



Sugar feeding by parasitoids inside and around vineyards varies with season and weed management practice

Miriam Kishinevsky^{a,*}, Tamar Keasar^b

^a Department of Evolutionary and Environmental Biology, University of Haifa, Haifa, Israel

^b Department of Biology and Environment, University of Haifa - Oranim, Tivon, Israel

ARTICLE INFO

Keywords:

Anthrone
Parasitoids
Seasonality
Sugar feeding
Weed management

ABSTRACT

Feeding on sugar-rich resources dramatically prolongs lifespan and increases foraging performance in most parasitoids. The physiological and fitness effects of sugar feeding were studied in many parasitoid species under lab conditions, but far fewer studies examined them in the field. Even less is known about the environmental conditions that affect the prevalence of sugar feeding in natural parasitoid communities. Such information can guide planning of suitable habitats for these important biological control agents. We compared sugar feeding frequencies in parasitoid communities along four seasons, in wine-producing vineyards and in nearby natural plots, using qualitative anthrone tests. We also tested how the vineyards' weed management practice (mechanical trimming or herbicide application) affected the parasitoids' sugar feeding rates. Sugar feeding was significantly more common in the natural areas compared to the vineyards in all seasons. Sugar feeding rates were highest during spring and lowest in the winter. They were not consistently affected by weed management treatment, but increased with the number of flowering plant species, especially in spring. Our results suggest that parasitoids are limited by sugar sources in agricultural plots even when undisturbed by farming activities (during winter in our case), and that 'softer' weed management treatments do not necessarily improve their prospects for sugar feeding. A more targeted approach, of planting or preserving a higher richness of flowering plants throughout the seasons, may provide parasitoids with more sugar sources and thus support them in agricultural plots.

1. Introduction

Agricultural systems are usually considered to be low in resources for natural enemies. Supplementing these resources can potentially enhance natural enemies' abundance and activity and thereby reduce damage caused by insect pests (Heimpel and Jervis, 2005; Rusch et al., 2017). Parasitoids are insects that lay their eggs in or on the body of another insect host, eventually killing it. They are important biological control agents but sometimes fail to reduce pest populations. Multiple studies suggest that the main reason for such failures is lack of resources, such as sugar or shelter, for the parasitoids' adult stage (Fiedler et al., 2008; Parolin et al., 2012; Begg et al., 2017).

Numerous laboratory studies showed that sugar meals can dramatically prolong parasitoid longevity and fecundity (Fadamiro and Heimpel, 2001; Wratten et al., 2003; Lee et al., 2004; Wyckhuys et al., 2008; Nafziger and Fadamiro, 2011). Longer-lived parasitoids are expected to disperse better, and to find and parasitize more hosts under field

conditions as well (Heimpel and Jervis, 2005). Jervis et al. (1993) observed a variety of plant species in natural and semi-natural areas and showed that parasitoids from different families actively visit flowers to feed on their nectar. Later studies found that extrafloral nectar and honeydew are also suitable sugar sources for parasitoids in the field (Géneau et al., 2012; Tena et al., 2016). Moreover, parasitoids may feed on honeydew produced by sap-feeding insects that are not necessarily their hosts (Evans and England, 1996; Tena et al., 2013). These findings support the importance of sugar feeding for adult parasitoids under field conditions.

Parasitoids may move from agricultural to natural areas to feed on floral nectar and return to the agricultural field once fed, sometimes covering considerable distances. For example, the parasitoid wasp *Dia-degma semiclausum* (Ichneumonidae) moved for up to 80 m after a sugar meal (Lavandero et al., 2005). Thus, spontaneous or planted flower-rich strips at field edges can potentially support parasitoids and their pest control services within the fields. To test this prediction, field studies

* Corresponding author at: Department of Integrative Biology, UW-Madison, 458 Birge Hall, Madison, WI, United States.

E-mail address: mashakish@gmail.com (M. Kishinevsky).

investigated how supplementary flower strips affected parasitoids in neighboring agricultural plots (Tylianakis et al., 2004; Bianchi and Wäckers, 2008). Lee and Heimpel (2008) showed that parasitoid sugar feeding rates are higher at the borders of cabbage fields, close to a flowering strip of buckwheat plants, than in no-flower control plots. However, they observed no difference in feeding rates between the two types of plots in the middle of the fields. In other studies, adding flower strips did not always improve parasitoid performance (Lee et al., 2006; Lee and Heimpel, 2008). Heimpel (2019) consequently suggested that parasitoids that feed on sugar disperse long distances away from the sugar source, and therefore provisioning of sugar sources at field edges does not necessarily benefit biological control within the agricultural plots.

