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Management of *Delia* spp. (Diptera: Anthomyiidae) through selectively timed planting of *Phaseolus vulgaris* (Fabaceae) in Atlantic Canada.

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Abstract

Delia spp., including *Delia platura* (Meigen) (Diptera: Anthomyiidae) are known pests of vegetable crops. Here, three studies were conducted to examine the relationship between *Delia* spp. and *Phaseolus vulgaris* L. (Fabaceae). Field studies documented a relationship between planting date and occurrence of *Delia* spp. damage on *P. vulgaris*. Plantings in mid-June resulted in higher crop yields (mean bean pods per plant) and reduced damage ratings compared with earlier plantings. Late-May and early-June planting dates were not favorable, as they resulted in high damage ratings and low plant survival. Late-June and July plantings resulted in low damage ratings but low crops yields. Growth chamber experiments examined oviposition preference of *D. platura* females at seven phenological stages for two varieties of *P. vulgaris*. Results indicated significantly higher oviposition rates on bean plants at early phenological growth stages, with no significant varietal preference shown by maggots. Laboratory experiments quantified the impact of *D. platura* larval infestation on two *P. vulgaris* varieties at two growth stages. Results indicated no significant difference in variety choice. Recommendation for planting *P. vulgaris* to coincide with *Delia* spp. phenology using degree day model is discussed.

Comment [SB1]: Comment 1

Introduction

Flies within the genus *Delia* (Diptera: Anthomyiidae) are known agricultural pests in Canada, Europe and the United States (van Schoonhoven and Voyest 1991). The following *Delia* spp. are commonly found in Nova Scotia and PEI: *D. florilega* (Zetterstedt), *D. antiqua* (Meigen), *D. platura* (Meigen), and *D. radicum* (Linneaus). *Delia* spp. larvae cause significant losses to commercial crops including *Brassica napus* L. by feeding on the root tissues, negatively affecting plant survival (Finch 1989; Broatch *et al.* 2006; Dosdall *et al.* 1994). *Delia platura*, the seedcorn maggot, causes crop losses in many vegetables, and can cause significant damage in common bean, *Phaseolus vulgaris* L. (Broatch *et al.* 2006; Finch 1989; Soroka and Dosdall 2011).

Nova Scotia and Prince Edward Island support three generations of *Delia* spp. in a growing season (Howard *et al.* 1994). The first generation will emerge from the soil in early spring, feeding on germinating seeds and the roots of established plants (Finch 1989). The first and second generations in each growing season are reported to cause the majority of damage to crops (Howard *et al.* 1994). To allow offspring easy access to food, adult females oviposit on soil near plants and germinating seeds (Finch 1989; Howard *et al.* 1994).

Females use olfactory cues as their primary method in choosing an oviposition site (Gouinguené and Städler 2006). A single adult female can oviposit between 50 and 270 eggs (Bennett *et al.* 2011; Soroka and Dosdall 2011). *Delia* spp. larvae will eclose approximately two days after oviposition (Throne and Eckenrode 1986). The developmental time for larvae is approximately two week during which feeding occurs on vegetation. After the larval stage, *Delia* spp. will pupate, emerging as adults approximately two weeks later. The documented degree days between oviposition of one generation to the next is 495 DD (Funderburk *et al.* 1984).

Traditional control of *Delia* spp. involves the use of insecticides, and has been shown to reduce plant damage, however, few options are available to organic growers (Montecinos *et al.* 1986; Valenciano *et al.* 2004). One method used to manage *Delia* species in canola in Alberta Canada is the cultural practice of selectively timed planting (Dosdall et al. 1996). Selectively timed planting can cause asynchrony between the phenology of the plant and pest. An optimal planting date would avoid damage from *Delia* larvae, while also optimizing crop yield. *Delia* adults typically emerge from the soil in early spring and will lay eggs on germinating seed over a two-week period. Larvae hatch from eggs and feed on newly germinated seedlings before pupating. Such a method, if timed appropriately, could be useful to snap bean producers in PEI and NS. The snap bean industry in NS and PEI is small with each province having less than 20 acres in production. Snap beans are produced for local markets with each grower planting between 1 and 3 acres. Growers typically start direct seeding bean at the end of May or early June, with additional sequential plantings every 10 – 14 days to ensure a continuous supply of fresh beans for the local market. At harvest, the plant is pulled from the field and the bean pods removed and placed in a bin for transport to the market. The pods are graded and any pods with bacterial spot or insect chewing are left in the field. Growers sow at a density of 4 cm between bean seeds. This spacing creates large plants each producing 15-20 bean pods.

Comment [SB2]: Comment 3

While most snap bean diseases attack the pods, *Delia* species will feed on the germinating seed and taproot tissues and, in severe cases, larvae will chew through the plant stem and tunnel up the stalk (Vea and Eckenrode 1976b). With no available controls, growers are left with little recourse but to accept loss. Selectively timed planting could allow establishment and maturation of the bean plant, well before attack by *Delia* species. Alternatively, a later planting date could avoid the first generation of *Delia* species altogether (Dosdall *et* *al.* 1996; Vea and Eckenrode 1976b). Selectively timed plantings are not a widely-used practice as further study is required to perfect this management strategy to synchronize planting with local *Delia* spp. populations (Finch 1989; Dosdall *et al.* 1996).

This research examines the cultural practice of selectively timed planting for management of *Delia* species in the Canadian Atlantic provinces of Nova Scotia and Prince Edward Island (PEI) in organically managed common bean, *P. vulgaris* . The major objectives of this study are to: (1) observe the relationship between planting date and occurrence of *Delia* species damage on *P. vulgaris*, (2) document host oviposition preference of *D. platura* females on two varieties and eight growth stages of *P. vulgaris*; and (3) to quantify the impact of *D. platura* larvae (at two levels of infestation) on two growth stages of *P. vulgaris*.

Materials and Methods

Monitoring *Delia spp.*

In each field, over both years, four blue 'dry touch' sticky traps (12 x 14 cm; Solida, Quebec, Canada) were attached to wooden stakes (1 x 4 cm and 1 m tall) with a bull clip 30 cm off the ground, as recommended by Finch (1989), and placed within the plot (see Fig.

