to Bar Harbor, in a new promotion video, with the legislative delegation Barrens and Farm presentations and in the Board of Pesticides Control educational session at Blueberry Hill Farm.

Meetings attended:

69th Annual meeting of Northeastern Weed Science Society, Williamsburg, VA, January 5-8, 2015.
Augusta Agricultural Trade Show, Augusta, ME, January 15, 2015.
Wild Blueberry Spring Grower Conference, Bangor, ME, March 20, 2015.
ICM Scouting Sessions, Warren, Jonesboro, Orland on April 28,29,30; May 26,27,28; June 30, July 1,2, 2015.
Organic Field Day Presentation, Penobscot, ME, July 7, 2015.
Wild Blueberry Summer Field Day & Meeting, Jonesboro, ME, July 15, 2015.
Wild Blueberry Health Summit, Bar Harbor, ME, August 5-7, 2015.
Legislative Tour, Machias and Jonesboro, ME, August 21-22, 2015.
Maine Board of Pesticides Control at Blueberry Hill Farm, Jonesboro, ME, August 27, 2015.
Maine Development Foundation Tour, Cherryfield, ME, October 8, 2015.
Wild Blueberry Research and Extension Workers Conference, Bar Harbor, ME, October 22, 2015.
Wild Blueberry Association of North America Annual Meeting. Bar Harbor, ME, October 23, 2015.

Presentations:

Evaluation of fall and spring combinations of preemergence herbicides to prevent weed resistance in wild blueberry fields. Northeastern Weed Science Society Annual Meeting, Williamsburg, VA, January 5-8, 2015.

Wild Blueberry Pest Management Update. Augusta Agricultural Trade Show, Augusta, ME, January 15, 2015.

A Systems Approach to Improving the Sustainability of Wild Blueberry Production; and 2014 Cultivated and Wild Blueberry Crop Report. Wild Blueberry Association of Nova Scotia Meeting, Truro, NS, March 14, 2015.

Worldwide Blueberry Production and MRLs in U.S. and overseas markets, harmonization; Preventing weed resistance in wild blueberry fields; and Pre- and post-emergence applications of herbicides for control of resistant fine leaf sheep fescue in wild blueberry fields in Maine. Wild Blueberry Spring Grower Conference, Bangor, ME, March 20, 2015.

A Systems Approach to Improving the Sustainability of Wild Blueberry Production; and MRLs in U.S. and overseas markets, harmonization. Annual General Meeting Bleuets New Brunswick Blueberries, Caraquet, NB, April 10, 2015.

ICM Scouting Sessions: Warren, Jonesboro, Orland on April 28, 29, 30; May 26 ,27, 28; June 30, July 1, 2, 2015.

Organic Field Day Presentation, Penobscot, ME, July 7, 2015.

Growing Season Effects on Wild Blueberry in Maine and Implications for Management. Wild Blueberry Summer Field Day & Meeting, Jonesboro, ME, July 15, 2015.

2015 Weed Management Research and Demonstration Plots. Wild Blueberry Summer Field Day & Meeting, Jonesboro, ME, July 15, 2015.

Maine Wild Blueberry Crop Estimate. Wild Blueberry Producers of Nova Scotia Summer Meeting, Great Village, NS, July 18, 2015.

Maine's Wild Blueberry Industry. Onawa Lake, ME, July 25, 2015.

Wild Blueberry Production in Maine. WBANA blogger immersion tour, Bar Harbor, ME, August 16-19, 2015.

Wild Blueberry Production in Maine and Overview of Integrated Crop Management. Legislative Tour, Machias and Jonesboro, ME, August 21-22, 2015.

Overview of Integrated Crop Management. Maine Board of Pesticides Control at Blueberry Hill Farm, Jonesboro, ME, August 27, 2015.

Wild Blueberry Trade Video. Ellsworth, ME, August 27-28, 2015.

Wild Blueberries in Maine. Big E, Springfield, MA, October 2-4, 2015.

Wild Blueberry Risk Management Strategies. Maine Development Foundation Tour, Cherryfield, ME, October 8, 2015.

Maine's Wild Blueberry Industry. Go Away Tours, Bar Harbor, ME, October 12, 2015.

Evaluation of Fall and Spring Applications of Herbicides Targeting Resistant Weeds in Wild Blueberry Fields. Wild Blueberry Research and Extension Workers Conference. Bar Harbor, ME, October 22, 2015.

2015 Maine Wild Blueberry Crop Report. Wild Blueberry Research and Extension Workers Conference. Bar Harbor, ME, October 22, 2015.

Single vs split applications of post-emergent herbicides for spreading dogbane (*Apocynum androsaemifolium*) control in wild blueberry fields. Wild Blueberry Research and Extension Workers Conference. Bar Harbor, ME, October 22, 2015.

2015 World Production Update. Wild Blueberry Association of North America Annual Meeting, Bar Harbor, ME, October 23, 2015.

Challenges to Establishing Harmonized Maximum Residue Levels (MRLs) for Facilitating Global Trade. Wild Blueberry Association of North America Annual Meeting, Bar Harbor, ME, October 23, 2015.

2015 World Production Update. Wild Blueberry Association of Nova Scotia Annual Meeting, Truro, NS, November 20-21, 2015.

2015 Maine Wild Blueberry Crop Report. Wild Blueberry Association of Nova Scotia Annual Meeting, Truro, NS, November 20-21, 2015.

Publications:

Yarborough, D.E. and J. Cote. 2015. Evaluation of fall and spring combinations of preemergence herbicides to prevent weed resistance in wild blueberry fields. Proceedings of the Northeastern Weed Science Society 69:42.

Yarborough, D.E. and J. Cote. 2015. Evaluation of fall and spring applications of herbicides targeting resistant weeds in wild blueberry fields. Wild Blueberry Research and Extension Workers Conference Proceedings. (Abstract 10).

Yarborough, D.E. and J. Cote. 2015. 2015 Maine Wild Blueberry Crop Report. Wild Blueberry Research and Extension Workers Conference Proceedings. (Abstract 14).

Cote, J. and Yarborough, D.E. 2015. Single vs split applications of post-emergent herbicides for spreading dogbane (*Apocynum androsaemifolium*) control in wild blueberry fields. Wild Blueberry Research and Extension Workers Conference Proceedings. (Poster Abstract 2).

Wild Blueberry Fact Sheets – 2015:

New:

Enhancing Wild Bees for Crop Pollination: Sowing Bee Pasture for New England's Wild Lowbush Blueberry by Eric M. Venturini, Francis A. Drummond, Aaron K. Hoshide, Lois Berg Stack & Alison Dibble (PDF).

Revised:

Fact Sheet #209 2015 Insect Control Guide for Wild Blueberries
Fact Sheet #239 2015 Weed Control Guide for Wild Blueberries
Fact Sheet #219 2015 Disease Control Guide for Wild Blueberries
Fact Sheet #220 Wild Blueberry Culture in Maine
Fact Sheet #216 Flower Primordia Development Stage with Temperature Tolerance
Using Irrigation Systems for Frost Protection
Agrichemical and Fertilizer Suppliers in Maine
Wild Blueberry Crop Statistics web page
2015 Maine Wild Blueberry Pesticide Chart 1 of 3 Insecticides
2015 Maine Wild Blueberry Pesticide Chart 2 of 3 Fungicides
2015 Maine Wild Blueberry Pesticide Chart 3 of 3 Herbicides

Wild Blueberry Website:
The Wild Blueberry website found at http://www.wildblueberries.maine.edu continues to be updated and has been revised to comply with the University of Maine content management system. It received 140,704 page views in 2015 and so is well used world-wide. The wild blueberry blog is being used to update growers on current activities including insect (both pollinator and SWD), and disease (mummy berry monitoring) posts at: http://mainewildblueberries.blogspot.com/

***Other program activities*:**
I am the principal investigator for the NIFA *Sustainable Production of Wild Blueberries*, which provided funds for a five year (2009-2014) multidisciplinary systems approach project for wild blueberries. We continued the project one additional year without funding to obtain a third crop cycle for the study and are compiling the results. I am responsible for obtaining, compiling and producing the proposals and reports for the Wild Blueberry Advisory Committee and a HATCH and NRSP4 Project reports for the REEport on-line database. I am the principal investigator on a Specialty Crop Block Grant (SCGB) *Improving Integrated pest management (IPM) to prevent weed resistance for Maine wild blueberry growers* (2015-2016) and as a co-investigator with Frank Drummond and Seanna Annis on a SCGB project: *Improving Integrated Pest Management Practices for the wild blueberry crop in Maine* (2014-2015).
I serve as the liaison for Maine in the IR-4, Minor Use Registration Program and convey project needs for all crops, as well as conduct projects. The objective of the program is to register least toxic alternative pesticides to replace materials that have been canceled so that our growers will be able to keep the minor crop production practices viable in Maine. I report on the wild blueberry crop to the New England Agricultural Statistics Service (NASS) on a weekly basis during the wild blueberry-growing season. NASS uses the information to provide updates on the web for the wild blueberry crop for all who are interested.
I serve as chair for all full professor promotions. I also serve on the graduate committee of: Alex Bajcz PhD student, Major advisor F. Drummond 2013 to present; Honors Thesis, Steven Valentino and Tyler Case M.S. student, Major advisor Seanna Annis 2015-2016. I serve on the peer committee for the School of Food and Agriculture and I am Chair of the AVS committee to hire a new faculty in Animal and Veterinary Science 2015 to 2016.

CONCLUSIONS: Growers are participating in IPM programs in the four primary wild blueberry growing counties: Washington, Hancock, Knox and Lincoln. The skills survey results indicate that growers are learning new skills and making positive changes in their management practices. A high percentage of participating growers indicated they had learned new skills and changed their practices in calibration, thereby reducing the rate of hexazinone used, being able to control blight, identifying and control weeds, being able to detect and control insects and the blueberry maggot fly, and using soil and leaf samples to determine fertilizer rates. Adoption of these management practices will enable growers to improve the efficiency of blueberry culture by reducing unnecessary pesticides and fertilizers. Developing alternative strategies for control of resistant weeds is necessary to prevent future losses in yield from weed competition. The introduction of the new pest, the spotted wing Drosophila, will present additional challenges in monitoring, identification and control to prevent losses from this pest. These practices are essential to counter the perception of the anti-pesticide and the anti-aerial spray protests that have taken place and intensified in recent years.

The most recent survey conducted from the newsletter mailing list indicates that growers need the information provided by the meetings, fact sheets and newsletters. It also indicates that many growers are using integrated management techniques. Adoption of Best Management Practices will enable growers to improve the efficiency of blueberry culture by reducing unnecessary pesticides and fertilizers. More efficient management will result in greater returns and a stable, sustainable industry.

RECOMMENDATIONS: Continue to support the Extension program to provide for the continuation of research based knowledge to be delivered to wild blueberry growers in Maine. Growers benefit in maintaining efficient production practices that allow them to be competitive with cultivated and Canadian production, and the public will benefit from production practices that allow growers to produce wild blueberries at an affordable price and volume so that consumers will be able to afford to eat more healthy wild blueberries. The benefits of a healthier society are incalculable.

INPUT SYSTEMS STUDY – SCRI GRANT OVERVIEW

13. TITLE: Systems approach to improving the sustainability of wild blueberry production, Year Six of a six-year study – experimental design.

EXPERIMENTAL DESIGN*:* In spring of 2010, a four-year study of the effects of different blueberry cropping input systems on a. crop growth, yield, quality and food safety, b. pest levels/dynamics and level of risks to growers, c. soil health, and d. economic and ecological costs/benefits was initiated. Overviews of the first five years of the study are presented in Report #19 of the 2010 Project Reports, Report #15 of the 2011 Project Reports, Report #13 of the 2012 Project Reports, Report #12 of the 2013 Project Reports and Report #11 of the 2014 Project Reports. In 2012, the study design was changed slightly for the second crop cycle of the project in order to give better representation of the ranges in variables examined in each management system, as well as greater statistical power. The second crop cycle had issues with grower cooperation and grower data submission. Therefore, the principal investigators decided a third crop cycle was necessary, and the study was extended for an additional crop cycle of data collection with some growers replaced.

In the second cycle, two one-acre blocks in four input systems (Organic and Low, Medium and High input conventional systems) were set up at four sites per management system for a total of eight blocks per system. We used the same two sites per system as in the first crop cycle but eliminated two blocks each; the two remaining blocks retained the original block designations. The other four blocks were set up two each on two additional sites, and growers were asked to perform their usual activities within these plots as part of the larger field landscape.

In the third cycle, the original two Low fields from the first cycle were replaced by two fields managed by a grower in the Union area; the two fields from the second cycle were retained. The Medium system retained the two original sites of the first cycle, but the sites added in the second cycle were replaced by two fields managed by two growers in the Union area. Three Organic growers were replaced due to management and record-keeping issues, leaving one grower common to all three cycles. There were no High input growers in mid-Maine, so all High sites

remained the same as in the second cycle. The "typical" management input parameters for each system, as determined at the start of the project, are presented in Table 1.

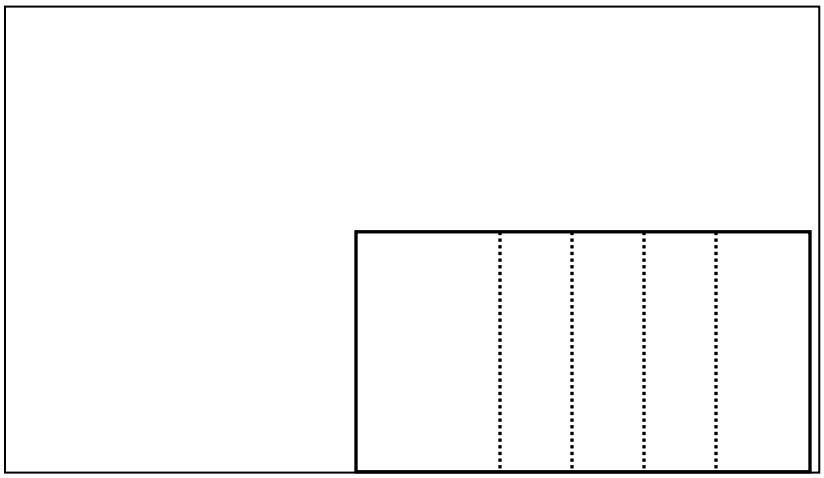
Table 1. Typical levels of inputs in four management systems for the production of wild blueberries.

	Management input systems			
Production Factors	**Organic**	**Low Input**	**Medium Input**	**High Input**
Pruning	Burned	Burn	Mowed	Mowed
Land leveling	Not land leveled	Not land leveled	Land leveled	Land leveled
pH management	pH managed	No pH management	pH managed	pH managed
Fertility	No fertilizer	No fertilizer	Reduced Fertility (every other cycle)	Fertility optimal
Pest, disease, and weed control	Cutting woody weeds	Herbicide, blueberry maggot, mummyberry control with standard pesticides	Scouting, standard and reduced risk pesticides	Scouting, reduced risk pesticides
Treatment of bare spots	Mulch	No mulch	No mulch	Mulch
Irrigation	No irrigation	No irrigation	No irrigation	Irrigation
Pollination	Bees 2 hives/acre	No added bees	Bees 2 hives/acre	Bees 6 hives/acre
Harvest method	Hand raked	Hand raked	Mechanical Harvest	Mechanical Harvest

Two one-acre blocks each were maintained on the following sixteen sites. The sites in the second cycle were numbered 1-16; replacement sites were numbered from #17:
Organic: Fields 1, 17, 18, 19;
Low input: Fields 7, 8, 20, 21;
Medium input: Fields 9, 10, 22, 23;
High input: Fields 13-16;

Each one-acre block contained a "sub-block" with four transects (Figure 1):

Figure 1. Example layout of an acre block, sub-block 15 x 30 m and 15 m transects.



METHODS: The following grower inputs were made to each system in 2015 and are found in Table 2.

Sampling

Weed cover assessment along the transects was conducted just prior to harvest in 2015. Disease assessments were conducted along the transects in May and across the entire block in September. Insect sampling was conducted multiple times per month from May through August over the entire block. In the fall of 2015, all growers were contacted for their crop year inputs and costs in order to build a preliminary partial budget spreadsheet for the two-year crop cycle, which is presented in Report #17.

Yield

All sites were harvested between July 27 and August 11, 2015 in order to remove all stakes and flags so that the growers could harvest the blocks. The blocks were harvested along the 15 m long transects, and went through the 1 m^2 weed cover plots. The Medium and High sites were harvested using two walk-behind harvesters with 2' wide heads, and the Low and Organic sites were harvested by hand using rakes of the same width as the harvesters. The berries were weighed on-site using two analog tray scales, and were not winnowed prior to weighing. Yields were converted to lbs/a; yield by system was analyzed using a Tukey's test (α=0.05), while yield by site was compared using the Standard Error of the Mean.

In the two previous crop years, composite subsamples of were brought back to UMaine for taste, color, nutrient analysis and for pathogens by the Food Science Department; their portion of the study ended in 2013, so subsamples were not collected in 2015.

Table 2. 2015 crop year inputs by site.

Input	Site	Pollination	pH	Fertility	Pest control	Disease control	Weed control	Irriga-tion
Organic	1	1.5 hives/a honeybees	Sulfur 2000 lb/a	None	None	None	Hand weeding, tree trimming	No
	17	1 quad/a bumblebees	None (400 lb/a S applied 2011)	Foliar spray worm leachate 1 gal:10 gal water= 50 gal over 1 ac	Scouting	Scouting	Hand weeding	No
	18	1 hive/a honeybees	None (2011)	None	Minimal scouting	Minimal scouting	Hand weeding, brush cutter	No
	19	0.8 hive/a honeybees 0.54 quad/a bumblebees	None	None	None	None	Hand weeding	No
Low	7	2.4 hive/a honeybees	None	CoRoN 1 gal/a	Imidan 1.3 lb/a	Proline 5.7 oz/a Fitness 6 oz/a	None	No
	8	1.5 hive/a honeybees	None	None	Imidan spot spray 1.3 lb/a	None	None	No
	20	2.1 hives/a honeybees 0.1 quad/a bumblebees	None	CoRoN+B 1 gal/a	Imidan 1.3 lb/a	Fitness 6 oz/a Pristine 18.5 oz/a	None	No
	21	2.6 hives/a honeybees 0.1 quad/a bumblebees	None	CoRoN+B 1 gal/a	Imidan 1.3 lb/a	Fitness 6 oz/a Pristine 18.5 oz/a	None	No
Medium	9	3 hives/a honeybees	None	Neptune's Harvest Fish/Seaweed 0.97 gal/a Calcium Nitrate 1.89 lb/a	Scouting Malathion 8F 1.28 pt/a Imidan 70W 1.03 lb/a	Tilt 6 oz/a (2x)	Poast 2.36 pt/a	No

Input	Site	Pollination	pH	Fertility	Pest control	Disease control	Weed control	Irriga-tion
Medium	10	3 hives/a honeybees	None	Neptune's Harvest Fish/Seaweed 0.92 gal/a Calcium Nitrate 1.79 lb/a	Scouting Imidan 0.9 lb/a	Tilt 6 oz/a	Poast 2.24 pt/a	No
	22	4 hives/a honeybees	None	None	Fly trapping	None	Mad Dog glyphosate spot spray 12 oz/a	No
	23	2 hives/a honeybees	None	CoRoN 1 gal/a	Imidan 1.3 lb/a	Tilt 6 oz/a	None	No
High	13	6 hives/a honeybees	None	None	Scouting Assail 4.25 oz/a Success 5 oz/a	Tilt 6 oz/a (2x) Pristine 20 oz/a	Poast 1 qt/a	Yes
	14	6 hives/a honeybees	None	None	Scouting Assail 4.25 oz/a Success 5 oz/a	Tilt 6 oz/a Proline 5.7 oz/a Pristine 20 oz/a	None	Yes
	15	6 hives/a honeybees	None	None	Scouting Assail 4.25 oz/a	Tilt 6 oz/a Proline 5.7 oz/a Pristine 20 oz/a	Poast 1 qt/a	Yes
	16	6 hives/a honeybees	None	None	Scouting Assail 4.25 oz/a Success 5 oz/a	Tilt 6 oz/a Pristine 20 oz/a Switch 11 oz/a	None	Yes

RESULTS/CONCLUSIONS:

Sampling

The results of each researcher's assessments are presented in their respective individual reports.

Yield

The yield results by system and site are presented in Figures 2-3. There were no significant differences in yield among systems, despite a range of almost 4,400 lbs/a (Figure 2); this is likely due to the large amount of variation in yields from site to site (Figure 3). Some researchers have also presented yield results in their individual reports as they relate to the researchers' respective project aspects (weeds, insects, disease, etc.).

The winter of 2014-15 was snowy and cold. Snowpack was sufficient in most fields but some plants were exposed by wind sweeping away snow across larger fields; spring also came late in 2015. The spring and summer through the end of August were cool, and May, July and August were very dry and although June rainfall average was sufficient, most of it came in one event (Figure 5), which slowed plant and fruit development. These environmental conditions led to field-level widespread damage to the blueberry plants; in some Medium and High fields (Fields 9, 10, 13, 14, and 23), vegetative growth was vigorous but there were little to no flower buds or fruit so yield was lower than normal for these fields, which reduced the system average (Figures 2-3). Figure 4 shows the yield by system for all three crop years in the project; in 2011 and 2013 there was a trend of increasing yield with increasing inputs. In 2015, this trend did not hold, mostly because the damaged fields tended to be Downeast where all of the High sites and two of the three damaged Medium sites were located. By contrast, the dry summer led to a large increase in yield for Low field #7 (see Figure 3); this field is poorly drained and the blueberry plants are normally short, occur only on hummocks and are low yielding. The dry summer resulted in this field being more productive, which increased the Low system average yield to a level similar to typical yields in the Medium and High systems (Figure 4). The Organic system had over twice the yield in 2015 as it did in 2011 and 2013 (Figure 4). This appears to be mostly due to improved management by the replacement organic growers in this production cycle, whose yields were equivalent to the Low input sites and were greater than the damaged Medium and High sites (see Figure 3 fields 17 and 19).

Figure 2. 2015 wild blueberry yields by input system (different letters denote significance at α=0.05).

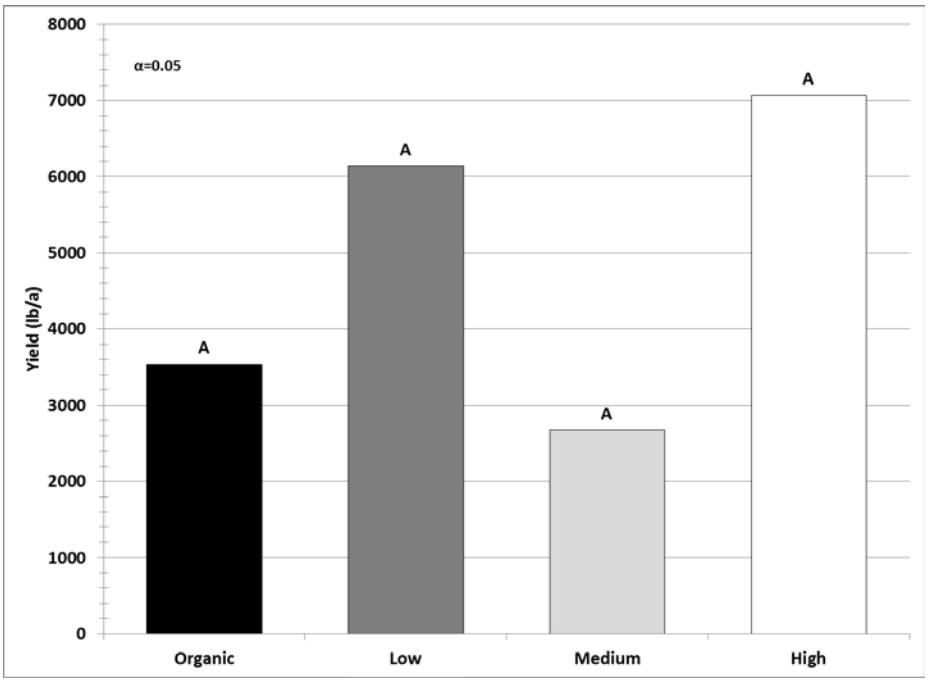


Figure 3. 2015 wild blueberry yields by site (Std. Error of Mean).

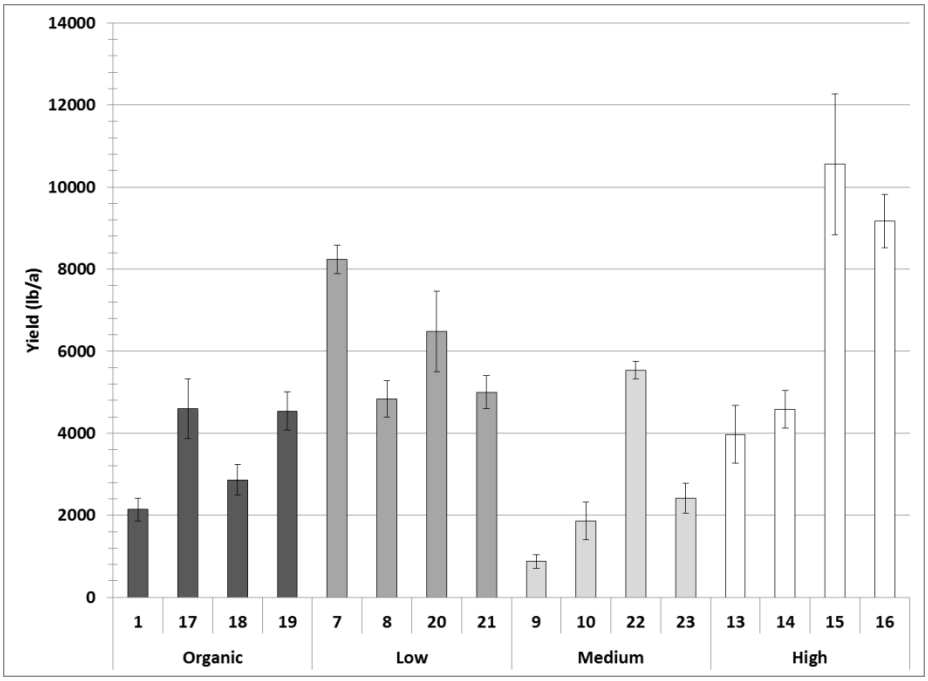


Figure 4. Yield by system for 2011, 2013 and 2015.

