



## Assessing annual and perennial flowering plants for biological control in asparagus

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### ABSTRACT

Despite the progress made to date to incorporate habitat diversification in farms, implementing it effectively for biological control programs in agriculture still faces many challenges. Our goal was to screen annual and perennial flowering plant species for their attractiveness to natural enemies and herbivores, and to select promising species to be used as flower enhancements in asparagus agroecosystems. Tested perennials were generally more attractive to predators than to parasitoid wasps, which were found in low numbers during our study. Among the annual plants, sweet alyssum (*Lobularia maritima* L. Desv.) was highly attractive to parasitoid wasps in common garden experiments and was selected for use in flowering borders, but it was not attractive to parasitoid wasps in asparagus field trials. We did not detect any effects of sweet alyssum on asparagus miner damage or on generalist herbivore abundance in asparagus fields. Thrips abundance was reduced in asparagus adjacent to sweet alyssum, but only at the field edge. Emergence rates from excised asparagus miner pupae were low with 18% (94) surviving, of which 77 were asparagus miners and 17 were parasitoids. In summary, although flower enhancements can often provide benefit for biological control of herbivores, our current results did not support this finding, but we suggest that continued effort is made to test the potential of perennial plants as flower enhancements in this agroecosystem as they require less maintenance than annuals.

### 1. Introduction

Increasing agroecosystem biodiversity at local or landscape scales is predicted to support natural enemies, which may in turn suppress herbivore populations and improve biological control (Barbosa, 1998; Bianchi et al., 2006; Blaauw and Isaacs, 2015; Chaplin-Kramer et al., 2011; Crowder and Jabbour, 2014; Gurr et al., 2017; Landis et al., 2000; Orr, 2009; Shanker et al., 2012; Werling et al., 2011). Despite the progress made to date to incorporate habitat diversification in farms, implementing it effectively for biological control programs in large scale agriculture is still faced with many challenges (Tschamtko et al., 2016). Designing effective biological control programs that rely on boosting local natural enemy populations can be achieved if pest densities are influenced by natural enemies and if the changes to the habitat do not lead to increased pest numbers. If these conditions are met, the next step is to diversify the habitat in such a way that it fits into the practical considerations of crop management (Tschamtko et al., 2016). Flower enhancement in and around agricultural fields is a common method of creating habitat diversity in farms to protect beneficial insects, but there are numerous important considerations that need to be tested before the successful deployment of flowers for conservation

biological control. For example, the timing of flower availability for natural enemies, flower species and morphology, availability of nectar/pollen or shelter, ease and cost of plant establishment, and interactions of flowers with the target and non-target insect groups need to be described for successful implementation (Foti et al., 2017; Wäckers, 2004).

Habitat enhancements with flowers are often designed for specific groups of organisms, such as natural enemies or pollinators, and they are likely to have various impacts on both beneficial and pest arthropods in or near agricultural fields (Winkler et al., 2010). Thus, it is necessary to assess the consequences of placing flowers near crops before implementing them in agricultural settings. Polyphagous herbivores, such as Japanese beetles (*Popillia japonica*, Coleoptera: Scarabaeidae) and thrips (Thysanoptera), can find agricultural fields increasingly attractive if surrounded by flowering borders. Specialist herbivores can also be problematic if for example the adult life stage benefits from nectar feeding. This is the case with asparagus miners, *Ophiomyia simplex* (Diptera: Agromyzidae), where sugar-rich diets increase life span of both adult asparagus miners and their parasitoids (Morrison et al., 2014), and both groups feed on nectar from asparagus and non-crop plants (Ferro and Gilbertson, 1982). It is important to

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note that not all flowers are equal in terms of attractiveness or resource availability; for example in greenhouse feeding trials buckwheat (*Fagopyrum esculentum* Moench), fava bean (*Vicia faba* L.), and sweet alyssum (*Lobularia maritima* L. Desv.) were poor resources for asparagus miner adults when compared to water, while Riddell's goldenrod (*Solidago riddellii* Frank) doubled adult miner longevity relative to water (Morrison et al., 2014). Sweet alyssum and buckwheat in particular are easy-to-grow annuals and highly attractive to parasitoids, making them common choices for natural enemy attraction (Arnó et al., 2018; Amoros-Jimenez et al., 2014; Balzan and Wackers, 2013; Gontijo et al., 2013; Hogg et al., 2011a,b; Johanowicz and Mitchell, 2000; Nafziger and Fadamiro, 2011; Pumarino and Alomar, 2012). Other annual species like partridge pea (*Chamaecrista fasciculata* (Michx. Greene)) have extrafloral nectaries which may make it attractive to beneficial insects (Rezende et al., 2014).