1). Two additional traps were located East and West of the plot, in the surrounding vegetation approximately 10 m away to capture the population prior to field preparation. Traps were collected and replaced each week. Only flies within the genus *Delia* were sexed and identified to species using a *Delia* taxonomic key (Savage *et al.* 2016). *Delia platura* and *Delia florilega* Zetterstedt males were counted while *D. platura* and *D. florilega* females were pooled as they are not reliably distinguishable morphologically. As the larvae are not distinguishable at the species level, any damage observed in the field studies could not be attributed to either of the *Delia* species present. As such, any reference to damage in the field studies will be attributed to *Delia* species. Degree days were calculated from 1 January each year, with a base of 3.9° Celsius (Funderburk *et al.* 1984).

Field Observations – 2015

Grower observation of plant loss early in the season in previous years and again in 2015 prompted closer observation in two Prince Edward Island (PEI) fields: Milton (46.306521° N, 63.277758° W) and Loyalist (46.309193° N, 64.358008° W). These fields were commercial organic agricultural fields surrounded by natural vegetation or other crops, e.g. radish, soybean or winter wheat. No insecticides were applied and weeding was done by hand. As per usual practice, *P. vulgaris* (Gold Rush and Lewis varieties, yellow and green, respectively) from Vesey's seed (York, PEI) were sown on various planting dates: 16 May, 26 May and 5 June in Loyalist and on 16 June, 27 June and 7 July in Milton (Table 2). A preliminary sample of damaged plants from the 16 and 26 May plantings in Loyalist found *Delia* species larvae. Plant counts were conducted when the plants reached the second trifoliate, stage 14, using the BBCH-scale developed by the Biologische Bundesanstalt, Bundessortenamt and Chemical industry (BBCH) working group (Meier 2001, Table 1) and again at flowering, stage 65, by randomly selecting 20 – 2 m long sections and counting the number of plants present. At these growth stages it was not possible to distinguish variety and the sampling occurred over all rows of the same planting date. As the fields had sequential plantings, the dates for these samplings ranged from 12 June to 20 August 2015. At harvest, the different varieties were easily distinguished. A sample of 30 plants per planting date and variety (where applicable) were collected from each field by walking down every second or third row (depending upon number of rows in the planting) and selecting a plant every 20 steps. Number of bean pods per plant was counted and damage from *Delia* spp. to the root was recorded as present or absent.

Field Trials – 2016

Based on the results from the 2015 field observations, replicated trials were conducted in three fields in Nova Scotia and PEI in 2016. Fields in PEI were located at Saint Ann (46.438631° N, 63.406906° W) and Wilmot Valley (46.384709° N, 63.701667° W) and the Nova Scotia site was located in Canning (45.170202° N, 64.437527° W). All field sites were commercial organic agricultural fields surrounded by natural vegetation or other crops, e.g. radish, soybean or winter wheat. No insecticides were applied to any site and weeding was done by hand.

In Canning and Saint Ann, blocks measuring 2.5 by 7.5 meters were established and contained six parallel rows (spaced 30 cm); where the two outer rows served as guard rows, and the 4 inner rows were used for treatments (Fig. 1). The guard rows were sown by hand with Gold Rush variety beans (2-3 cm deep and 5 cm apart) while the four inner rows were divided into four sections, each 1.5 m in length to create 16 plots. Additional guard plants, also Gold Rush variety and 6 per row were sown on either end of the treatment area. A 4 x 4 Latin square design (4 replicates of 4 planting dates) was sown by hand with Lewis seed on the following dates: 17 May, 31 May, 14 June, 28 June, 2016 for Canning and 25 May, 8 June, 22 June and 6 July, 2016 for Saint Ann. Planting dates were consistent with commercial grower planting timing in each area. At Wilmot Valley, the treatment block was planted in amongst the regular grower plantings and was 2 rows wide and 16 plots (each 1.5 m in length) into the field and sown with 30 seed of Lewis. Guard plants, variety Gold Rush, 6 in each row, were sown at each end of the treatment block. Planting dates were systematically randomized within each replicate and occurred on 1 June, 15 June, 29 June and 13 July 2016.

Plants were checked once or twice weekly and counted to assess survival. Any plant showing any sign of damage was sampled and examined to determine the cause. At harvest, bean plants were removed from the field and brought to the lab. Number of bean pods per plant and damage rating were recorded from all sites, fresh weight of the plant and root diameter was recorded from Wilmot Valley and Saint Ann. Root damage caused by *Delia* spp., was identified and categorized on a scale of 0-5 (Dosdall *et al.* 1994, Howard *et al.* 1994).

Laboratory Studies

Laboratory studies were conducted in a walk-in growth chamber (constant temperature of 20 ± 1°C, 60 ± 5% RH, and a 16:8 light/dark photoperiod) located at the Kentville Research and Development Center, Kentville, NS. *Phaseolus vulgaris* var. Gold Rush and Lewis and *D. platura* colonies were reared and all experiments conducted under these conditions.

Insects

Colonies were maintained in Bug Dorms measuring 30 x 30 x 30 cm (BioQuip, California, USA). Water was provided in 25 mL solo cups, with the lid cut to hold a 5 cm long dental wick. *Delia platura* pupae were obtained from Francois Fournier (Collège Montmorency, Laval, Québec, Canada) and placed in the Bug Dorms to develop. Upon eclosion, adults could mate and oviposit freely. Dry food, composed of sucrose, milk powder, yeast, and organic soy flour in a ratio of 10:10:2:1, was provided in a petri dish (McClanahan and Miller 1958). Oviposition substrate consisted of a petri dish with damp paper towel and a slice of rutabaga. Egg collection and rutabaga replenishment occurred twice weekly. Eggs were transferred to 500 mL plastic containers filled with 200 mL of larval diet (Ishikawa *et al.* 1983). Up to 200 eggs were added, and covered in damp vermiculite. Larvae hatched, developed and formed pupae in the larval food container. Pupae were then transferred to a petri dish with paper towel and damp vermiculite and placed in a Bug Dorm. Emerging adults were separated by generation.

Phaseolus vulgaris seeds were individually planted in pots (7.5 cm diam. by 10 cm deep) and watered daily. Bean seed required for studies at the germination stage were grown on a petri dish with damp paper towel and covered with vermiculite. Plants were selected for experimentation when they reached one of eight different phenological stages: 03, 07, 10, 12, 13, 21, 65, and 75 (Table 1).