Far fewer studies considered the effects of additional natural and man-made factors, other than flower strips and active provisioning of sucrose solution (Tena et al., 2015), on sugar feeding by parasitoids. Most notably, natural areas often contain a diverse complex of flowering plants, and are advocated as being rich in resources for natural enemies (Landis et al., 2000). Nevertheless, as far as we know, our previous study (Kishinevsky et al., 2018) provides the only estimate of the frequency of sugar feeding in parasitoid communities from non-crop habitats. Here, we therefore extend our previous work, and compare sugar feeding in natural and agricultural habitats of a vineyard agroecosystem. Sugar feeding may also vary with season. Segoli and Rosenheim (2013) found that *Anagrus* spp. (Mymaridae) from vineyards had generally low sugar feeding rates, which decreased along the months July-October. A similar trend was shown for *Aphytis melinus* (Aphelinidae), which had higher proportions of sugar-fed individuals during spring and summer than in autumn (Tena et al., 2013).

It is still unknown whether this seasonal trend also occurs in other parasitoids, and whether the frequency of sugar feeding shows a clear pattern over the whole year. Moreover, seasonal agricultural activities may also affect parasitoids' sugar feeding in crop habitats. For example, in our previous study, natural areas had higher proportions of sugar-fed parasitoids compared to adjacent agricultural fields (Kishinevsky et al., 2018). These wasps were sampled in the summer, a period of intensive agricultural activity (pest control and harvest) that could have disrupted the parasitoids' sugar resources within the agricultural plots. Possibly, sugar feeding in agricultural area recovers during the post-harvest season, when human interference is minimal and sugar resources for natural enemies may be restored. This could happen in crops that are inactive during the winter, because farmers tend to retain herbaceous vegetation that grows during fall and winter until early spring, and this vegetation can potentially provide sugar sources. To test this hypothesis, we compared sugar feeding rates across different seasons.

Finally, agricultural weed management is a farming practice that reduces the diversity and abundance of non-crop plants and hence could impact the availability of sugar sources. Traditionally, weeds are considered to negatively affect crops through competition and pest encouragement (Harlan and deWet, 1965). Growers therefore spray the weeds with herbicides or trim them mechanically, along with other practices (Akotsen-Mensah et al., 2012). Regular herbicide application usually results in low plant diversity, with mainly perennial, herbicide-resistant plants surviving the treatment. Trimming results in high plant diversity before each trimming event and low plant coverage available for insects immediately afterwards. It selects for early-flowering annual plants that complete their life cycle before the first trimming and for plants with vegetative propagation (Moller, 2019). Since each method results in potentially different plant communities in the agricultural plot, they may differ in their effect on sugar feeding by natural enemies. Trimming is usually considered a milder and more sustainable method than herbicide application, because it reduces soil erosion and does not cause water pollution (Melander et al., 2005). A direct comparison of different plant management strategies on parasitoid sugar feeding was never attempted, and this is one of the goals of the current study.

To address these knowledge gaps, we investigated sugar feeding by parasitoids across the seasons in natural and agricultural areas. Specifically, we asked: (a) Is sugar feeding more common in natural habitats than in agricultural plots in all seasons? and (b) Does weed management treatment effect parasitoid sugar feeding? Vineyards were chosen as the focal crop, since this crop has a clear seasonal phenology and an abundance of weeds growing beneath and between the vine rows.

2. Materials and methods

2.1. Study area and experimental design

The study was performed in northern Israel. Six wine-producing vineyards (cultivars Merlot and Cabernet Sauvignon), with at least one edge bordering a natural area, were selected. The vineyards were treated with insecticides according to the advice of an extension specialist. The vineyards' climate is Mediterranean, with cool rainy winters (700 mm annual precipitation) and hot summers. Rains usually occur between October and April, mostly concentrated between December and February. In Israel, the vineyards are non-active in winter, with no growth and no leaves present. Leaves start to bud in March, flowering occurs in April and fruit growth starts about two weeks after flowering. The grapes are harvested in late August-September and leaves fall in November.

