Active management of wildflower strips in commercial sweet cherry orchards enhances natural enemies and pest regulation services

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Abstract

To protect sweet cherry (Prunus avium L.) against pests in commercial orchards, pesticides are frequently used, but some have adverse environmental impacts. Natural enemies can deliver protection against pests but compared to the surrounding non-crop habitat their abundance is usually low in intensively managed agricultural systems. Wildflower interventions established for Conservation Biological Control as part of Integrated Pest Management (IPM) can reinstate habitat for natural enemies within cropped areas and enhance natural pest control. Over a three-year period, this more sustainable approach to crop protection was investigated in eight sweet cherry orchards protected under polytunnels in the West Midlands (UK). Wildflower strips were established in alleyways between rows of cherry trees and managed under two cutting regimes, Standard Wildflower Strips (SWS) (a single cut in late September) and Actively Managed Wildflower Strips (AMWS) (regularly cut to a height of 20 cm throughout the growing season). These were compared to unsown Control Strips (CS) (original vegetation dominated by grass species). To investigate natural enemy populations and pest regulation services, direct search, Vortis suction sampling, beat sampling, and aphid bait cards were used. Araneae (40.0% of records) and parasitoid wasps (22.7%) were the most frequent natural enemies recorded in alleyways, whilst Anystidae (51.8%) and Araneae (20.8%) were most abundant on cherry trees. Wildflower treatments almost doubled the abundance of natural enemies in alleyways, and increased abundance in cherry trees by ~15% compared to the CS. Wildflower strips increased predation of aphids (bait cards) in cherry trees by 25%. No difference in natural enemy abundance, richness or pest control was recorded between the two wildflower management regimes. Differences in natural enemy abundance and predation rates were detected despite the continued use of pesticides by growers (an average of 5.4 (± 0.4) applications per orchard per year). This study demonstrates that creating wildflower habitat in commercial sweet cherry orchards under polytunnels can boost natural enemies and the associated pest regulation services. Relative to CS, the novel grower-friendly approach of maintaining wildflower strips at a height of 20 cm

with regular cutting increased flower resource availability and pest regulation services, demonstrating the potential for growers to adopt this approach as part of a robust IPM strategy.

Keywords: bait cards, beneficial species, ecological intensification, ecosystem services, plant protection products, polytunnel

1. Introduction

Most modern sweet cherry (Prunus avium L.) production is highly intensive and, in some countries such as the UK and the USA, relies on polytunnels (plastic covers) to protect fruit from damage and disease (Lang, 2014; Bujdoso and Hrotko, 2017). However, the use of covers may enhance pests such as Panonychus ulmi (Lang, 2014), and Tetranychus urticae which damage leaves, reducing cherry production (Papadopoulos et al., 2017). Sweet cherry is also affected by other arthropod pests, which directly attack fruits causing up to 100% yield losses if left untreated (Daniel and Grunder, 2012). Key cherry pests include Myzus cerasi, Drosophila suzukii (Papadopoulos et al., 2017), and foliar feeding caterpillars (Alford, 2007). Growers are therefore heavily reliant on plant protection products to maintain commercially viable yields (Shaw et al., 2019b). However, some products are associated with negative effects on human health (Lamichhane, 2017), the environment, and beneficial arthropods (e.g. natural enemies and pollinators) (Geiger et al., 2010; Bonner and Alavanja, 2017; Lamichhane, 2017). As a consequence, in recent years, some pesticides have been withdrawn from use (e.g. thiacloprid for aphid control), whilst others (e.g. spinosad for D. suzukii control) rely on annual emergency approvals. The chemical pest control options available to cherry growers is becoming increasingly limited. Alternative and more sustainable approaches to sweet cherry production as part of Integrated Pest Management (IPM) are therefore demanded by the sector.

Natural enemies, including predators and parasitoids (Cross *et al.*, 1999; Solomon *et al.*, 2000), can provide high levels of pest regulation services (biological control) as part of an effective IPM strategy, reducing pest pressure and fruit damage (Papadopoulos *et al.*, 2017). However, they can only be abundant and diverse in cropped areas if supported through the provision of refuges and food resources (e.g. alternative prey, nectar, and pollen) (Wäckers and van Rijn, 2012). For example, some species of Araneae, Coleoptera, Dermaptera, Diptera (hoverfly larvae), Hemiptera, Neuroptera, Opiliones, Trombidiformes, and Hymenoptera (parasitoid wasps) contribute to pest regulation (Drummond *et al.*, 2010; Schüepp *et al.*, 2014; Rosas-Ramos *et al.*, 2020), and can all be supported through the provision of suitable habitat. This Conservation Biological Control (CBC) approach to IPM can therefore underpin the sustainable production of food (Begg *et al.*, 2017).

Introducing wildflower habitat as part of an IPM strategy can increase natural enemy abundance and the associated pest regulation services in a range of crops including apple (Campbell *et al.*, 2017; McKerchar *et al.*, 2020), blueberry (Blaauw and Isaacs, 2015), and wheat (Woodcock *et al.*, 2016; Hatt *et al.*, 2017). Wildflower interventions, employing native species adapted to local environments, can boost the abundance of beneficial arthropods, and the use of perennial species delivers greater consistency in floral resources throughout the year and between years (Isaacs *et al.*, 2009). Deploying native perennial wildflower habitat in sweet cherry orchards has the potential to support natural enemies and reduce the incidence of pests, reduce the need for pesticides, and increase crop yields (Poveda *et al.*, 2008; Redlich *et al.*, 2018; McKerchar *et al.*, 2020). However, there is concern that wildflower areas can harbour disease and be difficult to manage in cropped areas (Kleijn *et al.*, 2019).