Female Oviposition Preference

Choice experiments occurred in Bug Dorms under the same conditions used for maintaining the colony. Ten *D. platura* mating pairs were placed within a Bug Dorm with two bean plants (one Lewis and one Gold Rush) for 48 hours, and provided with a water source and adult food. Bean plants of the same phenological stage and planting date, of similar shape and colour were selected for each trial. Ten replicates for each of 8 phenological stages were completed for a total of 80 trials. Plants and flies were used only once. Following removal of adult flies, the soil, roots, and stem were examined for eggs and larvae, as described by Dosdall *et al.* (1994). Counts of eggs and larvae were recorded for each plant.

Larval impact trial

Phaseolus vulgaris plants (see plant growth conditions above) at two phenological stages, 12 and 13, were exposed to zero (control), five or ten *D. platura* larvae until flies emerged as adults. The trial began between 5 July and 19 July 2016 at asynchronous intervals; all plants were then grown to maturity (stage 75, 56 days after planting) when all plants were harvested. After harvest, each plant was processed by recording the number of bean pods, weight of each plant, and root damage (using the 0-5 scale) from July 28 to Sept 13, 2016.

Statistical Analysis

R Studio software was used to conduct all statistical analyses (R Studio 2015).

Field observations – 2015

As the planting dates were not repeated across the two fields (all 'early' plantings were in Loyalist and all 'later' plantings were in Milton) only the three planting dates within each field could be compared. Plant density (as mean number of bean plants per 2 m length), and percentage of surviving plants was analysed following transformation using asin(x) and ANOVA methods for differences between planting date (within each field), and between plant stage surveyed within each field and planting date. Mean number of bean pods per plant were analysed for differences between planting date and variety (where both varieties were planted) using ANOVA methods at $\alpha = 0.05$.

Field trials – 2016

Percentage of surviving plants was transformed using asin (x) and ANOVA methods to evaluate differences between planting date for each site. Mean damage rating, number of bean pods per plant, fresh plant weight, bean pod weight and root diameter were evaluated for differences between planting date using ANOVA within each site. Post-hoc means separation was determined using Tukey's Honestly Significantly Different (HSD) test at $\alpha = 0.05$. Degree day values were calculated using a threshold of 3.9°C (Broatch et al. 2006), starting accumulations from 1 January, to standardize the *Delia* species phenology across field sites in 2016 and cross-reference with plantings.

Laboratory trials

Female oviposition preference was analysed using two-way ANOVA to determine if bean variety and plant stage influenced number of eggs oviposited. Oviposition Preference Index (OPI) was calculated using (X-Z)/(X+Z) where X is number of eggs oviposited on Lewis var. plants and Y is number of eggs oviposited on Gold Rush var. plants to evaluate preference of females for Lewis against Gold Rush varieties (Little et al. 2017). Larval impact on phenological stage and variety was analysed using ordinal logistical regression.

Results

Monitoring *Delia* spp.

Delia platura and *D. florilega* were the most abundant species capture in both years (Fig. 2A, 2B and 2C). *Delia radicum* and *D. antiqua* were also present but in very low numbers (e.g. less than 10 per collection). Three generations of *Delia* spp. were clearly present in all fields. More males than females were captured early in the season (Figs. 2B and 2C) while more females than males were captured later in the season (Fig. 2A).

Comment [SB3]: Comment 4

Field observations – 2015

Plant density was significantly affected by planting date in Milton and Loyalist fields on Prince Edward Island (Table 2). In both fields, the first planting had significantly fewer plants per 2 m transect than the last planting when assessed at growth stage 14 ($F_{2,47} = 91.3$, $P < 0.0001$ and $F_{2,27} = 14.8$, $P < 0.0001$, Loyalist and Milton fields, respectively) and at growth stage 65 ($F_{2,67} = 6.96$, $P = 0.002$ and $F_{2,57} = 5.75$, $P = 0.005$, Loyalist and Milton fields, respectively). The 26 May planting at Loyalist experienced the greatest plant loss (plant death) and was replanted on 16 June. This planting had the lowest plant counts at growth stage 14 (0.85 plants) while the 7 July planting in Milton had the highest plant counts (21.5 plants) (Table 2). At growth stage 65, the 16 June planting in Milton had the fewest plants per transect (9.8 plants) while the replant of the 26 May planting in Loyalist (16 June) had the highest plant count at stage 65 (16.6 plants). Within each planting date, plant density was significantly different between assessments in Loyalist for the 5 June planting ($F_{1,48} = 5.63$, $P = 0.02$) and in Milton for the 16 June ($F_{1,28} = 40.0$, $P = 0.04$), and 7 July plantings ($F_{1,28} = 27.7$, $P < 0.0001$), in each case showing a decrease in plant density.

Mean number of bean pods at harvest was significantly different by planting date within each field. In Loyalist there were only two plantings with harvest data and both varieties. The 16 May planting for both Lewis and Gold Rush varieties produced significantly fewer beans per plant than the 16 June planting (replant of the 26 May planting) ($F_{1,182} = 144.82$, $P < 0.0001$ and $F_{1,182} = 15.33$, $P < 0.0001$, planting date and variety, respectively, Table 2). In the Milton field, there was no significant difference between the bean varieties ($F_{1,136} = 2.05$, $P = 0.16$) for mean number of bean pods/plant, but planting date was significantly different ($F_{2,136} = 41.5$, $P < 0.0001$, Table 2). Greatest number of bean pods/plant were harvested from the 27 June planting with an average of 17.6 bean pods from the Gold Rush and 15.2 bean pods from the Lewis varieties. The lowest number of bean pods was harvested from the 7 July planting with an average of 5.4 bean pods per plant. Estimates of the number of bean pods/acre show the 16 June (replant of the 26 May planting) in Loyalist and the 27 June planting in Milton to produce the greatest yields. Yields were estimated to be 1870 and 1539 x 1000 bean pods/acre for Gold Rush and Lewis varieties in Loyalist and 1577 and 1357 x 1000 bean pods/acre for Gold Rush and Lewis varieties in Milton.