All vineyards were sampled four times in different seasons – in February (Winter), April (Spring), August (Summer) and October (Autumn) along 2018. Insects were sampled from the spontaneous vegetation ('weeds') between the vine rows and from the vegetation in the neighboring natural plots, ~200 m away. The natural areas bordering the vineyards comprise Mediterranean phrygas, with scattered trees and shrubs and green herbaceous vegetation that starts growing in the winter, flowers mainly in the spring and dries out in the summer. The vine foliage was also sampled, except in February, when there were no leaves on the vines. Samples inside the vineyards were taken at least five meters from the plot borders. Vine foliage was sampled in the same rows as the spontaneous vegetation. An experiment that compared two weed management practices was conducted in four of the six vineyards. For this experiment, weeds were treated with chemical herbicides in one plot (>0.25 ha in area) within each vineyard, and mechanically trimmed in another plot of similar area. The weeds were trimmed in the first half of April and the main herbicide treatment was applied in the second half of December, to prevent weed germination. Insecticides were applied identically to both weed management treatments (see Table S1 for a detailed list of products used by the farmers). The two vineyards that were excluded from this experiment were only treated with herbicides and no trim plots were maintained, due to in compliance by the landowner with the experimental protocol. In each season, two samples were taken from each habitat/treatment

Table 1

The number of arthropod suction samples collected from each vineyard. Vineyards 1-4 were subdivided into two weed management plots (spraying and trimming), each of which was sampled separately in spring, summer and autumn. This resulted in a higher sampling effort in vineyards 1-4 than in vineyards 5-6. The vines carried no leaves in February, therefore vine foliage could not be sampled.

Location no.	Treatments of trimmed vs. sprayed	Season	Nature No. samples collected	Weeds in vineyard	Vine foliage
1-4	Yes	Winter	2	2	0
		Spring	4	4	2
		Summer	4	4	2
		Autumn	4	4	2
		Winter	2	2	0
5-6	No	Spring	2	2	2
		Summer	2	2	2
		Autumn	2	2	2

plot in each vineyard (see Table 1), and all the vineyards were sampled on the same day. Since time of day may affect sugar feeding (Heimpel et al., 2004), all samples in each of the six vineyards were collected within half an hour or less from each other. Parasitoids were collected using a 'Vortis' insect suction sampler (Burkard Manufacturing Co. Ltd, Rickmansworth, UK). We standardized sampling effort by vacuuming for one minute per sample. Each sample covered two segments of vineyard rows or of the natural vegetation, about 6 m (two vine rows) apart. We sampled approximately a 3×0.5 m area of vegetation in each segment. The sampled segments ran alongside vine rows within the vineyard, and along naturally occurring strips of green vegetation in the natural habitat. Samples were immediately stored on ice in the field and transported to a -20°C freezer within 8 h. Following sorting of the samples, parasitoids were sexed and identified to the species level or, in some cases, to genus/family and morphospecies. Identifications were based on Douth and Viggiani (1968); Masner (1976, 1980); Hayat (1983); Shaw and Huddleston (1991); Goulet and Huber (1993); Chiappini et al. (1996); Grissell et al. (1997); Woolley (1997); Noyes (2003); Pinto (2006); Ulrich (2006); Huber et al. (2009); Pricop (2013); and Aishan et al. (2015). We kept the samples on ice while sorting and identifying the insects to minimize enzymatic digestion of sugar in their guts (Phillips et al., 2018).

The ten most common morphospecies (each represented at least 2 % of the total parasitoid abundance) were chosen to illustrate the per-species sugar feeding rates. These abundant species were all found both in the agricultural and natural areas (Table 3).

As floral nectar is one of the most important sugar sources for parasitoids, we estimated its availability and diversity by counting the number of plant species that flowered in each sampling location in every season, in an area of 20 m^2 around the sampling site. Though potentially of great significance, the number of flowering plant individuals or flowers per species was not assessed. In addition to plant richness, we estimated the percentage cover of green vegetation in every sampling point. The cover percentage was estimated by two trained researchers and the reported value of cover is an average of the two estimates. The researchers estimated the cover of green vegetation in the row sampled inside the vineyard, and in the two rows flanking it, to account for variability between rows. In the natural area, an area of about 30×30 m around the sampling location was scanned and assessed for green vegetation cover.