Previously, wildflower strips have not been established in crops grown in polytunnels, and wildflower strips have never been actively managed throughout the growing season to deliver ecosystem services that underpin food production. Shorter wildflower strips rather than standard "tall" strips in alleyways is likely to facilitate the movement of workers when carrying

out orchard management tasks (e.g. pruning), minimising grower concerns (Kleijn *et al.*, 2019). The aim of this study was therefore to investigate the influence of cutting regime and the impact of wildflower strips established in polytunnels between rows of trees in commercial sweet cherry orchards on: 1) predator and parasitoid wasp abundance, richness, and diversity, and 2) pest regulation services. We hypothesized that the presence of wildflower strips in commercial sweet cherry orchards under polytunnels would increase natural enemy abundance and richness and the associated pest control services compared to unsown control strips. We also hypothesized that responses would not differ between wildflower cutting regimes.

2. Material and methods

2.1 Study site and experimental design

This three-year study (2017-2019) was carried out in the West Midlands, UK, at three sites in Herefordshire ($52^{\circ}10'46.7"N$, $3^{\circ}05'22.2"W$; $52^{\circ}11'25.6"N$, $2^{\circ}56'53.2"W$; and $52^{\circ}09'37.1"N$, $2^{\circ}35'38.2"W$) and one in Staffordshire ($52^{\circ}45'59.9"N$, $2^{\circ}09'48.3"W$). At each site, two orchards (defined as a separate parcel of land) were investigated. All orchards were managed intensively for commercial production and used polytunnel covers from April/May to late September. Orchards varied in size from 1.3 to 7.5 ha (mean 3.3 (\pm 0.7) ha) and were made up of adjacent rows of mixed compatible sweet cherry varieties. This study focussed on the economically important cultivar Kordia (Quero-García *et al.*, 2017). In each orchard, three Kordia rows were selected and alleyways adjacent to two of the tree rows received the random allocation of one of two wildflower treatments, whilst the third was a control, which consisted of the original alleyway vegetation. Control alleyways were dominated by grass species but included unsown wildflower species such as *Rumex obtusifolius*, *Taraxacum officinale*, and *Trifolium repens*. The three alleyway treatments were therefore:

- i) Control Strips (CS). Conventionally managed alleyways not sown with wildflowers that were cut regularly to a height of 10 cm from May to September, and then to a height of 8 cm in late September.
- ii) Standard Wildflower Strips (SWS). Cut annually in late September to a height of 8 cm.
- iii) Actively Managed Wildflower Strips (AMWS). Cut regularly (twice/three times per month) to a height of 20 cm from May to September, and then to a height of 8 cm in late September.

Alleyways were sown in October 2016 but due to poor establishment were re-sown in 2017 (Herefordshire sites in spring, and the Staffordshire site in autumn). Prior to sowing, alleyways were sprayed with glyphosate and cultivated to create a fine seed bed (Mateos-Fierro *et al.,* 2018). Seeds were mixed with sand to ensure even sowing by hand, after which strips were rolled. Sown species and sowing rates are provided in Table 1. Wildflower seeds were purchased from <u>www.wildflowersuk.com</u> and <u>www.wildseed.co.uk</u>.

Alleyways were 2 m wide (mean) and wildflower interventions were established in the central 1 m strip to avoid damage and soil compaction from vehicle movement. The strips were 95 m long, beginning at the edge of an orchard towards the centre. Based on the availability of Kordia rows in orchards, the distance between alleyway treatments varied from 26 to 43.5 m (mean 37.5 (\pm 2.7) m). The distance between orchards within each site also varied from 30 to 975 m (mean 478 (\pm 180.0) m). The typical landscape context of the sites was the domination of improved grasslands (46.1% (\pm 5.2)) and arable and horticulture areas (44.9% (\pm 5.9)), but also some broadleaved woodlands (5.5% (\pm 1.8)), suburban areas (2.0% (\pm 0.8)), and other land uses (1.5% (\pm 0.7)) were present (land cover broad habitat classes according to Land Cover Map 2015 (Rowland *et al.*, 2017)).

For data collection, experimental rows were split into five sections and assessments in alleyway vegetation and cherry trees were carried out in the centre of sections 1-4 (Figure 1). The fifth section acted as a buffer to the opposite edge due to five of the 30 alleyways being 95 m in length.

During the establishment year (2017), the wildflower treatments were managed with regular cutting to a height of 10 cm to prevent flowering and promote establishment of the sown species. This provided a baseline year for data collection.

2.2 Floral resource availability

In 2018 and 2019, the availability of floral resources for natural enemies was determined by recording floral unit abundance (single flower e.g. *Silene dioica* or flower cluster e.g. *Leucanthemum vulgare* (Carvell *et al.,* 2015)) in ten quadrats (0.5 x 0.5 m) randomly distributed along orchard alleyways. Floral units were recorded for each plant species (sown and unsown) in each alleyway treatment. Assessments were conducted once per month from June to September in 2018 and from June to August in 2019.

2.3 Natural enemies in alleyways

Data were collected for three consecutive years (2017-2019). In 2017, sampling was conducted twice per month (every two weeks) from July to September, whilst in 2018-19, sampling was done once per month, from June to September and from May to August, respectively.