Examination of the plant roots post-harvest found high percentages of plants showing damage from *Delia* feeding in the Milton field with > 70% of the roots showing damage across all plantings and varieties (Table 2). Lowest percentages of plants with damaged roots were in the Loyalist field and the 16 May planting, 15.1 and 28.1%, Gold Rush and Lewis varieties, respectively.

Field trials – 2016

Survival of bean plants was significantly different across planting dates in Wilmot Valley ($F_{3,11} = 6.65$, $P = 0.008$) and Saint Ann ($F_{3,12} = 10.11$, $P = 0.001$) but not Canning ($F_{3,12} = 0.98$, $P = 0.43$) (Table 3). At all sites, plant survival was highest for later planting dates. Percentage of plants surviving until harvest was significantly lower for the first and second planting dates, in both Saint Ann and Wilmot Valley with no plants surviving the first planting (1 June) in Wilmot Valley (Table 3). Recovery of the seed from these plots found rotten, partially germinated seed containing larvae. Larvae were reared to adulthood and identified as *D. platura* and *D. florilega*. The 1 June planting in Wilmot Valley was replanted on 29 July 2016.

A significant decrease in mean damage rating across planting dates was found in Wilmot Valley ($F_{3,11} = 48.07$, $P < 0.0001$) and Saint Ann ($F_{3,12} = 25.23$, $P < 0.0001$) but not Canning ($F_{3,12} = 3.49$, P

= 0.05). Across all sites, Wilmot Valley had the highest mean damage as the first planting date here, 1 June, experienced severe damage and failure to survive *Delia* attack. Examination of the ungerminated seed showed evidence of *Delia* spp. feeding. Later plantings at this site had significantly lower mean damage ratings. A similar pattern of damage early in growing season was observed at Saint Ann and Canning fields.

The number of bean pods per plant were significantly different across planting dates in Saint Ann ($F_{3,12}$ = 8.05, P = 0.003) but not in Canning ($F_{3,12}$ = 0.92, P = 0.46) or Wilmot Valley ($F_{3,11}$ = 3.27, P = 0.06) (Table 3). A higher number of bean pods were harvested from plantings occurring in the middle of the growing season with number of bean pods per plant being lower at the beginning of the season (22.4 and 6.7 pods per plant in Saint Ann, first and last planting, respectively). Mean bean pod weight was statistically different across planting dates at Saint Ann ($F_{3,12}$ = 8.41, P = 0.003) and Wilmot Valley ($F_{3,11}$ = 31.64, P < 0.0001). Lowest bean pod weights were observed on the last planting date for Wilmot Valley and the second last planting date at Saint Ann, 1.94 g and 3.03 g, respectively (Table 3). Highest bean pod weights were from the 25 May planting for

Saint Ann (6.13 g) and the 15 June planting for Wilmot Valley (4.03 g).

Larger plants were harvested from plantings occurring earlier in the season for both sites on PEI. Plant mass and root diameter were higher from early plantings, and lower from later plantings across both PEI field locations (plant mass Wilmot Valley, $F_{1,4} = 44.24$, $P < 0.0001$; Saint Ann, $F_{1,4} = 50.02$, $P < 0.0001$; root diameter, Wilmot Valley, $F_{1,4} = 3.54$, $P < 0.0001$; Saint Ann, $F_{1,4} = 3.06$, $P < 0.0001$).

Laboratory Trials

Delia platura females showed no significant variety preference during oviposition ($F_{1,150} = 0.14$, $P = 0.71$, Fig. 3). Oviposition Preference Index values ranged from -0.13 to 0.004 across plant stages, suggesting that for any given plant stage the difference in number of eggs laid was at most 13% more on Gold Rush over Lewis. When varieties were pooled, a significant difference in oviposition rate between phenological stages was disclosed ($F_{7,71} = 5.41$, $P < 0.001$, Fig. 3). Significantly higher oviposition occurred at phenological stages 3 and 7, two to four days post germination, when compared with later stages ($P < 0.05$). There were no significant differences in larval damage to varieties of *P.*

vulgaris tested ($F_{1, 7} = 0.55$, $P = 0.48$, Table 4). A significantly higher damage rating was observed on the roots of plants infested with larvae over the controls ($P < 0.05$, Residual Deviance 186.3, Akaike Information Criteria (AIC) 204.3). Lower damage ratings were experienced by plants infested with larvae at phenological stage 12, first true leaf, than on stage 13, third true leaf. Although compelling, this result was not statistically significant. Further, the larvae did not complete development and were not recovered from the pots at the end of the trial. We cannot say for certain that the feeding pressure within each treatment (5 or 10 larvae) was consistent throughout the trial. Plant loss from other factors, pathogens (n = 2) and lack of germination (n=1), also occurred in the laboratory.

Comment [BS4]: Comment 7

Comment [BS5]: Comment 8

Discussion

Organic bean producers in Nova Scotia and Prince Edward Island can experience severe plant loss early in the growing season. It is recognized that *D. platura* prefer to oviposit on early growth stage *P. vulgaris* (Gouinguené and Städler 2006), and *P. lunatus* (Weston and Miller (1989) and that the most vulnerable time for a bean plant is from planting to 3 – 4 weeks of growth (van Schoonhoven & Voyest 1991; Vea & Eckenrode 1976b). Lack of lignification during early development leaves *P.vulgaris* vulnerable to attack, while healthy, lignified *P. vulgaris* experience little damage from *Delia* spp. (Turnock *et al.* 1992). In this study, examination of damaged *P. vulgaris* plants disclosed damage characteristic of *Delia* spp. feeding (Howard *et al.* 1994; Turnock *et al* 1992; Vea *et al* 1975), described as superficial chewing of the taproot tissues and chewing through the plant stem and tunneling up the stalk (Vea and Eckenrode 1976b and Finch 1989). Additionally, these fields had high *Delia* species trap catches during May and June (average 50 to 350 per trap per collection date), and *Delia* larvae were recovered from damaged roots and stems, making *Delia* spp. the likely cause for the crop losses observed by growers. Further, significant damage by *D. platura* to *P. vulgaris* was documented under controlled conditions with plants having damage similar to that observed in field studies. The Canning site had a heavier soil type and retained moisture more than the field sites on PEI. As a result, some seed failed to germinate and upon recovery were found rotting and infected with fungal pathogens. This type of damage accounted for approximately 2% of the plants in this trial and was easily distinguishable from damage by *Delia* species.