The identified parasitoids were tested for gut sugars using qualitative cold anthrone tests (Van Handel, 1985; Heimpel et al., 2004). Anthrone is a yellow liquid that changes color in the presence of fructose and fructose-yielding sugars (such as sucrose) (Halhoul and Kleinberg, 1972). The low amounts of fructose and sucrose in the insect hemolymph are not detected by this test and therefore the color-change response indicates that the insect had fed on sugar (Van Handel, 1984). To perform the test, each individual parasitoid was placed on a slide, covered by a drop of anthrone, and gently pressed to expose its gut content. If the anthrone changed color to blue within an hour, the individual was recorded as "anthrone positive". All samples were inspected by two observers, blind to each other's scores. Disagreements between observers were rare (<1 % of samples), and these samples were not included in the results. Some false positive and negative scores are likely to result from this method. Yet, the frequencies of these errors are expected to be similar across treatments and seasons, and therefore should not change our conclusions.

2.2. Statistical analysis

We considered each individual as an independent Bernoulli experiment, since each anthrone test has only two possible outcomes (fed/unfed). Therefore, we used a mixed generalized linear model (GLMM) (Bolker et al., 2009) for binomially distributed data to analyze the effects of season, land use (natural vs. agricultural), sex, vegetation cover, flowering plant richness and habitat inside the vineyard (vine vs.

herbaceous vegetation) on sugar feeding. We also analyzed the interaction between season and habitat, to discern possible effects of seasonal agricultural activities on sugar feeding within the vineyards, and the interaction between season and the number of flowering plant species. Sampling site was defined as random-intercept factor in this model (model 1), to account for the repeated-measures design and for the dependence of samples collected from each site. Tukey's post-hoc tests were performed to determine the differences between the seasons. We defined the species richness of flowering plants as the subset of the flowering species at each site that could potentially be suitable for parasitoid sugar feeding (Table S2). Plant species with concealed floral nectaries (members of the families Fabaceae, Caryophyllaceae, Lamiales) and species that do not produce nectar were therefore excluded.

We used a subset of the samples and a second GLMM (model 2) to analyze the effect of the weed management treatment. All winter samples (treatments began after winter sampling) and the two vineyards that did not apply the weed trimming treatment were excluded. This left us with three seasons and four paired vineyards for the weed treatment analysis (Table 1). Model 2 had the same structure as model 1, with the addition of weed management treatment as a fixed factor. We chose to use two GLMMs, although this somewhat complicates our analyses, because this allows us to test most variables of interest with the full dataset (model 1).

A third GLMM analysis (model 3) was performed on the 10 most dominant species (presented in Table 3), which together constitute 38 % of individuals collected, to assess the effect of species on feeding rates. The effect on anthrone scores by species, and the interactions of season, land use (natural vs. agricultural) and sex with species were tested as in model 1. Sampling site was defined as random-intercept factor.

Parasitoid community composition could explain differences in sugar feeding, since parasitoids from different species can have different sugar requirements. We used a PERMANOVA test (Anderson, 2001) to analyze whether the species composition of parasitoids is affected by the season, land use, and weed treatment. Since habitat did not affect the frequency of sugar feeding (see results below), we did not test for its effect on community composition.

R version 3.4.0 (R Core Team, 2013) was used for all analyses. The package 'lme4' (Bates et al., 2014) was used for the GLMMs and the package 'vegan' (Oksanen et al., 2008) was used for the PERMANOVA test.