The abundance and richness of predators and abundance of parasitoid wasps in the orchard alleyways was investigated using direct searches combined with VortisTM suction sampling (Burkard Manufacturing Company Ltd, UK) (Brook *et al.*, 2008). In each section (1-4), direct searches involved recording natural enemies observed during a two-minute active search over a 0.5 x 0.5 m area. Vortis suction sampling consisted of 15, 10-second suction samples over the same area.

Predators were identified to family, whilst parasitoid wasps were not identified further. Only predatory species within each taxonomic group were considered since some species are non-zoophagous (do not feed on animals) (e.g. *Psyllobora vigintiduopunctata*, Coleoptera: Coccinellidae). Additionally, the species with different feeding behaviours during their life cycle were only recorded during the zoophagous stage (e.g. hoverfly larvae). Formicidae (ants) were recorded but the majority were *Lasius niger*, an aphid-attending species (Stutz and Entling, 2011). Ant data were therefore not included in further analyses.

2.4 Natural enemies in cherry trees

Natural enemies in cherry trees were sampled twice per month in 2017 from May to October, and once per month in 2018 and 2019, from May to October, and from May to August, respectively.

To investigate the occurrence of natural enemies (abundance and richness) in cherry trees, two complimentary techniques were used, direct search and beat sampling. The canopy section from the base (~1 m above the ground) to a height of ~2 m was assessed on the side of the tree that faced the alleyway treatment (e.g. wildflower strip). Direct search assessments were carried out on the middle tree of each section for two minutes (Woodcock *et al.,* 2016). Following the direct search, beat sampling was used with a 1 m PVC stick to tap five different

branches on each tree whilst holding a white plastic tray underneath (45 x 35 x 2.5 cm) (Miliczky and Horton, 2005). All arthropods were identified to family except for parasitoid wasps.

2.5 Aphid depletion

As the study orchards were managed for commercial production, pesticides were applied by growers throughout the study. A total of three acaricides and ten insecticides were used to control the main cherry arthropod pests (Table S1). Within acaricides, spirodiclofen accounted for 75% of the total number of applications. Indoxacarb, spirotetramat, acetamiprid and cyazypyr (19.5%, 15.9%, 14.2% and 14.2%, respectively) were applied to control insect pests. The number of applications ranged from three to ten per orchard per year, with a mean of 5.4 (\pm 0.4). Each year, spray programmes started in March, prior to the cherry blossom period, and continued through to July before harvest.

Colonies of *M. cerasi* were monitored by direct search on the side of cherry trees that faced the alleyway treatments in 2017 and 2018, whilst to detect *D. suzukii* larval presence in fruit at the end of June and mid-July, two sugar floatation tests were carried out in 2017 (Shaw *et al.*, 2019a). Abundances of *M. cerasi* and *D. suzukii* were very low and thus insufficient to measure differences between treatments (results not presented). Consequently, to assess pest regulation services, in 2019, sentinel aphid bait cards were used to measure the predator/scavenger activity of natural enemies (Geiger *et al.*, 2010; McKerchar *et al.*, 2020). PVA glue (Pritt PVA Craft Glue) and white PVC cards (760 Micron, CR80) were used. Cards were freshly prepared on the day of deployment (around 12:00 hrs). Aphids were frozen before being glued. In total, ten adult and late stage nymphs (third and fourth) of *Acyrthosiphon pisum* (pea aphid) were glued to cards by their rear legs or abdominal sternum (Figure 2A).

Eight trees were selected adjacent to each alleyway treatment at approximately 5, 14, 24, 33, 43, 52, 62, and 71 m from the orchard edge. One bait card per tree was attached to the inner part of the tree with 2 mm wide, black, cable ties approximately 1.8 m above the ground (Figure 2B).

Three rounds of bait cards were deployed during the summer (June, July, and August), corresponding with the highest predicted arthropod activity (Bradshaw and Holzapfel, 2010). The number of aphids depleted was determined every 24 h for five days (Figure 2C). The number of aphids dried and shrunken and therefore not available to natural enemies were also recorded and excluded from analysis.

2.6 Statistical analysis

Data were analysed with generalized linear mixed effect models using the package Ime4 (Bates *et al.*, 2014) in R (version R-3.6.1) (R Core Team, 2019). Tukey *post-hoc* tests were used to investigate pair-wise differences between fixed factors (multcomp package (Hothorn *et al.*, 2008)).

2.6.1 Floral resource availability

Floral units were analysed with a generalized linear mixed effect model with negative binomial error structures (function = GLMER.NB). Alleyway treatment and year were set as fixed factors, whilst quadrats nested within orchards nested within sites were defined as random effects (*Floral units ~ Alleyway treatment + Year + (random: Site/Orchard/Quadrat*).

To provide an overall response of natural enemies to alleyway treatment, data obtained from the alleyway vegetation and cherry trees were combined for each section surveyed (i.e. data from direct search and Vortis sampling were combined for alleyways, and data from direct search and beat sampling were combined for cherry trees). Shannon diversity values were calculated for each section in each habitat (alleyways and trees) based on the natural enemy families recorded.

To investigate responses in natural enemy abundance, family richness and Shannon diversity, generalized linear mixed effect models with negative binomial error structures (function = GLMER.NB) were used. Natural enemy abundance, family richness and Shannon diversity were therefore the response variables in the three separate models for each habitat (six models in total). As year was expected to influence the response of natural enemies due to wildflower cutting treatments not being applied in 2017, it was included as a fixed effect, with an interaction term with alleyway treatment. Orchards nested within sites were specified as a random effects. The interaction term was studied by comparing two models (with and without interaction between treatment and year), for each of the three response variables (natural enemy abundance, family richness and Shannon diversity) for each habitat (alleyways and trees), using Akaike's Information Criterion (AIC). The model with the lowest AIC was deemed as the most parsimonious in each of the six cases (Burnham and Anderson, 2002).