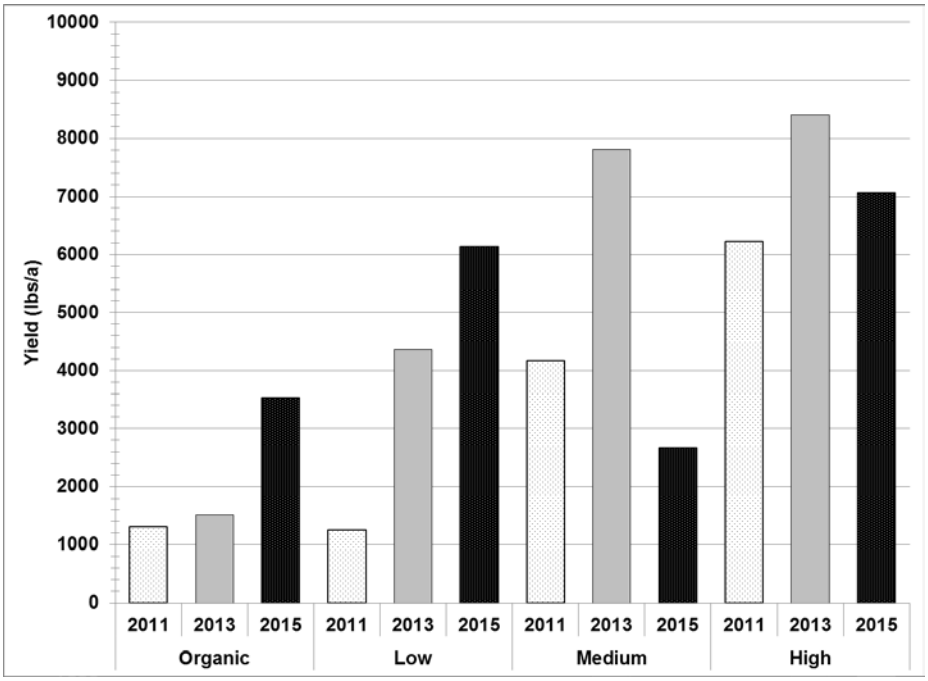
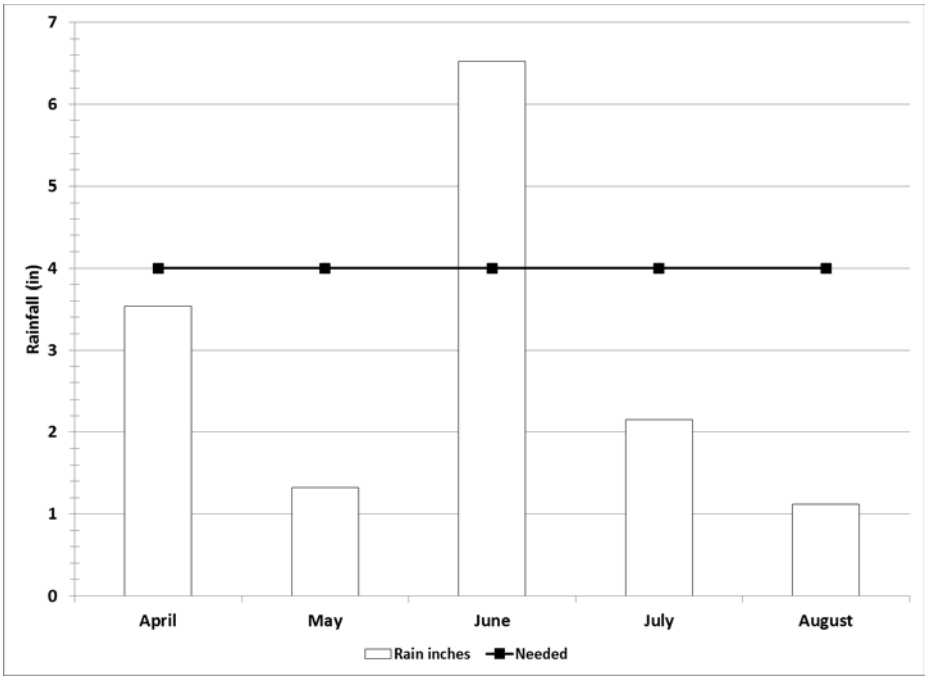


Figure 5. Monthly rainfall totals at Blueberry Hill Farm for 2015.



In the previous two crop years we stated that the lack of a difference in yield between the Medium and High input systems suggested that there is a point of diminishing returns where adding more inputs to a system no longer results in significantly improved yield, and that this point lies somewhere in the range of inputs between these two systems. The third crop year

illustrated that in the wild blueberry production system, growers may be exposed to a higher level of weather related risk, and this risk can negate all efforts at protecting and improving the crop. This higher level of risk due to exposure to weather and pests cannot be eliminated or counteracted by any type or level of input. Losses due to weather can be complete and catastrophic, as was illustrated this year by the High system losing money in two out of four fields despite having the most inputs and historically very profitable fields, as well as the Medium system losing money in all four fields (see partial Enterprise budgets in Report #17).

RECOMMENDATIONS: Recommendations regarding each component of the project, such as disease, insects, weeds, etc. are presented in each researcher's respective reports. Recommendations pursuant to overall yield are presented in the partial Enterprise budgets report. This is the final year of this study. All data from all components of the study will be compiled and analyzed. Predictive models for calculating interactions between components of the wild blueberry production system pursuant to inputs will be generated to determine the relative importance of each, and an interactive budget will be developed for to growers so that they can estimate monetary returns for different levels and combinations of inputs.

INPUT SYSTEMS STUDY

ENTOMOLOGY: Frank Drummond, Professor of Insect Ecology/Entomology
Judith Collins, Assistant Scientist of Insect Pest Management
Elissa Ballman, Research Associate in Invasive Species/Entomology

14. TITLE: Systems approach to improving the sustainability of wild blueberry production, Year 6.

Blueberry stem measurements

METHODS: Between 4 and 8 May, 2015, all the stems from each of ten, 15.2 x 15.2 cm (6 x 6 in) quadrats per field were cut at ground level, brought into the laboratory, and counted to determine stem density, stem length, and branching. Ten stems were randomly selected from each sample to determine the number of flower-bud clusters per stem.
Analysis of Variance (CRD) and LS Means Differences ($P \leq 0.05$) were used to compare stem density, stem length, flower-bud clusters per stem, and branching among the treatments. Subplots were pooled within main plots. Data was transformed by using the square root to stabilize variance prior to analysis.

RESULTS: Stem density did not vary significantly among the production system treatments ($F_{(3,12)} = 2.80$, $P = 0.085$)(Figs. 1 & 2). Significantly more flower-bud clusters were found on stems from the high input sites ($F_{(3,12)} = 8.22$, $P = 0.0031$)(Figs. 3 & 4). There was also a significant difference in stem length (Figs. 5 & 6). Stems from high and medium input sites were taller than those from low or organic input sites ($F_{(3,12)} = 4.50$, $P = 0.0246$). There was not a significant difference in branching ($F_{(3,12)} = 1.37$, $P = 0.3004$)(Figs. 7 & 8).

Fig. 1. The mean stem density, by site, for each production system. Line segments are standard error of the mean.

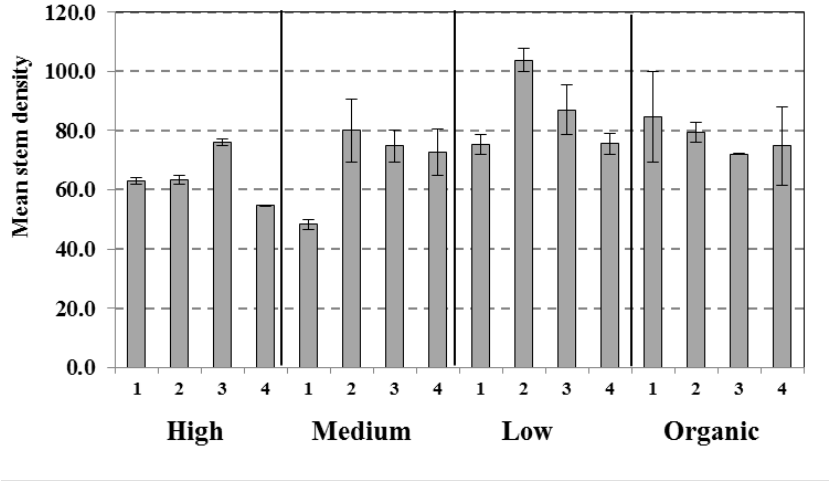


Fig. 2. The mean stem density, by production system. Line segments are standard error of the mean.

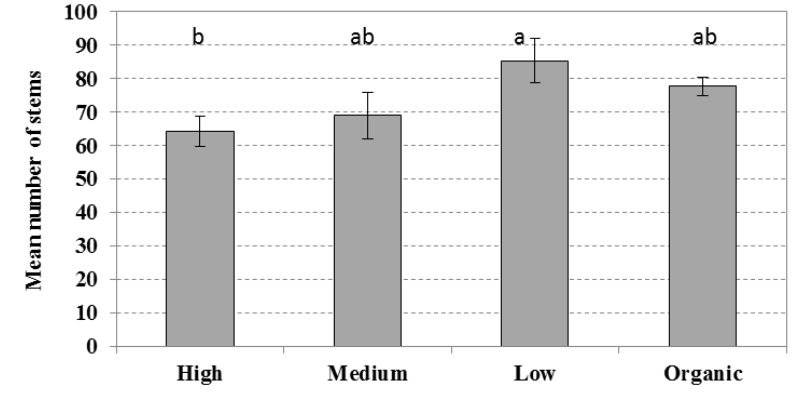


Fig. 3. The mean number of flower-bud clusters per stem, by site, for each production system. Line segments are standard error of the mean.

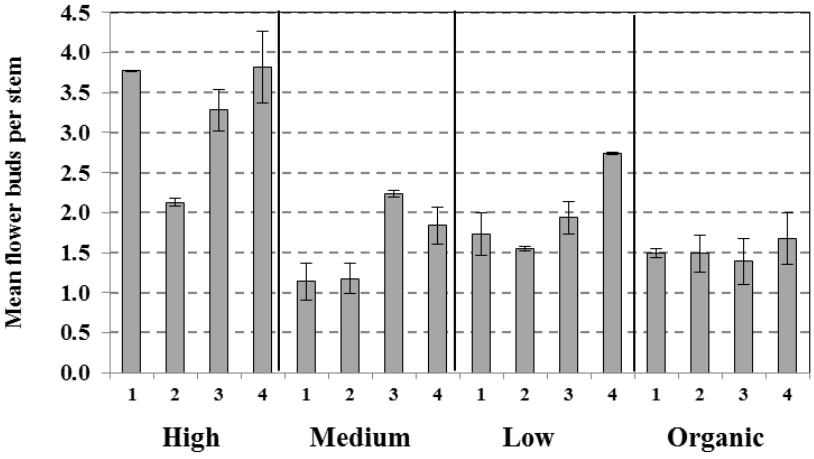


Fig. 4. The mean number of flower-bud clusters per stem, by production system. Line segments are standard error of the mean.

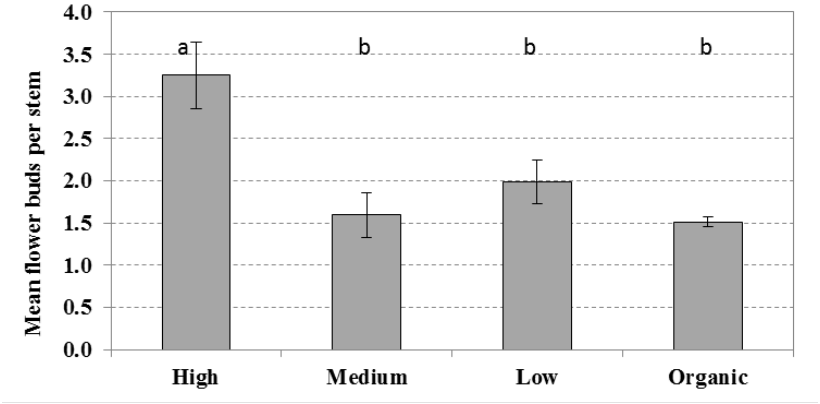


Fig. 5. The mean stem length (cm), by site, for each production system. Line segments are standard error of the mean.

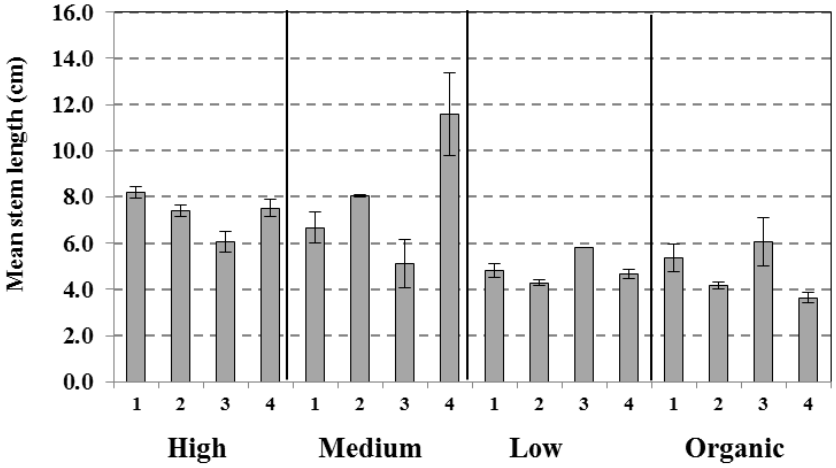


Fig. 6. The mean stem length (cm), by production system. Lines are standard error of the mean.

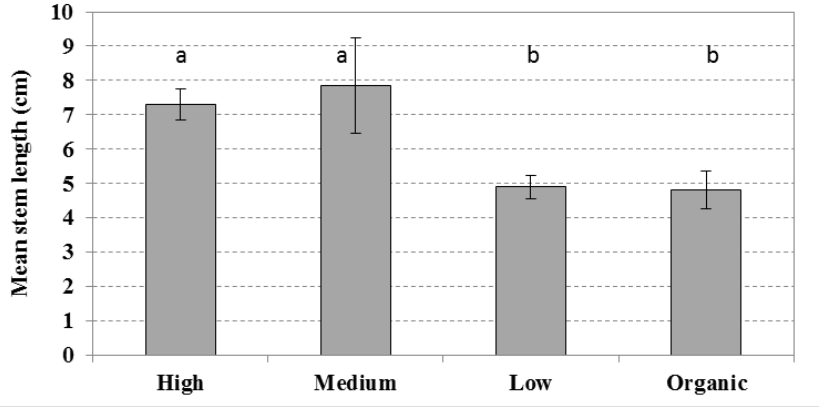


Fig. 7. The number of branches per stem, by site, for each production system. Line segments are standard error of the mean.

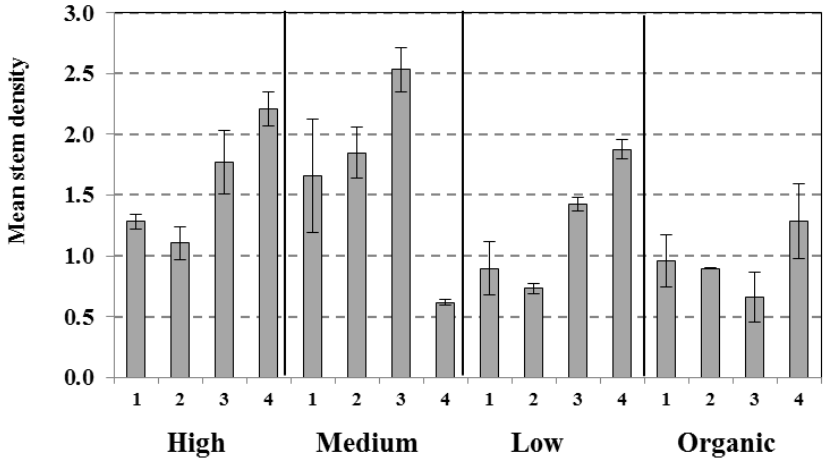
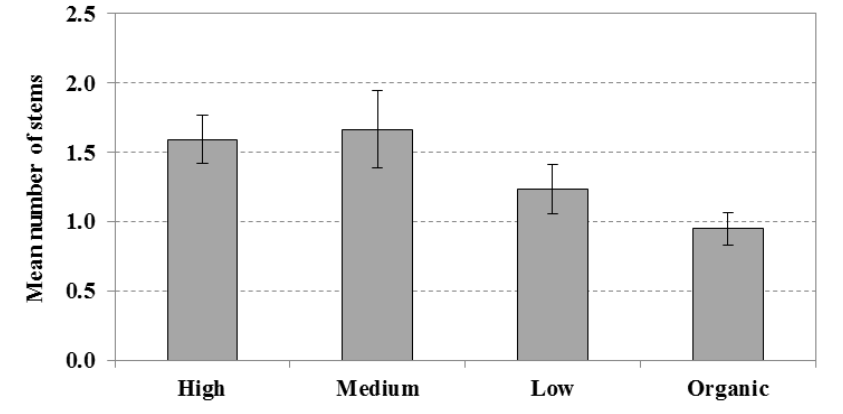


Fig. 8. The mean number of branches per stem, by production system. Line segments are standard error of the mean. No significant differences among the treatments, thus no letters on bars.



Soil samples

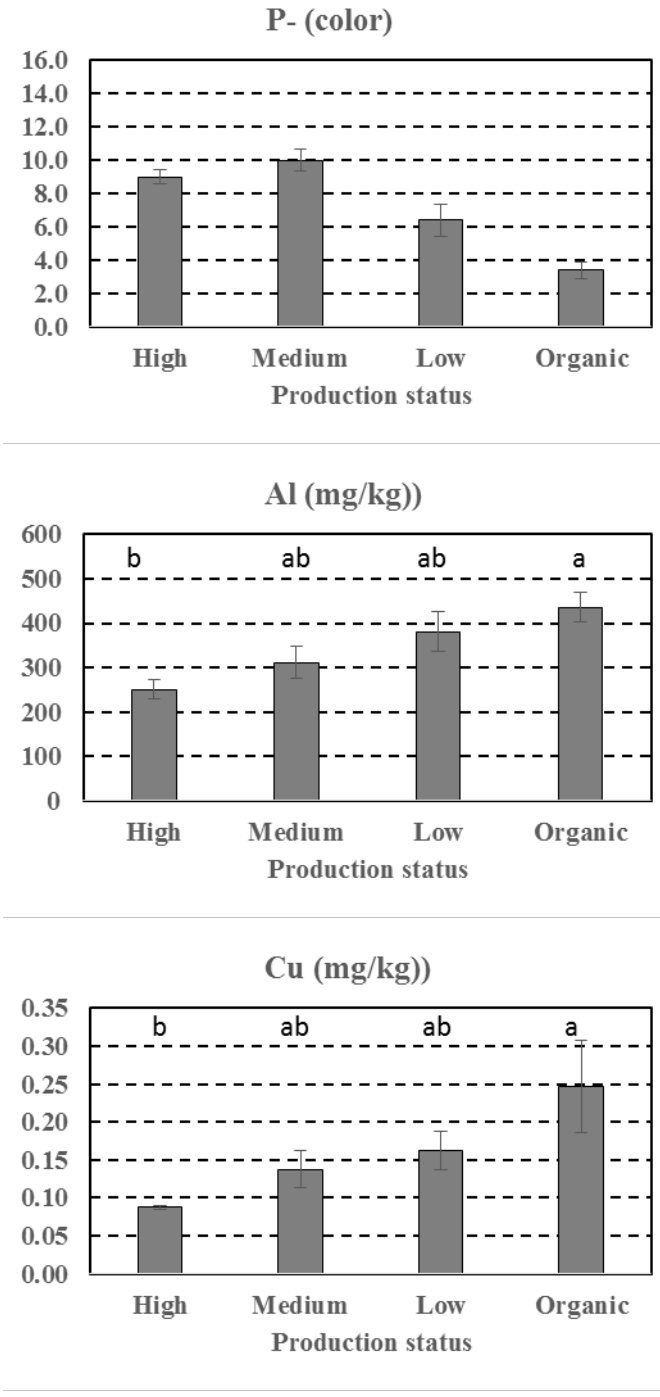
METHODS: Soil samples were collected from each site. Samples were sent to the Maine Soil Testing Service for Analysis. Soil pH was measured in distilled water. Organic matter was measured by loss on ignition (LOI) at 375^{o}C. Nutrients were extracted in pH 4.8 ammonium acetate (modified Morgan extract). P was determined via colorimetry by an Ion Analyzer. All other nutrients were measured by ICP-OES. Effective cation exchange capacity (ECEC) is calculated by summation of base cations plus readily exchangeable acidity. Unless otherwise specified, all nutrients are expressed as parts per million in the dry soil. The results of the soil analysis are shown in Table 1. Analysis of Variance (CRD) and LS Means Differences ($P \leq 0.05$) were used to compare soil nutrient levels among the treatments. Subplots were pooled within main plots. Data were transformed by using the square root to stabilize variance prior to analysis.

RESULTS: A multivariate analysis of the soil fertility measures suggested that there was no evidence for differences in overall soil health when all measures were taken into account simultaneously ($F_{(3,12)} = 1.485$, $P = 0.268$)(Table 1). There was marginal evidence of a production system x treatment interaction suggesting that specific production systems were higher or lower in specific nutrients or soil characteristics than others ($F_{(51,204)} = 1.303$, $P = 0.102$, corrected for autocorrelations by Greenhouse-Gier factor). Subsequent exploration of individual soil characteristics and nutrients suggested that Phosphorous (as determined via colorimetry) was lower in the organic system compared to the low, medium, and high systems ($F_{(3,12)} = 19.36$, $P = 0.0001$). Aluminum ($F_{(3,12)} = 5.35$, $P = 0.0143$) and copper ($F_{(3,12)} = 3.60$, $P = 0.0462$) were higher in the organic system compared to the high input system (Fig. 9).

Table 1. Soil sample analysis.

											mg/kg								meq/100 gn
Field #	**Status**	**soil pH**	**buffer pH**	**% LOI**	**Ca**	**K**	**Mg**	**P-ICP**	**P-color**	**Al**	**B**	**Cu**	**Fe**	**Mn**	**Na**	**S**	**Zn**	**acidity**	**ECEC**
1	Organic	4.2	4.76	13.0	86	92	25	11.0	2.5	421	0.32	0.27	106	22	20	150	1.6	4.7	5.7
7	Low	4.6	5.11	9.3	69	60	17	8.4	3.5	457	0.18	0.13	41	16	16	96	1.7	3.5	4.2
8	Low	4.8	5.12	9.4	185	79	36	14.8	7.5	365	0.14	0.19	18	24	10	72	4.9	3.3	4.8
9	Medium	4.4	4.60	12.1	323	73	59	12.5	9.4	205	0.13	0.09	14	19	29	91	3.2	5.0	7.4
10	Medium	4.6	4.79	12.6	176	56	41	14.7	10.5	336	0.17	0.10	22	15	32	49	3.0	4.3	5.8
13	High	4.4	4.47	12.0	258	78	43	13.4	9.1	262	0.28	0.09	28	7.9	31	33	2.8	5.3	7.2
14	High	4.3	4.51	12.4	197	79	45	14.0	9.9	224	0.25	0.09	16	4.8	26	44	2.5	5.3	6.9
15	High	4.2	4.44	12.2	203	73	56	11.5	7.9	211	0.18	0.09	25	4.9	25	45	3.3	5.5	7.3
16	High	4.5	4.70	12.2	256	83	52	13.5	9.1	306	0.21	0.08	29	11	25	59	2.8	4.6	6.6
17	Organic	4.4	4.57	15.0	103	61	27	7.9	3.1	533	0.18	0.14	50	8.2	16	147	1.4	5.1	6.0
18	Organic	5.1	5.29	9.2	211	75	39	9.6	4.8	381	0.14	0.17	23	11	10	54	1.5	2.7	4.3
19	Organic	4.4	4.80	11.9	172	112	34	14.6	3.2	409	0.33	0.41	54	57	32	159	3.7	4.5	6.1
20	Low	4.6	4.53	20.4	711	119	134	13.8	7.4	260	0.28	0.11	49	43	42	19	5.0	4.9	10.1
21	Low	4.6	4.93	11.2	134	55	24	14.6	7.2	440	0.25	0.22	52	32	10	105	2.3	3.9	5.0
22	Medium	4.8	5.00	11.5	341	98	69	15.2	8.5	340	0.22	0.18	37	26	17	53	3.0	3.6	6.2
23	Medium	4.3	4.76	11.8	111	86	28	20.1	11.6	364	0.18	0.18	25	30	12	148	2.0	4.6	5.7
	measured->		6.0	6.02	4.8	1090	230	114	16.9	15.0	45	0.21	0.83	2.3	34	9.3	14	3.3	
	OK range->		*5.8-6.0*	*6.00-6.04*	*4.2-4.8*	*50-115*	*185-245*	*106-120*	*16-19*	*14-16*	*40-48*	*0.15-0.25*	*0.7-0.9*	*1.5-2.5*	*32-38*	*8-12*	*12-15*	*2.0-2.5*	

Fig. 9. Comparison of levels of soil phosphorous (as determined via colorimetry), Al, and Cu among the production systems. Line segments are standard error of the mean.



Leaf nutrients

METHODS: Samples of the foliage were collected from each clone at peak bloom. Ten stems were randomly collected, flowers were removed and the leaves were collected and dried for analysis by the University of Maine MAFES Soils Laboratory. Multiple analysis of variance was used to assess differences in leaf nutrient profiles as affected by the production system. The leaf nutrient data as well as the soil fertility data will be used at a later date to assess the contribution of nutrition to yield after flower density and pollinator density are accounted for.

RESULTS: The foliar nutrients (Table 2) did show evidence for a production systems effect ($F_{(3,12)} = 3.498$, $P = 0.0497$). The organic system had higher nutrient levels in foliage than the high input system. Overall, the ranking of nutrients from high to low was: organic, low, medium, high. The low ranking of the high input system is not surprising considering that there was no fertilizer applied in 2015 compared to many of the fields in the other production systems (Table 3, next section).

Table 2. Analysis of foliar nutrients.

		%	%	%	%	%	ppm	ppm	ppm	ppm	ppm	ppm
Field #	**Status**	**N**	**Ca**	**K**	**Mg**	**P**	**Al**	**B**	**Cu**	**Fe**	**Mn**	**Zn**
1	Organic	2.2358	0.2727	0.6467	0.1215	0.2588	63.842	14.175	6.9492	50.358	2025	22.2
7	Low	2.3083	0.2404	0.7311	0.1202	0.2793	60.842	19.908	7.1867	51.308	1611.3	25.308
8	Low	2.205	0.2686	0.635	0.1359	0.2623	51.592	18.733	6.5133	38.392	1360	23.8
9	Medium	2.1267	0.3118	0.5749	0.1337	0.2404	39.25	15.175	5.7908	36.592	576.5	26.067
10	Medium	1.9725	0.2501	0.5659	0.1332	0.2481	34.117	19.358	6.7733	36.367	648.75	25.817
13	High	2.0908	0.2861	0.5947	0.1507	0.2384	49.875	24.725	6.2925	53.9	449.08	31
14	High	1.9117	0.2836	0.5933	0.1491	0.2346	35.025	25.192	5.7567	40.183	162.19	25.475
15	High	1.8825	0.2954	0.5263	0.1596	0.2164	46.717	21.833	5.8008	47.567	363.75	22.508
16	High	1.9783	0.3071	0.5737	0.1512	0.2222	45.358	20.992	5.51	45.283	505.08	24.475
17	Organic	1.8617	0.3733	0.5904	0.1547	0.2166	66.633	20.225	6.355	60.458	1077.3	20.175
18	Organic	1.6383	0.3955	0.6075	0.1709	0.1842	58.867	17.15	5.6892	46.392	1120.4	18.375
19	Organic	2.335	0.3062	0.6807	0.1343	0.2377	64.742	14.883	6.6992	47.9	2539.2	25.908
20	Low	2.0392	0.286	0.682	0.1633	0.2532	51.983	22.667	5.4075	46.592	716.08	21.542
21	Low	1.8933	0.3222	0.6614	0.1507	0.219	69.508	23.583	5.3358	52.575	2202.5	20.617
22	Medium	2.175	0.2843	0.6037	0.1557	0.2244	52.833	16.975	5.3008	40.992	1079.5	20.175
23	Medium	2.0825	0.3731	0.6955	0.1607	0.207	69.392	29.208	5.7275	49.908	2247.5	19.008

Production Inputs

METHODS: Production statistics were provided by growers cooperating in the Systems Project. Additional information collected on each site included number of bee hives, fertility, and pesticide inputs (insecticides, fungicides, herbicides).

RESULTS: Table 3 is the crop cycle inputs in the systems project, 2015.

Table 3. Additional site information; number bee hives, fertility, and pesticide inputs.