3. Results

3.1. Parasitoid sugar feeding

Altogether 1725 parasitoids of 198 species were sampled, identified and tested for sugar feeding. The proportion of anthrone positive parasitoids was significantly affected by season (model 1: GLMM, $\chi^2_3 = 44.013$, $P < 0.0001$). Post-hoc tests showed that sugar-feeding proportions were lower in the winter than in spring, summer and fall, but similar in the other seasons (Fig. 1, Table S3). Sugar feeding was also affected by land use (natural vs. agricultural) (GLMM, $\chi^2_1 = 28.635$, $P < 0.0001$) with 27 % fewer parasitoids that fed (averaged across seasons) in the agricultural plots than in the natural areas (Fig. 1). The interaction between season and land use was not significant (GLMM, $\chi^2_3 = 1.255$, $P = 0.739$). No difference in sugar feeding rates was found between male and female parasitoids (GLMM, $\chi^2_1 = 0.034$, $P = 0.851$), nor between the vine foliage and the vegetation growing between the vine rows (GLMM, $\chi^2_1 = 2.559$, $P = 0.109$). The number of plant species that flowered and were also potential nectar sources for parasitoids positively affected the proportion of anthrone-positive scores, alone and in interaction with season (Fig. 2, number of plant species: GLMM, $\chi^2_1 = 4.543$, $P = 0.033$, interaction between the number of flowering plant species and season: GLMM, $\chi^2_3 = 25.415$, $P < 0.0001$). This interaction reflects the higher richness of potential nectar plants, and their larger effect on the sugar feeding scores, in spring compared to other seasons.

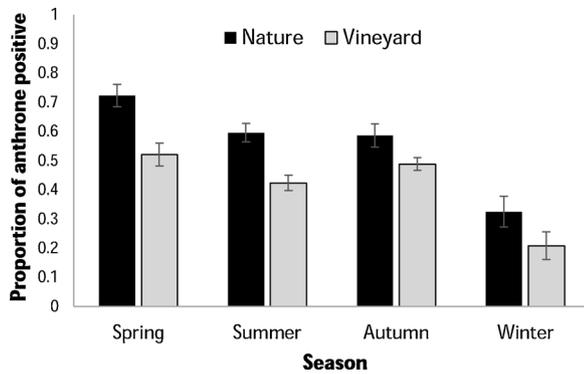


Fig. 1. Mean (\pm SE) proportions of anthrone-positive individuals in natural areas and vineyards (vine foliage and vegetation beneath the vines combined) in the four seasons.

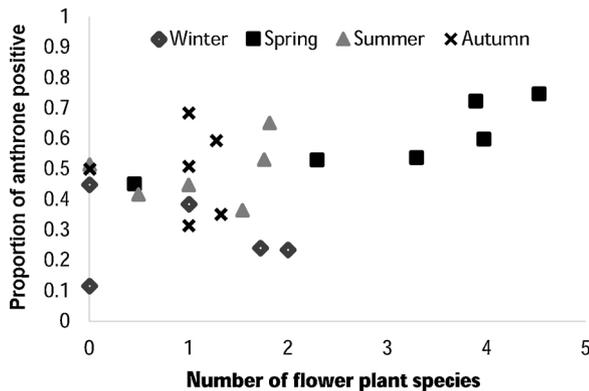


Fig. 2. Proportions of anthrone-positive individuals plotted against the mean number of flowering plant species that could potentially be suitable for parasitoid nectar feeding. The number of plant species was averaged over the natural area, vineyard vegetation and vine foliage for each vineyard-season combination.

The percentage cover of green vegetation in the sampling location did not affect parasitoid sugar feeding (GLMM, $\chi^2_1 = 0.088$, $P = 0.765$). All plant species found that were potential nectar sources for parasitoids are listed in the supplementary material (Table S2) according to habitat and season.

3.2. Effects of weed control treatments

Weed control treatments also affected feeding proportions, but this effect differed between the seasons (Fig. 3). Sprayed plots had higher proportions of anthrone-positive individuals than trimmed plots in spring, and lower proportions in autumn (interaction between weed control treatments and season, model 2: GLMM, $\chi^2_2 = 14.401$, $P < 0.001$). Trimmed plots contained more suitable nectar-producing plant species than sprayed plots in all seasons (Table 2).

3.3. Community composition and species-wise sugar feeding frequencies

Representatives of 48, 74, 108 and 113 species were collected in winter, spring, summer and autumn, respectively. Parasitoid community composition was affected by season (Adonis (PerMANOVA) test: $F_{3,50} = 3.399$, $r^2 = 0.161$, $P = 0.001$) but not by land use (natural vs. agricultural) (Adonis: $F_{1,50} = 1.179$, $r^2 = 0.018$, $P = 0.168$), their interaction (Adonis: $F_{1,48} = 0.874$, $r^2 = 0.042$, $P = 0.787$) or by weed control treatment (Adonis: $F_{1,18} = 0.99$, $r^2 = 0.041$, $P = 0.409$).