Producers wanting to ensure a continuous supply of fresh beans for the market will often use sequential plantings, starting when the ground has thawed and repeat every 10-14 days. Such plantings, typically done without consideration of *Delia* spp. phenology, are at risk of coinciding with peak larval eclosion from the egg. The importance of selecting a planting date to maximize crop growth while avoiding pest damage has been noted by others (Balasubramanian *et al.* 2004, van Schoonhoven and Voyest 1991, Valenciano *et al.* 2004). An optimal planting date would result in a high number of plants producing a large number of bean pods per plant. Many studies evaluating the use of selectively timed plantings will refer to a period of time based on the Julian calendar, e.g. mid-June or early summer (Dosdall et al. 1995). Selection of a Julian-based planting date faces challenges when transferred to a different region due to climatic issues, requiring the study to be repeated in each region where the crop-pest complex is present. We propose using the degree-day model for *D. platura*, the predominant *Delia* species present in these fields to best determine the planting date most likely to result in avoidance of the larvae.

Degree day models for *D. platura* have been developed in the United States (Funderburk *et al.* 1984) and Korea (Lee *et al.* 2000) using field observations. In the laboratory, developmental rates for *D. platura* and *D. florilega* were determined by Sanborn *et al.* (1982) and Throne and Eckenrode (1986). Such studies can produce a range of base development temperatures, e.g. 0.6- 7.9°C, creating the need to evaluate each model in a specific regions. The field study by Funderburk *et al.* (1984) and the laboratory study by Sanborn *et al.* (1982) both found the same development temperature for *D. platura*, e.g. 3.9°C. In Canada, Broatch *et al.* (2006) used this threshold and found it effective to predict *D. platura* populations in canola in western Canada. Previous work with *Delia* species in NS have found this threshold to reasonably predict *Delia* phenology.

The phenology of the female is more critical than the male when trying to predict and avoid damage from the larvae. For the *Delia* species in NS and PEI, comparison of *D. platura* and *D. florilega* female phenology and planting dates from 2016 within each site (Figure 4) shows how the various plantings were likely to be impacted by *Delia* larvae. The first planting in Wilmot Valley (Figure 4B) was decimated (0% survival) by *Delia* larvae. This planting occurred at 277 $DD_{3.9}$ and at the peak flight of the first generation. That creates the situation where females have been emerging and ovipositing eggs on the soil for a few days, many have likely have started to eclose and larvae are ready to feed on any newly germinating seed. The second planting at Wilmot Valley (at 400 $DD_{3.9}$) experienced high plant death (~ 73%) as well, occurring when

Comment [BS6]: Comment 10

Comment [BS7]: Comment 11

oviposition would be starting to decline, but the eggs and larvae are still eclosing and feeding. By the third planting date (765 $DD_{3.9}$), *Delia* larvae are starting to pupate and only 28% loss was realised. The fourth planting (765 $DD_{3.9}$) experienced 34% loss, occurring shortly after the second emergence peak, when larvae would again be seeking food. The fifth planting (1024 $DD_{3.9}$) experienced 19% loss. Similar patterns were found in Canning and Saint Ann (Figure 4A and 4C). When seeding occurred during the first generation emergence, newly germinating seed were at risk of attack. Seed planted later in the season, even if *Delia* populations were on the rise, had the advantage of warmer temperatures to advance the lignification process. Based on these results, it would be recommended that growers monitor the degree days and plant their bean seed to avoid the first and second generations, i.e. between 500 and 600 $DD_{3.9}$ for the first planting in areas where populations are high or about 400 $DD_{3.9}$ where populations are low (see Saint Ann, PEI, Figure 4C). Avoidance of the second generation would require planting around 900 $DD_{3.9}$. However, planting at this time means that plants will have shorter days and fewer heat units for development, as *P. vulgaris* has a relatively high temperature threshold for growth, e.g. 10°C (de Medeiros *et al.* 2000). Planting earlier than 300 $DD_{3.9}$ results in the

Comment [BS8]: Comment 13

bean seed not having enough heat units to grow and produce lignin to withstand larval feeding.

Data collected from our field and laboratory experiments suggest a potential for *P. vulgaris* to tolerate *Delia* spp. attack. Tolerance, the ability of a plant to compensate quickly after attack or to withstand attack, has been observed in many crops and a variety of insect pests, including the common bean (Blatt *et al.* 2008; Ojwang 2010; Vea and Eckenrode 1976a). In our study, the Wilmot Valley field site showed no significant change in number of bean pods per plant between the 15 June and 29 July plantings even though damage rating was 3.4 for the 15 June planting where only 21% of the plants survived, compared with the 29 July planting with a mean damage rating of 1.6 and 71% of plants surviving. Laboratory experiments found severe *D. platura* damage on plants infested at early phenological stages of growth, while low damage ratings were realised from older *P. vulgaris* plants. Consistent with Turnock *et al.* (1992), such tolerance could be due to lignification of these older plants.

Field evidence from 2015 suggested that *Delia* spp. may exhibit a varietal preference, an observation supported by Vea and Eckenrode (1976a). However, no significant difference in oviposition or extent of damage between bean varieties was observed in the current laboratory studies. Oviposition site is chosen by *Delia* spp. female flies through the detection of plant volatiles by olfactory senses (Gouinguené and Städler 2006; Hough-Goldstein 1985; Spencer *et al* 1995). The lack of oviposition site preference found in this study could speak to similarity in olfactory profiles between the two varieties of bean. As such, there is no benefit to using one of these varieties over the other to reduce *Delia* damage. One issue which may have skewed our results is the large number of flies used (ten males and ten females per replicate). This may have caused competition between females for an oviposition site, possibly altering preferences.

Conclusion

Laboratory experiments found no varietal preference by *D. platura* females for either Lewis or Gold rush varieties of *P. vulgaris*. In the lab, high oviposition rates and high damage from *D. platura* occurred on early phenological plant stages. In the field, selectively timed planting to avoid the first and second generations of *Delia* can be an effective strategy to avoid loss from *Delia* spp. feeding. However, to obtain high crop yields, a balance must be achieved between late planting to manage pest damage and early planting to ensure high yield. Using the phenology of *Delia* spp. as an indicator of when to plant will provide growers with a selective planting date where young *P. vulgaris* plants are not subject to high *Delia* spp. larvae. Further study to compare plantings based on Julian day with degree day recommendations are required to verify these results.