Input	Site	Pollination	Fertility	Pest control	Disease control	Weed control
Organic	1	1.5 hives/a honeybees	None	None	None	Hand weeding, tree trimming
	17	1 hive/a bumble bees	Foliar spray worm leachate 1 gal:10 gal water= 50 gal over 1 ac	Scouting	Scouting	Hand weeding
	18	1.5 hives/a honeybees	None	Minimal scouting	Minimal scouting	Hand weeding, brush cutter
	19	0.8 hives/a Honeybees 0.54 hives/a Bumble bees				
Low	7	2.4 hives/a honeybees	CoRoN 1 gal/a	Imidan 1.3 lb/a	Proline 5.7 oz/a Fitness 6 oz/a	None
	8	1 hive/a honeybees	None	Imidan spot spray 1.3 lb/a	None	None
	20	2.1 hives/a honeybees 0.1 hive/a bumble bees	CoRoN+B 1 gal/a	Imidan 1.3 lb/a	Fitness 6 oz/a Pristine 18.5 oz/a	None
	21	2.6 hives/a honeybees 0.1 hive/a bumble bees	CoRoN+B 1 gal/a	Imidan 1.3 lb/a	Fitness 6 oz/a Pristine 18.5 oz/a	None
Medium	9	3 hives/a honeybees	Neptune's Harvest Fish/Seaweed 0.97 gal/a Calcium Nitrate 1.89 lb/a	Scouting Malathion 8F 1.28 pt/a Imidan 70W 1.03 lb/a	Tilt 6 oz/a (2x)	Poast 2.36 pt/a
	10	3 hives/a honeybees	Neptune's Harvest Fish/Seaweed 0.92 gal/a Calcium Nitrate 1.79 lb/a	Scouting Imidan 0.9 lb/a	Tilt 6 oz/a	Poast 2.24 pt/a
	22	4 hives/a honeybees	None	Fly trapping	None	Mad Dog glyphosate spot spray 12 oz/a
	23	2 hives/a honeybees	CoRoN 1 gal/a	Imidan 1.3 lb/a	Tilt 6 oz/a	None
High	13	6 hives/a honeybees	None	Scouting Assail 4.25 oz/a Success 5 oz/a	Tilt 6 oz/a (2x) Pristine 20 oz/a	Poast 1 qt/a

Input	Site	Pollination	Fertility	Pest control	Disease control	Weed control
High	14	6 hives/a honeybees	None	Scouting Assail 4.25 oz/a Success 5 oz/a	Tilt 6 oz/a Proline 5.7 oz/a Pristine 20 oz/a	None
	15	6 hives/a honeybees	None	Scouting Assail 4.25 oz/a	Tilt 6 oz/a Proline 5.7 oz/a Pristine 20 oz/a	Poast 1 qt/a
	16	6 hives/a honeybees	None	Scouting Assail 4.25 oz/a Success 5 oz/a	Tilt 6 oz/a Pristine 20 oz/a Switch 11 oz/a	None

Blueberry maggot fly monitoring

METHODS: To monitor blueberry maggot fly (BMF), two baited yellow Pherocon® AM traps were placed in each block. Traps were checked at 6 to 9 day intervals. Any captured BMF were counted and removed from the traps. To measure fruit infestation, we raked four quarts of berries from each block. To collect BMF pupae, the berries were combined and distributed in a 1 to 2-inch deep layer in screened boxes suspended over ca. 2 inches (5 cm) of fine sand. Hardware cloth (1/4 in (6.4 mm)) was used as a screening material. In late-October, BMF pupae were separated from the sand. Analysis of Variance (CRD) and LS Means Differences ($P \leq 0.05$) were used to compare seasonal density of adults and the number of pupae per quart of fruit among the treatments. Data for seasonal density of adults was log transformed to stabilize variance prior to analysis. Data for pupae/qt was transformed by the square root.

RESULTS: Overall, throughout the Maine blueberry growing regions, we observed that blueberry maggot fly was very abundant. There was a significant difference in the seasonal density of adults (integration of abundance over the season = # fly days) with organic and high input sites being the highest ($F_{(3,15)} = 5.18$, $P = 0158$)(Fig. 10). This also held true for fruit infestation. Organically-managed sites had significantly more pupae per quart of fruit ($F_{(3,15)} = 4.38$, $P = 0.0267$)(Fig. 11).

Fig. 10. Seasonal density of adults, by production system. Line segments are standard error of the mean.

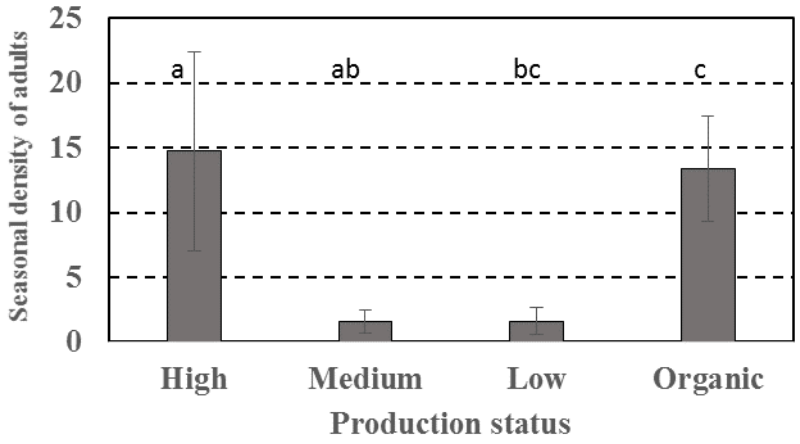
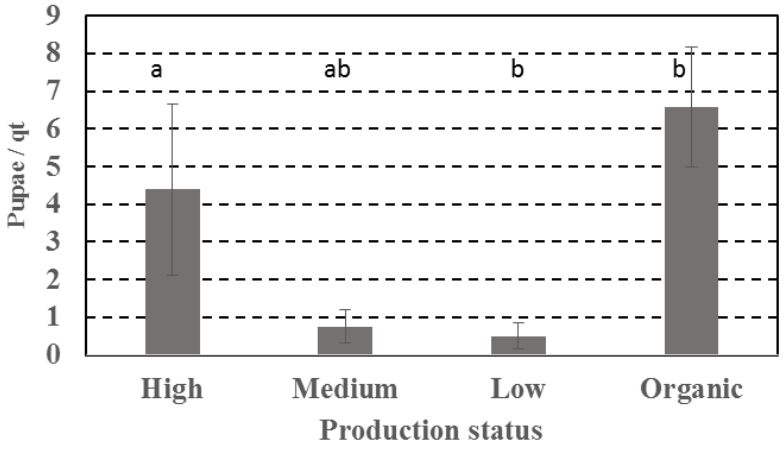


Fig. 11. The mean BMF pupae, by production system. Line segments are standard error of the mean.

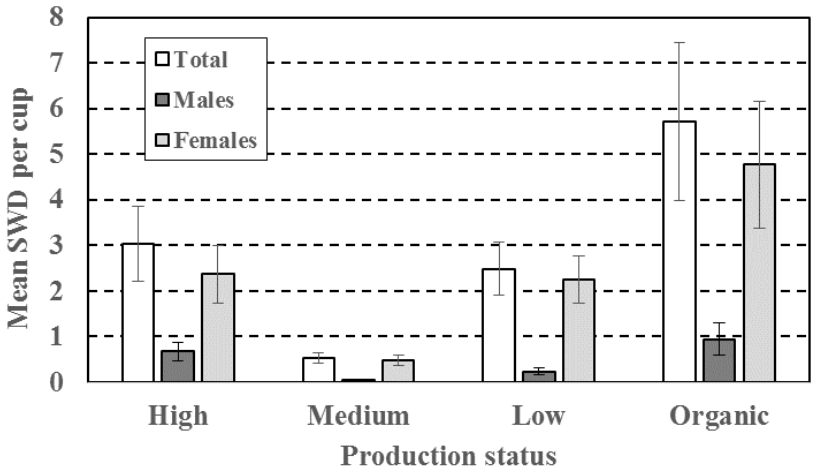


Influence of production system on spotted wing drosophila abundance

METHODS: Beginning on 16 or 24 Jul and continuing until fields were harvested, two traps were set in each block per field (4 traps at each site) and monitored at 3 to 7 day intervals for the presence of spotted wing drosophila (SWD) adults. Four additional trap sites were set on 21 Jul at Blueberry Hill Farm. Traps were constructed from Solo®, 16 fl. oz, red polystyrene cups with opaque lids. Seven to 10, 3/16-inch holes were punched on the side of each container near the top, evenly spaced around the rim. Bait consisted of live yeast (1 tbsp) + sugar (4 tbsp) + 12 oz water (makes enough for 4 traps). The traps were hung 1-2 ft above the top of the canopy using 36' plant stands. On each sample date, traps set from the previous week were collected and returned to the laboratory where male, female, and total abundance of SWD were determined and recorded. Using this data we calculated the mean SWD per trap captured from each site. Analysis of the data was conducted in a similar manner to that described in the blueberry maggot fly section above.

RESULTS: Overall, SWD levels were low prior to harvest in 2015. Figure 12 shows the effect of production system on adult SWD captures for males, females, and total SWD captured. Although there appears to be a trend towards more SWD in organically-managed fields, the difference was not significant (ANOVA, CRD, males, $P = 0.6418$; females, $P = 0.4648$; total SWD, $P = 0.5065$). This does not take into account the effect of any insecticide applications. Information on applications is still being collected.

Fig. 12. SWD adult abundance over the season. Line segments are standard error of the mean.



Abundance of natural enemies and pest insects, sweep-net survey

METHODS: Samples were taken prior to bloom between 4 and 18 May. Twelve sets of ten sweeps each were taken from each field (two, 1 acre blocks/field) with a 12-inch diameter sweep net. Samples were distributed through the blocks (six/block). The number of insects and spiders of each species was counted and then returned to the same plot. Data were analyzed using Analysis of Variance (CRD) ($P \leq 0.05$). Mean separation was by Least Square Means. Subplots were pooled within main plots. Data for blueberry spanworm larvae were transformed by the square root prior to analysis.

RESULTS: Ants and spiders were the most abundant natural enemies (Fig. 13). There was a significant difference in the number of ants between the production system treatments ($F_{(3,12)} = 11.27$, $P = 0.0008$). Significantly more ants were found in the organic compared to low, medium, and high-input systems. There was no significant difference in the number of spiders among production systems ($F_{(3,12)} = 0.99$, $P = 0.4293$). A very small number of lady beetles and ground beetles (both larvae and adults), and click beetle adults were also found in the samples, but the numbers were too low to statistically analyze the data.

Pest abundance was very low, with no pest exceeding threshold numbers during the season. The most abundant pest insects found in sweep-net samples were grasshoppers and blueberry spanworm larvae (Fig. 14). Significantly more spanworm larvae were found in the high input fields ($F_{(3,12)} = 4.69$, $P = 0.0217$). And, although an ANOVA indicated no significant difference, LSD mean separation did indicate that significantly more grasshoppers were present in the low and organic input compared with the medium-input system ($F_{(3,12)} = 3.15$, $P = 0.0647$). Small numbers of strawberry rootworm adults, blueberry leaf beetles, misc. cutworms, and unidentified caterpillars were also found in the samples.

Fig. 13. Relative abundance of natural enemies in sweep-net samples. Line segments are standard error of the mean. No significant differences among the treatments for spiders, thus no letters on bars.

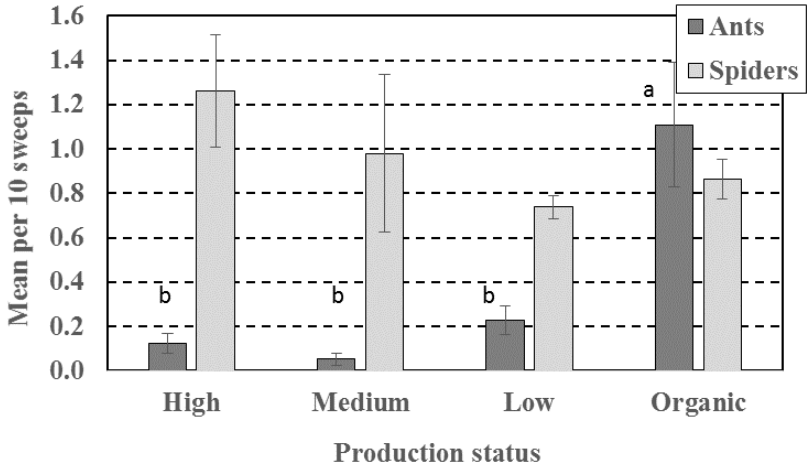
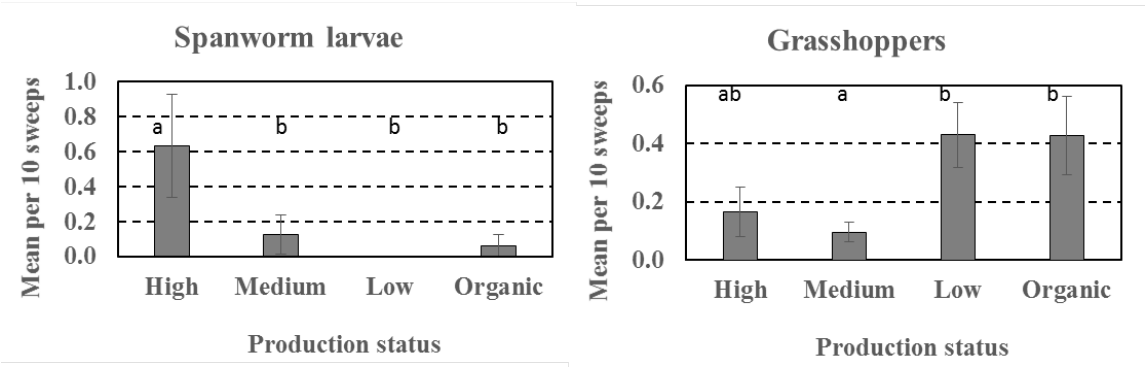


Fig. 14. Relative abundance of pest insects in sweep-net samples. Line segments are standard error of the mean.



CONCLUSIONS: Stem density was higher in low input systems than high input systems, this may be a function of average clone age, but also soil type. However, the number of bud clusters and stem length were greater in high input and high and medium input systems, respectively. Soil fertility did not differ in general among the production systems; although, phosphorous, aluminum, and copper did vary between the organic production system and the three levels of conventional input systems. When all of the data is in a database, analyses will be conducted to assess the influence of stem density, flowers / stem, fruit-set and soil nutrients on yield among the production systems.

Insect pests were affected by production system. Organic farms had higher levels of blueberry maggot fly (BMF) and maggot infestation; however, this was not significantly different from the high input system. The organic systems having higher BMF is not unexpected. The number of maggots in fruit was fairly low for the organic farms, given that no insecticides were used for control. However the high input observation was also not unexpected since a management practice of using BMF thresholds will result in higher non-damaging levels of BMF. This is

borne out by the maggot infestation in the high input system. The damage is at 4 maggots/qt which is the current detection level for maggot infestation (ca. 0.8% infestation).
The trends for SWD were that in general populations were low across Maine in 2015 and no trend in population level in fields was associated with production system. Other patterns that were observed with insects across the production systems were that blueberry spanworm was higher in the high input system. We hypothesize that this might be due to fewer natural enemies. The low level of spanworm in the organic system is most likely due to suppression by natural enemies. On the other hand, while not significant, a trend in grasshopper abundance was seen to be higher in low and organic production systems. This has been observed before and is generally thought to be due to higher levels of grasses in these fields. Grasses are the primary food of most grasshopper species in Maine. Arthropod natural enemies were variable in their response to production system. Spider predators were fairly abundant across all production systems, while ants were predominantly associated with organic production. This corroborates a three-year study by Dr. Beth Choate several years ago. She found similar patterns across production systems.

INPUT SYSTEMS STUDY

DISEASE MANAGEMENT: Seanna Annis, Associate Professor, School of Biology and Ecology
Rachael Martin, Research Asst. School of Biology & Ecology
Tamara Levitsky, Research Asst. School of Biology & Ecology

15. TITLE: Systems approach to improving the sustainability of wild blueberry production, 2015, Year 6 of a six-year study, disease management results.

OBJECTIVE: In 2015 we continued to examine differences in leaf loss and disease levels between field management types (organic, low, medium and high input).

METHODS: Mummy berry blight, caused by *Monilinia vaccinii-corymbosi,* and Botrytis blight (*Botrytis cinerea)* were rated between May 20 to May 29 at each site by block. Within each block, 30 random stems were rated along each of four 45 ft transects. The number of stems with mummy berry, Botrytis, red leaf, top kill and frost damage were counted, and the percentage of affected stems was calculated.
Blueberry cover, leaf loss, leaf spot and stem diseases were rated by block in each field between September 17 and 29, 2015. Within each block, 5 sampling plots of $0.25m^2$ were rated by 2 surveyors visually estimating percentages of blueberry coverage, blueberry leaf loss, blueberry stems with Phomopsis, and blueberry leaf area with the following leaf spot diseases: Septoria leaf spot, powdery mildew, and leaf rust. Any red leaf and False Valdensia disease were also noted. Fall disease ratings were averaged across the surveyors by sampling plot and then across all five sampling plots within a block before analysis. Disease incidence was measured as the percentage of plots with different diseases calculated for each block within a field.
Data were analyzed at the management input level for blueberry cover and disease measures in SAS (Statistical Analysis Software - SAS Cary, NC) using non-parametric one way ANOVA and

then with mixed model procedures (PROC GLIMMIX). Percentage data were arcsine transformed prior to analysis. Least Square means were used to determine specific differences among system types ($\alpha = 0.05$). Data was analyzed at the field level with untransformed data for correlations amongst different measures of disease, blueberry cover, and leaf loss using Spearman's rank correlation in SAS (PROC CORR). Untransformed data is shown in all graphs.

RESULTS: The presence of mummy berry was minimal. Disease levels ranged from less than 1% to 15.5%, with four fields having no symptoms. There were no significant differences in levels of mummy berry between management types (Fig. 1). There was a higher level of disease in the medium input management system, but this was related to two fields which showed the highest levels of mummy berry found. Botrytis symptoms were not found, however the May rating took place before bloom was fully underway, which is when flowers are most susceptible. There were no significant differences in levels of frost, top kill, bare ground or red leaf between management systems (data not shown).

The organic input system had significantly lower blueberry cover than the other input systems (Fig. 2). There was a significant interaction of management type and field where individual fields were sometimes significantly different from other fields of the same input type. The organic fields had the most variation in blueberry cover.

Leaf loss was significantly lower in the low input system compared to the other systems and the high input system had significantly higher levels than either the organic or low input systems (Fig. 3). Leaf loss ranged from 11 to 61%, and there was a significant interaction between management type and field with considerable variation between fields of each management type. Most leaves were affected by multiple leaf spots, but Septoria was not detected in most fields. Septoria infections may have been low this year or most of the leaves affected by Septoria may have fallen before we were able to rate. Powdery mildew was present in all fields and most plots were affected. However, there was a low overall percentage of leaf area affected and there were no significant differences between management types by percent of plots affected or by percentage of leaf area (Fig. 4). False Valdensia leaf spot was not detected in any of the fields this year. Red leaf was detected at very low levels in some fields (data not shown).

Leaf rust was present in all fields with over 90% of plots having plants with symptoms (Fig. 5A). There were no significant differences between the percentages of plots affected by leaf rust between management systems. The percentage of leaf area affected by rust ranged from 1.8 to 27% of leaf area (Fig. 5B). Low input fields had significantly less leaf rust on average than the other input systems. However, there was a significant interaction between management type and field. There was a significant correlation between increased levels of Rust and increased leaf loss ($R=0.95$, $P<.0001$, Fig. 6). We have found the same correlation in prune years 2012 and 2014, but not in the crop year 2013.

No organic plots were affected with Phomopsis stem blight but fields under the other input management systems had highly variable incidence of Phomopsis (Fig. 7A). High input systems had significantly higher plot areas affected by Phomopsis than the other input systems (Fig. 7B). There was a significant interaction of management type and field where some of the high input fields were not significantly different in Phomopsis levels than medium or low input fields.

CONCLUSIONS/RECOMMENDATIONS: Management inputs can affect the level of leaf and stem diseases present during the crop year and this may be partially affected by the disease levels in the prune year. It is difficult to detect the effect of leaf loss in the prune year unless it is early in the year. Leaf loss was measured after harvest in all of the fields and condition of the fields, when and how they were harvested may affect leaf loss. Disease levels are highly variable by year, within the year and among fields within the same management systems. Further analysis of factors that can affect leaf loss and Phomopsis levels will be studied.

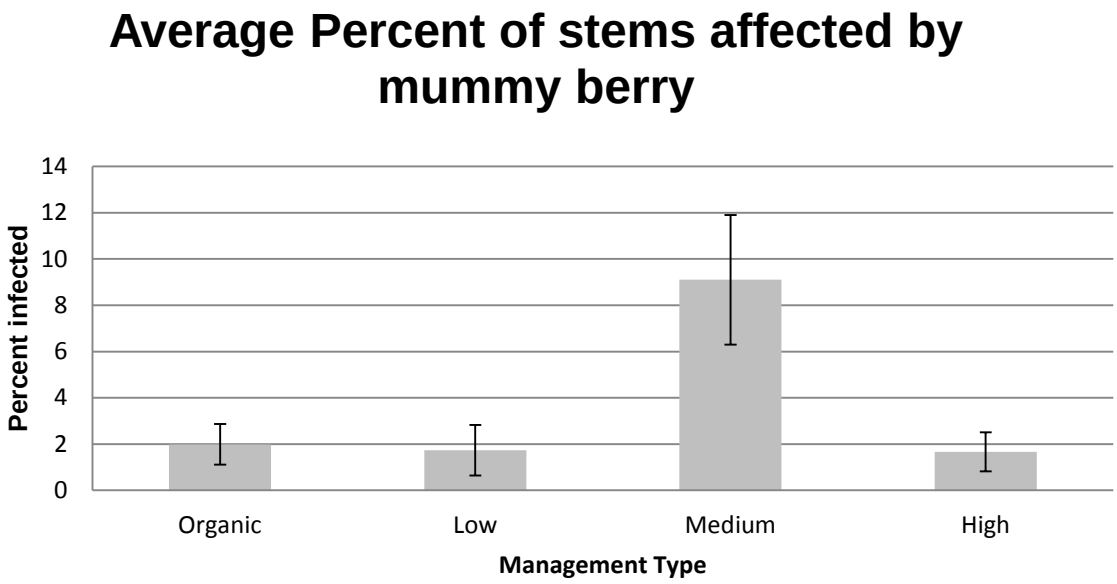


Figure. 1. – Average percentage of stems affected with mummy berry by management input system for 2015. Error bars indicate standard error of the mean. There were no significant differences in the percent of plots affected by mummy berry by management type.

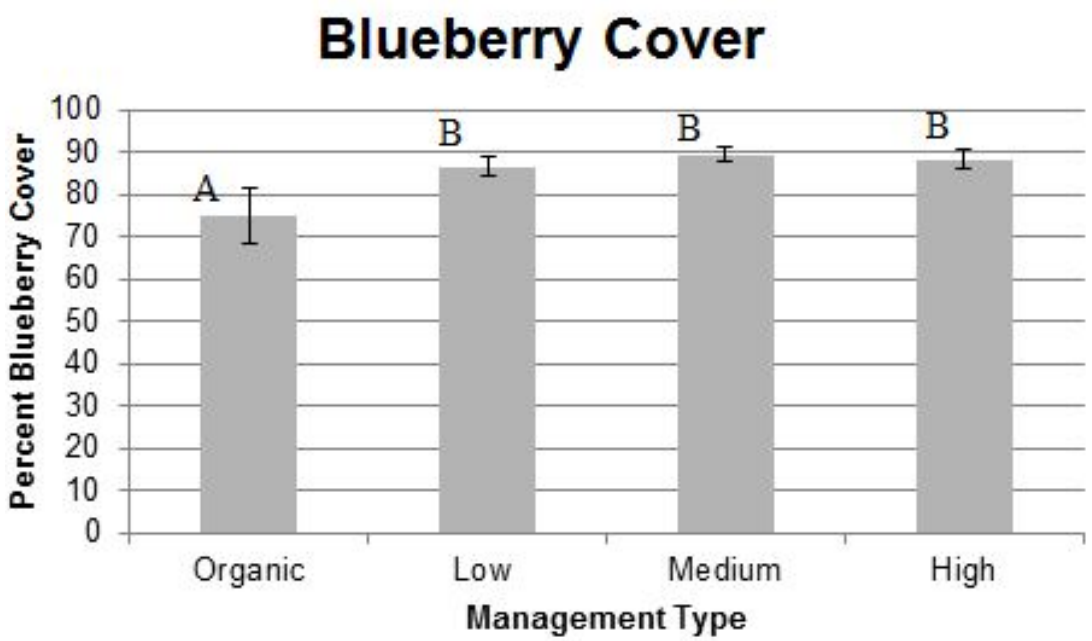


Figure 2. - Average percent of blueberry cover by management input system for September 2015. Error bars indicate standard error of the mean. Bars with different letters indicate statistically significant differences at $\alpha = 0.05$.

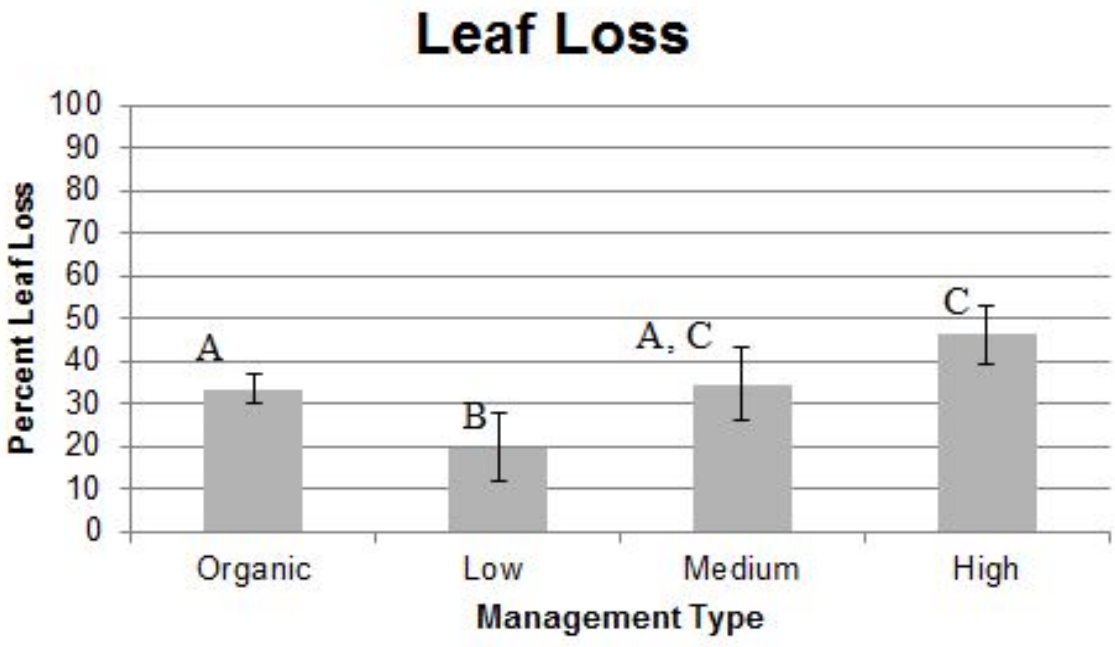
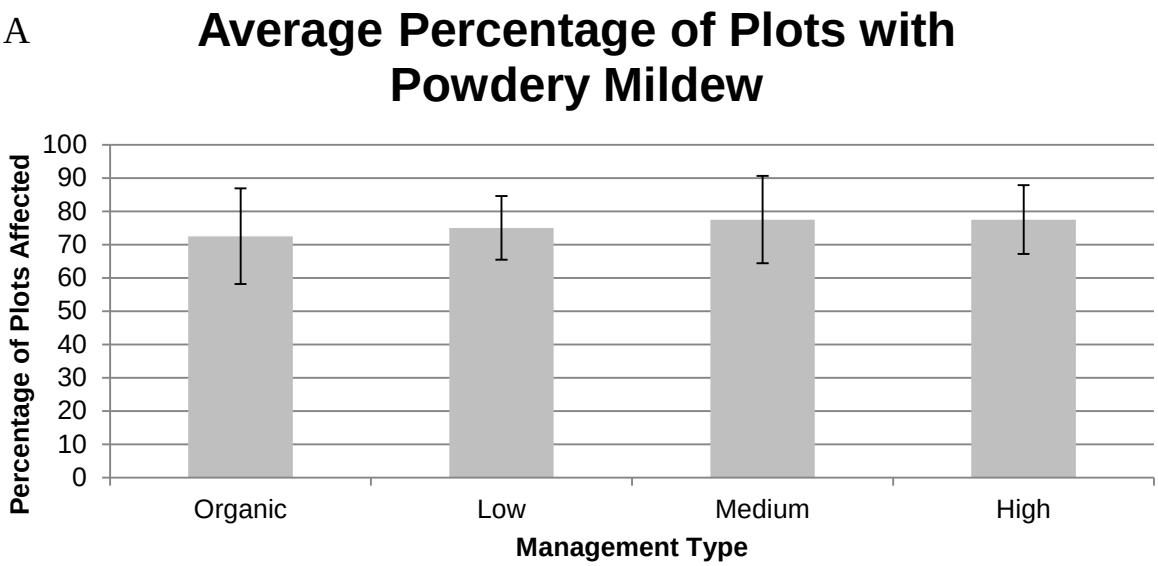


Figure 3. – Average percent leaf loss by management input system for September 2015. Error bars indicate standard error of the mean. Bars with different letters indicate statistically significant differences at $\alpha = 0.05$.