The influence of each of the fixed terms was obtained using the AIC. Fixed factors were individually removed from the models (global models) and a difference of AIC was calculated (Δ AIC). AIC > 2 was accepted to empirically support significance (Burnham and Anderson, 2002).

2.6.3 Aphid depletion

Aphids partially eaten or removed from bait cards were considered depleted. Values of depletion were calculated by subtracting the number of complete aphids remaining on the cards from their initial number. To investigate differences in aphid depletion according to alleyway treatment, a generalized linear mixed effect model was used with the difference in the number of aphids depleted/not depleted as the response variable (function = GLMER, family = binomial). To investigate the influence of alleyway treatment on values for each survey round, month surveyed was considered in the model as fixed effect. In addition, the interaction between alleyway treatment and month was also studied. Orchards nested within sites were set as random effects.

3. Results

3.1 Floral resource availability

The mean number of floral units recorded per quadrat in 2018 and 2019 was greater in the Standard Wildflower Strips (SWS) (Tukey test: Z = 8.74, P < 0.001) and Actively Managed Wildflower Strips (AMWS) (Tukey test: Z = 7.22, P < 0.001) compared to the Control Strips (CS) (Figure 3). There were 554% more floral units associated with the SWS treatment compared to CS, whilst the increase of floral units associated with AMWS was 365% greater than with CS. No significant difference was found between wildflower treatments.

3.2 Abundance of natural enemies

3.2.1 In alleyways

A total of 6,635 natural enemies were recorded during the direct search and Vortis sampling between 2017-19 in the orchard alleyways (Table 2). Of those, 89.9% were recorded by Vortis sampling and the remainder (10.1%) by direct search. The most abundant groups were Araneae and parasitoid wasps, followed by Coleoptera. The sum of these three groups accounted for 84.8% of the total natural enemies recorded in the alleyway vegetation.

Araneae included seven families; Linyphiidae with 84.8% relative abundance, Theridiidae 9.0%, Lycosidae 4.1%, and Araneidae 1.1%. Clubionidae, Thomisidae, and Tetragnathidae were recorded less frequently, with 0.6%, 0.3%, and 0.2%, respectively. Three Coleoptera families were identified; Staphylinidae (63.3%), Carabidae (30.5%), and Coccinellidae (6.2%). Hemiptera, Neuroptera, and Opiliones included two families each. Anthocoridae constituted the highest percentage of Hemiptera (91.8%), whilst Nabidae accounted for 8.2%. Within Neuroptera, Hemerobiidae accounted for 95.2%, compared to only 4.8% for Chrysopidae. The majority of Opiliones recorded belonged to Phalangiidae (99.8%), and only 0.2% were Leiobunidae.

3.2.2 In cherry trees

A total of 6,783 individual arthropods were recorded during direct search and beat sampling of cherry trees over the three-year period (Table 2). Of those, 93.1% were recorded by beat sampling, and 6.9% through direct search. The most abundant groups were Anystidae and Araneae followed by Opiliones. Anystidae accounted for half of the total records, whilst these three groups summed 83.2% of the total natural enemy records.

Araneae was represented by ten families. The most frequent being Linyphiidae (relative abundance; 34.4%), Theridiidae (29.8%), and Araneidae (26.5%). Together, they composed 90.6% of the total Araneae records. Thomisidae accounted for 5.6%, whilst Philodromidae and Tetragnathidae had 2.1% and 1.1%, respectively. Four families contributed less than 1%

of the total Araneae abundance, including Clubionidae, Dictynidae, Metidae, and Salticidae (0.4%, 0.1%, 0.1%, and 0.1%, respectively). Three natural enemy families were recorded within Coleoptera; Coccinellidae 76.8%, Staphylinidae (21.1%), and Carabidae 2.1%. Two families were recorded for Neuroptera and Opiliones. Chrysopidae made up the majority of Neuroptera (96.9%) with only 3.1% Hemerobiidae. Likewise, the abundance of Opiliones was dominated by Phalangiidae (99.0%), with only 1.0% Leiobunidae. All Hemiptera were Anthocoridae and no Nabidae were recorded in the cherry trees.

3.3 The influence of alleyway treatment on natural enemies

3.3.1 In alleyways

The response of total natural enemies to alleyway treatment was not consistent between years (Table 3). In 2017 (establishment year), all three alleyway treatments had similar values of natural enemy abundance, however, by 2018 both wildflower treatments, SWS and AMWS, were associated with a greater abundance of natural enemies compared to the CS treatment (Figure 4B; Table 3). In 2019, the difference was even greater. The *post-hoc* test revealed that total natural enemy abundance was greater in SWS (Tukey test: Z = 6.34, P < 0.001) and AMWS (Tukey test: Z = 8.22, P < 0.001) compared to CS, but there was no significant difference between wildflower treatments (Figure 4A).

Family richness was also influenced by the interaction between alleyway treatment and year but Shannon diversity was not (Table 3). A greater family richness was recorded in SWS (Tukey test: Z = 4.79, P < 0.001) and AMWS (Tukey test: Z = 6.18, P < 0.001) compared to CS (Figure 5A). Similarly, higher Shannon diversity was recorded in SWS (Tukey test: Z = 3.55, P < 0.01) and AMWS (Tukey test: Z = 3.64, P < 0.001) compared to CS (Figure 5B). In both cases, there was no difference between wildflower treatments.