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Table 1. Selected phenological stages of *Phaseolus vulgaris* (BBCH, 2001).

Code	Description
0	Dry seed
3	Radicle emergence from seed coat (two days post germination)
7	Hypocotyl break through seed coat (four days post germination)
10	Cotyledons emerged, unfolded
12	First true leaf emerged, unfolded
13	First trifoliate leaf emerged, unfolded (third true leaf)
14	Second trifolate leaf emerged, unfolded (fourth true leaf)
21	Growth of first side shoot
65	Blossoms in Flower (over 50% of flowers open)
75	Bean harvest (over 50% of pods at full length)

Table 2: Mean plant density (± SE) at two growth stages, mean number (± SE) of bean pods per plant at harvest, estimated yield/acre and percentage of plants at harvest showing damage from *Delia* species for *Phaseolus vulgaris var.* Gold Rush and Lewis, sown at various dates at two locations on Prince Edward Island in 2015. Means within column (for plant density) and means within column (Number bean pods/plant) and variety (Loyalist field only) with same letter not significantly different using ANOVA methods followed by Tukeys HSD mean separation test ($P < 0.05$).

Comment [BS9]: Comment 15

Field	Plant date	Harvest date	Plant density* GS 14	Plant density* GS 65	Variety	Mean # bean pods/plant (± SE)	Estimated[1] pods/acre (x 1000)	% plants damaged roots
Loyalist	16 May	12 Aug	10.6 (0.97) a	11.5 (0.62) a	Gold Rush	6.8 (0.79) a	524	15.2
					Lewis	3.3 (0.45) A	247	28.1
	26 May[+]	24 Sep	0.8 (0.32) b	16.6 (0.99) b	Gold Rush	16.9 (0.91) b	1870	44.1
					Lewis	13.9 (0.72) B	1539	55.7
	5 June	--[++]	18.1 (1.33) c	15.1 (0.53) b	Gold Rush	--	--	45.0
Milton	16 June	4 Sep	11.3 (0.93) A	9.8 (0.65) A	Gold Rush	9.9 (1.60) A	584	78.9
	27 June	4 Sep	14.4 (1.86) A	13.5 (1.47) B	Gold Rush	17.6 (1.62) B	1577	73.9
					Lewis	15.2 (1.20) B	1357	86.8
	7 July	12 Sep	21.5 (1.09) B	13.3 (0.96) B	Lewis	5.4 (0.65) C	473	83.3

*Plant densities taken from fields when plants too young to allow variety differentiation

[1]Estimated yield determined by multiplying the mean number of bean pods/plant by the plant density at growth stage 65 and using a conversion factor of 6634 which is based upon a row spacing of 0.61 m and assuming that the plant density is the same for both varieties

[+]May 26 planting was replanted on June 16, plant density from stage 14 is the May 26 planting and from stage 65 is the June16 replacement planting

[++]Grower harvested bean pods on 10 September before sample could be taken, leaving plants in the field. A sample of these plants allowed evaluation of *Delia* species damage for this planting

Table 3. Mean (± SE) percent survival, mean (± SE) number of bean pods per plant, and mean (± SE) damage rating of *Phaseolus vulgaris* at all field locations in NS and PEI in 2016. Letters within column, for each site, denote significant differences within each measured variable using ANOVA methods and Tukeys HSD mean separation test ($P < 0.05$).

Comment [BS10]: Comment 15

Field	Planting date	Percent plant survival	Damage rating	Number bean pods/plant	Plant mass (g)	Bean pod weight (g)	Root diameter (mm)
Canning, NS	17 May	55.0 (9.5)	2.4 (0.3)	4.9 (0.5)	--	--	--
	31 May	55.0 (9.5)	1.5 (0.3)	7.4 (1.9)	--	--	--
	14 June	70.0 (23.8)	1.9 (1.0)	5.9 (2.2)	--	--	--
	28 June	75.0 (5.0)	0.0 (0.0)	4.1 (0.1)	--	--	--
Wilmot Valley, PEI	1 June*	0.0 (0.0)	--	--	--	--	--
	15 June	20.8 (5.2) B	3.4 (0.2) A	18.9 (2.0)	189.31 (2.9) A	4.28 (0.8) A	8.50 (0.7) A
	29 June	71.8 (4.2) A	1.6 (0.1) B	20.2 (0.8)	101.67 (12.0) C	1.78 (0.2) C	6.76 (0.1) BC
	13 July	65.6 (6.4) AB	1.3 (0.1) BC	23.4 (0.8)	137.43 (8.3) B	3.11 (0.3) B	7.00 (0.1) AB
	29 July	80.5 (9.2) A	0.5 (0.1) C	18.4 (0.7)	72.54 (7.4) C	1.94 (0.2) C	5.84 (0.1) BC
Saint Ann, PEI	25 May	16.7 (4.1) b	3.4 (0.4) a	22.4 (5.1) a	237.32 (69.6) a	6.13 (0.5) a	9.45 (0.8) a
	8 June	20.0 (4.9) b	3.7 (0.3) a	21.6 (1.4) a	188.65 (23.0) ab	5.01 (0.6) a	8.40 (0.4) a
	22 June	64.2 (11.4) a	1.4 (0.2) b	9.2 (2.1) b	54.29 (17.0) b	3.03 (0.2) b	5.43 (0.5) b
	6 July	70.0 (4.3) a	0.6 (0.2) b	6.7 (0.7) b	57.31 (8.3) b	4.93 (0.3) a	5.38 (0.3) b

*denotes planting date where no plants germinated and evidence of *Delia* spp. larval infestation was found.

Table 4. Number of *Phaseolus vulgaris* plants (two varieties pooled) showing damage (0-5 rating) when infested at two phenological stages (12 and 13, first and third true leaf, respectively) with either 5 or 10 *Delia platura* larvae in a growth chamber experiment, n=12 for each treatment. Significant difference observed between first and third true leaf phenological stages ($P < 0.05$, Residual Deviance 186.3, AIC 204.3).