A



B

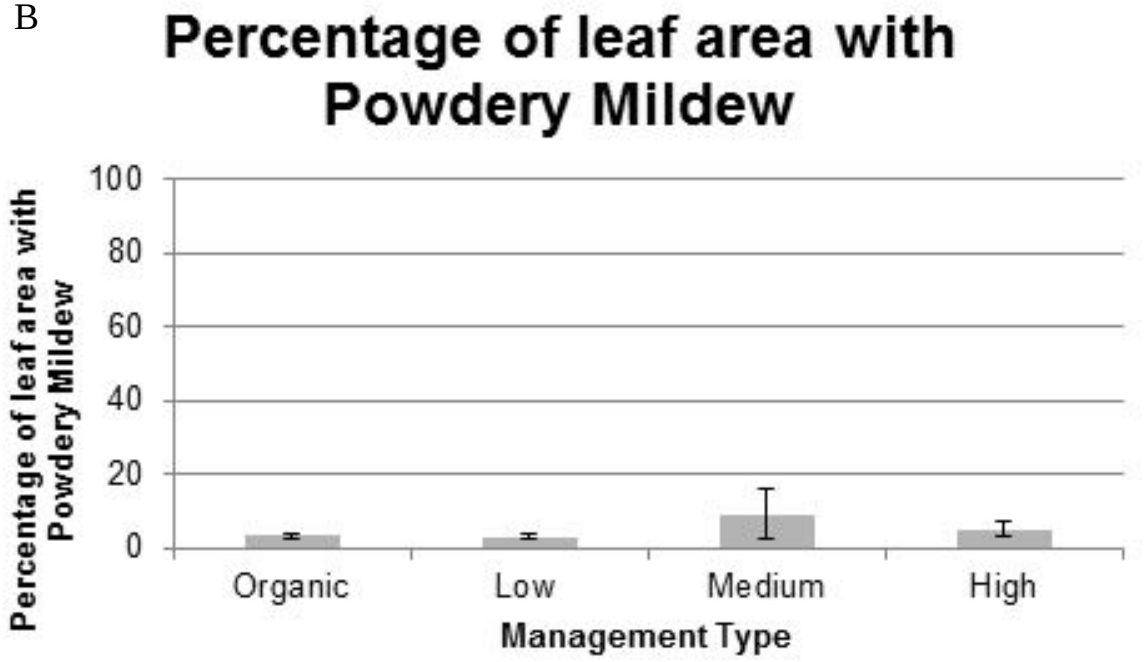
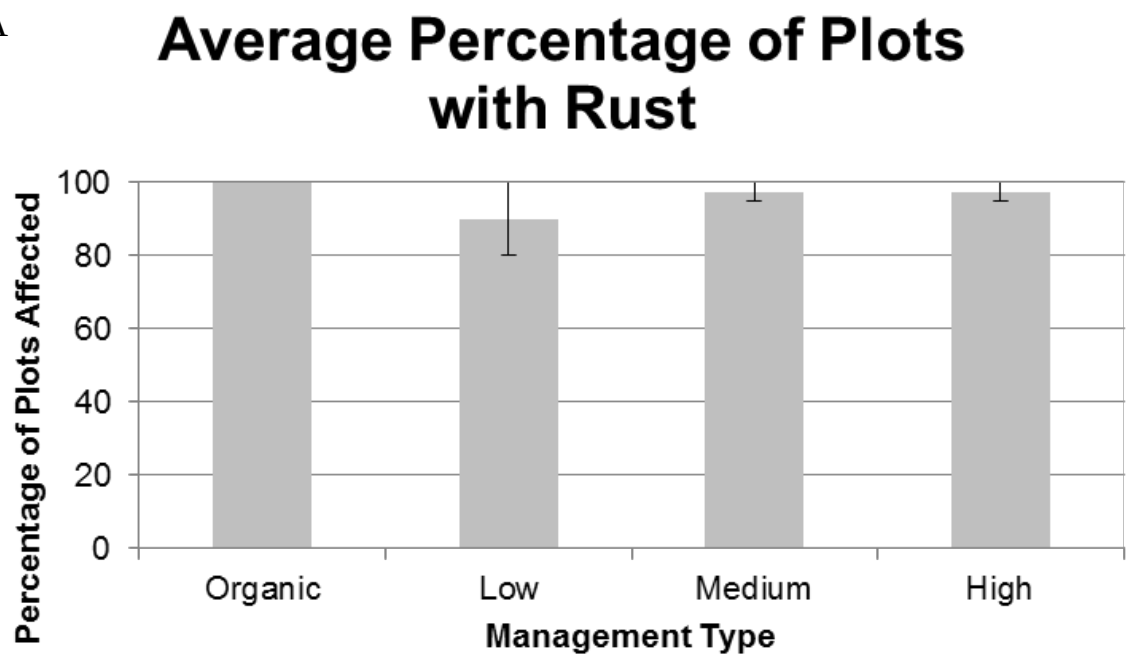


Figure 4. -Average percent of plots with Powdery Mildew (A) and average percent of leaf area affected by Powdery Mildew (B) by management input system for September 2015. Error bars indicate standard error of the mean. There were no significant differences in the percent of plots affected or in the percentage of leaf area by management type.

A



B

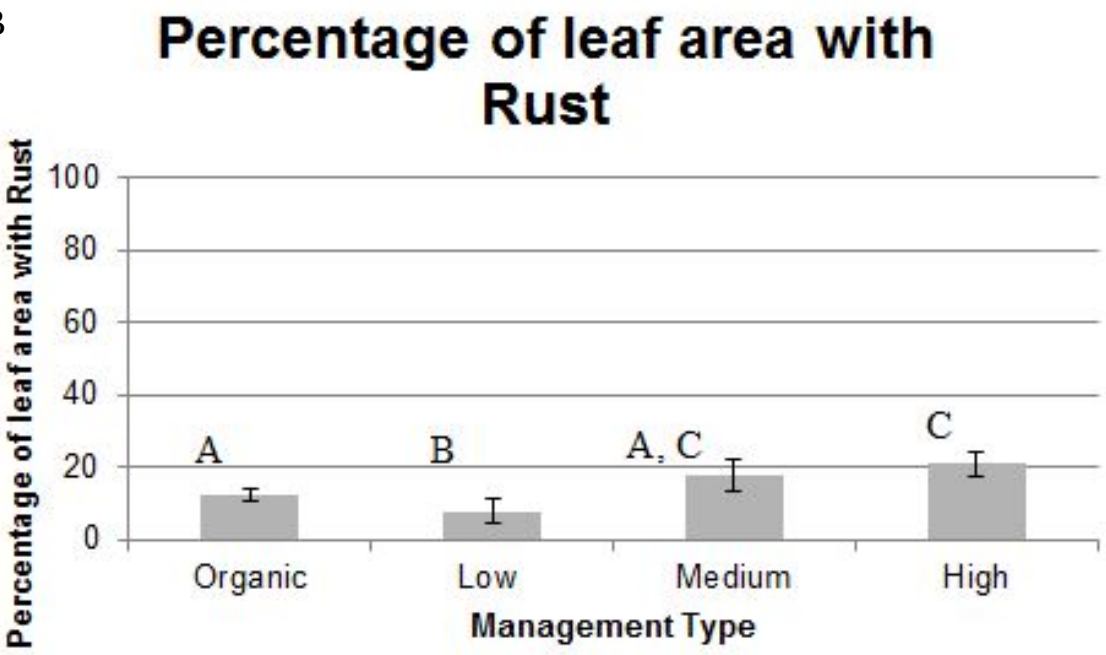


Figure 5. –Average percent of plots with leaf rust (A) and average percent of leaf area affected by rust (B) by management input system for September 2015. Error bars indicate standard error of the mean. Bars with different letters indicate statistically significant differences at $\alpha = 0.05$.

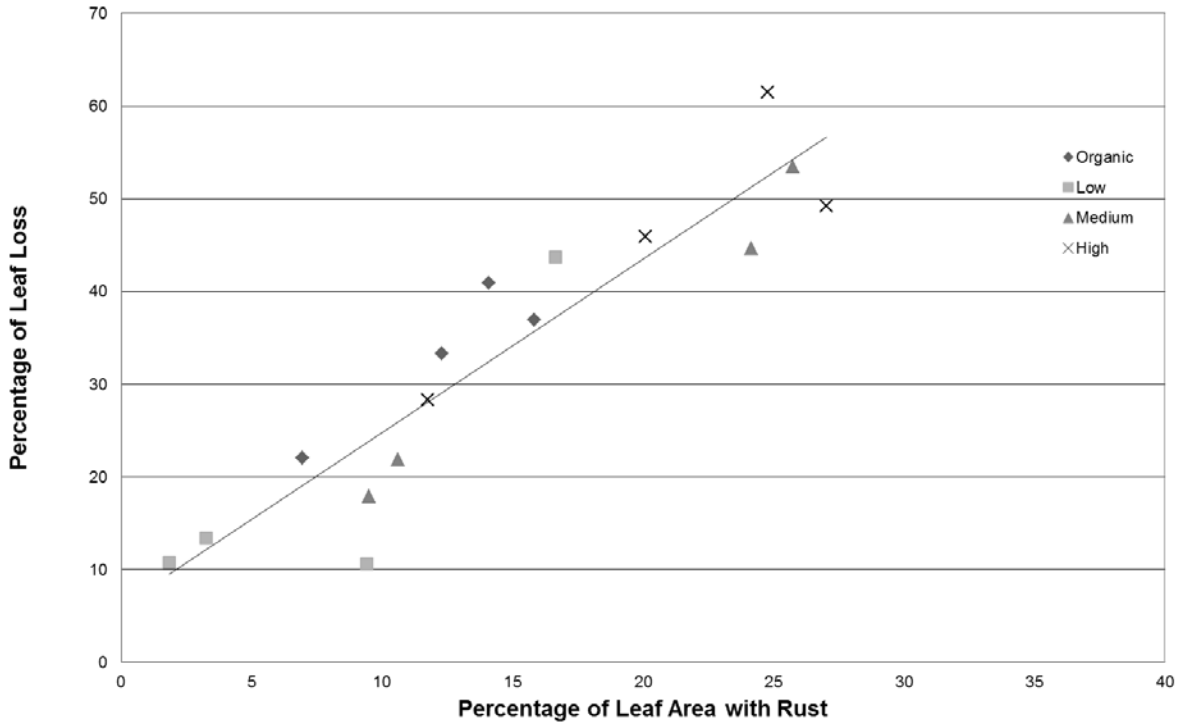
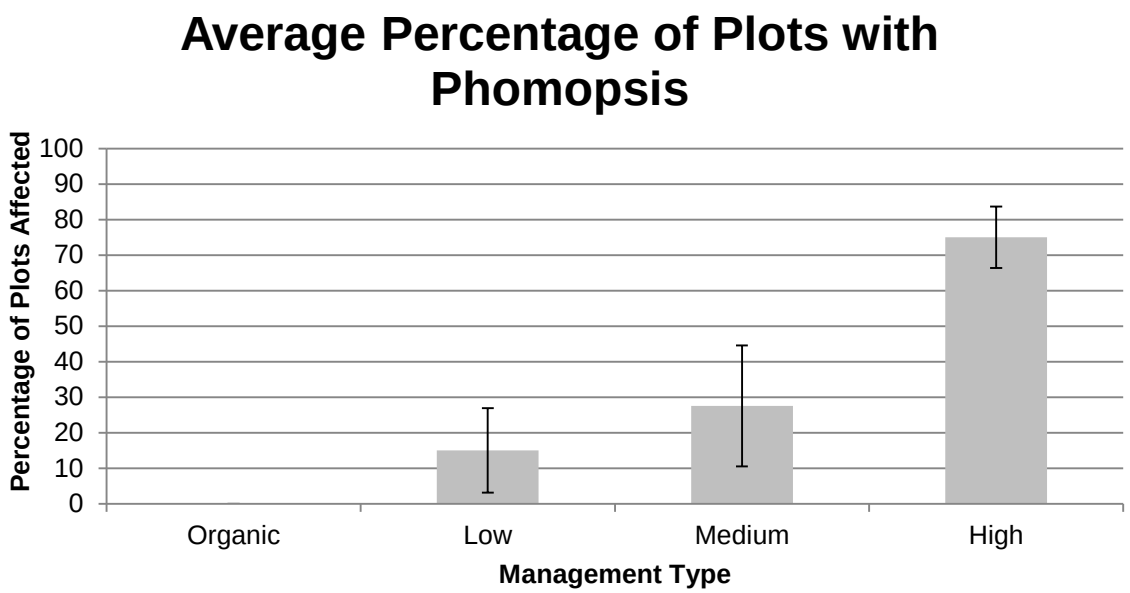


Figure 6. - Average percentage of leaf area with rust compared with the average leaf loss per field. ($R = 0.95, p<0.0001$). Diamonds represent organic input systems, squares represent low input systems, triangles represent medium input systems and Xs represent high input systems.

A



B

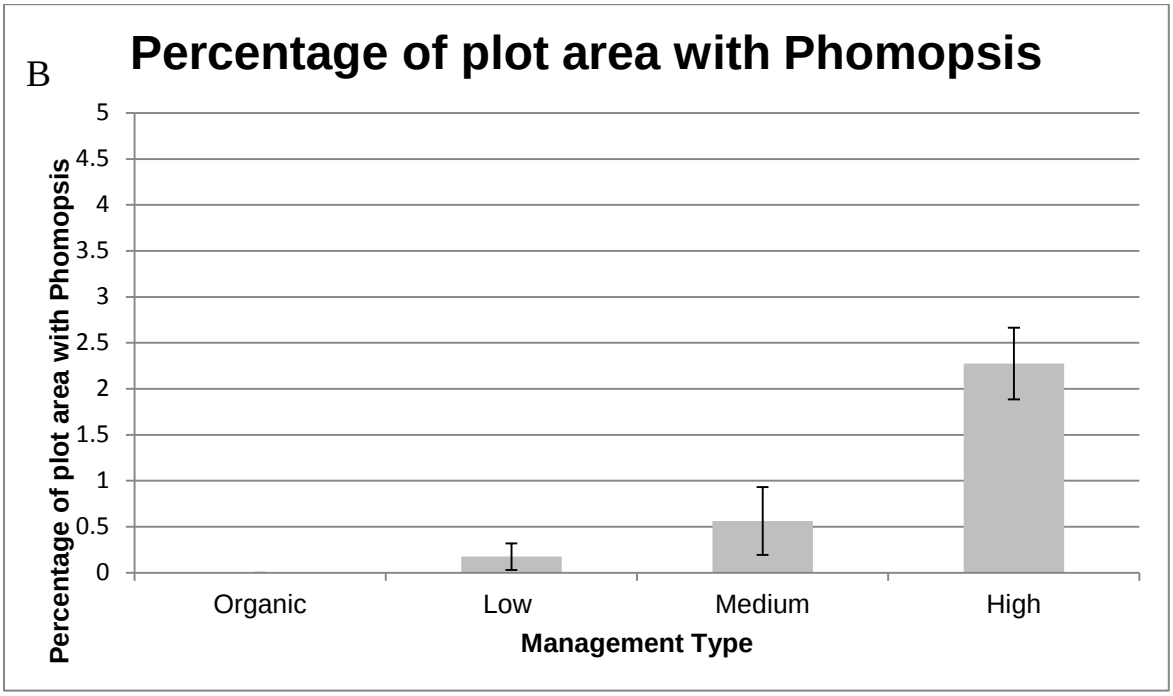


Figure 7. - Average percent of plots with Phomopsis (A) and average percent of leaf area affected by Phomopsis (B) by management input system for September 2015. Error bars indicate standard error of the mean. Bars with different letters indicate statistically significant differences at $\alpha = 0.05$.

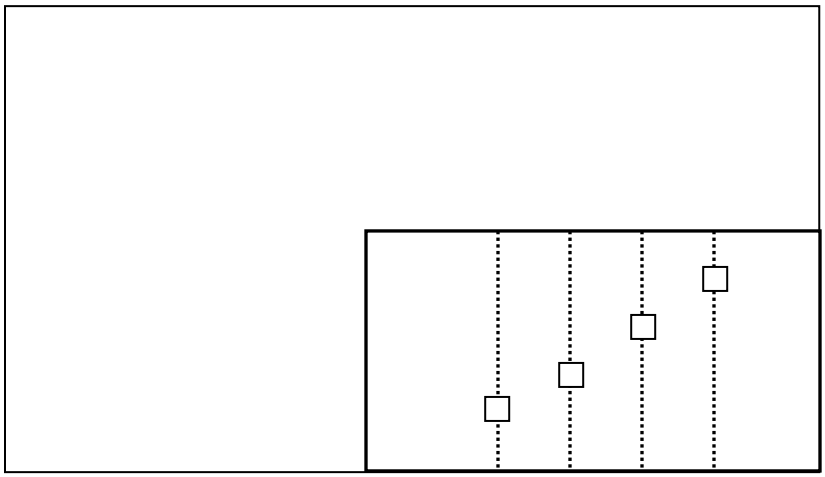
INPUT SYSTEMS STUDY

WEED MANAGEMENT: David E. Yarborough, Professor of Horticulture
Jennifer L.D. Cote, Assistant Scientist

16. TITLE: Systems approach to improving the sustainability of wild blueberry production, Year Six of a six year study, weed management results.

METHODS: The third crop cycle study design and 2015 crop year inputs are listed in Table 2 of the overall Experimental Design (Report #13). In 2014 16 trial sites were once again set up containing two 1 acre blocks each with 15 x 30 m sub-blocks; along the 30 m baseline (the outer long edge of the block) of each sub-block, four transects were located 5 m apart in order to set up 1 m^2 sample plots to assess weed cover. One 1 m^2 sample plot was staked on each transect 3 m apart so that the sample plots ranged diagonally across the subplot (Figure 1). In this cycle, seven sites were replaced due to either a lack of grower cooperation, or in order to encompass a wider geographical range.

Figure 1. Example layout of a block, sub-block, transects and weed sample plots (not to scale).



Blueberry cover, woody weed cover, broadleaf weed cover and grass cover were assessed in all 1 m^2 sample plots just prior to harvest, July 21-August 11, 2015. Cover was assessed using the Daubenmire Cover Class scale, which was converted to percent; weed species were also identified. The data were analyzed using the Nested General Linear Model (SAS 9.4) and Tukey's HSD tests for significant differences (α=0.05). Overall blueberry cover and weed cover comparisons were made among all four input systems.
All sites were harvested July 27 to August 11 to compare yields among input systems. Detailed methods and general comparisons are included in the overall Experimental Design Report. Weed cover as a determining factor for yield is presented two different ways in the Results section. Woody, broadleaf and grass weed covers were combined for an overall % weed cover (if the sum was >100, then the value was 100%). First, a contrast comparison was performed on the weed cover (x) vs. yield (y) for each input system, to determine what type of relationship, if any, existed between weeds and yield (e.g. linear, quadratic, cubic, etc.). Weed cover was also arcsine transformed to normalize the data for analysis, and compared to yield to determine how

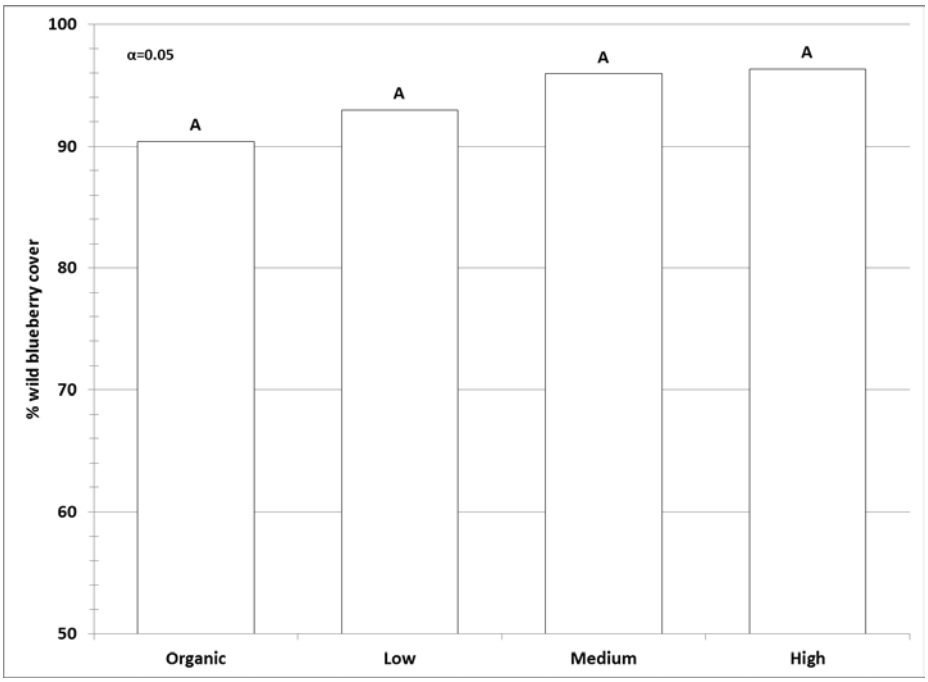
much variation in yield was due to input system and how much was due to weed cover. For the purposes of this report, the arcsine-transformed weed data are graphed as percent cover.

RESULTS:

Blueberry and weed cover

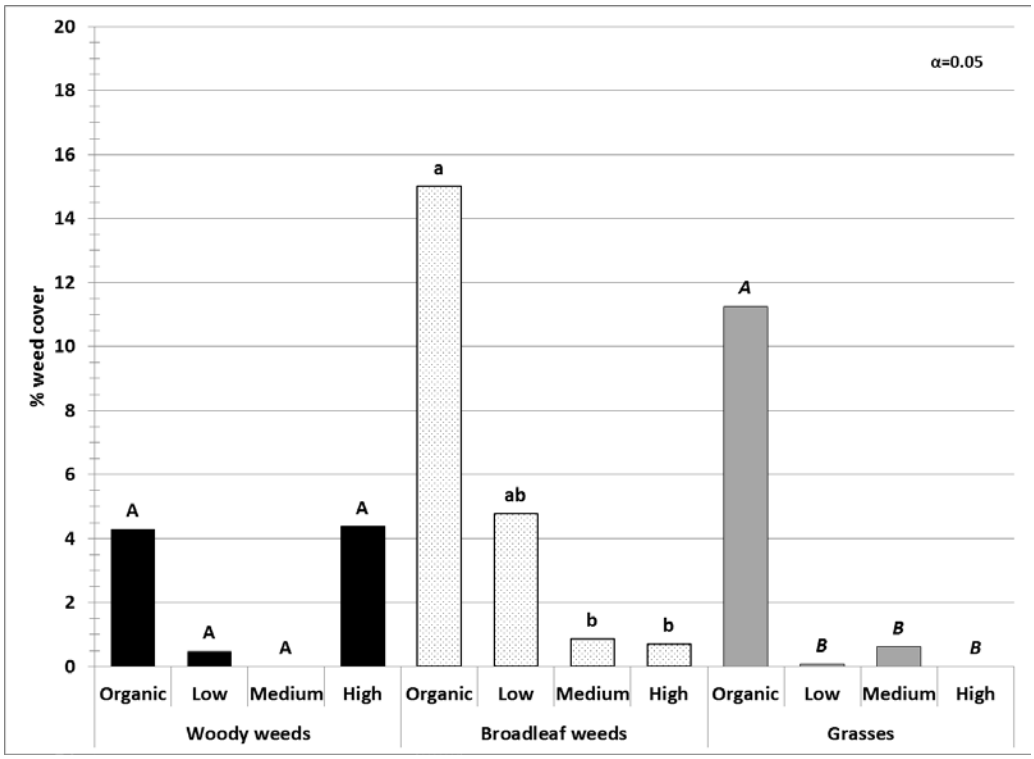
There were no significant differences in wild blueberry cover among the four systems (Figure 2). In 2015 the trend in blueberry cover closely matched that of 2014, and followed a slightly different trend compared to 2011-13. In previous years, by August the Medium system had the highest cover, followed by the High system. In this production cycle, the High system had the highest cover both years. In previous crop cycles, all Medium sites were managed by one company and were located Downeast; in this cycle, they were managed by three companies and ranged from the Union area to Downeast.

Figure 2. Wild blueberry cover by input management system in crop year 2015 (Tukey's HSD, different letters denote significance at α=0.05).



In 2015, levels of woody weeds, broadleaf weeds and grasses and relationships among systems were almost identical to 2014 (Figure 3). As in 2014, woody weed cover was very low overall (<5 %); and there were no significant differences among systems. Broadleaf weed cover followed the same trend as in 2014 and the previous cycle; the Organic system had the highest broadleaf weed cover and was significantly higher than the Medium and High systems, while the Low system was not significantly different from any other system. Grass cover also followed the trend seen in 2014 and the previous cycle, where the Organic system had significantly more grasses than the conventional management systems.

Figure 3. Woody weed, broadleaf weed and grass cover among input systems in crop year 2015 (Tukey's HSD, different letters denote significance at α=0.05).



Yield vs. weeds

When weed cover by input system was compared to yield to find out what type of relationship weeds and yield had (e.g. linear, quadratic, cubic, etc.), there were no significant relationships (Figure 4). In 2013 the High system had a positive correlation between weed cover and yield; in 2015 the correlation was negative. The correlation between weed cover and yield was slightly negative in the Medium system in 2013; in 2015 it was strongly positive, but no site had over 5% weed cover. In both 2013 and 2015, there was a positive correlation between weed cover and yield in the Low system and a negative correlation in the Organic system, although the effect was stronger in both systems in 2015.

When input system, site and weed cover were both used as independent predictor variables in a regression to predict what was driving variations in yield, there was a negative relationship between weed cover (arcsine transformed but presented as % cover) and yield (Figure 5). Input system was significant at $\alpha=0.04$ and site was significant at $\alpha<0.0001$, while weed cover was not significant at $\alpha=0.67$. Input system accounted for the greatest portion of the variation in yield at 56 % ($R^2=0.5577$), site accounted for 15 % ($R^2=0.1510$), and weed cover accounted for very little of the variation in yield at 1 % ($R^2=0.0110$). In 2013, input system accounted for the same amount of variation at 56 %, but weed cover accounted for more at 18 %.

We also pooled weed cover of all input systems for each crop year and plotted it against yields for each year (Figure 6). In all three crop years there was a negative correlation between weed cover and yield; however, the relationship was weaker in 2015 compared to 2011 and 2013.

Figure 4. The relationships between weed cover by input system and yield in 2015 (α=0.05; no significant relationships).