In the ten most dominant species tested, parasitoid species had a significant effect on the rate of sugar feeding (model 3: GLMM, $\chi^2_9 =$

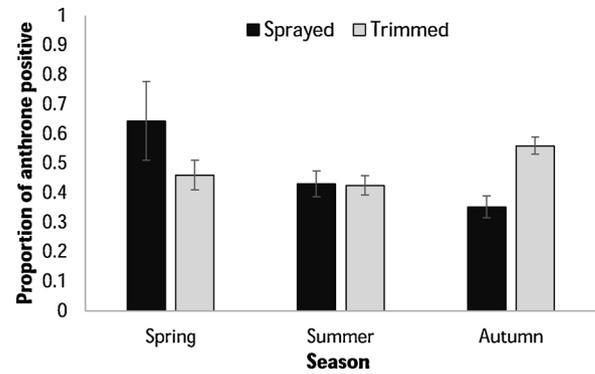


Fig. 3. Mean (\pm SE) proportion of anthrone-positive individuals in the two weed management treatments: chemical herbicide (sprayed) vs. mechanical trimming (trimmed). Vine foliage and vegetation beneath the vines are combined. The treatments were applied after the winter sampling using a paired experimental design in four vineyards.

39.42, $P < 0.001$). The interactions between species and season, land use and sex were not significant (GLMM: $\chi^2_9 = 30.03$, $P = 0.051$; $\chi^2_9 = 9.21$, $P = 0.418$; $\chi^2_1 = 1.326$, $P = 0.249$ respectively).

4. Discussion

Parasitoid sugar feeding was greatly affected by season and land use. The clear seasonal trend we found (Fig. 1) can be explained by declines in both nectar providing plants and honeydew-producing insects from spring to winter. Assessing the abundance and richness of flowers, aphids and other sap-feeding insects was beyond the scope of our study. Nevertheless, we noticed that these potential sugar sources were common in our sampling sites from spring till autumn. During winter, on the other hand, green vegetation is present but flowers and sap-feeding insects are scarce. Similarly, Segoli and Rosenheim (2013), who studied Californian vineyards, found more sugar feeding in the summer than in autumn. Our results broaden these findings by showing that sugar feeding peaks in spring and gradually diminishes towards winter. It is important to remember that the present study encompassed one year and six vineyards only. To strengthen our understanding of seasonal dynamics of parasitoid sugar feeding, longer-term studies should be performed. More research is also needed to find the exact reason for the seasonal trend in parasitoid sugar feeding, particularly taking into account the number of flowers and sap-feeding insects available for parasitoids.

Although the availability of sugar sources is a simple and plausible explanation to the seasonal differences in both agricultural and natural areas, they could also reflect seasonal changes in the parasitoids' community composition or foraging behavior. Different parasitoid species may vary in their life history traits and dietary requirements, and some species may not use sugar resources at all. Though not found in our samples, *Goniozus nephantidis* (Hymenoptera: Bethyliidae) is a rare example of a parasitoid that does not seem to benefit from sugar meals under lab conditions (Hardy et al., 1992). Even within a species, season could affect the wasps' intensity of searching for sugar sources, since temperature can affect their lifespan (McDougall and Mills, 1997; Colinet et al., 2007; Williams and Roane, 2007). In agreement with this possibility, the rates of sugar feeding differed across the dominant species, and varied between seasons within some of the species (Table 3), though the interaction between season and species was not significant. A field experiment with added plants to achieve year-long flowering could disentangle the contributions of sugar availability vs. composition of parasitoids to the observed seasonal differences in sugar feeding rates.

Sugar feeding was markedly lower in agricultural plots than in natural areas, consistent with our previous findings from pomegranate orchards (Kishinevsky et al., 2018). In fact, we recorded anthrone-positive

Table 2

Mean \pm SE numbers of nectar-producing plant species that are potentially available for parasitoids (NPP), the percentage of these species out of the total number of flowering plant species found (in brackets), and the percentage of green vegetation cover, in the different treatments and seasons (weed management treatments started after the winter sampling). The data pertain to the four vineyards where the weed management experiment was done.