3.3.2 In cherry trees

A greater number of natural enemies were found in cherry trees adjacent to the wildflower treatments, with the most parsimonious model including the interaction, although the difference between AIC was lower than 2 (Table 3). Total natural enemy abundance was greater in 2018 and 2019 under both wildflower treatments compared to CS (Figure 4D, Table 3). SWS (Tukey test: Z = 2.96, P < 0.01) and AMWS (Tukey test: Z = 3.05, P < 0.01) were associated with significantly more natural enemies than CS (Figure 4C). During the three-year study, there were no significant differences in total natural enemy abundance between SWS and AMWS.

There was a tendency for Family richness and Shannon diversity to be greater in wildflower treatments compared to CS, but this was not statically significant (Figures 5C, and 5D; Table 3). The response of family richness and Shannon diversity to alleyway treatment was constant between years (Table 3).

3.4 The influence of alleyway treatment on pest control (aphid depletion)

Overall, across the three rounds of bait cards deployed, aphid depletion was strongly influenced by alleyway treatment (GLMER: Δ AIC Alleyway treatment: Month surveyed = 25.3). A significantly greater depletion of aphids was recorded from cards deployed on trees adjacent to SWS (Tukey test: *Z* = 3.41, *P* < 0.01) and AMWS (Tukey test: *Z* = 5.04, *P* < 0.001) compared to CS (32.0% (± 2.4), 28.9% (± 2.5), and 24.3% (± 2.5), respectively). This was equivalent to an increased depletion of 31.9% with AMWS and 18.9% in SWS compared to CS (control as 100%).

Survey month was also shown to influence pest regulation services; a significantly greater number of aphids were depleted in AMWS and SWS compared to CS in June and July but not in August (Figure 6). There was a higher depletion rate in June (Tukey test: Z = -6.88, P < 0.001) and July (Tukey test: Z = -4.73, P < 0.001), compared to August; but not between June and July (Tukey test: Z = -2.23, P = 0.07).

4. Discussion

This three-year study has demonstrated, for the first-time, that by creating wildflower habitat in sweet cherry orchards managed intensively under polytunnels, natural enemies and their pest regulation services can be boosted. Importantly, maintaining wildflower strips at 20 cm by cutting regularly from May to September, not only increased the abundance of floral units compared to standard practice, they also improved levels of pest regulation which were comparable to wildflower strips not actively cut. This positive outcome was achieved despite the continued use of pesticides, demonstrating the potential for growers to adopt this approach as part of a robust IPM strategy. This novel wildflower strip management is also likely to increase uptake by growers as in contrast to standard wildflower strips, it will reduce any impacts on the delivery of pesticide sprays, facilitate the movement of growers and workers along alleyways, decrease the potential for punnets to be contaminated with plant debris during harvest, and reduce the potential for humidity to be increased.

4.1 The influence of the wildflower interventions on natural enemies

Both wildflower treatments (AMWS and SWS) enhanced predator abundance and richness, and parasitoid wasp abundance compared to alleyways managed conventionally (CS). Alleyways sown with wildflowers create a more complex semi-natural habitat compared to unsown control alleyways (Balzan *et al.*, 2014) and it is likely that in addition to the increased provision of pollen and nectar, the wildflower strips also provided alternative prey and shelter for natural enemies (Blaauw and Isaacs, 2012; Campbell *et al.*, 2017). The benefits of providing wildflower strips to boost the abundance of natural enemies in sweet cherry orchards was more apparent in the alleyway vegetation than in the adjacent cherry trees. However, wildflowers serve the purpose of attracting natural enemies into orchards which then spill over to the cherry trees (Woodcock *et al.*, 2016).

The impact of the perennial wildflower strips on natural enemy abundance and richness was consistent across both wildflower management treatments. From 2018, sown species dominated these alleyways and were able to flower under the management treatments, compared to CS, which were dominated by grass species. Consequently, the abundance and richness of natural enemies in alleyways and cherry trees was increased in AMWS and SWS. The similar values of natural enemy abundance, richness and diversity observed with the AMWS and SWS treatments suggests that wildflower strips can be actively managed without significant negative impacts on natural enemies and their pest regulation services. This could bring benefits to growers and workers by allowing easier movement along alleyways for management activities such as pruning and monitoring pests. We have therefore developed a management approach aligned more closely to current grower practice whilst delivering benefits that underpin sustainable production. Compared to other wildflower options (Kleijn *et al.,* 2019), barriers to the uptake of actively managed wildflower interventions are therefore likely to be minimal.

4.2 The influence of wildflower interventions on pest regulation services

The greater abundance, richness, and diversity of natural enemies associated with wildflower habitats is expected to have underpinned the observed higher depletion rates in the aphid baited cards (pest regulation services), as found in other studies (Marc and Canard, 1997; Blaauw and Isaacs, 2015; Campbell *et al.*, 2017; Dainese *et al.*, 2017). Greater natural enemy

diversity can provide a more effective and resilient pest regulation service, since different natural enemies can attack the same (Dainese *et al.*, 2017), or different pests (Marc and Canard, 1997). Consequently, the greater depletion of aphids (25.4%) with wildflower strips compared to CS has clearly demonstrated that wildflower strips in sweet cherry orchards should be considered as part of an IPM programme to increase pest control. In turn, this could reduce the need for growers to use pesticides (Hatt *et al.*, 2017), which can otherwise mask responses (McKerchar *et al.*, 2020). In this study, natural enemies were enhanced in wildflower treatments despite the continued use of pesticides. Reducing pesticide inputs in sweet cherry orchards might further benefit natural enemies and result in better pest regulation services, bringing positive outcomes for CBC as part of IPM programmes.