While annuals may be attractive to natural enemies and have a long history as insectary plants, perennial species may be preferable for growers because they do not require replanting and may provide permanent habitat or overwintering sites for natural enemies. Several species of perennial plants native to Michigan have been screened for attractiveness to natural enemies; those flowering in mid- to late-summer to coincide with second-generation asparagus miner activity include butterfly weed (*Asclepias tuberosa* L.), bee balm (*Monarda fistulosa* L.), hoary vervain (*Verbena stricta* Vent.), early goldenrod (*Solidago juncea* Ait.), and mountain mint (*Pycnanthemum virginianum* (L.) T. Durand & B.D. Jacks, ex B.L. Rob. & Fernald). Each of these species showed high levels of attractiveness to natural enemies and the ability to grow in sandy well-drained and occasionally dry conditions characteristic of asparagus fields (Fiedler and Landis, 2007; per. comm.). Perennials are an optimal choice for flower enhancements in perennial cropping systems or in non-rotated annual crops, since they require little maintenance after establishment.

Here we focused on testing different flower enhancements in asparagus (*Asparagus officinalis* L.), a crop with few key pests and a well described natural enemy community (Ingrao et al., 2017; Morrison et al., 2014). Our main goal was to test the efficacy of flower enhancements to improve biological control by assessing abundance of parasitoid wasps visiting floral enhancements, parasitism rate of asparagus miner pupae, and asparagus miner damage. In addition, we assessed the parasitoid, predator, and herbivore arthropod communities associated with the selected flowering plants and with asparagus plots adjacent to flowering strips. We also considered the practicality of these enhancements for crop production thus we aimed to design schemes that may complement current crop management programs. In asparagus, natural enemies and herbivores are typically concentrated along the field borders and edges (Ingrao et al., 2017; Morrison and Szendrei, 2013) which lends itself to placing flower enhancements near field edges. For our study, annual and perennial flowering species were selected for common garden screening based on their attractiveness to natural enemies and/or lack of resources for one of the key pests, the asparagus miner (Fiedler and Landis, 2007; Morrison et al., 2014). The most promising annual plant species was subsequently selected to test flower enhancements next to asparagus fields.

## 2. Materials and methods

### 2.1. Annual garden experiment

An annual common garden experiment was established at a research farm in 2015 in Hart, MI. A 12 × 12 m field was treated with Round-Up® Weed and Grass Killer (1 L/ha glyphosate, Scotts Miracle-Gro™, Marysville OH) on 10 May 2015, and rototilled on 19 May 2015. Twenty-five 1 × 1 m plots with 1 m inter-plot spacing were established with five replicates of five treatments in a randomized complete block design. Treatments were sweet alyssum (*L. maritima* var. 'Carpet of Snow', non-native, seeded 20 May 2015, 0.28 g/m<sup>2</sup>, mixed with 100 g

sand and surface sown, Seedland Inc., Wellborn, FL), fava bean (*V. faba* L. var. 'Windsor', non-native, seeded 28 May 2015, 17.9 g/m<sup>2</sup>, 2 cm seed depth, Johnny's Selected Seeds, Fairfield, ME), partridge pea (*C. fasciculata*, native, stratified for 14 days according to supplier directions and seeded 28 May 2015, 2.2 g/m<sup>2</sup>, 0.5 cm seed depth, Prairie Moon Nursery, Winona MN), buckwheat (*F. esculentum* var. 'Mancan', non-native, seeded 11 June 2015, 6.7 g/m<sup>2</sup>, 2 cm seed depth, Sustainable Seed Co., Chico, CA), and a control consisting of naturally occurring weeds.

Floral canopies were vacuum sampled for arthropods weekly for eight weeks, beginning 21 July 2015 (modified Craftsman® 19.2 V vacuums, Heavy Duty Hand-Held DC Vac/Aspirator, BioQuip® Products Inc., Rancho Dominguez, CA). During the weekly sampling, the entire floral canopy was sampled in each plot for 1 min during mid-morning. Collected arthropods were transferred to plastic zip-top bags and frozen until identification. Arthropods were identified to order, family, or species (Table S1). Specimens that were identified to order only (e.g., Coleoptera or Diptera) were not categorized by functional group and were not included in analyses. Concurrent with arthropod sampling, percent of the plot flowering was estimated for each plot, based on the maximum potential flowering for the plants present in the plot (i.e., if every plant in the plot was producing the maximum amount of flowers, 100% of the plot was flowering).

Supplementary data associated with this article can be found, in the online version, at <https://doi.org/10.1016/j.biocontrol.2018.08.013>.

### 2.2. Perennial garden experiment

Several species of perennial plants native to Michigan have been screened in previous studies for attractiveness to natural enemies (Fiedler and Landis, 2007); in 2016 we selected some that flower in mid- to late-summer to coincide with second-generation asparagus miner activity. The perennial common garden was established in the same field as the 2015 annual garden experiment. The field was expanded to 15 × 40 m, sprayed with Round-Up® Weed and Grass Killer (1 L/ha glyphosate) on 25 May 2016 and rototilled on 5 June 2016. Thirty 1 × 1 m plots with 3 m inter-plot spacing were established with six replicates of five native perennial plant treatments randomized within each block. Three plant plugs per plot were transplanted and mulched on 6 June 2016: bee balm, spotted bee balm (*Monarda punctata* L.), butterfly weed, early goldenrod, and hoary vervain; all plugs were obtained from Wildtype Native Plant Nursery (Mason, MI). Spotted bee balm did not survive the winter and was replaced by 2-year-old transplants of mountain mint acquired from a previously established planting in Clarksville, MI.