	Winter		Spring		Summer		Autumn	
	NPP	Cover	NPP	Cover	NPP	Cover	NPP	Cover
Natural area	1 \pm 0.57 (50%)	96 %	3.75 \pm 1.25 (55 %)	100 %	1.75 \pm 0.48 (100 %)	24 %	1.25 \pm 0.25 (100 %)	40 %
Sprayed vineyard	0.66 \pm 0.33 (100 %)	50 %	2.00 \pm 0.57 (89 %)	5%	0.75 \pm 0.48 (67 %)	13 %	0.75 \pm 0.25 (62 %)	19 %
Trimmed vineyard			3.75 \pm 0.63 (79 %)	76 %	1.25 \pm 0.75 (67 %)	10 %	1.25 \pm 0.25 (79 %)	38 %

Table 3

Abundance and percentages of anthrone-positive reactions in each season for 648 individuals of the ten most abundant species. The species are tabulated by decreasing "Percentage in vineyard", the percentage of all individuals from the species that were collected within the agricultural plots. Percentages of positive responses were calculated after pooling individuals over all vineyards and habitats in each species-season combination, when this pooled sample contained three or more individuals. Species abundance information and anthrone scores for the remaining 1077 sampled individuals are reported in Table S4.

Parasitoid species	Percentage in vineyard	Winter		Spring		Summer		Autumn	
		Abundance	Anthrone positive						
<i>Baryscapus</i> sp. (Eulophidae)	85 %	0	–	0	–	1	–	39	72 %
<i>Lymaenon litoralis</i> (Mymaridae)	82.7 %	3	33 %	21	81 %	33	33 %	24	50 %
<i>Telenomus</i> sp.1 (Platygastridae)	79.1 %	5	60 %	0	–	110	63 %	14	57 %
<i>Oligosita</i> sp. (Trichogrammatidae)	78.4 %	0	–	0	–	23	61 %	28	43 %
<i>Synopeas</i> sp. (Platygastridae)	76.7 %	1	–	0	–	2	–	70	74 %
<i>Diglyphus isaea</i> (Eulophidae)	73.7 %	3	33 %	28	68 %	6	33 %	1	–
<i>Trichogramma</i> sp. (Trichogrammatidae)	69.7 %	0	–	0	–	15	46 %	51	29 %
<i>Ceranis menes</i> (Eulophidae)	52.1 %	1	–	15	80 %	6	66 %	49	34 %
<i>Telenomus</i> sp.2 (Platygastridae)	51 %	5	20 %	2	–	24	58 %	20	80 %
<i>Aphidiinae</i> sp. (Braconidae)	43.8 %	1	–	41	46 %	3	66 %	3	100 %

responses in fewer than 50 % of the individuals from the agricultural area. The actual frequency of sugar feeding within the vineyards is probably even lower, because sugar-fed individuals have higher longevity and hence are likely over-represented in our samples (Kishinevsky et al., 2018). Since land use did not affect the parasitoids' species composition, this result cannot be explained by differences in community composition. Instead, it suggests that individuals do not freely move back and forth between natural and agricultural habitats, although they are separated by short distances, as shown by Derocles et al. (2014) and Navasse et al. (2018).

While season and habitat affected sugar feeding rates as main factors, we found no interactive effects of these variables. For example, no agricultural activities were conducted for several months before our winter samples in the vineyards. Nevertheless, fewer parasitoids fed on sugar within the vineyards than in the natural areas (Fig. 1). This result suggests that sources for sugar remain lower in the agricultural field throughout the year, even though farming activities fluctuate in intensity. The cold anthrone test does not allow us to determine whether the parasitoids fed on nectar, honeydew or both (this can be achieved by HPLC sugar analysis, as in Steppuhn and Wäckers, 2004). It is possible that the relative proportions of nectar vs. honeydew feeding changed across habitats and seasons. We found a positive connection between the number of flowering plants and parasitoid sugar feeding (Fig. 2). This correlation was especially clear during spring time when the number of flowering plants was highest.