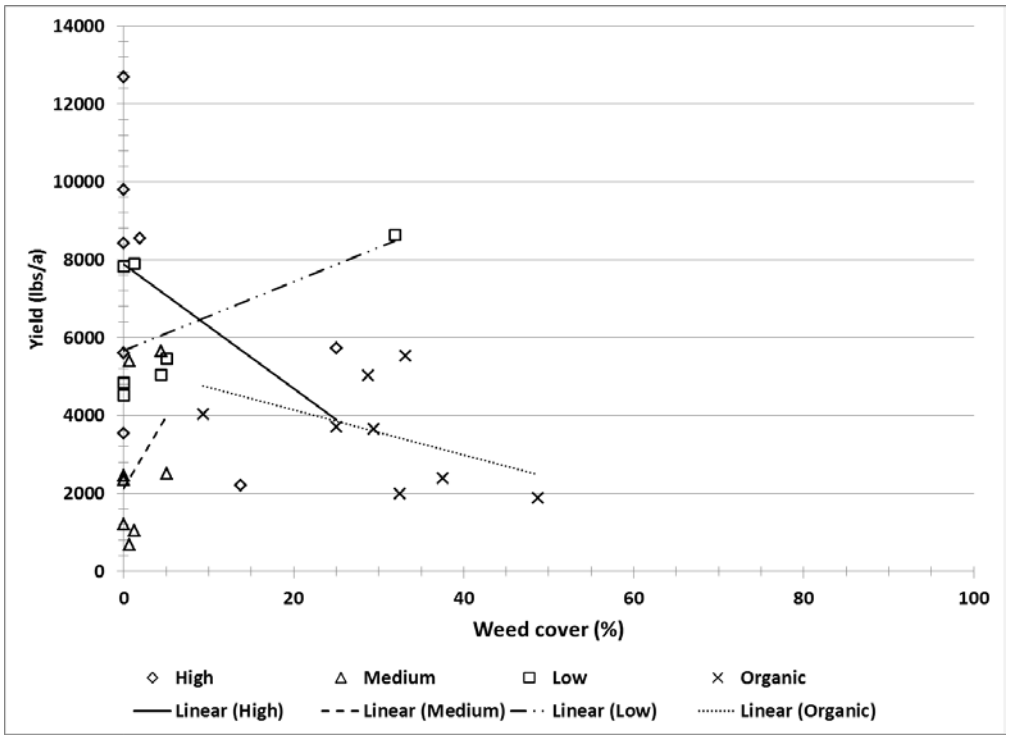


Figure 5. Weed cover, site and input system as predictors of variation in yield, 2015 (overall R^2=0.7197; input system R^2=0.5577; site R^2=0.1510; weed cover R^2=0.0110).

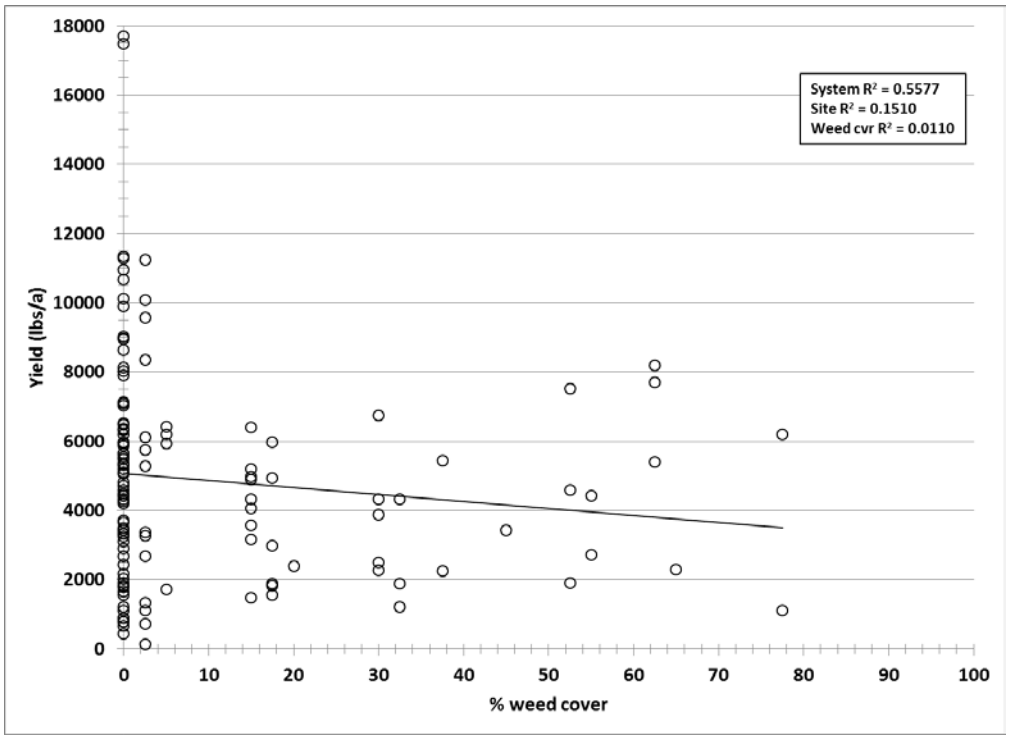
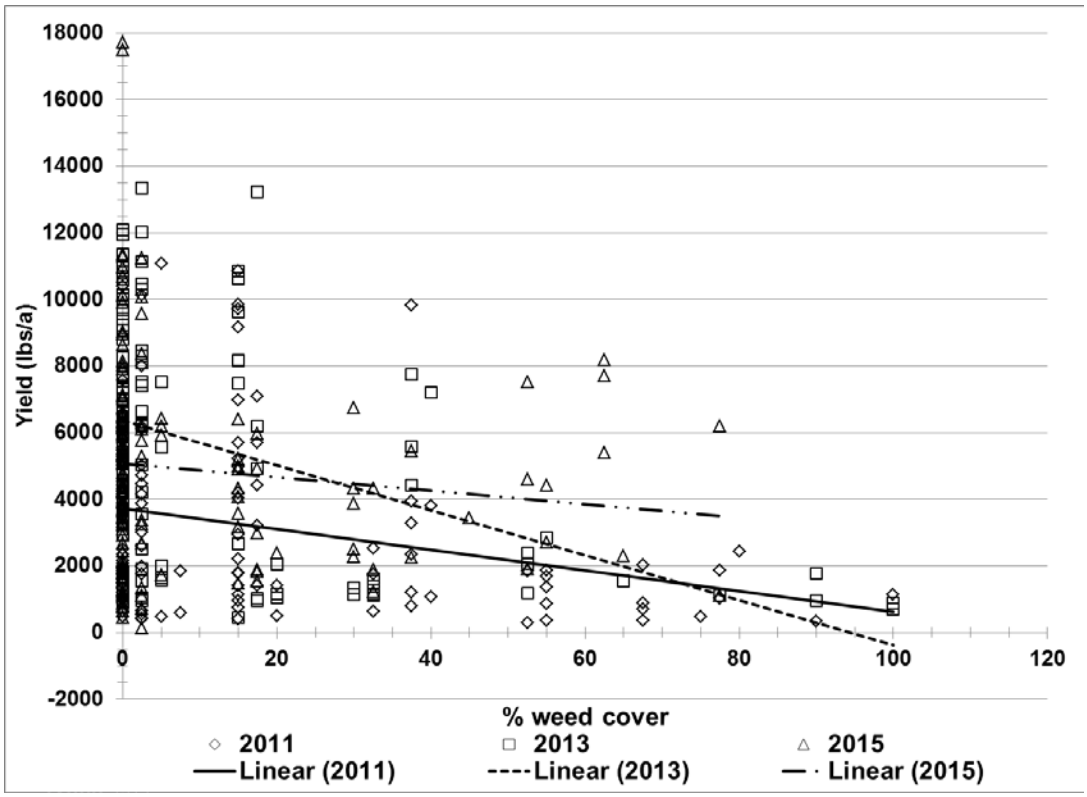


Figure 6. The relationship between weed cover across all input systems and yield in crop years 2011, 2013 and 2015.



CONCLUSIONS: In this final crop year of the project, wild blueberry cover and weed cover followed the same basic trends as the previous two cycles. Wild blueberry cover was comparable among systems, and overall weed cover was highest in the Organic system compared to the conventional management systems (Photos 1, 2A-B, 3-4). As in 2014, woody weed cover in the High system was equal to the Organic system. In prune year 2014 we stated that there was less grass cover in the Organic system compared to the 2012-13 crop cycle, and that the difference was likely due to better management and more inputs by the replacement organic growers. This was observed again this year; grass cover increased slightly from 10% in 2014 to 11% in 2015, but was lower than the 18% grass cover in 2013. In contrast with previous cycles where the Low system had the most variability in wild blueberry cover, the Organic system had the most variability in 2015 (data not presented). This was probably due to the combination of the replacement of two Low sites in the third cycle with fields with better blueberry cover, and Low field #7 having improved blueberry cover in 2015 resulting from dry summer conditions allowing plants to spread into normally excessively wet areas (see Photo 2A-B).
We would expect to see a negative correlation where increasing weed cover would lead to decreasing yield. When data for all systems are pooled, we see this relationship as demonstrated in Figures 5 and 6. However, site differences or moisture conditions may have led the Low and Medium system yields to increase with increasing weed cover, as seen in Figure 4. The positive correlation of weeds to yield in the Low system may be because improved growing conditions for blueberry in Low field #7 likely provided improved growing conditions for weeds as well as

the blueberries, and many of the weeds were located in bare spots between the clones. In this cycle, the transects for block O2A were also located in a slightly flatter area that had more consistent blueberry cover and so had better yield. The positive correlation in the Medium system may be due to weather related damage from the winter and spring of 2015, which led to little to no flowers or fruit in 2015 (see overall Experimental Design report for a more in-depth discussion).

Photo 1. Example of blueberry and weed cover in an organic plot in early August. Although weed cover was higher than the conventional systems, yield was higher than in the Organic system in previous crop years.

Photo 2. Example of wild blueberry and weed cover for the Low system in late July in (**A**) 2015 versus (**B**) 2013. Note bare low areas in **B**, with blueberry restricted to high ground, versus filling in of depressions in **A** by blueberry and weeds such as bunchberry.

Photo 3. Example of wild blueberry and weed cover in the Medium system in late July; note lack of fruit in this winter-damaged field.

Photo 4. Example of wild blueberry and weed cover in the High input system in late July; note lack of fruit in this winter-damaged field.

RECOMMENDATIONS: This is the final year of this study. All data from all components of the study will be compiled and analyzed. Predictive models for calculating interactions between components of the wild blueberry production system pursuant to inputs will be generated to determine the relative importance of each, and an interactive budget will be developed for to growers so that they can estimate monetary returns for different levels and combinations of inputs.

INPUT SYSTEMS STUDY

ECONOMICS: David E. Yarborough, Professor of Horticulture
Jennifer L.D. Cote, Assistant Scientist

17. TITLE: Systems approach to improving the sustainability of wild blueberry production, preliminary economic comparison for 2014-15.

METHODS: Project layout is described in the overall Experimental Design Report. The number of acres, wild blueberry yields and cost of harvest, and all variable input costs were obtained from the cooperators for each input level and location for the 2014-2015 crop production cycle. At the time of this report, per pound prices for berries had not been determined. Therefore, field prices per pound were estimated for all growers except one Medium grower at $0.50/lb for processed berries and by each grower's estimate for organic berries. For the purposes of this report, the interest on capital and fixed costs were also estimated pursuant to the last time they were calculated. The project economist is currently updating the fixed costs and once they are finalized, these budgets will be modified. These partial budget Excel spreadsheets will be available to wild blueberry growers on the www.wildblueberries.com website to determine the cost and returns per acre and per pound.
Average yield versus average monetary returns per acre were analyzed for all three crop years using a Tukey's test (α=0.05), with the range in returns per acre for each system/year combination. The same analysis was conducted to compare the four systems with all years' data pooled. The yield data provided by the growers were used for both analyses, except for the 2013 data for Organic fields #2 and #4, as these two growers did not provide any data for 2013, so the average site yields harvested and recorded by University personnel were used.

RESULTS: This production cycle was anomalous in that there was widespread field-level damage from the winter of 2014-15, as well as a cold late spring and drought in July and August. These factors combined to make some historically high-producing fields yield much less than usual, while one normally excessively wet Low input field had much higher yield than usual due to the dry summer making growing conditions more favorable. This resulted in some historically profitable fields losing money over the 2014-15 production cycle because the yield profit did not cover the cost of inputs. For example, in the two Medium fields carried over from cycle 2 to cycle 3, although many costs such as weed control, insect control, disease control and fertilization remained about the same and harvest costs declined, pollination costs almost doubled and yield declined drastically (from 5,955 lbs/a in Field #9 to 1,301 lbs/a, and from 6,389 lbs/a in Field #10 to 2,642 lbs/a). As a result, annual return for Field #9 went from a profit of $114,432 in 2013 to a loss of $28,033 in 2015, while Field #10 went from a profit of $224,609

in 2013 to a loss of $892 in 2015. The same trend held true for the two High fields, Fields #13 and #14, which lost money. In both fields, many input costs remained similar and harvest costs declined from cycle 2 to cycle 3. In Field #13 pollination cost increased by almost $250/acre and yield declined by about 2,500 lbs/acre, while in Field #14 pollination cost actually declined by approximately $70/acre but yield declined by 4,000 lbs/acre. Both of these fields were profitable in cycle 1 and 2, but observations in these fields revealed winter damage and late spring injury, which resulted in some fields having little to no or patchy flowers or fruit but with vigorous vegetative growth (including the four fields mentioned). The loss in yield for all growers was compounded by a lower field price for 2015; estimates range from $0.43/lb to $0.50+/lb, but no more than $0.60/lb. By contrast, in the 2013 budgets conventional growers listed as estimated or received from $0.65 to $0.73/lb and in 2011 estimated or received $0.80/lb.

The Organic growers in this cycle tended to apply more inputs and diversify their yields more compared to the previous cycles. For example, in this cycle all four Organic growers sold part of their harvest to a processor and kept a portion for either fresh pack or frozen direct sale to the consumer. Therefore, not only did they have the added security of having a buyer for their harvest, even if the processor paid significantly less than their direct sale prices, but the premium prices they were able to get for their direct sale berries offset reductions in yield due to weather. In addition, increases in inputs were reflected in the significantly higher yields reported by Organic growers in cycle 3 compared to the previous cycles. For example, Field #1 had a yield of approximately 450 lbs/acre in 2011, 575 lbs/acre in 2013 and 2,165 lbs/acre in 2015. This grower applied sulfur just prior to the start of the study, did mechanical weed control and had minimal supplementation of pollinators over cycles 1 and 2 (2 hives for 10 acres). By cycle 3 the sulfur had taken effect and the grower applied more sulfur to bring down the soil pH, thereby controlling weeds, did mechanical weed control and put out 15 hives over 10 acres. The investment in long-term weed control by sulfur and the added pollination input resulted in a four-fold increase in yield under less than optimal environmental conditions.

When average per acre returns were viewed on average per acre yields, several trends emerged. Although the Low system's 2015 yield was the best of the three production cycles, the system lost money overall every cycle (Figure 1). This appeared to be largely due to a combination of rockier sites with poorer, shallower soils compared to the Medium and High systems and fewer inputs for disease and pest control; in 2015, the increased yield was offset by a low per pound price for processed berries. The Organic system also had its best yield in 2015 and yields overall were comparable with the Low system (and with the Medium and High systems in 2015). The large increase in average returns, as well as the large range, in 2015 is due to new organic cooperators who practiced better management in the 2014-15 production cycle, and to the addition of value-added products which increased their income. The upper limit of the range of returns was at over $10,300 per acre (see Organic 1 value-added budget) which illustrates that an organic farm can be lucrative despite fewer inputs, more weed, disease and pest pressure, and lower yields than conventional systems, because the grower can obtain a much higher price per pound for organic value-added products. The Medium and High systems were both adversely affected by the poor weather in winter and spring 2015; in the first two production cycles they had significantly higher yields overall than the Low and Organic systems, but because of the winter damage and dry summer conditions in 2015, all systems had comparable yields in 2015. All four Medium fields lost money in 2015, two of which were carried over through all three cycles and were historically profitable fields. Two of the four High fields also lost money, and were also historically profitable. Despite these losses, when data from all crop years were

pooled and average per acre returns were viewed on average per acre yield, the anomalous year 2015 did not negate the overall trend observed over the first two production cycles (Figure 2). The Medium and High systems had significantly higher yields over time compared to the Low and Organic systems, and the point of diminishing returns lies between the level of inputs and costs for the Medium and High systems. The Low system consistently lost money due to fewer inputs combined with poorer sites, and although the Organic system had the lowest yield overall, it was more lucrative than the conventional systems due to the premium prices organic growers receive for their fresh pack and value-added berries.
Partial budgets are included for the four systems and four field locations included at the end of this report. The variability of each individual site and returns for fresh pack and value added can be seen from these budgets.

Figure 1. Average grower per acre yields and per acre monetary returns by system for all crop years; the range in per acre returns is overlaid as error bars. Different letters denote significance at α=0.05 for systems yield comparisons within the same year.

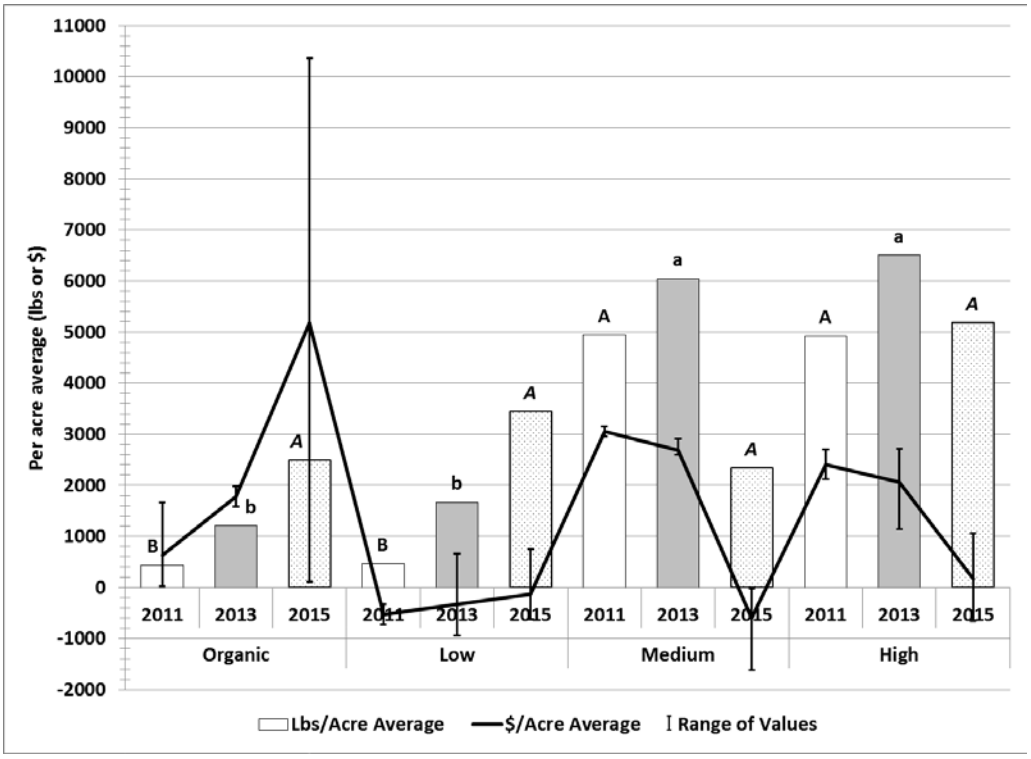
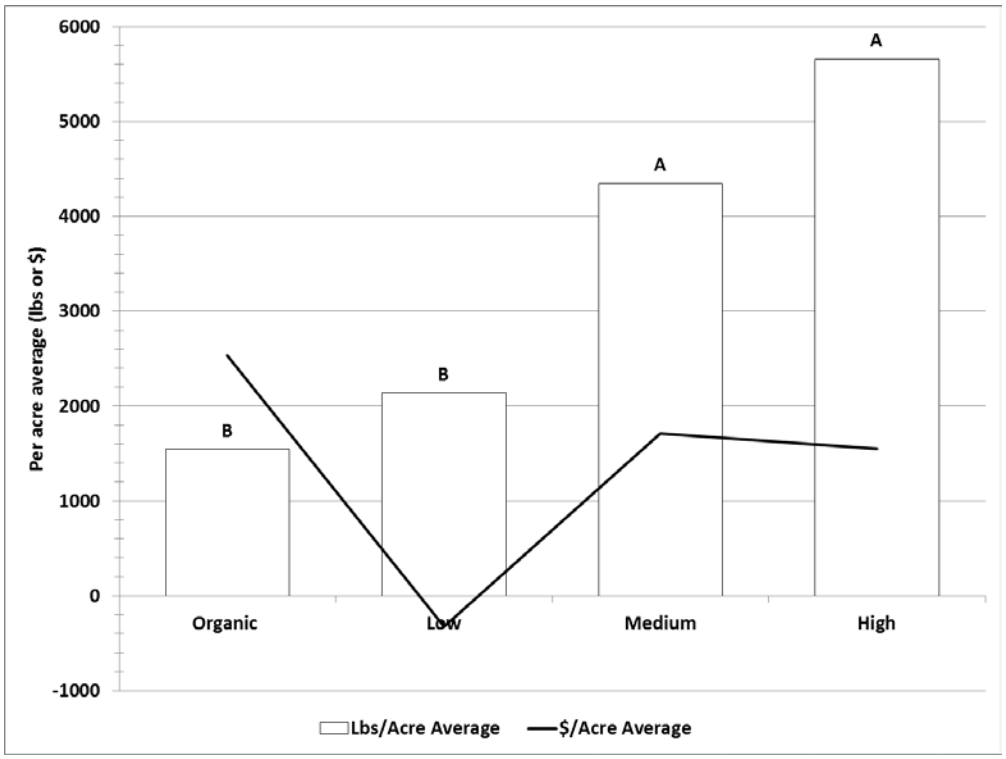


Figure 2. Average per acre yield and per acre returns by system with all crop years' data pooled. Different letters denote significance at α=0.05 for yield comparisons.



CONCLUSIONS/DISCUSSION: In the two previous cycles we saw a relationship regarding inputs versus yield, where there appeared to be a point of diminishing returns for per acre input costs versus returns to the company. This cycle showed us that weather and environmental conditions are important factors influencing yield in the wild blueberry cropping system. Although drought can be mitigated somewhat by irrigation, wild blueberry cannot be protected from cold weather events by the use of greenhouses, high tunnels, etc. as in some other cropping systems. Growers are exposed to a variable level of risk among years, as illustrated in 2015 where growers who applied the highest levels of inputs to their fields did not get the expected returns because of environmental circumstances beyond their control. This supports the decision-making tactic of determining the point of diminishing returns when deciding what or how many inputs to apply to a field, so that losses can be minimized when "acts of God" occur. A consistent level of inputs over time can minimize long-term losses as well as long-term gains, as is illustrated in Figures 1 and 2 for the Low system versus the Medium and High systems. This study also shows that Organic growers occupy a niche that is not being utilized by most conventional growers, namely fresh pack and value-added products, and they obtain a higher price for organic produce which serves to maximize profits from smaller yields and has fewer input costs.

RECOMMENDATIONS: No recommendations at this time. The data will be analyzed by the project Economist, and final interactive budget spreadsheets will be generated for growers to use.

Wild Maine Blueberry Budget		
Organic 1 Processed		
Number of Acres (Crop)	4.15	
Yield (Lbs./Acre)	2,166.00	
Price/Lb. ($)	1.58	
REVENUE/ACRE ($)	3,422.28	
VARIABLE COSTS	**($/Acre)**	**($/Pound)**
Pruning:		
Burning	0.00	0.00
Mowing	23.70	0.01
Average Pruning	23.70	0.01
Weed Control	88.00	0.04
Fertilization	0.00	0.00
Pollination	25.33	0.01
Pest Monitoring	0.00	0.00
Insect Control	0.00	0.00
Disease Control	0.00	0.00
Irrigation	0.00	0.00
Sulfur (pH)	82.15	0.04
Harvest:		
Raking	0.00	0.00
Mechanical	493.75	0.23
Average Harvest	493.75	0.23
Packing and Marketing	0.00	0.00
Interest on Capital	42.60	0.02
Blueberry Tax	16.25	0.0075
TOT. VARIABLE COSTS	771.78	0.36
FIXED COSTS:		
Machinery & Equipment	60.10	0.01
Taxes	32.83	0.01
TOTAL FIXED COSTS	92.93	0.02
TOTAL COSTS	864.71	0.40
RETURNS ABOVE COSTS SHOWN	2,557.58	1.18
AVERAGE TOTAL ANNUAL RETURN TO MANAGEMENT		
($/FARM)	**10,613.94**	

Wild Maine Blueberry Budget		
Organic 1 Freshpack		
Number of Acres (Crop)	0.57	
Yield (Lbs./Acre)	2,166.00	
Price/Lb. ($)	4.40	
REVENUE/ACRE ($)	9,530.40	
VARIABLE COSTS	**($/Acre)**	**($/Pound)**
Pruning:		
Burning	0.00	0.00
Mowing	23.70	0.01
Average Pruning	23.70	0.01
Weed Control	88.00	0.04
Fertilization	0.00	0.00
Pollination	25.33	0.01
Pest Monitoring	0.00	0.00
Insect Control	0.00	0.00
Disease Control	0.00	0.00
Irrigation	0.00	0.00
Sulfur (pH)	82.15	0.04
Harvest:		
Raking	0.00	0.00
Mechanical	493.75	0.23
Average Harvest	493.75	0.23
Packing and Marketing	1,776.12	0.82
Interest on Capital	42.60	0.02
Blueberry Tax	32.49	0.015
TOT. VARIABLE COSTS	2,564.14	1.18
FIXED COSTS:		
Machinery & Equipment	60.10	0.01
Taxes	32.83	0.01
TOTAL FIXED COSTS	92.93	0.02
TOTAL COSTS	2,657.07	1.23
RETURNS ABOVE COSTS SHOWN	6,873.33	3.17
AVERAGE TOTAL ANNUAL RETURN TO MANAGEMENT		
($/FARM)	**3,917.80**	

Wild Maine Blueberry Budget		
Organic 1 Value-added		
Number of Acres (Crop)	5.20	
Yield (Lbs./Acre)	2,166.00	
Price/Lb. ($)	6.00	
REVENUE/ACRE ($)	12,996.00	
VARIABLE COSTS	**($/Acre)**	**($/Pound)**
Pruning:		
Burning	0.00	0.00
Mowing	23.70	0.01
Average Pruning	23.70	0.01
Weed Control	88.00	0.04
Fertilization	0.00	0.00
Pollination	25.33	0.01
Pest Monitoring	0.00	0.00
Insect Control	0.00	0.00
Disease Control	0.00	0.00
Irrigation	0.00	0.00
Sulfur (pH)	82.15	0.04
Harvest:		
Raking	0.00	0.00
Mechanical	493.75	0.23
Average Harvest	493.75	0.23
Packing and Marketing	1,752.72	0.81
Interest on Capital	42.60	0.02
Blueberry Tax	32.49	0.015
TOT. VARIABLE COSTS	2,540.74	1.17
FIXED COSTS:		
Machinery & Equipment	60.10	0.01
Taxes	32.83	0.01
TOTAL FIXED COSTS	92.93	0.02
TOTAL COSTS	2,633.67	1.22
RETURNS ABOVE COSTS SHOWN	10,362.33	4.78
AVERAGE TOTAL ANNUAL RETURN TO MANAGEMENT		
($/FARM)	**53,884.12**	