Floral canopies were vacuum sampled weekly for six weeks, beginning 18 July 2017. Due to time constraints, the floral canopy in each treatment was sampled for 30 s during mid-morning. Collected arthropods were transferred to plastic zip-top bags and frozen until identification. Arthropods were identified to order, family, or species (Table S2). Concurrent with arthropod sampling, floral area was estimated for each plot by measuring the average size of a flower or inflorescence, counting the number of flowers or inflorescences in the plot or subsection of the plot, and multiplying flower or inflorescence size by flower or inflorescence number. Floral area was chosen to estimate flowering, rather than percent of the plot flowering as was used in the annual garden, to give an absolute value of flowering rather than a value relative to species.

### 2.3. Border strip experiment

In 2017, four pairs of border strips (10 × 1 m) were established next to a mature experimental asparagus field (1300 m<sup>2</sup>, established in 2011, not treated with pesticides during this experiment). Each pair of border strips consisted of sweet alyssum and a mowed control. The border strips were established at least 20 m (average distance = 110 m) from

the perennial garden plots, which were established in 2016 at the same research farm in Hart, MI (Suppl. Fig. S1). The strips were located 1 m from the edge of the asparagus plots, in the field border comprised of occasionally mowed weeds. Treatment strips were prepared by applying Round-Up® Weed and Grass Killer (1 L/ha glyphosate) on 23 May 2017, and rototilling on 30 May 2017. On 8 June 2017, strips were seeded with sweet alyssum as described in Section 2.1. Three strips with low germination rates were identified on 22 June 2017, and were reseeded with identical methodology as above. On 27 June and 5 July 2017, Assure® Grass Killer (0.17 L/ha quizalofop-p-ethyl, DuPont™, Wilmington DE) was applied to flowering strips. On 12 July 2017, the control strips were mowed to remove flowers and maintain consistent vegetation structure across control strips.

Arthropods within border strips and the asparagus field were sampled weekly for six weeks beginning 18 July 2017. To sample border strips, the flower canopy in flower enhancements or the available vegetation in mowed control plots was vacuumed for 30 s during mid-morning, which sampled the majority of the plot area. Collected arthropods were transferred to plastic zip-top bags and frozen until identification. Arthropods were identified to order, family, or species; in addition parasitoid wasps from vacuum samples were identified to family, genus, or species when possible. Concurrent with arthropod sampling, floral area was estimated for each plot in the same manner as described in Section 2.1.

To sample in the asparagus field, 1-m-wide sections of the asparagus canopy were sampled along a 15 m transect perpendicular to the center of each treatment strip; arthropods were observed along each transect at 0, 5, 10, and 15 m into asparagus plots. Highly mobile arthropods such as flies or wasps were counted on the asparagus plants, and the remainder were sampled by shaking the asparagus over a beat sheet (1 × 1 m). Sampled arthropods were identified and recorded.

Asparagus miner damage sampling and pupal collection to determine parasitism rate in the asparagus field occurred on 13 September 2017. Sampling took place at 0, 5, and 10 m into asparagus plots along a transect perpendicular to the center of each treatment or control strip. At each sample point, a 1 m length of row was sampled for the number of asparagus stems and the number of stems with mines. Twenty mined stems were collected from each transect point, asparagus miner pupae were extracted, placed individually in ventilated 59 mL plastic Solo® cups (Dart Container Corp., Mason MI), and stored in a growth chamber at 26 °C, 75% relative humidity, 16:8 L:D for four weeks, refrigerated at 4 °C for another four weeks, then left at room temperature for a final four weeks. As they emerged, parasitoids and miners were counted and identified to genus or species using voucher specimens from the A.J. Cook Arthropod Research Collection (Michigan State University, East Lansing MI).

#### 2.4. Statistical analyses

Treatment effects on season-long arthropod community composition in all experiments were analyzed with non-metric multidimensional scaling (NMDS, function = ‘monoMDS’, ‘adonis’, package = ‘vegan’, (Oksanen et al., 2015)) in R version 3.2.2 (R Core Team, 2015). Composition of arthropod communities in asparagus plots also included distance into plot effects. For abundance analyses, arthropods were divided into the functional categories of parasitoid, predator, or herbivores (see Tables 1 and 2, Tables S1 and S2 for categories by taxa). Arthropods that are not a focus of pest control in asparagus, such as pollinators, were not analyzed, nor were arthropods that could not be identified precisely enough to assign a functional category.