Comment [BS11]: Comment 15

Plant Growth Stage	Larvae	Damage rating 0	1	2	3	4	5
12	0	12	0	0	0	0	0
(First true leaf)	5	1	0	1	3	2	3
	10	0	0	1	2	2	7
13	0	12	0	0	0	0	0
(Third true leaf)	5	5	6	0	0	0	1
	10	2	2	3	1	4	0

Figure captions

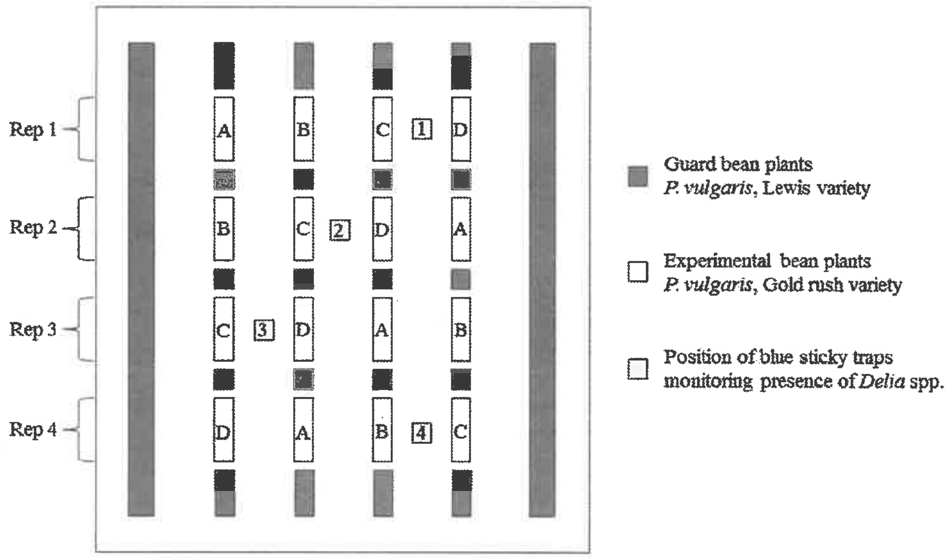
Figure 1. Plot design for 2016 field trials at Canning, Nova Scotia and Wilmot Valley, PEI. Letters A through D represent planting dates. Dark grey areas indicate guard plants (Gold Rush *P. vulgaris* var). Numbered boxes show location of blue sticky traps for monitoring adult populations of *Delia* spp.

Figure 2. Mean number of *D. platura* and *D. florilega* females (A) and *D. platura* males (B) and *D. florilega* males (C), identified per trap from four traps collected weekly from organic commercial bean fields on Prince Edward Island (2015 and 2016) and Nova Scotia (2016).

Figure 3: Mean number (± SE) *D. platura* eggs per plant when given choice of two varieties of *P. vulgaris*: Gold Rush and Lewis varieties in a growth chamber study. Phenological growth stage (varieties pooled) followed by the same letter not significantly different ($P < 0.05$). For most growth stages n=10, with the exception of growth stages 12 and 13 (n = 9) and 75 (n = 8).

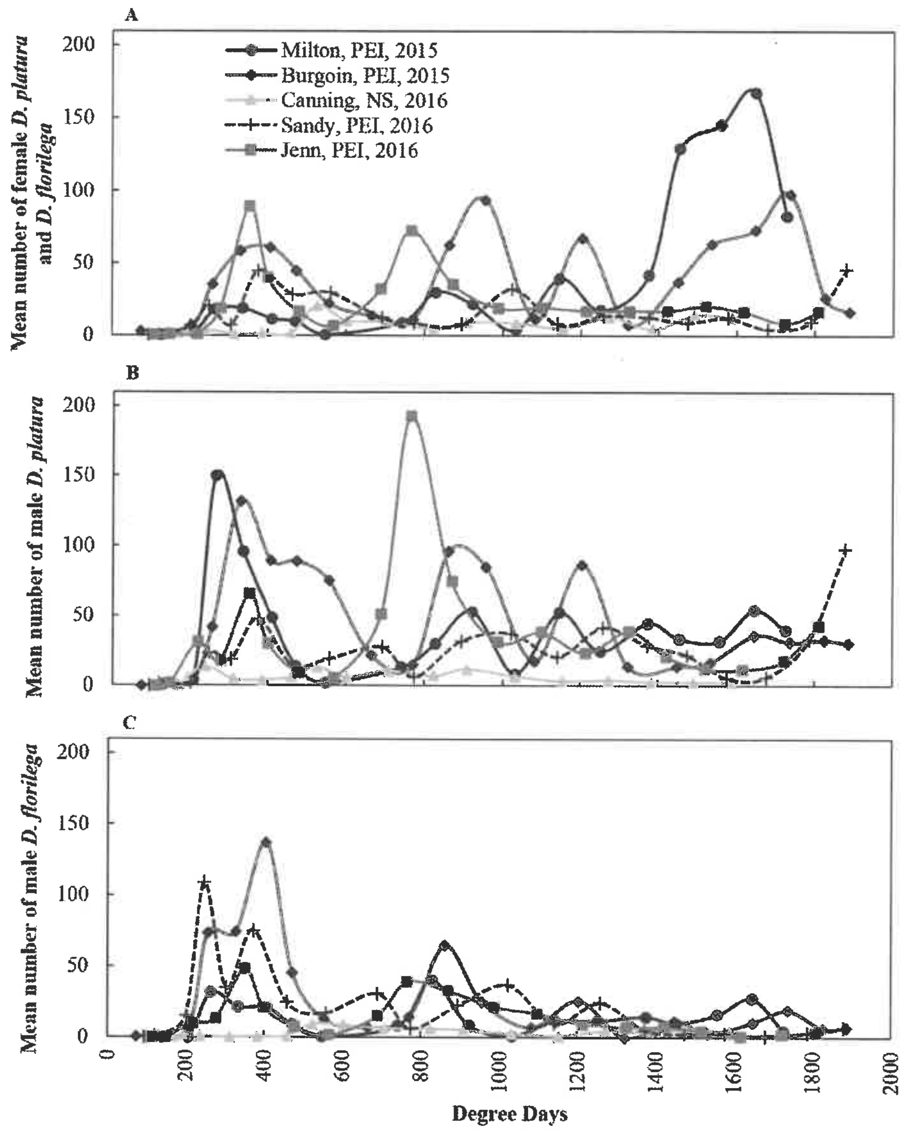
Figure 4: Mean number of *D. platura* and *D. florilega* females per blue sticky trap from organic bean fields in Canning, Nova Scotia (A), Wilmot Valley (B) and Saint Ann (C) on PEI in 2016 by accumulated degree days

(base 3.9, start date of 1 January). Numbers 1-5 denote planting times in conjunction with *Delia* phenology.