Wild Maine Blueberry Budget		
Organic 17 Processed		
Number of Acres (Crop)	1.00	
Yield (Lbs./Acre)	3,714.00	
Price/Lb. ($)	1.53	
REVENUE/ACRE ($)	5,682.42	
VARIABLE COSTS	**($/Acre)**	**($/Pound)**
Pruning:		
Burning	0.00	0.00
Mowing	70.43	0.02
Average Pruning	70.43	0.02
Weed Control	527.57	0.14
Fertilization	60.00	0.02
Pollination	310.00	0.08
Pest Monitoring	15.00	0.00
Insect Control	0.00	0.00
Disease Control	0.00	0.00
Irrigation	0.00	0.00
Sulfur (pH)	240.00	0.06
Harvest:		
Raking	1,227.60	0.33
Mechanical	0.00	0.00
Average Harvest	1,227.60	0.33
Packing and Marketing	0.00	0.00
Interest on Capital	42.60	0.01
Blueberry Tax	27.86	0.0075
TOT. VARIABLE COSTS	2,521.06	0.68
FIXED COSTS:		
Machinery & Equipment	60.10	0.01
Taxes	32.83	0.01
TOTAL FIXED COSTS	92.93	0.02
TOTAL COSTS	2,613.99	0.70
RETURNS ABOVE COSTS SHOWN	3,068.44	0.83
AVERAGE TOTAL ANNUAL RETURN TO MANAGEMENT		
($/FARM)	**3,068.44**	

Wild Maine Blueberry Budget		
Organic 17 Freshpack		
Number of Acres (Crop)	2.50	
Yield (Lbs./Acre)	3,714.00	
Price/Lb. ($)	4.00	
REVENUE/ACRE ($)	14,856.00	
VARIABLE COSTS	**($/Acre)**	**($/Pound)**
Pruning:		
Burning	0.00	0.00
Mowing	70.43	0.02
Average Pruning	70.43	0.02
Weed Control	527.57	0.14
Fertilization	60.00	0.02
Pollination	310.00	0.08
Pest Monitoring	15.00	0.00
Insect Control	0.00	0.00
Disease Control	0.00	0.00
Irrigation	0.00	0.00
Sulfur (pH)	240.00	0.06
Harvest:		
Raking	1,227.60	0.33
Mechanical	0.00	0.00
Average Harvest	1,227.60	0.33
Packing and Marketing	2,492.40	0.67
Interest on Capital	42.60	0.01
Blueberry Tax	0.00	0.00
TOT. VARIABLE COSTS	4,985.60	1.34
FIXED COSTS:		
Machinery & Equipment	60.10	0.01
Taxes	32.83	0.01
TOTAL FIXED COSTS	92.93	0.02
TOTAL COSTS	5,078.53	1.37
RETURNS ABOVE COSTS SHOWN	9,777.47	2.63
AVERAGE TOTAL ANNUAL RETURN TO MANAGEMENT		
($/FARM)	**24,443.68**	

Wild Maine Blueberry Budget		
Organic 18 Processed		
Number of Acres (Crop)	1.00	
Yield (Lbs./Acre)	1,087.00	
Price/Lb. ($)	1.10	
REVENUE/ACRE ($)	1,195.70	
VARIABLE COSTS	**($/Acre)**	**($/Pound)**
Pruning:		
Burning	252.00	0.23
Mowing	0.00	0.00
Average Pruning	252.00	0.23
Weed Control	52.50	0.05
Fertilization	0.00	0.00
Pollination	100.00	0.09
Pest Monitoring	0.00	0.00
Insect Control	0.00	0.00
Disease Control	0.00	0.00
Irrigation	0.00	0.00
Sulfur (pH)	0.00	0.00
Harvest:		
Raking	540.00	0.50
Mechanical	0.00	0.00
Average Harvest	540.00	0.50
Packing and Marketing	0.00	0.00
Interest on Capital	42.60	0.04
Blueberry Tax	8.15	0.0075
TOT. VARIABLE COSTS	995.25	0.92
FIXED COSTS:		
Machinery & Equipment	60.10	0.01
Taxes	32.83	0.01
TOTAL FIXED COSTS	92.93	0.02
TOTAL COSTS	1,088.18	1.00
RETURNS ABOVE COSTS SHOWN	107.52	0.10
AVERAGE TOTAL ANNUAL RETURN TO MANAGEMENT		
($/FARM)	**107.52**	

Wild Maine Blueberry Budget		
Organic 18 Freshpack		
Number of Acres (Crop)	5.00	
Yield (Lbs./Acre)	1,087.00	
Price/Lb. ($)	3.75	
REVENUE/ACRE ($)	4,076.25	
VARIABLE COSTS	**($/Acre)**	**($/Pound)**
Pruning:		
Burning	252.00	0.23
Mowing	0.00	0.00
Average Pruning	252.00	0.23
Weed Control	52.50	0.05
Fertilization	0.00	0.00
Pollination	100.00	0.09
Pest Monitoring	0.00	0.00
Insect Control	0.00	0.00
Disease Control	0.00	0.00
Irrigation	0.00	0.00
Sulfur (pH)	0.00	0.00
Harvest:		
Raking	540.00	0.50
Mechanical	0.00	0.00
Average Harvest	540.00	0.50
Packing and Marketing	815.00	0.75
Interest on Capital	42.60	0.04
Blueberry Tax	0.00	0.00
TOT. VARIABLE COSTS	1,802.10	1.66
FIXED COSTS:		
Machinery & Equipment	60.10	0.01
Taxes	32.83	0.01
TOTAL FIXED COSTS	92.93	0.02
TOTAL COSTS	1,895.03	1.74
RETURNS ABOVE COSTS SHOWN	2,181.22	2.01
AVERAGE TOTAL ANNUAL RETURN TO MANAGEMENT		
($/FARM)	**10,906.10**	

Wild Maine Blueberry Budget		
Organic 19 Processed		
Number of Acres (Crop)	3.0	
Yield (Lbs./Acre)	3,013.00	
Price/Lb. ($)	1.59	
REVENUE/ACRE ($)	4,790.67	
VARIABLE COSTS	**($/Acre)**	**($/Pound)**
Pruning:		
Burning	0.00	0.00
Mowing	264.86	0.09
Average Pruning	264.86	0.09
Weed Control	1,033.33	0.34
Fertilization	0.00	0.00
Pollination	304.05	0.10
Pest Monitoring	0.00	0.00
Insect Control	0.00	0.00
Disease Control	0.00	0.00
Irrigation	0.00	0.00
Sulfur (pH)	0.00	0.00
Harvest:		
Raking	432.44	0.14
Mechanical	0.00	0.00
Average Harvest	432.44	0.14
Packing and Marketing	0.00	0.00
Interest on Capital	42.60	0.01
Blueberry Tax	0.00	0.00
TOT. VARIABLE COSTS	2,077.28	0.69
FIXED COSTS:		
Machinery & Equipment	60.10	0.01
Taxes	32.83	0.01
TOTAL FIXED COSTS	92.93	0.02
TOTAL COSTS	2,170.21	0.72
RETURNS ABOVE COSTS SHOWN	2,620.46	0.87
AVERAGE TOTAL ANNUAL RETURN TO MANAGEMENT		
($/FARM)	**7,861.38**	

Wild Maine Blueberry Budget		
Organic 19 Freshpack		
Number of Acres (Crop)	0.7	
Yield (Lbs./Acre)	3,013.00	
Price/Lb. ($)	3.75	
REVENUE/ACRE ($)	11,298.75	
VARIABLE COSTS	**($/Acre)**	**($/Pound)**
Pruning:		
Burning	0.00	0.00
Mowing	264.86	0.09
Average Pruning	264.86	0.09
Weed Control	1,033.33	0.34
Fertilization	0.00	0.00
Pollination	304.05	0.10
Pest Monitoring	0.00	0.00
Insect Control	0.00	0.00
Disease Control	0.00	0.00
Irrigation	0.00	0.00
Sulfur (pH)	0.00	0.00
Harvest:		
Raking	432.44	0.14
Mechanical	0.00	0.00
Average Harvest	432.44	0.14
Packing and Marketing	0.00	0.00
Interest on Capital	42.60	0.01
Blueberry Tax	0.00	0.00
TOT. VARIABLE COSTS	2,077.28	0.69
FIXED COSTS:		
Machinery & Equipment	60.10	0.01
Taxes	32.83	0.01
TOTAL FIXED COSTS	92.93	0.02
TOTAL COSTS	2,170.21	0.72
RETURNS ABOVE COSTS SHOWN	9,128.54	3.03
AVERAGE TOTAL ANNUAL RETURN TO MANAGEMENT		
($/FARM)	**6,389.98**	

Wild Maine Blueberry Budget		
Low 7		
Number of Acres (Crop)	30.00	
Yield (Lbs./Acre)	4,039.30	
Price/Lb. ($)	0.50	
REVENUE/ACRE ($)	2,019.65	
VARIABLE COSTS	**($/Acre)**	**($/Pound)**
Pruning:		
Burning	531.94	0.13
Mowing	57.80	0.01
Average Pruning	589.74	0.15
Weed Control	198.75	0.05
Fertilization	202.72	0.05
Pollination	247.20	0.06
Pest Monitoring	0.00	0.00
Insect Control	71.28	0.02
Disease Control	18.46	0.00
Irrigation	0.00	0.00
Sulfur (pH)	0.00	0.00
Harvest:		
Raking	1,090.62	0.27
Mechanical	0.00	0.00
Average Harvest	1,090.62	0.27
Packing and Marketing	0.00	0.00
Interest on Capital	42.60	0.01
Blueberry Tax	60.59	0.0150
TOT. VARIABLE COSTS	2,521.96	0.62
FIXED COSTS:		
Machinery & Equipment	60.10	0.01
Taxes	32.83	0.01
TOTAL FIXED COSTS	92.93	0.02
TOTAL COSTS	2,614.89	0.65
RETURNS ABOVE COSTS SHOWN	(595.24)	(0.15)
AVERAGE TOTAL ANNUAL RETURN TO MANAGEMENT		
($/FARM)	**(17,857.19)**	

Wild Maine Blueberry Budget		
Low Input 8		
Number of Acres (Crop)	7.00	
Yield (Lbs./Acre)	3,592.4	
Price/Lb. ($)	0.50	
REVENUE/ACRE ($)	1,796.20	
VARIABLE COSTS	**($/Acre)**	**($/Pound)**
Pruning:		
Burning	0.00	0.00
Mowing	60.00	0.02
Average Pruning	60.00	0.02
Weed Control	110.00	0.03
Fertilization	50.00	0.01
Pollination	126.00	0.04
Pest Monitoring	0.00	0.00
Insect Control	27.00	0.01
Disease Control	0.00	0.00
Irrigation	0.00	0.00
Sulfur (pH)	0.00	0.00
Harvest:		
Raking	0.00	0.00
Mechanical	484.97	0.13
Average Harvest	484.97	0.13
Packing and Marketing	0.00	0.00
Interest on Capital	42.60	0.01
Blueberry Tax	53.89	0.0150
TOT. VARIABLE COSTS	954.46	0.27
FIXED COSTS:		
Machinery & Equipment	60.10	0.01
Taxes	32.83	0.01
TOTAL FIXED COSTS	92.93	0.02
TOTAL COSTS	1,047.39	0.29
RETURNS ABOVE COSTS SHOWN	748.81	0.29
AVERAGE TOTAL ANNUAL RETURN TO MANAGEMENT		
($/FARM)	**5,241.70**	

Wild Maine Blueberry Budget		
Low Input 20		
Number of Acres (Crop)	14.00	
Yield (Lbs./Acre)	1,810.40	
Price/Lb. ($)	0.50	
REVENUE/ACRE ($)	905.20	
VARIABLE COSTS	**($/Acre)**	**($/Pound)**
Pruning:		
Burning	513.26	0.28
Mowing	0.00	0.00
Average Pruning	513.26	0.28
Weed Control	123.61	0.07
Fertilization	102.34	0.06
Pollination	235.71	0.13
Pest Monitoring	0.00	0.00
Insect Control	25.57	0.01
Disease Control	89.94	0.05
Irrigation	0.00	0.00
Sulfur (pH)	0.00	0.00
Harvest:		
Raking	298.72	0.17
Mechanical	0.00	0.00
Average Harvest	298.72	0.17
Packing and Marketing	0.00	0.00
Interest on Capital	42.60	0.02
Blueberry Tax	13.58	0.0075
TOT. VARIABLE COSTS	1,445.33	0.80
FIXED COSTS:		
Machinery & Equipment	60.10	0.01
Taxes	32.83	0.01
TOTAL FIXED COSTS	92.93	0.02
TOTAL COSTS	1,538.26	0.85
RETURNS ABOVE COSTS SHOWN	(633.06)	(0.35)
AVERAGE TOTAL ANNUAL RETURN TO MANAGEMENT		
($/FARM)	**(8,862.85)**	

Wild Maine Blueberry Budget		
Low Input 21		
Number of Acres (Crop)	14.00	
Yield (Lbs./Acre)	4,326.20	
Price/Lb. ($)	0.50	
REVENUE/ACRE ($)	2,163.10	
VARIABLE COSTS	**($/Acre)**	**($/Pound)**
Pruning:		
Burning	612.42	0.14
Mowing	0.00	0.00
Average Pruning	612.42	0.14
Weed Control	128.42	0.03
Fertilization	81.79	0.02
Pollination	274.71	0.06
Pest Monitoring	0.00	0.00
Insect Control	25.57	0.01
Disease Control	89.22	0.02
Irrigation	0.00	0.00
Sulfur (pH)	0.00	0.00
Harvest:		
Raking	832.79	0.19
Mechanical	0.00	0.00
Average Harvest	832.79	0.19
Packing and Marketing	0.00	0.00
Interest on Capital	42.60	0.01
Blueberry Tax	32.45	0.0075
TOT. VARIABLE COSTS	2,119.97	0.49
FIXED COSTS:		
Machinery & Equipment	60.10	0.01
Taxes	32.83	0.01
TOTAL FIXED COSTS	92.93	0.02
TOTAL COSTS	2,212.90	0.51
RETURNS ABOVE COSTS SHOWN	(49.80)	(0.01)
AVERAGE TOTAL ANNUAL RETURN TO MANAGEMENT		
($/FARM)	**(697.15)**	

Wild Maine Blueberry Budget		
Medium Input 9		
Number of Acres (Crop)	44.00	
Yield (Lbs./Acre)	1,301.00	
Price/Lb. ($)	0.50	
REVENUE/ACRE ($)	650.50	
VARIABLE COSTS	**($/Acre)**	**($/Pound)**
Pruning:		
Burning	0.00	0.00
Mowing	115.32	0.09
Average Pruning	115.32	0.09
Weed Control	116.27	0.09
Fertilization	138.02	0.11
Pollination	390.00	0.30
Pest Monitoring	5.70	0.00
Insect Control	45.25	0.03
Disease Control	15.43	0.01
Irrigation	0.00	0.00
Sulfur (pH)	0.00	0.00
Harvest:		
Raking	0.00	0.00
Mechanical	306.59	0.24
Average Harvest	306.59	0.24
Packing and Marketing	0.00	0.00
Interest on Capital	42.60	0.03
Blueberry Tax	19.52	0.0150
TOT. VARIABLE COSTS	1,194.70	0.92
FIXED COSTS:		
Machinery & Equipment	60.10	0.01
Taxes	32.83	0.01
TOTAL FIXED COSTS	92.93	0.02
TOTAL COSTS	1,287.63	0.99
RETURNS ABOVE COSTS SHOWN	(637.13)	(0.49)
AVERAGE TOTAL ANNUAL RETURN TO MANAGEMENT		
($/FARM)	**(28,033.50)**	

Wild Maine Blueberry Budget		
Medium Input 10		
Number of Acres (Crop)	77.00	
Yield (Lbs./Acre)	2,642.00	
Price/Lb. ($)	0.50	
REVENUE/ACRE ($)	1,321.00	
VARIABLE COSTS	**($/Acre)**	**($/Pound)**
Pruning:		
Burning	0.00	0.00
Mowing	141.86	0.05
Average Pruning	141.86	0.05
Weed Control	65.97	0.02
Fertilization	141.54	0.05
Pollination	384.94	0.15
Pest Monitoring	6.51	0.00
Insect Control	34.49	0.01
Disease Control	36.50	0.01
Irrigation	0.00	0.00
Sulfur (pH)	0.00	0.00
Harvest:		
Raking	0.00	0.00
Mechanical	345.62	0.13
Average Harvest	345.62	0.13
Packing and Marketing	0.00	0.00
Interest on Capital	42.60	0.02
Blueberry Tax	39.63	0.0150
TOT. VARIABLE COSTS	1,239.66	0.47
FIXED COSTS:		
Machinery & Equipment	60.10	0.01
Taxes	32.83	0.01
TOTAL FIXED COSTS	92.93	0.02
TOTAL COSTS	1,332.59	0.50
RETURNS ABOVE COSTS SHOWN	(11.59)	(0.00)
AVERAGE TOTAL ANNUAL RETURN TO MANAGEMENT		
($/FARM)	**(892.43)**	

Wild Maine Blueberry Budget		
Medium Input 22		
Number of Acres (Crop)	29.00	
Yield (Lbs./Acre)	3,330.50	
Price/Lb. ($)	0.43	
REVENUE/ACRE ($)	1,432.12	
VARIABLE COSTS	**($/Acre)**	**($/Pound)**
Pruning:		
Burning	514.11	0.15
Mowing	140.38	0.04
Average Pruning	654.49	0.20
Weed Control	376.51	0.11
Fertilization	52.17	0.02
Pollination	382.20	0.11
Pest Monitoring	9.40	0.00
Insect Control	9.83	0.00
Disease Control	0.00	0.00
Irrigation	0.00	0.00
Sulfur (pH)	0.00	0.00
Harvest:		
Raking	773.58	0.23
Mechanical	636.53	0.19
Average Harvest	705.06	0.21
Packing and Marketing	0.00	0.00
Interest on Capital	42.60	0.01
Blueberry Tax	24.98	0.0075
TOT. VARIABLE COSTS	2,257.24	0.68
FIXED COSTS:		
Machinery & Equipment	60.10	0.01
Taxes	32.83	0.01
TOTAL FIXED COSTS	92.93	0.02
TOTAL COSTS	2,350.17	0.71
RETURNS ABOVE COSTS SHOWN	(918.05)	(0.28)
AVERAGE TOTAL ANNUAL RETURN TO MANAGEMENT		
($/FARM)	**(26,623.56)**	

Wild Maine Blueberry Budget		
Medium Input 23		
Number of Acres (Crop)	5.70	
Yield (Lbs./Acre)	2,106.00	
Price/Lb. ($)	0.50	
REVENUE/ACRE ($)	1,053.00	
VARIABLE COSTS	**($/Acre)**	**($/Pound)**
Pruning:		
Burning	0.00	0.00
Mowing	98.16	0.05
Average Pruning	98.16	0.05
Weed Control	174.69	0.08
Fertilization	108.80	0.05
Pollination	200.00	0.09
Pest Monitoring	0.00	0.00
Insect Control	44.09	0.02
Disease Control	35.18	0.02
Irrigation	0.00	0.00
Sulfur (pH)	0.00	0.00
Harvest:		
Raking	0.00	0.00
Mechanical	379.08	0.18
Average Harvest	379.08	0.18
Packing and Marketing	0.00	0.00
Interest on Capital	42.60	0.02
Blueberry Tax	15.80	0.0075
TOT. VARIABLE COSTS	1,098.40	0.52
FIXED COSTS:		
Machinery & Equipment	60.10	0.01
Taxes	32.83	0.01
TOTAL FIXED COSTS	92.93	0.02
TOTAL COSTS	1,191.33	0.57
RETURNS ABOVE COSTS SHOWN	(138.33)	(0.07)
AVERAGE TOTAL ANNUAL RETURN TO MANAGEMENT		
($/FARM)	**(788.45)**	

Wild Maine Blueberry Budget		
High Input 13		
Number of Acres (Crop)	112.54	
Yield (Lbs./Acre)	3,635.00	
Price/Lb. ($)	0.50	
REVENUE/ACRE ($)	1,817.50	
VARIABLE COSTS	**($/Acre)**	**($/Pound)**
Pruning:		
Burning	0.00	0.00
Mowing	63.00	0.02
Average Pruning	63.00	0.02
Weed Control	177.52	0.05
Fertilization	188.72	0.05
Pollination	762.00	0.21
Pest Monitoring	25.50	0.01
Insect Control	121.11	0.03
Disease Control	233.59	0.06
Irrigation	55.00	0.02
Sulfur (pH)	0.00	0.00
Harvest:		
Raking	0.00	0.00
Mechanical	385.31	0.11
Average Harvest	385.31	0.11
Packing and Marketing	0.00	0.00
Interest on Capital	42.60	0.01
Blueberry Tax	54.53	0.0150
TOT. VARIABLE COSTS	2,108.88	0.58
FIXED COSTS:		
Machinery & Equipment	60.10	0.01
Taxes	32.83	0.01
TOTAL FIXED COSTS	92.93	0.02
TOTAL COSTS	2,201.81	0.61
RETURNS ABOVE COSTS SHOWN	(384.31)	(0.11)
AVERAGE TOTAL ANNUAL RETURN TO MANAGEMENT		
($/FARM)	**(43,249.68)**	

Wild Maine Blueberry Budget		
High Input 14		
Number of Acres (Crop)	63.29	
Yield (Lbs./Acre)	2,971.00	
Price/Lb. ($)	0.50	
REVENUE/ACRE ($)	1,485.50	
VARIABLE COSTS	**($/Acre)**	**($/Pound)**
Pruning:		
Burning	0.00	0.00
Mowing	63.00	0.02
Average Pruning	63.00	0.02
Weed Control	199.89	0.07
Fertilization	188.72	0.06
Pollination	762.00	0.26
Pest Monitoring	25.50	0.01
Insect Control	121.11	0.04
Disease Control	220.83	0.07
Irrigation	70.00	0.02
Sulfur (pH)	0.00	0.00
Harvest:		
Raking	0.00	0.00
Mechanical	314.93	0.11
Average Harvest	314.93	0.11
Packing and Marketing	0.00	0.00
Interest on Capital	42.60	0.01
Blueberry Tax	44.57	0.0150
TOT. VARIABLE COSTS	2,053.15	0.69
FIXED COSTS:		
Machinery & Equipment	60.10	0.01
Taxes	32.83	0.01
TOTAL FIXED COSTS	92.93	0.02
TOTAL COSTS	2,146.08	0.72
RETURNS ABOVE COSTS SHOWN	(660.58)	(0.22)
AVERAGE TOTAL ANNUAL RETURN TO MANAGEMENT		
($/FARM)	**(41,807.79)**	

Wild Maine Blueberry Budget		
High Input 15		
Number of Acres (Crop)	82.11	
Yield (Lbs./Acre)	7,619.00	
Price/Lb. ($)	0.50	
REVENUE/ACRE ($)	3,809.50	
VARIABLE COSTS	**($/Acre)**	**($/Pound)**
Pruning:		
Burning	0.00	0.00
Mowing	63.00	0.01
Average Pruning	63.00	0.01
Weed Control	273.46	0.04
Fertilization	188.72	0.02
Pollination	762.00	0.10
Pest Monitoring	25.50	0.00
Insect Control	55.06	0.01
Disease Control	255.83	0.03
Irrigation	70.00	0.01
Sulfur (pH)	0.00	0.00
Harvest:		
Raking	0.00	0.00
Mechanical	807.61	0.11
Average Harvest	807.61	0.11
Packing and Marketing	0.00	0.00
Interest on Capital	42.60	0.01
Blueberry Tax	114.29	0.0150
TOT. VARIABLE COSTS	2,658.07	0.35
FIXED COSTS:		
Machinery & Equipment	60.10	0.01
Taxes	32.83	0.01
TOTAL FIXED COSTS	92.93	0.02
TOTAL COSTS	2,751.00	0.36
RETURNS ABOVE COSTS SHOWN	1,058.51	0.14
AVERAGE TOTAL ANNUAL RETURN TO MANAGEMENT		
($/FARM)	**86,913.85**	

Wild Maine Blueberry Budget		
High Input 16		
Number of Acres (Crop)	132.19	
Yield (Lbs./Acre)	6,500.00	
Price/Lb. ($)	0.50	
REVENUE/ACRE ($)	3,250.00	
VARIABLE COSTS	**($/Acre)**	**($/Pound)**
Pruning:		
Burning	0.00	0.00
Mowing	63.00	0.01
Average Pruning	63.00	0.01
Weed Control	180.64	0.03
Fertilization	188.72	0.03
Pollination	762.00	0.12
Pest Monitoring	25.50	0.00
Insect Control	121.11	0.02
Disease Control	252.80	0.04
Irrigation	70.00	0.01
Sulfur (pH)	0.00	0.00
Harvest:		
Raking	0.00	0.00
Mechanical	689.00	0.11
Average Harvest	689.00	0.11
Packing and Marketing	0.00	0.00
Interest on Capital	42.60	0.01
Blueberry Tax	97.50	0.0150
TOT. VARIABLE COSTS	2,492.87	0.38
FIXED COSTS:		
Machinery & Equipment	60.10	0.01
Taxes	32.83	0.01
TOTAL FIXED COSTS	92.93	0.02
TOTAL COSTS	2,585.80	0.40
RETURNS ABOVE COSTS SHOWN	664.20	0.10
AVERAGE TOTAL ANNUAL RETURN TO MANAGEMENT		
($/FARM)	**87,800.60**	

INPUT SYSTEMS STUDY – *ANCILLARY STUDY*

DISEASE MANAGEMENT: Seanna Annis, Assoc. Professor, School of Biology and Ecology
Erika Lyon, MS graduate student, School of Biology & Ecology
Rachael Martin, Research Asst., School of Biology & Ecology

18. TITLE: Ancillary projects in disease research.

OBJECTIVES:
- Determine possible sources and methods of spread of Valdensia leaf spot; and
- Determine the timing of spore release of leaf spot causing fungi using spore traps.

METHODS: Fields suspected to have Valdensia leaf spot in 2015 were visited in July to August to survey for disease. If the field showed Valdensia leaf spot symptoms, 5 to 7 stems in diseased areas about 10 ft apart were collected and placed in individual plastic bags. Isolates were obtained by surface sterilizing ten infected leaves per sample and plating them out on half-strength oatmeal medium amended with antibiotics. Plated leaves were incubated at 17°C under 12 hr light to induce spore formation by *Valdensia*. *Valdensia* spores were transferred to new plates and put into storage for genetic analysis. Fields previously known to have Valdensia leaf spot were visited in May to look for sexual structures on diseased leaves and later in the season to determine how well control methods were working.
Spore traps were placed in the prune fields at Blueberry Hill Research Farm, East Machias and Wesley in May 2015. These locations also had weather stations collecting humidity, leaf wetness and air temperature data. We collected spore tapes containing the trapped airborne spores every week until mid-October 2015. Spore trap tapes were cut in half and half was frozen for future DNA work and the other half was mounted on glass slides. Tapes from 4 hour intervals were examined for rust spores and rust spores were counted. Plant samples with signs of powdery mildew and leaf rust were collected in August and September. Fungal structures for powdery mildews or rusts were scraped off of leaves and their DNA was extracted. Specific sections of this DNA will be sequenced and used to design primers specific for these fungal organisms.

RESULTS: Five new fields were reported with Valdensia leaf spot this year. The spots ranged from less than a foot across to approximately 40 ft. A collection of isolates from Maine and Canada were DNA fingerprinted and the data analyzed. It appears the fungus is being spread by humans to new fields and once in a new field is reproducing clonally. We did not find any sexual structures in the plant debris examined this year. We did find that sclerotia of a few mm long are able to produce asexual spores under the wet conditions.
The tracking of spore release project is in its preliminary stages. Microscope slides from the spore trap tapes are still being examined. We have isolated DNA from rust samples from 2 locations and sequenced sections of the DNA and demonstrated it is from rust. We are currently working on isolating DNA from powdery mildew. In 2016, we hope to be designing primers specific to the powdery mildew fungus and rust fungus.

CONCLUSIONS/RECOMMENDATIONS: Valdensia leaf spot is still spreading among lowbush blueberry fields. Wet weather conditions around bloom provide an early start for this

disease. Extra care must be taken to wash equipment and remove all leaves before moving equipment among fields.