Parasitoid, predator, and herbivore abundances in vacuum samples from the annual garden, perennial common garden, and border strips were each summed across all sample dates, log-transformed and analyzed with linear mixed-effects models (function = ‘lmer’, package = ‘lme4’ (Bates et al., 2015)), including treatment as a fixed effect and spatial block as a random effect. Common garden models also

**Table 1**

Total number of arthropods found in border strip vacuum samples. Abundances are the total found across the season, vacuum sampled weekly for 6 weeks.

Arthropod taxa	Mowed control	Sweet alyssum	Functional category
Pteromalidae	46	51	Parasitoid
<i>Chorebus rondanii</i>	4	6	Parasitoid
Neochrysocharis	13	21	Parasitoid
Araneae	3	9	Predator
Anthocoridae	12	6	Predator
Neuroptera (larva)	2	2	Predator
Cantharidae	4	1	Predator
Miridae	35	43	Herbivore
<i>Lygus</i> spp.	17	9	Herbivore
Chrysomelidae	1	0	Herbivore
Aphidoidea	0	4	Herbivore
Membracidae	8	30	Herbivore
Lepidoptera (larva)	3	3	Herbivore
Thysanoptera	16	8	Herbivore
<i>Popillia japonica</i>	1	0	Herbivore
Diptera (non-Syrphidae)	372	394	Uncategorized
Apoidae	0	1	Uncategorized
Formicidae	0	1	Uncategorized
Coleoptera	9	21	Uncategorized

**Table 2**

Total number of arthropods found in asparagus plots adjacent to border strips. Abundances are the total found across the season, beat sheet sampled weekly for 6 weeks.

Arthropod taxa	Mowed control	Sweet alyssum	Functional category
Parasitoid wasp	10	12	Parasitoid
Syrphidae	3	3	Predator
Nabidae	3	3	Predator
Araneae	22	16	Predator
Vespidae	1	6	Predator
Anthocoridae	11	16	Predator
Coccinellidae (adult)	2	1	Predator
Chrysopidae	6	5	Predator
Thysanoptera (predatory)	0	1	Predator
<i>Podisus maculiventris</i>	4	6	Predator
<i>Ophiomyia simplex</i>	6	11	Herbivore
Miridae	36	31	Herbivore
<i>Lygus</i> spp.	11	12	Herbivore
Aphidoidea	44	45	Herbivore
Membracidae	3	4	Herbivore
Lepidoptera (larva)	5	0	Herbivore
Pentatomidae	2	6	Herbivore
<i>Crioceris asparagi</i> (adult)	9	4	Herbivore
<i>Crioceris asparagi</i> (larva)	4	0	Herbivore
<i>Crioceris asparagi</i> (egg)	29	5	Herbivore
<i>Popillia japonica</i>	416	319	Herbivore
Thysanoptera	511	405	Herbivore
Coleoptera	5	3	Uncategorized
Anthicidae	64	47	Uncategorized

included floral display area as fixed effects. All models were checked for overdispersion. Two taxa comprised more than 20% of collected arthropods from the border strip experiment, and were individually analyzed by the same methods as described above; these taxa were Membracidae, which made up 21% of arthropods in control strips and 6% of arthropods in flower strips; and tarnished plant bugs (*Lygus lineolaris*, Hemiptera: Miridae), which made up 28% of arthropods in control strips, 38% of arthropods in flower strips.

Predator and herbivore abundances from asparagus transect beat sheet samples were summed across all sample dates, log-transformed and analyzed with linear mixed-effects models, including treatment, distance into plot, and a treatment-by-distance interaction as fixed effects and block as a random effect. The interaction term was dropped when not significant. Two taxa comprised more than 20% all collected