4.3 Response of natural enemy taxonomic groups to the wildflower interventions

Predatory mites (Anystidae) were the most frequently recorded natural enemy in cherry trees and enhanced pest regulation services (depletion from bait cards) could have been provided by this group of predators. *Anystis baccarum*, a cosmopolitan generalist species has shown to provide important pest control in UK apple orchards (Cuthbertson *et al.*, 2014). The greater depletion of aphids on trees adjacent to wildflower treatments may also have been the result of enhanced natural enemies such as Anthocoridae, Chrysopidae, Coccinellidae, and Phalangiidae, which were in greater abundance in wildflower strips compared to CS. Araneae (spiders) have been identified as important natural enemies in the orchards, but they do not usually scavenge (Harwood and Obrycki, 2005). Consequently, whilst it is unlikely they contributed to the depletion of aphids from bait cards, they might be expected to provide pest regulation services not directly measured in this study (Bogya, 1999).

Araneae benefited most from wildflowers, as they were associated with a greater abundance and family richness with these treatments, most likely due to their dependence on natural habitats (Schmidt and Tscharntke, 2005; Schüepp *et al.*, 2014). Araneae are heterogeneous generalist predators with a wide range of hunting behaviours (Bogya, 1999; Solomon *et al.*, 2000). Of the total 11 families identified in alleyways and cherry trees, Linyphiidae, Theridiidae, and Araneidae were the most abundant on trees, which is consistent with previous studies in UK apple orchards (Chant, 1956; McKerchar *et al.*, 2020). Individuals of these families use webs to catch prey; whilst Lycosidae, a ground-dwelling spider only recorded in alleyway vegetation, is an active predator (Solomon *et al.*, 2000). Other less abundant families such as Philodromidae, Clubionidae, and Salticidae are also active predators (Solomon *et al.*, 2000).

When pests are scarce alternative resources are essential to enhance and maintain natural enemies in orchards (Wäckers and van Rijn, 2012; Blaauw and Isaacs, 2015). Wildflower habitats provide alternative prey, pollen (protein), nectar (sugar), and shelter (Wäckers and van Rijn, 2012; Blaauw and Isaacs, 2015), explaining the increased abundance of Coleoptera, Hemiptera, Neuroptera, Formicidae, Opiliones, and parasitic wasps in association with wildflower strips compared to CS. Generalist natural enemies (e.g. Anystidae, Araneae, Coleoptera, and Opiliones) were more abundant than specialist groups (e.g. parasitoid wasps, and Syrphidae) (Balzan *et al.*, 2014), as they may be able to survive feeding on alternative prey (Solomon *et al.*, 2000; Harwood and Obrycki, 2005; Drummond *et al.*, 2010). Generalists are important in orchards as they provide a wide range of pest regulation services (Marc and Canard, 1997; Cuthbertson *et al.*, 2014; Schüepp *et al.*, 2014).

5. Conclusions

This study demonstrated, for the first-time, that wildflower interventions in sweet cherry orchards can enhance natural enemies, leading to an increase in pest regulation services under polytunnel systems. The novel approach of maintaining wildflower strips in alleyways to a height of 20 cm throughout the growing season resulted in similar pest regulation services to standard wildflower strips and is therefore more likely to be adopted by growers. The

response of natural enemies to wildflower strips and their ability to provide pest regulation services could be further enhanced if implemented on a larger scale; especially when combined with an IPM regime where pesticide inputs are reduced. This should be the focus of further research.

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Figures

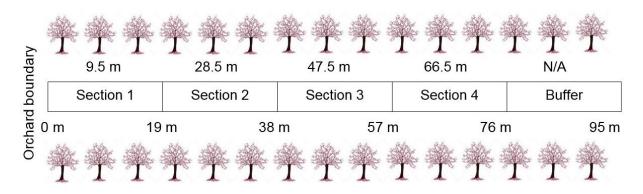


Figure 1. Division of the alleyways into four sections for sampling, including the buffer (not assessed). Top values give the location of the mid-section points and cherry trees and bottom values give the end of the sections by distance from the orchard boundary (m).



Figure 2. A) Bait card with ten dead aphids glued to the surface. B) Bait card deployed ~ 2 m above the ground in cherry tree canopy (highlighted with a red circle). C) Bait card with all ten aphids removed.

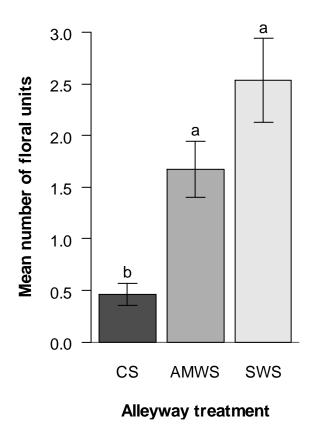


Figure 3. Mean number (\pm SE) of floral units per quadrat (0.5 x 0.5 m) according to alleyway treatment across all surveys and years (2017-19). The same superscript letters indicate no significant difference according to the Tukey test (P > 0.05). CS (Control Strips), AMWS (Actively Managed Wildflower Strips), SWS (Standard Wildflower Strips).