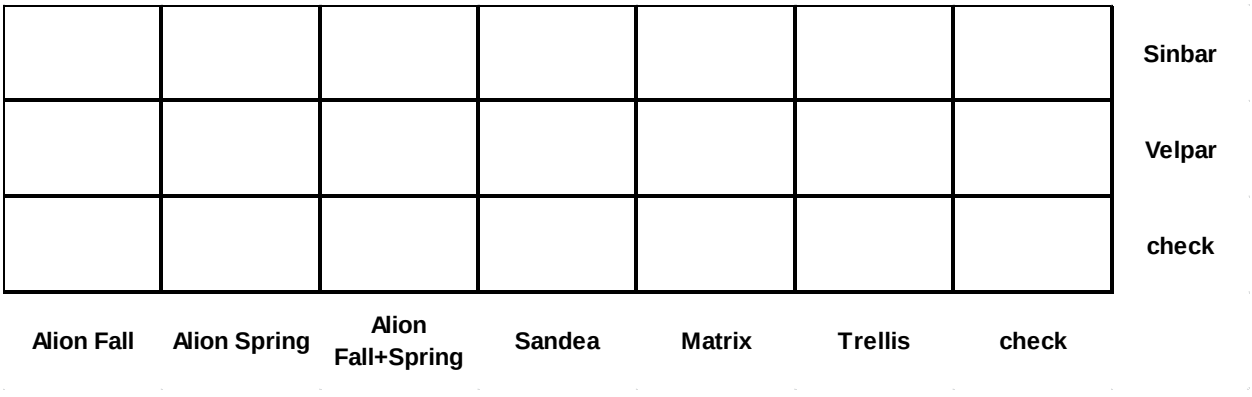
INPUT SYSTEMS STUDY – *ANCILLARY STUDY*

WEED MANAGEMENT: David E. Yarborough, Professor of Horticulture
Jennifer L.D. Cote, Assistant Scientist

19. TITLE: Evaluation of fall and spring combinations of preemergence herbicides to prevent weed resistance in wild blueberry fields, 2013-15.

METHODS: In the fall of 2013, a trial was initiated to test the efficacy of several unregistered and/or untested herbicides on wild blueberries in Maine, in conjunction with the industry standards Velpar and Sinbar. Nine sites across the wild blueberry growing region of Maine were sprayed in November 2013 and/or preemergence in May 2014, but seven were dropped due to lack of weeds from the growers' previous weed control. The two sites that were carried through into the crop year were the Northfield site and Blueberry Hill Farm in Jonesboro. The main 18'x54' treatments were as follows: Alion (5 oz/a fall; 6.5 oz/a spring; fall + spring at respective rates), Sandea (1 oz/a fall), Matrix (2 oz/a fall), Trellis (1.33 lb/a spring) and an untreated check. In spring 2014, 18'x72' strips of Sinbar (2 lb/a), Velpar (1 lb/a) or an untreated check were applied at right angles to the test herbicides for a total of 21 treatments (Figure 1). In the crop year, wild blueberry cover, broadleaf weed cover, and grass cover were assessed by sampling two 1 m^2 quadrats per treatment on 15 June 2015 for all treatments as well as the growers' spray regimes outside the trial areas. Cover was assessed using a Daubenmire cover scale converted to percent. The plots were harvested on 14 August 2015 by hand-raking all berries within two 1 m^2 quadrats per treatment. The berries were weighed prior to winnowing and weights were converted to lbs/acre for comparison. Main effects on cover of the herbicides alone, with Velpar or with Sinbar were compared to the check and the grower samples using t-tests for pairwise comparisons (α=0.05), and the treatments were compared to the check, Sinbar or Velpar (e.g. Velpar combinations to the check and to Velpar) using non-parametric one-way exact tests (α=0.05). Main effects of yield were compared to each other using t-tests (α=0.05), and treatment yields were compared to each other using a Tukey's test (α=0.05).

Figure 1. Example of plot layout (not to scale).

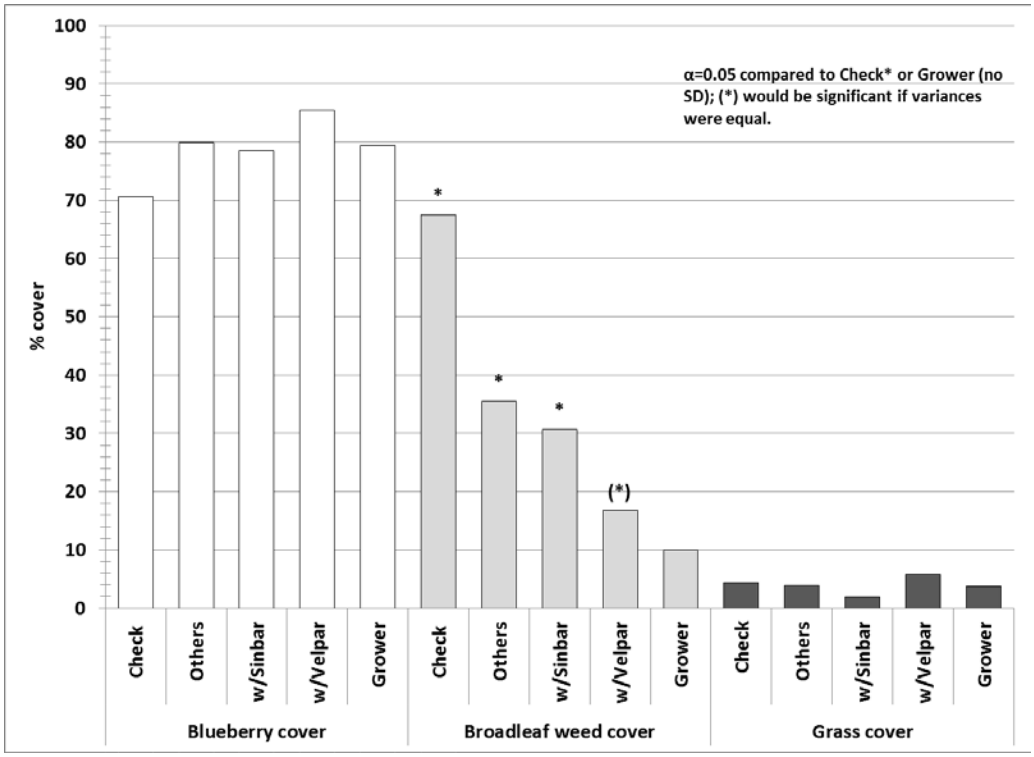


RESULTS:

Weed cover

There were no significant differences in wild blueberry cover for main effects of the test herbicides ("others") with or without the industry standards, when compared to the untreated check or grower spray regimens (Figure 2). Grass cover was <10% overall and there were also no significant differences for main effects (Figure 2). Broadleaf weed cover in the herbicides alone and with Sinbar were significantly lower compared to the check; although broadleaf weed cover in the Velpar combinations was lower than with Sinbar, alone or the check, the difference was not significant due to unequal variances (Figure 2).

Figure 2. Main effects of herbicides alone, with Sinbar and with Velpar on wild blueberry cover, broadleaf weed cover and grass cover compared to the check or grower in the crop year (α=0.05).



When all of the Alion treatments were compared to the check, and the Sinbar combinations were compared to Sinbar alone, there were no significant differences (Figure 3). Fall Alion + Velpar resulted in significantly higher blueberry cover compared to Velpar alone. There were no significant differences in blueberry cover for any Matrix, Sandea or Trellis treatment, alone or with Sinbar/Velpar, when compared to the check, Sinbar alone or Velpar alone (Figure 4).

Figure 3. Wild blueberry cover in the Alion treatments. All treatments are compared to the check; Sinbar combinations are compared to Sinbar alone, and Velpar combinations are compared to Velpar^ alone (α=0.05).

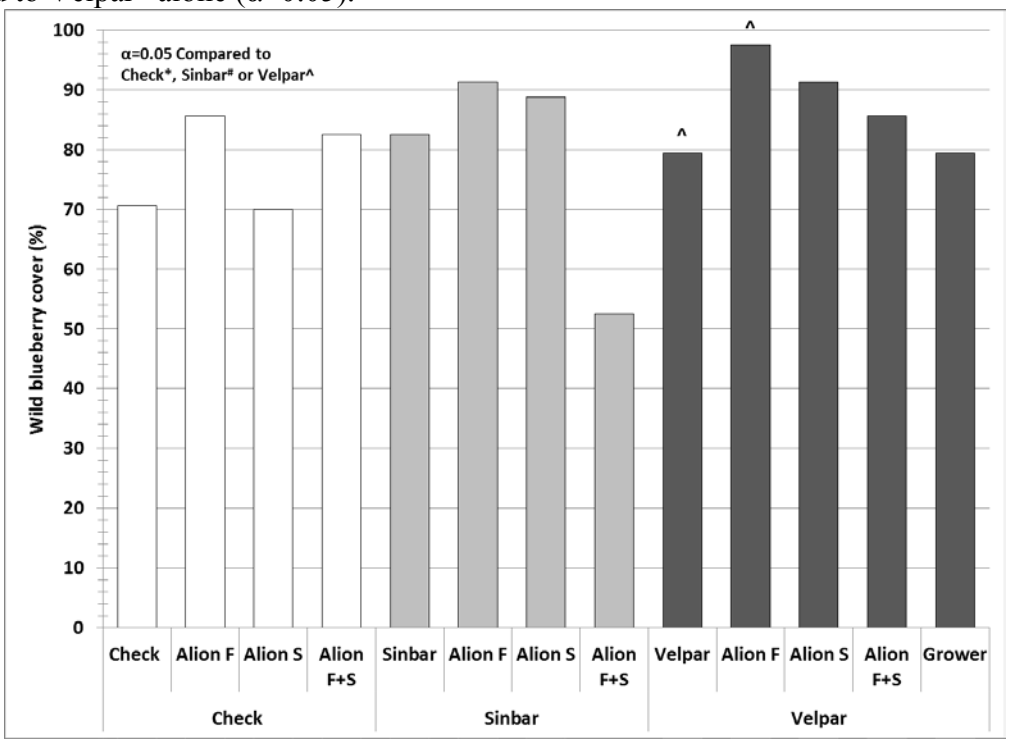
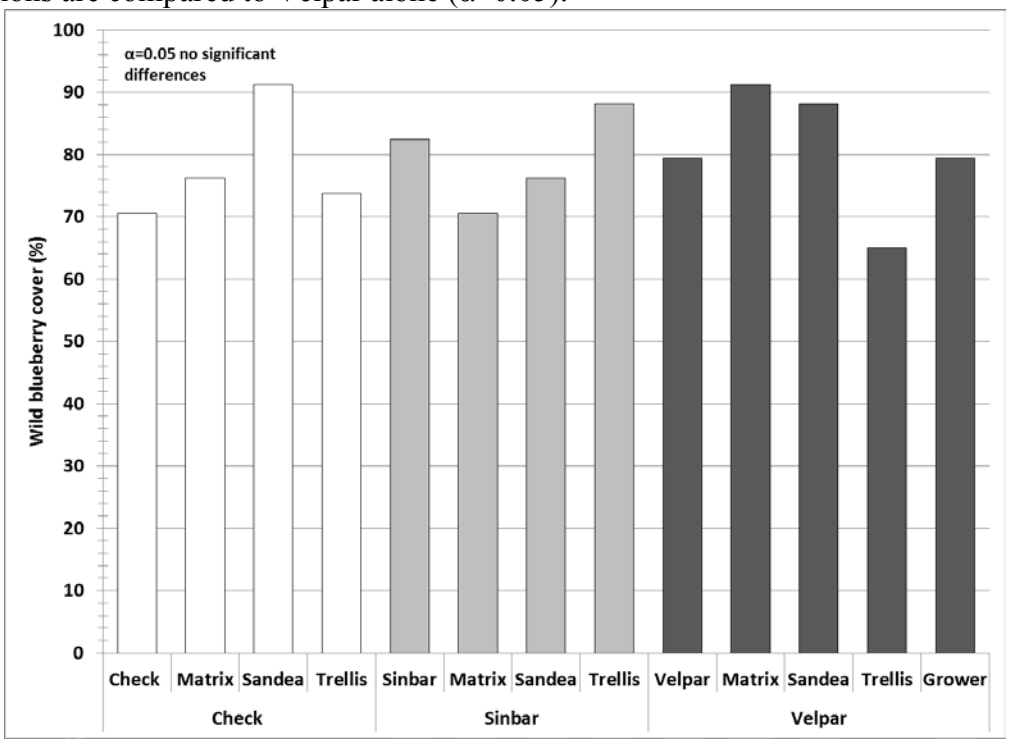


Figure 4. Wild blueberry cover in the Sandea, Matrix and Trellis treatments. All treatments are compared to the check; Sinbar combinations are compared to Sinbar alone, and Velpar combinations are compared to Velpar alone (α=0.05).



Broadleaf weed cover was significantly lower in the fall Alion+Sinbar, spring Alion+Sinbar, spring Alion+Velpar, fall+spring Alion+Velpar, and grower treatments compared to the check (Figure 5). In addition, the fall Alion+Sinbar was also significantly lower than Sinbar alone and had over 5x less broadleaf weed cover than fall Alion alone or with Velpar. Trellis alone, with Sinbar and with Velpar as well as Sandea+Velpar and the grower treatments also had significantly less broadleaf weed cover than the check (Figure 6). Trellis+Sinbar had significantly lower broadleaf weed cover compared to Sinbar alone, while Matrix+Sinbar and Velpar+Matrix appeared to release broadleaf weeds which resulted in more broadleaf weeds than the check, Sinbar or Velpar alone.

Figure 5. Broadleaf weed cover in the Alion treatments. All treatments are compared to the check*; Sinbar combinations are compared to Sinbar$^{\#}$ alone, and Velpar combinations are compared to Velpar alone (α=0.05).

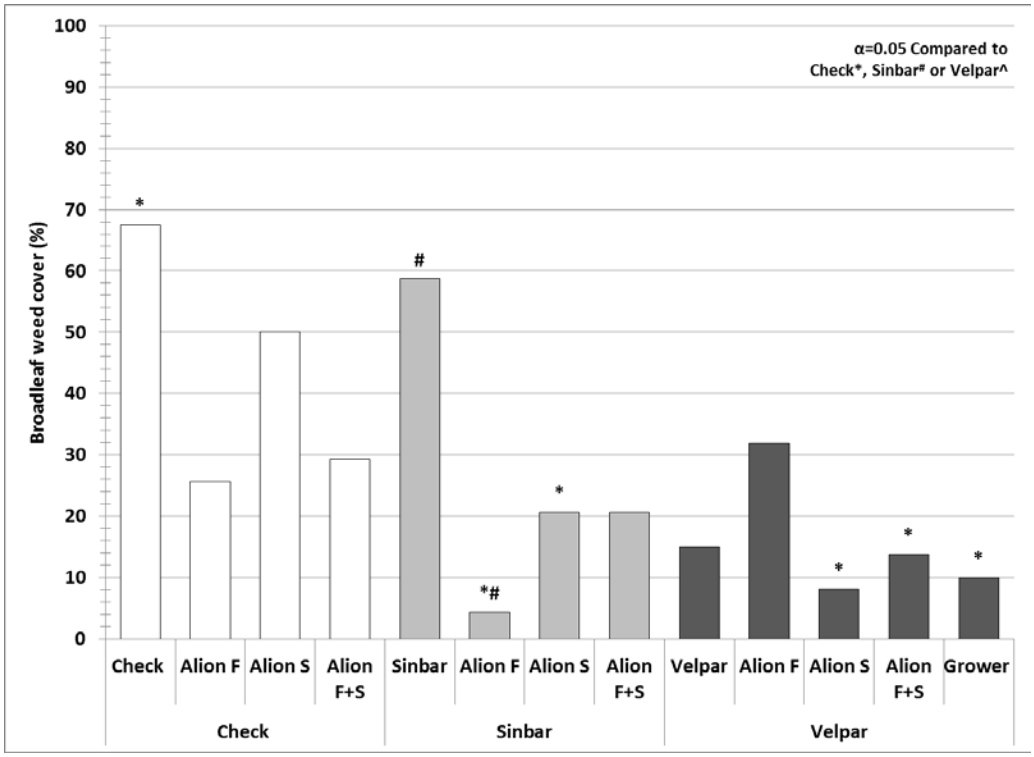
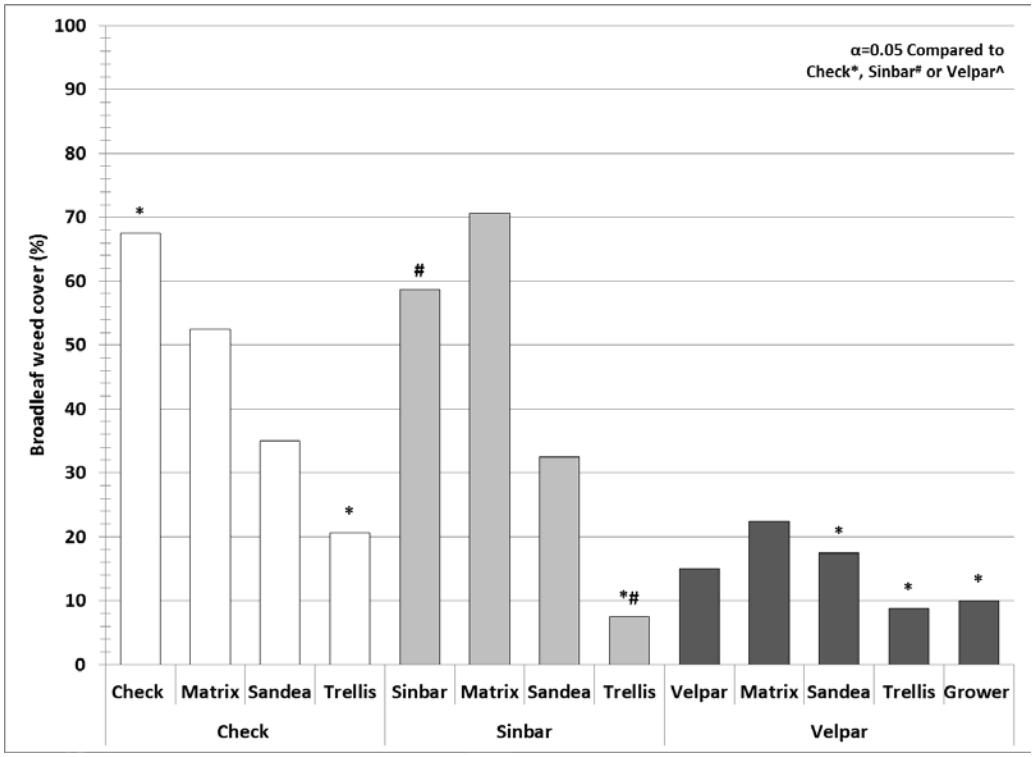


Figure 6. Broadleaf weed cover in the Sandea, Matrix and Trellis treatments. All treatments are compared to the check*; Sinbar combinations are compared to Sinbar# alone, and Velpar combinations are compared to Velpar alone (α=0.05).



Grass cover was less than 20% overall, with no significant differences for any herbicide alone or in combination compared to the check, Sinbar or Velpar (Figures 7-8). It should be noted that the fall + spring Alion treatments did not improve grass control compared to fall or spring only. Alion did not improve grass control in combination with Sinbar, but it did with Velpar (Figure 7). Trellis was not effective on grasses whatsoever, and without Sinbar it released grasses resulting in more grass cover than the check (Figure 8). Matrix + Velpar also released grasses compared to the check. In contrast, Sandea + Velpar eliminated grasses but when in combination with Sinbar it was not more effective than Sinbar alone or Sandea alone.

Figure 7. Grass cover in the Alion treatments. All treatments are compared to the check; Sinbar combinations are compared to Sinbar alone, and Velpar combinations are compared to Velpar alone (α=0.05).

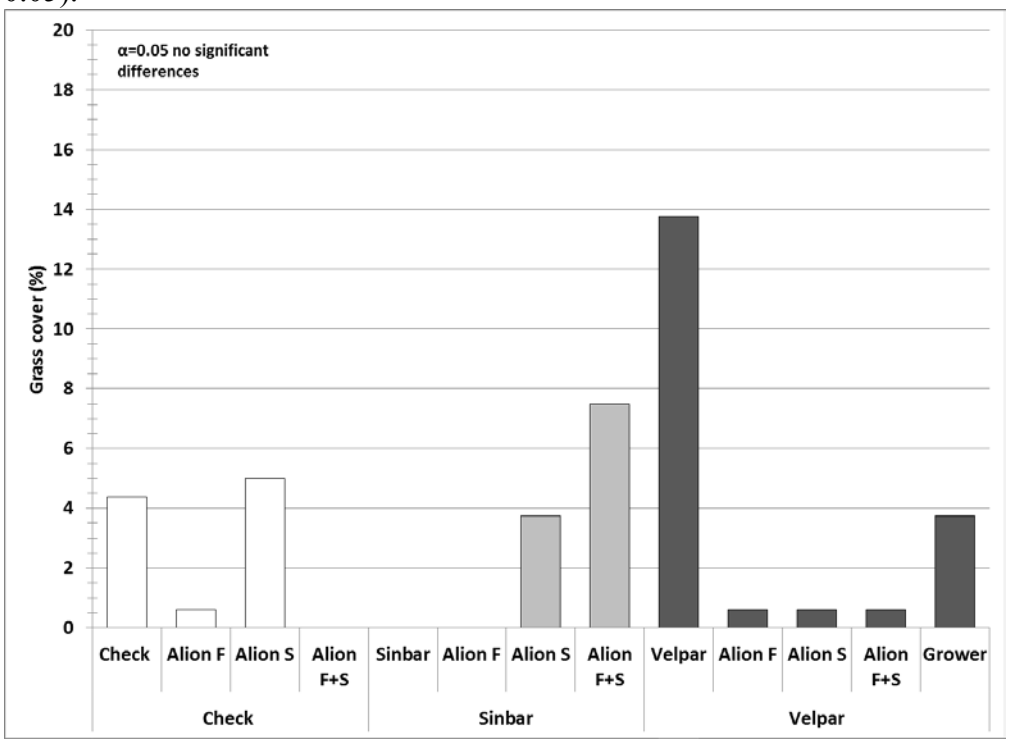
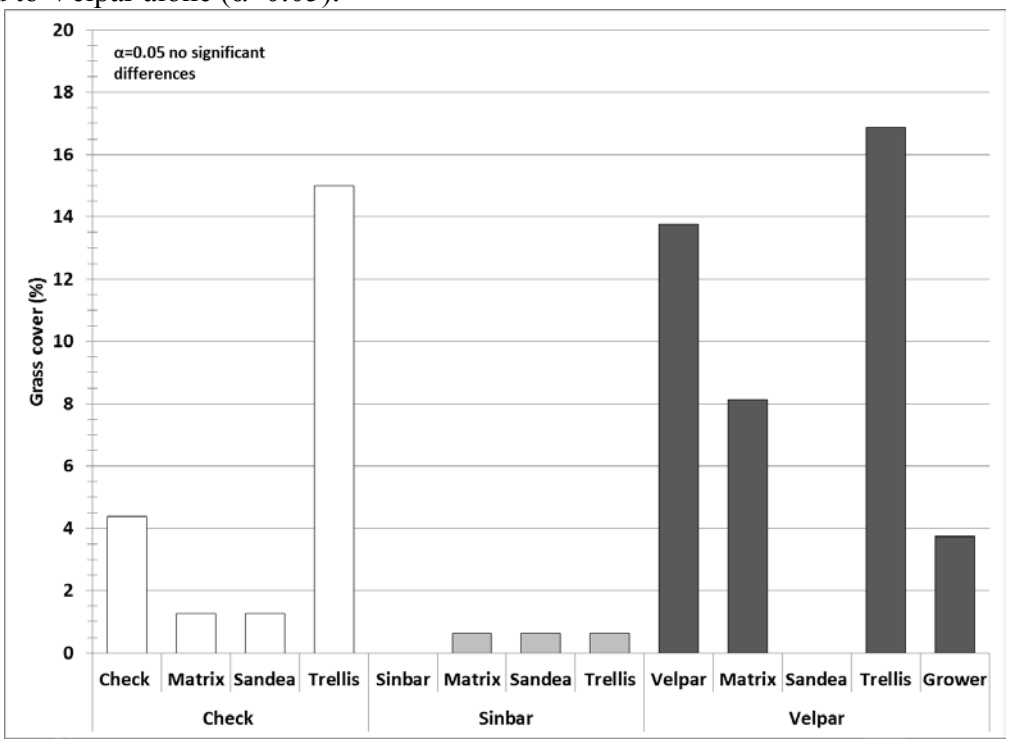


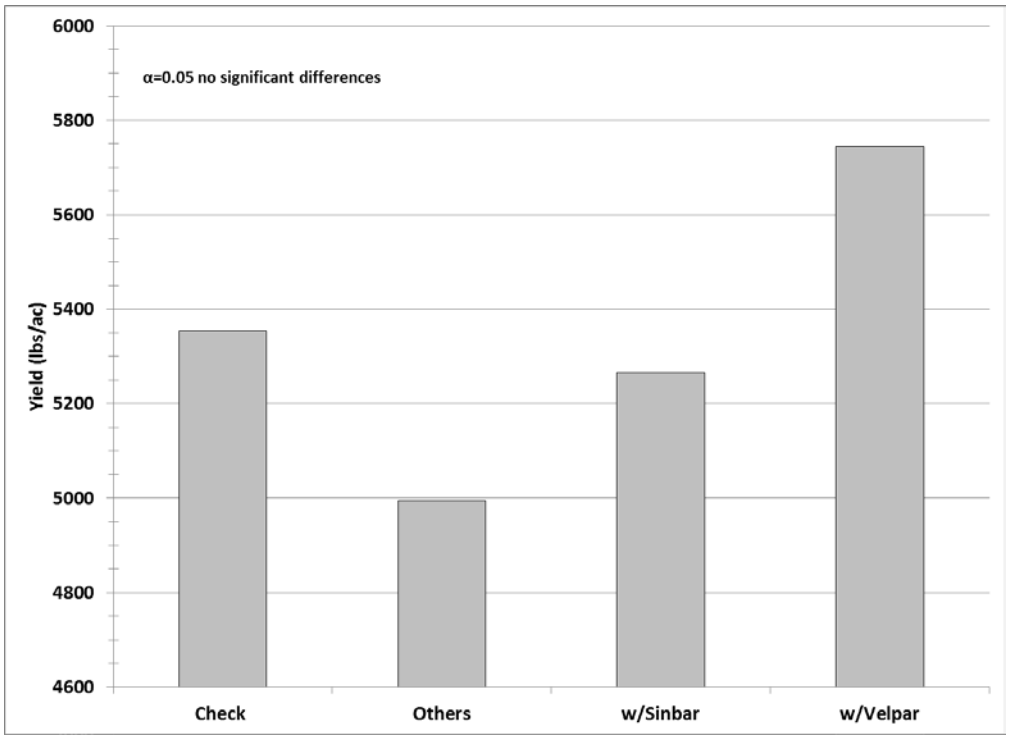
Figure 8. Grass cover in the Sandea, Matrix and Trellis treatments. All treatments are compared to the check; Sinbar combinations are compared to Sinbar alone, and Velpar combinations are compared to Velpar alone (α=0.05).



Yield

When the main effects of the herbicides on yield were compared, although there were no significant differences, the Velpar combinations had the highest yield, followed by the untreated check (Figure 9). The herbicides alone resulted in the lowest yield, but at approximately 5000 lbs/acre it was still considered "average". Figure 10 shows the comparison of yield among all treatments. Given the large differences in yields, for example Matrix + Sinbar at 3288 lbs/acre versus fall + spring Alion + Velpar at 7082 lbs/acre, significant differences should have been detected. The fact that no significant differences were detected is likely due to the large variances and lack of replication among samples obscuring any treatment effects.