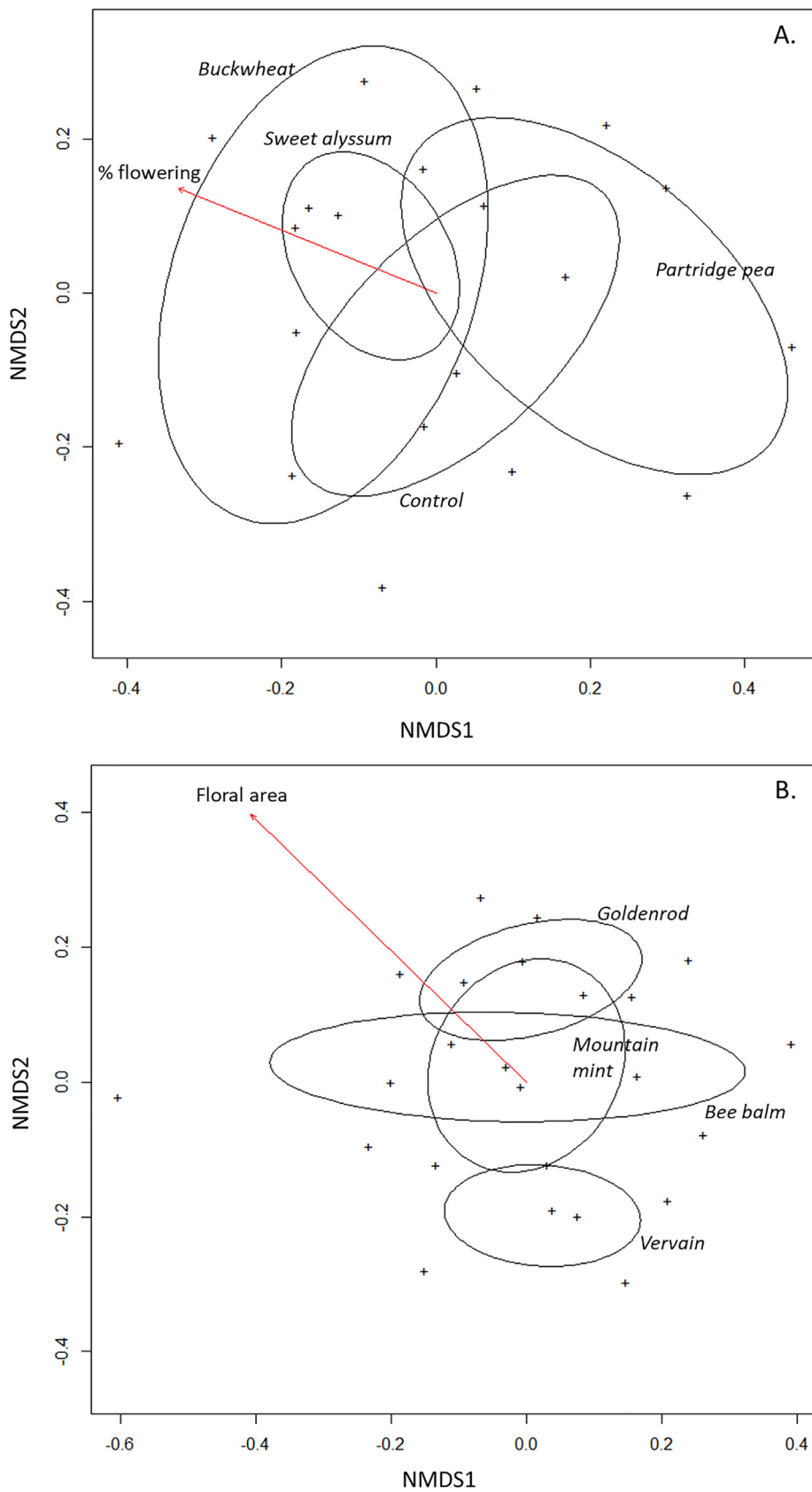


Fig. 1. Ordination plots for arthropod communities in the annual (A) and perennial (B) common gardens. Arthropods were vacuum sampled weekly for eight (A) or six (B) weeks. Ellipses represent 95% confidence intervals for the arthropod community associated with each treatment (labeled in italics). Mean floral area across the sampling period is shown as a vector.

arthropods from asparagus transects, and were individually analyzed by the same methods as described above. These taxa were Japanese beetles, which made up 34–35% of collected arthropods; and thrips, which made up 43–44% of collected arthropods. In all cases, when treatment effects were significant at  $\alpha = 0.05$ , Tukey's HSD was performed to determine differences among treatments.

Proportion of stems damaged by asparagus miner along asparagus transects was analyzed with a generalized linear model (function = 'glm') including treatment, distance into plot, and a treatment-by-distance interaction as fixed effects and block as a random effect. Proportion parasitized asparagus miner pupae was not statistically analyzed due to low emergence rates; number of miners and parasitoids emerged are reported.

### 3. Results

#### 3.1. Annual garden experiment

Fava bean treatments did not survive in high enough numbers to be included in analyses. The flowering seasons of the remaining annual species were similar, with flowering over 70% for August 2015 (Suppl. Fig. S2A). The beginning of the second generation of asparagus miners, the biological control target for this study, began on 29 July 2015 (MSU Enviroweather, 2018). Arthropod communities differed significantly across flowering species in the annual garden ( $F_{3,16} = 15.1$ ,  $R^2 = 0.74$ ,  $P < 0.01$ , Stress = 0.2, Fig. 1A). Annual plant species attracted significantly different numbers of parasitoids ( $\chi^2 = 17.5$ ,  $P < 0.01$ ) and herbivores ( $\chi^2 = 22.1$ ,  $P < 0.01$ ), but not predators ( $\chi^2 = 0.8$ ,  $P = 0.9$ ). Sweet Alyssum had at least three times more parasitoid wasps and twice as many herbivores compared to the other treatments (Fig. 2A). No asparagus miners were collected from any treatments in the annual garden.