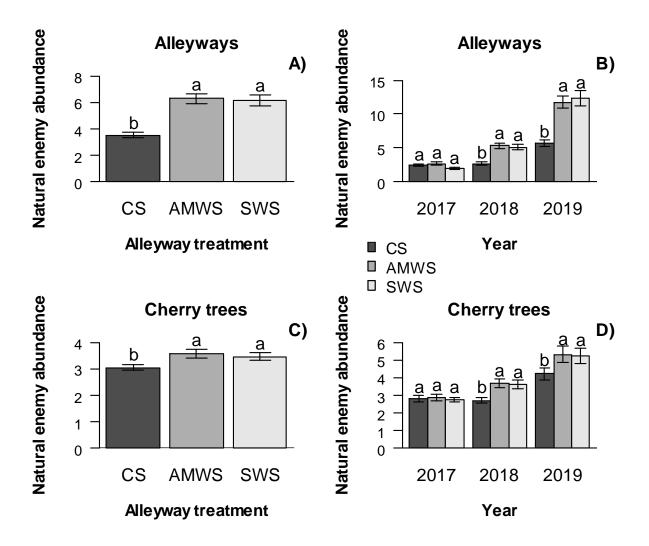


Figure 4. Mean numbers (\pm SE) of natural enemy abundance per section recorded throughout the three-year study according to A) and C) alleyway treatment, and B) and D) year and alleyway treatment in alleyways and cherry trees. The same superscript letters indicate no significant difference (Tukey test, *P* > 0.05); for each category (year) in B) and D). CS (Control Strips). AMWS (Actively Managed Wildflower Strips), SWS (Standard Wildflower Strips).

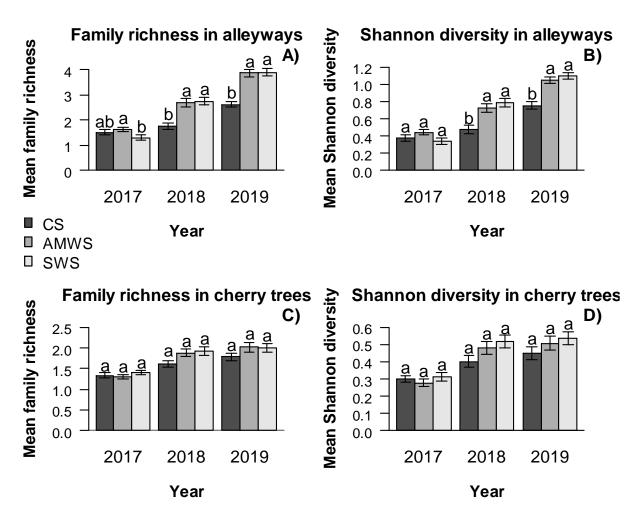


Figure 5. Mean values (\pm SE) of family richness (A, C) and Shannon diversity (B, D) of natural enemies per section in alleyways and cherry trees according to year and alleyway treatment. The same superscript letters for each category (year) indicate no significant difference (Tukey test, *P* > 0.05). CS (Control Strips). AMWS (Actively Managed Wildflower Strips), SWS (Standard Wildflower Strips).

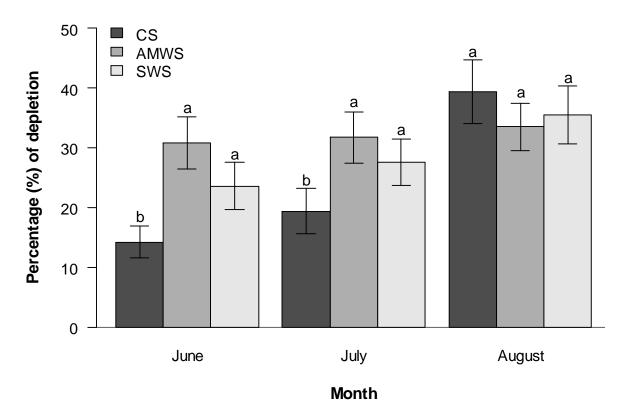


Figure 6. Mean percentage (\pm SE) of *Acyrthosiphon pisum* aphids (dead) depleted from bait cards placed in cherry trees according to month and alleyway treatment. The same superscript letters indicate no significant difference for each category (month) (Tukey test, *P* > 0.05). CS (Control Strips). AMWS (Actively Managed Wildflower Strips), SWS (Standard Wildflower Strips).

Tables

Table 1. Seed mix composition and sowing rate used to establish the wildflower strips. w.t. (wild type). Total sowing rate = 1.97 g m^{-2} .

Scientific name	Common name	Sowing rate	Sowing rate	% by
		(seeds m²)	(g m²)	weight
Achillea millefolium	Yarrow	200	0.033	1.69
Centaurea nigra	Knapweed	200	0.444	22.60
Dactylis glomerata (w.t.)	Cock's-foot	100	0.100	5.10
Leontodon hispidus	Rough hawkbit	200	0.222	11.30
Leucanthemum vulgare	Ox-eye daisy	200	0.100	5.08
Lotus corniculatus (w.t.)	Bird's-foot trefoil	200	0.400	20.34
Prunella vulgaris	Selfheal	200	0.200	10.17
Silene dioica	Red campion	200	0.200	10.17
Trifolium pratense (w.t.)	Red clover	200	0.267	13.56

Table 2. Numbers of arthropods, percentage abundance, and the mean (\pm SE) per alleyway (all four sections) across all years (2017-19).