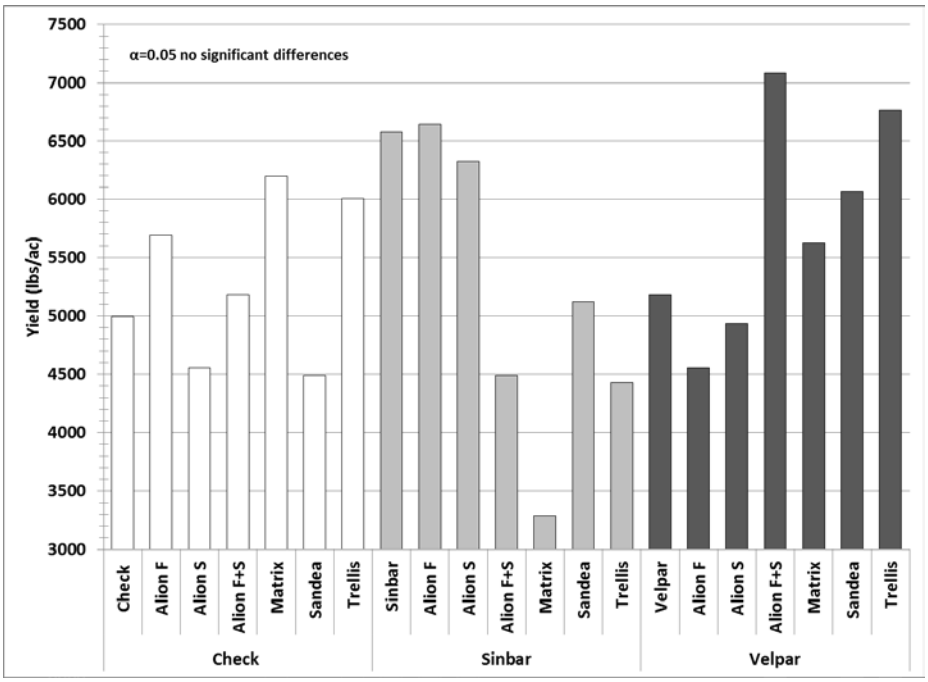
Figure 9. Main effects of the herbicides alone, with Sinbar or with Velpar on blueberry yield (α=0.05).



When applied alone, Matrix and Trellis resulted in the highest yields, and fall Alion application had the best yield of the three Alion treatments (Figure 10). The addition of Sinbar improved yield in the fall and the spring Alion treatments, but did not improve fall + spring Alion; and yield in the Sinbar only treatment was comparable to fall or spring Alion combinations. In contrast, adding Velpar to Alion reduced the fall or spring application yields compared to the herbicide with Sinbar, but greatly increased the fall + spring Alion combination.

The addition of Velpar to Matrix did not affect yield, but yield was greatly reduced with the addition of Sinbar (Figure 10). Finally, yield was improved when Sinbar or Velpar was added to Sandea.

Figure 10. Treatment comparisons for the effects of the test herbicides alone, with Sinbar or with Velpar on blueberry yield (α=0.05).

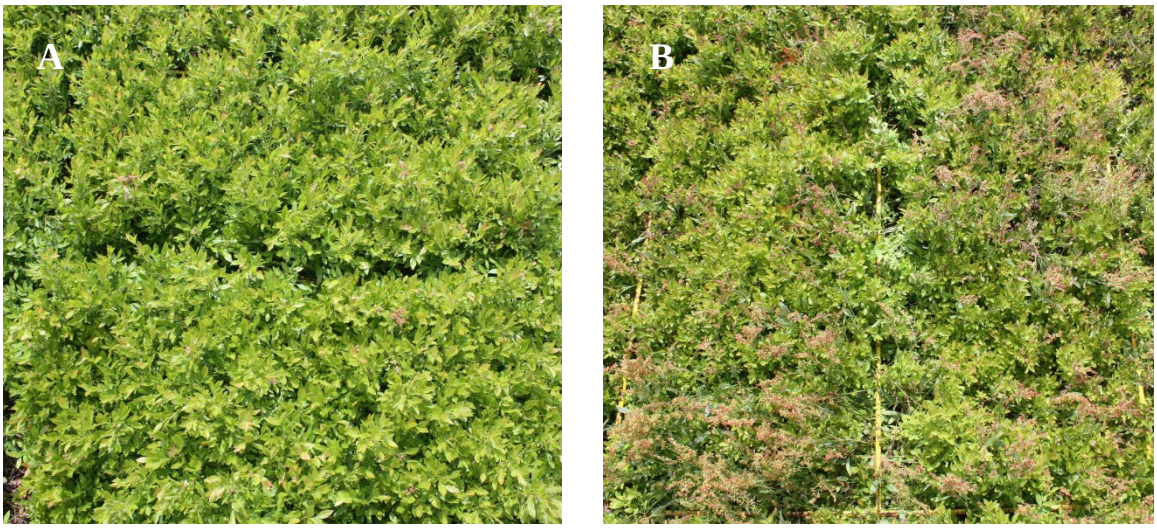


CONCLUSIONS:

Weed cover

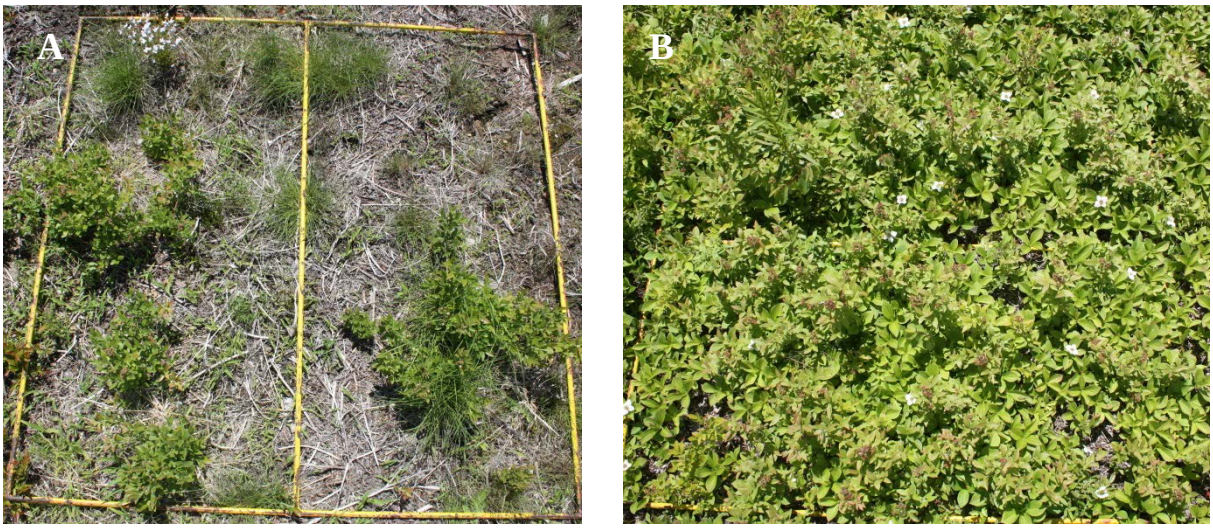
In the prune year we concluded that Alion, in combination with Sinbar, is effective in controlling both broadleaf weeds and grasses whether applied in the fall or spring. In the crop year, fall application of Alion was most effective on broadleaf weeds with or without Sinbar but it was not as effective with Velpar. A fall plus spring application of Alion did not improve broadleaf weed or grass control (Photos 1A-B) (see also yield section).

Photo 1. **A**) Fall Alion + Sinbar versus **B**) fall + spring Alion + Sinbar. The double application did not improve weed control.



Last year we stated that Trellis also appeared effective on both broadleaf weeds and grasses in combination with Sinbar. The trend held true this year, and Trellis was also effective on broadleaf weeds alone and with Velpar. However, Trellis alone or with Velpar actually resulted in more grasses than no treatment at all (Photo 2A-B).

Photo 2. Trellis + Velpar (**A**) resulted in more grasses than the check (**B**), but was more effective on broadleaf weeds.



In the prune year, Sandea alone or with Sinbar was effective on grasses, but did not offer long-term suppression of broadleaf weeds. In the crop year, Sinbar alone resulted in as good long-term grass control as with Sandea, but Sandea alone or with Velpar controlled grasses better than no treatment or Velpar alone. Sandea did not control broadleaf weeds significantly better than no treatment in the crop year unless used with Velpar, which indicates that it needed the Velpar combination to extend the broadleaf weed carryover control.

In 2014, Matrix exhibited fair control of weeds, but was not significantly more effective than the standards alone. The combinations of fall and spring treatments provided for improved weed control versus what growers were currently using. In 2015, Matrix was not effective on broadleaf weeds alone or with Sinbar. It was also more effective with Velpar but was less effective than the other Velpar combinations, except for the fall Alion treatment. Matrix was also no more effective on grasses than Sinbar alone, and only moderately more effective than the check when used alone, and less effective than no treatment or when combined with Velpar.
The growers' applications were as or more effective than many of the test herbicides for broadleaf weed control, but although not significant, several of the test herbicides performed better on grasses. Therefore, if growers have weeds that aren't managed by their current practices, these alternate chemistries can enable growers to prevent problem or resistant weeds.

Yield
The summer of 2015 was cool through June and July and August were very dry, which may have reduced overall yield compared to a more typical year. The relationship between Alion application and yield appeared to be driven by broadleaf weed control (see Figure 5 and Figure 10). For example, in the fall Alion and fall + spring Alion treatments had the highest broadleaf weed cover (for alone vs with Sinbar vs with Velpar) yet resulted in the lowest yield; the relationship wasn't as clear in the spring Alion treatments. Therefore, reducing problem broadleaf weeds with Alion can result in increased yields, and in a productive field the increased monetary return from yield could be worth the cost of the extra fall application.
Yield in the Trellis treatments remained high regardless of the increase in grasses compared to the check. We believe this is due to the broadleaf weed control of the Trellis treatments combined with the low overall grass cover in 2015, and expect that in a field with heavy grass pressure, yields would be reduced.
The use of Matrix with Sinbar resulted in reduced yield and as with Alion, it appeared to be from a lack in broadleaf weed control (see Figure 6 and Figure 10). Sandea had the best yield when combined with Velpar, which had the lowest amount of both grasses and broadleaf weeds compared to Sandea alone or with Sinbar (see Figure 6 and Figure 10)

RECOMMENDATIONS: Alion, applied it in the fall will to avoid phytotoxicity, and when combined with a spring Sinbar application will provide for the most effective combination of both broadleaf weed and grass control. The manufacturer is pursuing Alion registration in wild blueberry and a label and recommendations to growers will be provided as soon as it is approved for use.
If you have a broadleaf weed problem, Trellis may be used alone or with Sinbar or Velpar. However, if you have primarily a grass problem, the addition of Trellis was no more effective with Sinbar than Sinbar alone, and without Sinbar it led to an increase in grass cover. The lack of grass control suggests that in fields with heavy grass pressure, yield would be reduced.
If you are using Sinbar to control grasses, Sandea is unnecessary, but it provided good carryover grass control into the crop year when used alone or with Velpar. In this trial Sandea did not control broadleaf weeds long-term without Velpar, but other trials have indicated that Sandea can control certain difficult weeds, so if you have a broadleaf weed problem the effective use of Sandea will depend on which weeds are present. The use of Sandea on problem weeds in combination with Velpar can increase yield as shown in this trial. Sandea needs to be applied in

the fall, or if this is not possible then as early as possible in the spring to avoid phytotoxicity issues and reduced yield.
Although Matrix was mediocre in this trial, it has exhibited stronger control of weeds in other trials. Variability in efficacy may be related to weather post-application. The label states that for best results, at least ½ inch rain or irrigation is required within a week of application. Therefore, this product should be used when irrigation is available or rain is expected within a week. As we continue to test this product, we will examine the relationship between Matrix efficacy and rainfall after application. We do not recommend using Matrix with Sinbar in fields with even moderate broadleaf weed pressure, as broadleaf weeds will be released and yield reduced.

INPUT SYSTEMS STUDY – *ANCILLARY STUDY*

WEED MANAGEMENT: David E. Yarborough, Professor of Horticulture
Jennifer L.D. Cote, Assistant Scientist

20. TITLE: Post-harvest control of red sorrel in a non-crop blueberry field, 2013-2015 - crop year evaluation.

METHODS: In the fall of 2013, we initiated a follow-up trial to the 2012-14 post-harvest red sorrel control trial, to determine whether red sorrel control in wild blueberry fields could be achieved by treating the weed after post-harvest pruning. The trial was set up on Wyman's Camp Meadow Hill Lot off the Baseline Road in Deblois, ME. The plots were set out in a Completely Randomized Design with ten 3-m^2 replications per treatment, which were as follows: 1. Untreated check; and 2. Roundup 2% v/v. The Roundup was applied on 31 October 3013 using a backpack boom sprayer with 20 GPA TeeJet nozzles. Wyman's sprayed their herbicide treatments on the plots as well: Sandea 1 oz/a and Velossa 1.5 lb/a in late April/early May.
In 2015, the plots were evaluated for wild blueberry cover, broadleaf weed cover, grass cover, and red sorrel cover on 2 July 2015. Cover data were determined by using the Daubenmire Cover Class system converted to percent, and were analyzed using t-tests (α=0.05). Yield could not be taken from the plots because the plots had been set up after pruning in the fall to encompass red sorrel, and when the plants emerged in the spring we found that the red sorrel occurred in bare spots so there was little to no blueberry in the plots.

RESULTS AND CONCLUSIONS: In 2014, we found that there were no significant differences between the treatments for any cover or wild blueberry phytotoxicity. This held true in the crop year as well; there were no significant differences between the check and Roundup treatment for wild blueberry, red sorrel, or other weed cover (Figure 1). Red sorrel cover in the Roundup plots increased from less than 10% in the prune year to over 40% cover in the crop year, while broadleaf weeds also increased from <5% to over 30% cover. In a previous trial, we noted that Roundup released other broadleaf weeds such as blue toadflax (*Nuttallanthus canadensis*) and spreading dogbane (*Apocynum androsaemifolium*). This was not observed during the prune year of this trial, but in the crop year broadleaf weed cover was dominated by weeds that tend to colonize bare spots, such as blue toadflax, rabbitfoot clover (*Trifolium arvense*) and horseweed (*Conyza canadensis*) (Photo 1).

Figure 1. Wild blueberry, red sorrel, broadleaf weed and grass cover in the 2015 crop year.

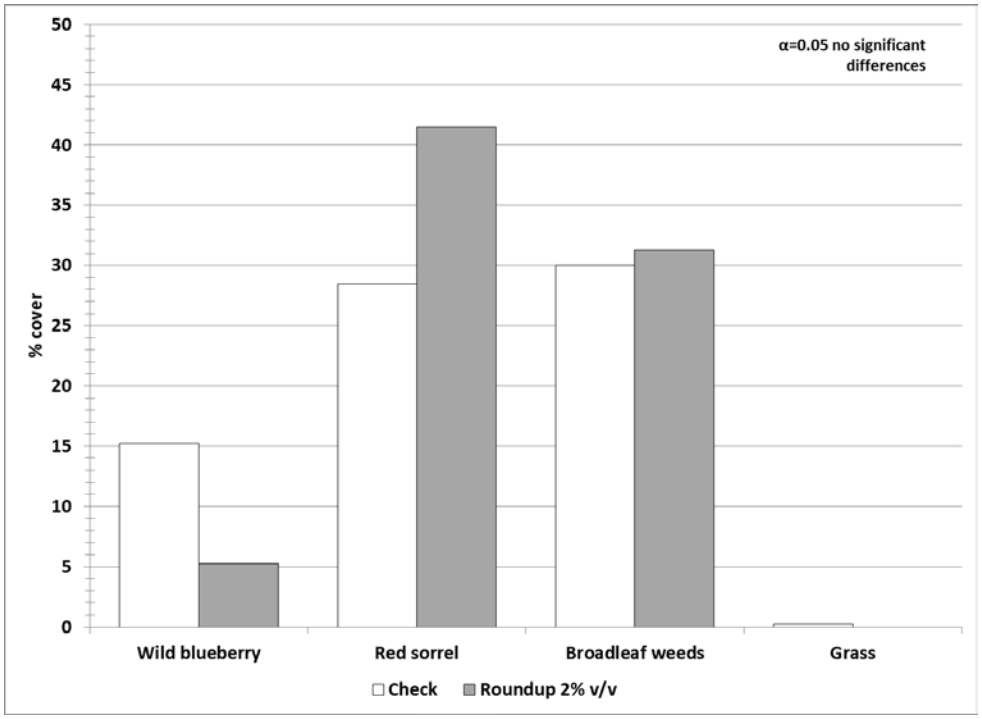


Photo 1. Trial site in the crop year, showing red sorrel as well as blue toadflax, rabbitfoot clover and horseweed.

RECOMMENDATIONS: Roundup does not appear to be an effective in controlling red sorrel, and can release other problem weeds. Therefore, we do not recommend Roundup for red sorrel control. A third red sorrel control trial was initiated in May 2015, looking at herbicides with different modes of action, and a fourth trial was initiated in November 2015 to examine the effectiveness of herbicides applied in fall after pruning on red sorrel cover and other weeds in next year's crop field.

INPUT SYSTEMS STUDY – *ANCILLARY STUDY*

WEED MANAGEMENT: David E. Yarborough, Professor of Horticulture
Jennifer L.D. Cote, Assistant Scientist

21. TITLE: Evaluation of spring applications of herbicides targeting red sorrel in wild blueberry fields.

METHODS: Several herbicides, both registered and unregistered, are currently under review for use on wild blueberry. Alion (6.5 oz/a), Chateau (12 oz/a), Sandea (1 oz/a) and Trellis (1.33 lb/a) are pre-emergence herbicides, while Matrix (4 oz/a) may be used pre- or post-emergence. We treated red sorrel (*Rumex acetosella*) to evaluate these herbicides on this established problem weed. Red sorrel is reported to be resistant to several herbicides, competes with blueberry, and hinders harvest– the control methods examined in the previous trials were ineffective, so we are looking at alternate herbicides . The treatments were applied to red sorrel on ten 1-m^2 plots on 14 May 2015 and five of the plots were treated with Velossa (0.4 gal/a) on the same day by the grower. Wild blueberry and red sorrel cover and phytotoxicity, as well as percent cover of broadleaf weeds and grasses, were evaluated in June and September. Cover data were determined by using the Daubenmire Cover Class system converted to percent; phytotoxicity data were gathered using a scale of 0-10 (0=no damage, 10=100% damaged/dead) converted to percent. The treatments were compared using Tukey's tests (α=0.05) to determine significant differences.

RESULTS: There were no significant differences in wild blueberry cover in the red sorrel trial at either evaluation (Figure 1). Phytotoxicity on wild blueberry was initially highest with Sandea and was significantly higher than all other treatments except Matrix alone, but no differences in phytotoxicity were observed by September. Although there were initially no differences in red sorrel cover, phytotoxicity was highest in the Chateau-grower treatment and was significantly higher compared to the check and other herbicides alone, except for Chateau alone (Figure 2); there were no significant differences in phytotoxicity among grower-treatments in June. In September, red sorrel cover was almost eliminated in the grower-treatments. The only significant difference in red sorrel cover in September was between Alion alone and the non-Alion grower-treatments, but adding Velossa to the treatments resulted in almost twice the injury to red sorrel (Photos 1-4). The phytotoxicity noted in the untreated check was due to a background level of senescing red sorrel. Weed cover was 0-14% cover overall, and there were no differences in broadleaf or grass cover (Figure 3).

Figure 1. Wild blueberry cover and phytotoxicity following spring treatments for red sorrel control.

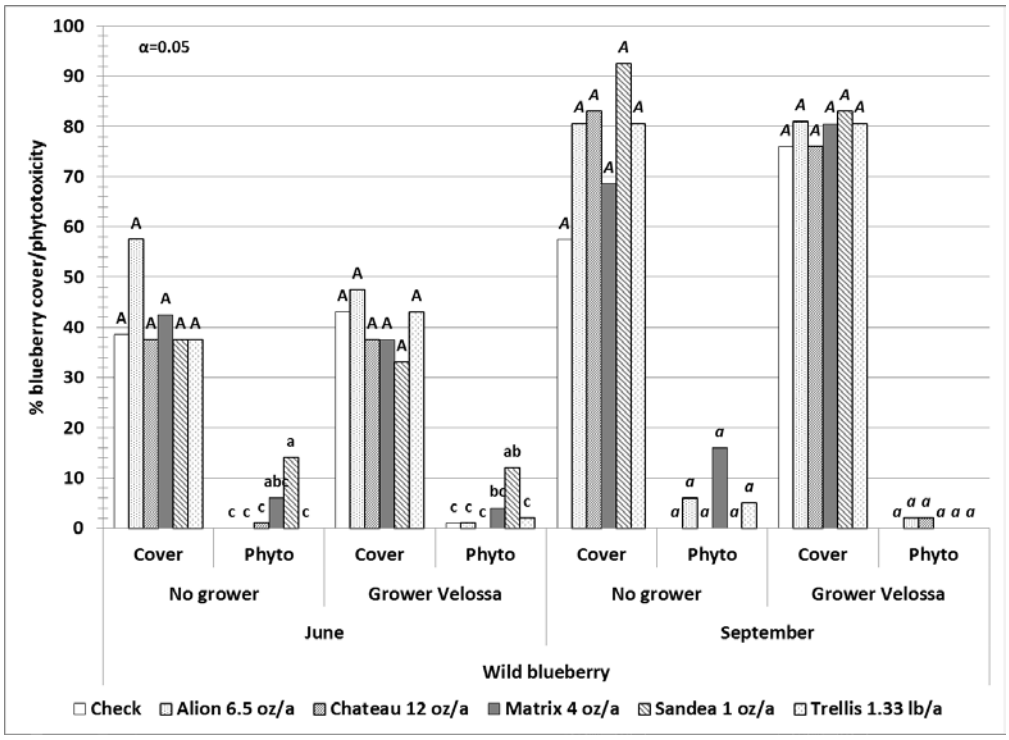


Figure 2. Red sorrel cover and phytotoxicity following spring applications of herbicides.

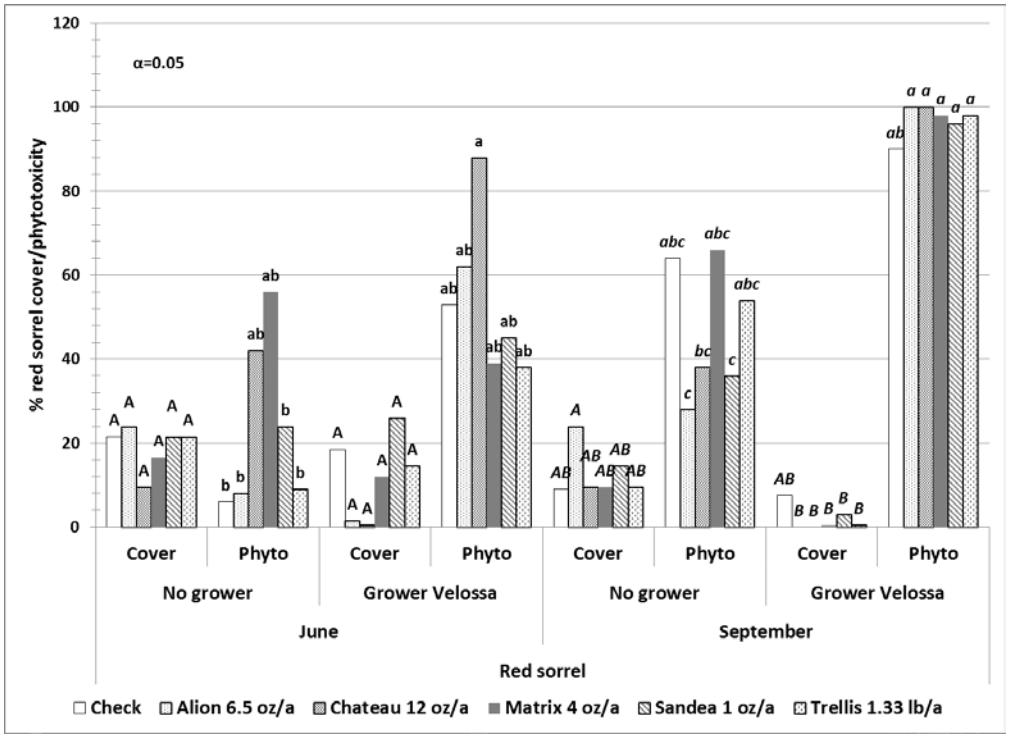


Figure 3. Broadleaf weed and grass cover following spring applications of herbicides for red sorrel control.

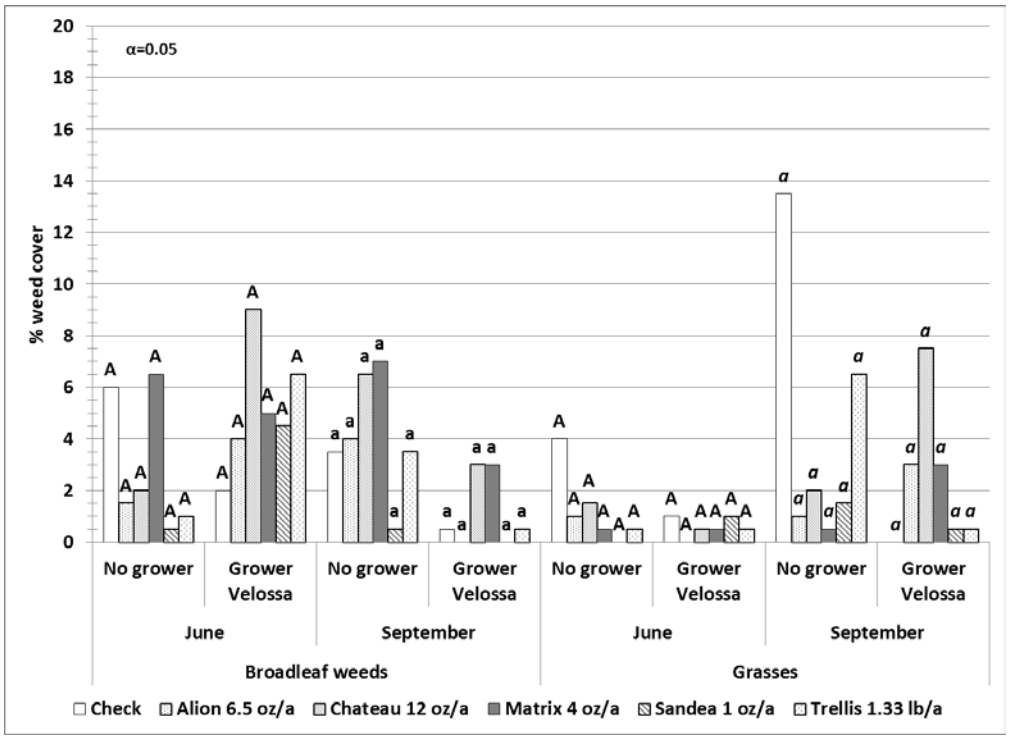


Photo 1. An untreated check plot in September, showing green and senescing red sorrel.

Photo 2. Alion treatment in September, with higher red sorrel cover than the check.

Photo 3. Chateau with Velossa eliminated red sorrel by September.

Photo 4. The grower's application alone almost eliminated red sorrel by September.

CONCLUSIONS/RECOMMENDATIONS: The addition of Alion and Chateau improved the effectiveness of red sorrel control when combined with Velossa, particularly early in the growing season when the blueberry plants are small and more easily outcompeted, and should be evaluated further. Injury to blueberries was observed on spring treatments form past experiments with both Alion and Chateau, so these treatments should be applied in the Fall. Fall applications of Alion, Chateau, Matrix and Sandea were set out in a prune field and crop field in November 2015. Control of red sorrel in both fields will be evaluated over the next one to two years, respectively. Trellis was dropped from the trial as it wasn't more effective than other products alone or with Velossa.