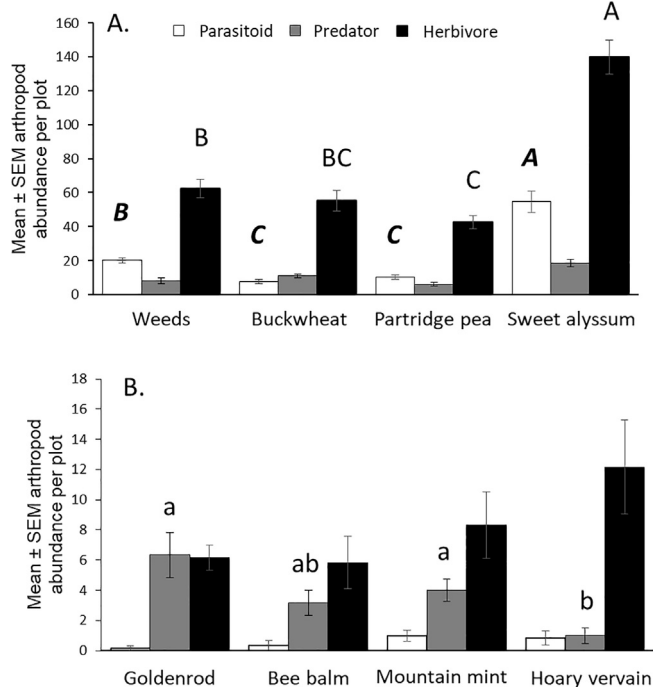


Fig. 2. Mean  $\pm$  SEM arthropod abundance in flowering plots from annual (A) and perennial (B) common gardens. Arthropods were summed across vacuum samples for eight (A) or six (B) weeks, and averaged across replicates. Where letters are present above bars, different letters indicate significant differences across plant species using a Tukey's HSD ( $\alpha = 0.05$ ).

#### 3.2. Perennial garden experiment

Butterfly weed did not produce flowers in sufficient numbers to be included in analyses. The flowering seasons of the remaining perennial species differed, with bee balm and hoary vervain flowering early and goldenrod and mountain mint flowering later (Suppl. Fig. S2B). The beginning of the second generation of asparagus miners, the biological control target for this study, began on 26 July 2017 (MSU Enviroweather, 2018). Arthropod communities differed significantly across flowering species in the perennial garden ( $F_{3,20} = 2.1$ ,  $R^2 = 0.24$ ,  $P < 0.01$ , Stress = 0.3, Fig. 1B). Perennial plant species attracted different numbers of predators ( $\chi^2 = 30.1$ ,  $P < 0.01$ ), but not parasitoids ( $\chi^2 = 3.8$ ,  $P = 0.3$ ) or herbivores ( $\chi^2 = 3.1$ ,  $P = 0.4$ ). Early goldenrod and mountain mint were roughly 5 times more attractive to predators than hoary vervain, while bee balm attracted an intermediate number of predators (Fig. 2B). The majority of the parasitoid wasps collected were Pteromalidae (12 individuals) and most of these were found on mountain mint (6 individuals, Suppl. Table S2).

#### 3.3. Border strip experiment

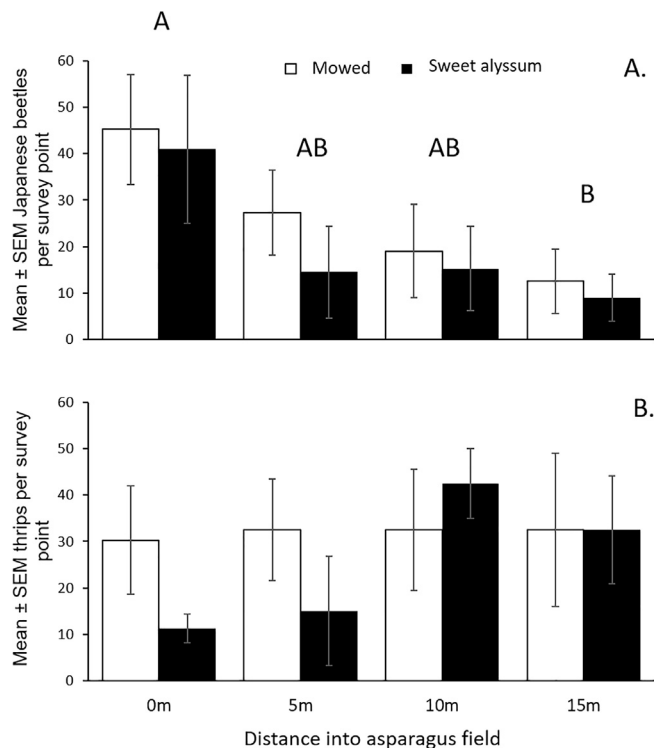
In the border strips, overall arthropod community composition as analyzed with NMDS did not differ between sweet Alyssum and control strips ( $F_{1,6} = 1.3$ ,  $R^2 = 0.18$ ,  $P = 0.3$ , Stress = 0.08, Table 1). Predators were more abundant in sweet Alyssum strips ( $\chi^2 = 22.8$ ,  $P < 0.01$ ); herbivores did not differ between treatments ( $\chi^2 = 1.8$ ,  $P = 0.2$ ). Asparagus miner adults were not found in sweet Alyssum or in control strips. Membracids and tarnished plant bugs were a substantial portion of the observed herbivores and were analyzed separately. While abundances of membracids did not differ between treatments (control mean  $\pm$  SEM:  $6.5 \pm 5.8$ ; flowering mean  $\pm$  SEM:  $3 \pm 0.9$ ,  $\chi^2 = 1$ ,  $P = 0.3$ ), there were twice as many tarnished plant bugs in the flowering (mean  $\pm$  SEM  $17.5 \pm 3.3$ ) relative to control strips (mean  $\pm$  SEM:  $8.5 \pm 3.2$ ,  $\chi^2 = 3.9$ ,  $P < 0.05$ ). Parasitoid wasps were not statistically different across treatments ( $\chi^2 = 3.3$ ,  $P = 0.07$ , Fig. 3); most parasitoids were Pteromalidae (69% of parasitoid wasps, Table 1).