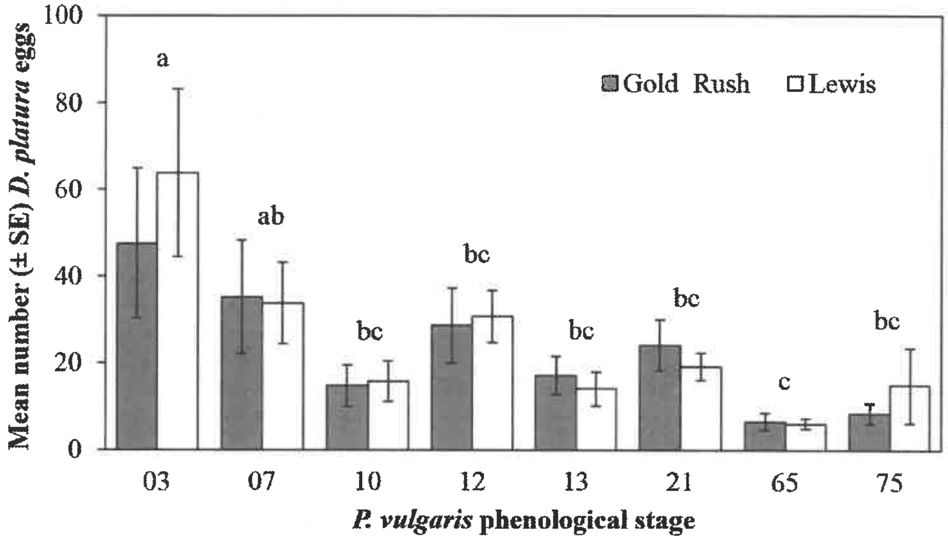
Plot design for 2016 field trials at Canning, Nova Scotia and Wilmot Valley, PEI. Letters A through D represent planting dates. Dark grey areas indicate guard plants (Gold Rush P. vulgaris var). Numbered boxes show location of blue sticky traps for monitoring adult populations of Delia spp.

158x94mm (240 x 240 DPI)



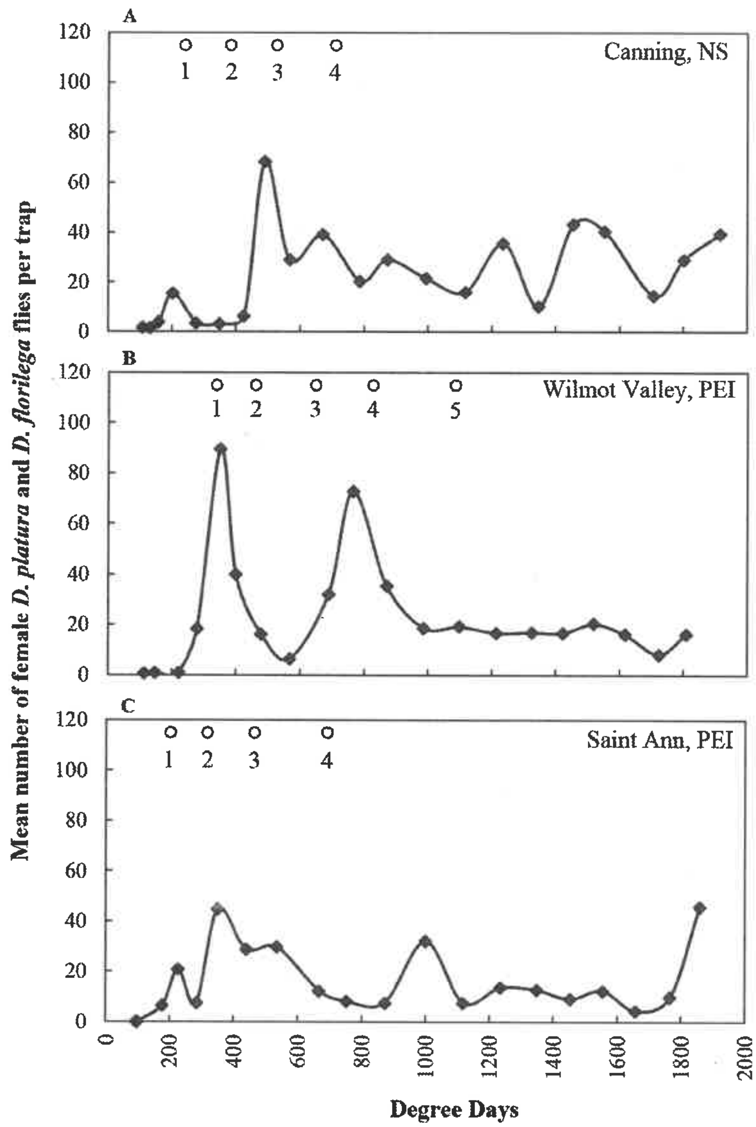
Mean number of D. platura and D. florilega females (A) and D. platura males (B) and D. florilega males (C), identified per trap from four traps collected weekly from organic commercial bean fields on Prince Edward Island (2015 and 2016) and Nova Scotia (2016).

169x207mm (240 x 240 DPI)



Mean number (± SE) D. platura eggs per plant when given choice of two varieties of P. vulgaris: Gold Rush and Lewis varieties in a growth chamber study. Phenological growth stage (varieties pooled) followed by the same letter not significantly different ($P < 0.05$). For most growth stages n=10, with the exception of growth stages 12 and 13 (n = 9) and 75 (n = 8).

147x87mm (240 x 240 DPI)



Mean number of D. platura and D. florilega females per blue sticky trap from organic bean fields in Canning, Nova Scotia (A), Wilmot Valley (B) and Saint Ann (C) on PEI in 2016 by accumulated degree days (base 3.9, start date of 1 January). Numbers 1-5 denote planting times in conjunction with Delia phenology.

136x202mm (240 x 240 DPI)