Taxonomic	Alleyway			Cherry tree			
group	Number of	Percentage	Mean (±	Number of	Percentage	Mean (±	
	individuals	abundance (%)	SE)	SE) individuals abundance		SE)	
Anystidae	-	-		3,512	51.8	7.0 (± 0.4)	
Araneae	2,652	40.0	8.5 (± 0.8)	1,408	20.8	2.8 (± 0.1)	
Parasitoid	1 500	22.7	4.9.(.0.4)	216	47	06(+01)	
wasps	1,509	22.7	4.8 (± 0.4)	316	4.7	0.6 (± 0.1)	
Coleoptera	1,463	22.0	4.7 (± 0.3)	95	1.4	0.2 (± 0.03)	
Opiliones	444	6.7	1.4 (± 0.2)	726	10.7	1.4 (± 0.1)	
Hemiptera	305	4.6	1.0 (± 0.1)	419	6.2	0.8 (± 0.1)	
Neuroptera	187	2.8	0.6 (± 0.1)	131	1.9	0.3 (± 0.04)	
Syrphidae	75	1.1	0.2 (± 0.1)	166	2.4	0.3 (± 0.1)	
Forficulidae	-	-		10	0.1	0.02 (±0.01)	

Table 3. Comparisons in the generalized linear mixed models with and without interaction between alleyway treatment and year for the natural enemies in alleyways and cherry trees using the Akaike Information Criterion (AIC). Models with the lowest AIC (in italics) for each pair of response variable was deemed as the most parsimonious. Interaction between alleyway treatment and year represented by *Alleyway treatment: Year*. Fixed factors (explanatory variables) are removed in each reduced model to determine significant differences. Models include degrees of freedom, and the difference between models (Δ AIC). Δ AIC > 2 was accepted to be significantly different. Values in bold are significant.

Habitat studied	Response	Model	Explanatory variables	Degrees of	AIC	ΔΑΙC
	variable		(fixed factors)	freedom		
Natural enemies	Total natural	Total number of natural enemies ~ Alleyway	Global model		6,287.8	0.0
in alleyways	enemies	treatment: Year + (random: Site/Orchard)			,	
			Alleyway treatment: Year	4	6,339.4	51.6
		Total number of natural enemies ~ Alleyway treatment + Year + (random: Site/Orchard)	Global model		6,338.4	0.0
			Alleyway treatment	2	6,401.4	63.0
			Year	2	6,886.0	547.6

E e secilita	Number of natural enemy families ~				
Family	Alleyway treatment: Year + (random:	Global model		4,303.2	0.0
richness	Site/Orchard)				
		Alleyway treatment: Year	4	4,322.2	19.0
	Number of natural enemy families ~				
	Alleyway treatment + Year + (random:	Global model		4,322.2	0.0
	Site/Orchard)				
		Alleyway treatment	2	4,371.8	49.6
		Year	2	4,670.8	348.6
		Year	2	4,670.8	348.6
Shannon	Shannon diversity value ~ Alleyway treatment: Year + (random: Site/Orchard)	Year Global model	2	4,670.8 2,253.3	348.6 0.0
Shannon diversity	Shannon diversity value ~ Alleyway treatment: Year + (random: Site/Orchard)		2		
		Global model		2,253.3	0.0

			Year	2	2,358.8	107.0
Natural enemies in cherry trees	Total natural enemies	Total number of natural enemies ~ Alleyway treatment: Year + (random: Site/Orchard)	<i>Global model</i> Alleyway treatment: Year	4	<i>9,286.0</i> 9,286.4	0.0 0.4
		Total number of natural enemies ~ Alleyway treatment + Year + (random: Site/Orchard)	<i>Global model</i> Alleyway treatment Year	2 2	9,286.4 9,290.1 9,379.6	0.0 3.7 93.2
	Family richness	Number of natural enemy families ~ Alleyway treatment: Year + (random: Site/Orchard)	<i>Global model</i> Alleyway treatment: Year	4	6,064.8	0.0

	Number of natural enemy families ~				
	Alleyway treatment + Year + (random:	Global model		6,060.5	0.0
	Site/Orchard)				
		Alleyway treatment	2	6,062.1	1.6
		Year	2	6,141.4	80.9
Shannon diversity	Shannon diversity value ~ Alleyway treatment: Year + (random: Site/Orchard)	Global model		2,904.2	0.0
		Alleyway treatment: Year	4	2,898.2	-6.0
	Shannon diversity value ~ Alleyway treatment + Year + (random: Site/Orchard)	Global model		2,898.2	0.0
		Alleyway treatment	2	2,896.9	-1.3
		Year	2	2,938.1	39.9

1 Supplementary Table

- 2
- 3 Table S1. Mean number (± SE) of spray applications of insecticides and acaricides per orchard
- 4 and per year, and the percentage of application of the pesticide type.

Pesticide type	Active ingredient	Mean number of	% application
resticide type	Active ingredient	applications	pesticide type
Acaricide	Etoxazole	0.1 (± 0.1)	12.5
Acaricide	Maltodextrin	0.5 (± 0.1)	12.5
Acaricide	Spirodiclofen	0.1 (± 0.1)	75.0
Insecticide	Acetamiprid	0.7 (± 0.1)	14.2
Insecticide	Bifenthrin	0.3 (± 0.2)	5.3
Insecticide	Cyazypyr	0.7 (± 0.2)	14.2
Insecticide	Indoxacarb	0.9 (± 0.1)	19.5
Insecticide	Lambda-cyhalothrin	0.04 (± 0.04)	0.9
Insecticide	Pirimicarb	0.1 (± 0.1)	1.7
Insecticide	Pyrethrin	0.2 (± 0.1)	3.5
Insecticide	Spinosad	0.6 (± 0.1)	12.4
Insecticide	Spirotetramat	0.8 (± 0.2)	15.9
Insecticide	Thiacloprid	0.6 (± 0.1)	12.4

5