In the asparagus field, overall arthropod community composition as analyzed with NMDS was not significantly different between border strip treatments ( $F_{1,24} = 0.7$ ,  $R^2 = 0.02$ ,  $P = 0.6$ , Stress = 0.3, Table 2), or with distance into plot ( $F_{3,24} = 1.8$ ,  $R^2 = 0.1$ ,  $P = 0.05$ ). Overall predator and herbivore abundances within asparagus plots did not differ by treatment (predator:  $\chi^2 = 0.002$ ,  $P = 0.9$ ; herbivore:  $\chi^2 = 1.8$ ,  $P = 0.2$ ) or distance (predator:  $\chi^2 = 1.6$ ,  $P = 0.2$ ; herbivore:  $\chi^2 = 1.0$ ,  $P = 0.3$ ). Japanese beetle abundance was about twice as high on the field edge relative to inner rows ( $\chi^2 = 9.5$ ,  $P = 0.02$ , Fig. 4A). Thrips abundance was not affected by distance into plot when adjacent to control strips, but was reduced by 65% at the field edge relative to



Fig. 3. Mean  $\pm$  SEM abundance of parasitoid wasps, predators, and herbivores within border strips. Arthropods were vacuum sampled, summed across samples for six weeks, and averaged across replicates. Asterisk indicates significant ( $\alpha = 0.05$ ) differences in abundances between treatments.





**Fig. 4.** Mean  $\pm$  SEM abundance of Japanese beetles (A) and thrips (B) per survey point in asparagus field transects. Arthropods were counted on asparagus plants or in a beat sheet, summed across samples for six weeks, and averaged across replicates. Different letters above bars indicate significant differences in abundances of Japanese beetles by distance into plot tested with Tukey's HSD ( $\alpha = 0.05$ ). Contrasts within distance into plot from field edge for thrips abundance were not significantly different across treatments.

interior rows when adjacent to flowering strips (interaction:  $\chi^2 = 4.2$ ,  $P = 0.04$ , Fig. 4B).

Neither border strip treatment ( $\chi^2 = 0.1$ ,  $P = 0.7$ ) nor distance into plot ( $\chi^2 = 2.9$ ,  $P = 0.09$ ) significantly influenced asparagus miner damage rate. Overall, 43% of asparagus stems had miner damage, with 56% of stems at the field edge sustaining damage and 36–38% of stems 5 m and 10 m into the field sustaining damage. Of 525 pupae excised from damaged asparagus stems, 94 produced either asparagus miners or parasitoids (18% overall emergence rate). Miner emergence rate was 17% in control treatments (55 miners/324 collected pupae) and 11% in flower treatments (22 miners/201 collected pupae). Of surviving pupae, parasitism rate was 20% in control treatments (14 parasitoids/69 living pupae) and 12% in flower treatments (3 parasitoids/25 living pupae; Suppl. Table S3).

#### 4. Discussion

Although flower enhancements often provide benefits for biological control (Blaauw and Isaacs, 2014; Tschumi et al., 2016) our current results do not support this finding. In this study, we made progress in assessing annual and perennial plants for their suitability to grow and be used as flower enhancements in asparagus agroecosystems. Since none of these plants have been tested in or near asparagus fields previously, we had no prior information on which arthropods would use these and if they would be utilized by those natural enemies that we identified as important biological control agents in our system (Ingrao et al., 2017; Morrison et al., 2014). Our measurements indicated that arthropod communities found on flowers differed across plant species. This was not likely a function of floral area, based on the distance between the floral area vector and community composition in the ordination space. While peak flowering time was not included in the

multidimensional scaling analysis, differences in arthropod community composition across perennial species may be better explained by within-season variation in flowering times. For example bee balm and hoary vervain flowered earlier, while goldenrod and mountain mint flowered later. Goldenrod and mountain mint attracted more predators on average relative to bee balm or hoary vervain, although bee balm attracted more total predators. Bee balm was especially attractive to minute pirate bugs (Anthocoridae), an insect group that was previously noted on multiple perennial flowering plant species in Michigan (Fiedler and Landis, 2007). While these insects are generalist predators (Baez et al., 2004), and are frequently used for augmentative biological control, they are not known to feed on key asparagus pests, such as the asparagus miner. Thus, the annuals and perennials we tested in the asparagus agroecosystem may not be attracting those natural enemies that can suppress asparagus pests and we will need to test different ones in the future that are more suitable for the natural enemies of key pests.

Despite the fact that our plant selection was based on prior information about their benefit to biological control agents, our results were often inconsistent. For example sweet alyssum in garden plots was attractive to parasitoids, but as border strips it was not more attractive than mowed control areas. Some differences between these two experiments were that the flowers in the common garden were further away from asparagus and were planted in a 1 by 1 m square, whereas the flower strips were placed in a long rectangle along the asparagus field border close to the crop. This potentially indicates the importance of flower enhancement arrangement, size, position relative to crop field and surrounding habitat context, with relatively small changes causing potentially significant consequences for biological control. We also detected that in the flower strip experiment sweet alyssum was not more attractive than the mowed control area possibly due to differences in plant architecture: sweet alyssum plots had complex and vertical architecture, with plants reaching  $\sim 0.5$  m in height whereas mowed control treatments had relatively simple architecture, reaching less than 0.1 m. Similarly, the positive predator response to sweet alyssum in border strip experiment did not transfer into the garden experiment setting. The greater number of predators in sweet alyssum relative to control border strips may have been due to the presence of alternative prey, which if not a key pest of the focal crop may ultimately benefit conservation biological control efforts (Jonsson et al., 2010; Pumarino and Alomar, 2012).

Ultimately the goal of biological control is that the boost natural enemies get from flower enhancements translates to a reduction in herbivores on the crop. One of the main herbivores, the asparagus miner, feeds on flowers as adults and although it was present during our experiments, it was not found on any of our tested plants or the weedy plots in the common gardens. This is an important finding that needs further confirmation using different sampling methods, but if our findings are correct then these flowers would not cause increased herbivore pressure in asparagus when used as flower enhancements. In our experiments herbivore abundance within asparagus plots was not significantly reduced in the presence of sweet alyssum border strips. This does not support our expectation based on previous research which found some predator taxa, including those found in our experiment (e.g., Chrysopidae, Coccinellidae), inhabiting asparagus border habitat and potentially moving into the field to feed on herbivores (Ingrao et al., 2017). While not sampled in our study, predators often found on the ground such as Linyphiidae (sheetweb spiders), Staphylinidae (rove beetles) and Carabidae (ground beetles) have been documented as feeding on asparagus miners (Ingrao et al., 2017), and may benefit from the habitat and protection offered by flowering borders. Spiders were common within our floral strips and this group has numerous taxa that consume asparagus miners (Ingrao et al., 2017). Spider webs can often be seen with adult asparagus miners, in and around asparagus fields (Z. Szendrei, per. obs.). We also found many members of Anthicidae (flower beetles) which have not been described previously as predators of asparagus herbivores but they are a large omnivorous taxa that could

potentially provide biological control in this system.

## 5. Conclusion

In summary, our study leaves many areas open for future investigation. For example, the size and configuration of flower enhancements may have an impact on biological control. We will also need to investigate how landscape complexity around focal fields impacts natural enemies, especially considering that in our systems wooded borders seem to favor natural enemy abundance (Ingrao et al., 2017). Beneficial insect abundances can increase on perennial species multiple years after planting, which should be tested in the future (Blaauw and Isaacs, 2014). In general, our recommendation is to continue to study the potential of using flower enhancements in this system and include new plant species in the screening process, with a focus on perennials which require less input from growers than annuals. In addition, we need to study how natural enemies use the flowering resources because they may initially be attracted to fields (Gontijo et al., 2013), but arthropods may not disperse into crops (Quinn et al., 2017). In previous experiments we detected no intraguild predation among asparagus predators (Ingrao et al., 2017), and although this prior work did not investigate the impact of flower enhancements on interactions within the third trophic level, it is possible that arthropod interactions change as resources are altered. It will also be important to test flower species mixes that can provide resource complementarity and longevity through the season.

## 6. Author statement

AB contributed to data curation, formal analysis, investigation, methodology, project administration, resources, writing – original draft, and writing – review & editing. MG contributed to conceptualization and funding acquisition. ZS contributed to conceptualization, funding acquisition, project administration, resources, writing – original draft, and writing – review & editing.

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