Mechanical trimming, a weed control practice that is considered more sustainable than the application of herbicides (Melander et al., 2005), did not enhance sugar feeding of parasitoids. Interestingly, the richness of nectar-producing plant species was consistently higher in the trimmed plots than in the sprayed ones (Table 2), yet the trimmed plots had higher proportions of sugar-fed parasitoids in autumn only. This could be due to differences in the abundance of flowering plants with

accessible nectar, or in their species composition. It seems that variables related to flowering plant species, which warrant further study, affect sugar feeding more than the weed management practice *per se*. The importance of the number of flowering plant species and flower diversity suggests a possible limitation of previous studies, which added a single nectariferous plant species to agricultural systems as a cover crop or border plant (Fiedler et al., 2008). Different parasitoid species may benefit from different flowers, as shown by Arnó et al. (2018), and local herbaceous plants are potentially effective sugar sources (Fiedler et al., 2008). Therefore, preserving natural strips with numerous species of accessible flowering plants may be a better method to provide parasitoids with sugar sources and to enhance biodiversity in agroecosystems. This, according to our results, should be done inside and not around the agricultural field.

Declaration of Competing Interest

The authors report no declarations of interest.

Acknowledgments

The study was supported by the Israeli Ministry of Agriculture and Rural Development (grant number 131-1973). We are grateful to Dr. Serguei V. Triapitsyn from the Department of Entomology, University of California, Riverside, CA, USA and Dr. Zoia Yefremova of the Steinhardt Zoological Museum at Tel Aviv University, Israel, for assistance with parasitoid determination. The Tabor winery supervised the weed management treatments and allowed us to access the vineyards and sample them. We thank Shlomo Cain and Idan Shapira who took part in the field work. We also thank Shivani Krishna, Keren Perry, Gabriella Moller and Daniella Moller for assisting with lab work.

Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at doi:<https://doi.org/10.1016/j.agee.2020.107229>.

References

- Aishan, Z., Triapitsyn, S.V., Hu, H.Y., 2015. Review of *Tumidiclava Girault* (Hymenoptera: Trichogrammatidae) from Xingjiang, China, with description of two new species and taxonomic notes on other Holarctic taxa. *Zootaxa* 3949, 393–407.
- Akotsen-Mensah, C., Boozer, R.T., Fadamiro, H.Y., 2012. Influence of orchard weed management practices on soil dwelling stages of plum curculio, *Conotrachelus nenuphar* (Coleoptera: Curculionidae). *Florida Entomol.* 95, 882–890.
- Anderson, M.J., 2001. A new method for non-parametric multivariate analysis of variance. *Austral Ecol.* 26, 32–46.
- Arnó, J., Oveja, M.F., Gabarra, R., 2018. Selection of flowering plants to enhance the biological control of *Tuta absoluta* using parasitoids. *Biol. Control* 122, 41–50.
- Bates, D., Maechler, M., Bolker, B., Walker, S., 2014. lme4: Linear Mixed-Effects Models Using Eigen and S4. R Package Version 1.
- Begg, G.S., Cook, S.M., Dye, R., Ferrante, M., Franck, P., Lavigne, C., Lövei, G.L., Mansion-Vaquie, A., Pell, J.K., Petit, S., 2017. A functional overview of conservation biological control. *Crop Prot.* 97, 145–158.
- Bianchi, F.J., Wäckers, F.L., 2008. Effects of flower attractiveness and nectar availability in field margins on biological control by parasitoids. *Biol. Control* 46, 400–408.
- Bolker, B.M., Brooks, M.E., Clark, C.J., Geange, S.W., Poulsen, J.R., Stevens, M.H.H., White, J.S.S., 2009. Generalized linear mixed models: a practical guide for ecology and evolution. *Trends Ecol. Evol.* 24, 127–135.
- Chiappini, E., Triapitsyn, S., Donev, A., 1996. Key to the Holarctic species of *Anagrus* Haliday (Hymenoptera: Mymaridae) with a review of the Nearctic and Palaearctic (other than European) species and descriptions of new taxa. *J. Nat. Hist.* 30, 551–595.
- Colinet, H., Boivin, G., Hance, T., 2007. Manipulation of parasitoid size using the temperature-size rule: fitness consequences. *Oecologia* 152, 425–433.
- Derocles, S.A., Le Ralec, A., Besson, M.M., Maret, M., Walton, A., Evans, D.M., Plantegenest, M., 2014. Molecular analysis reveals high compartmentalization in aphid–primary parasitoid networks and low parasitoid sharing between crop and noncrop habitats. *Mol. Ecol.* 23, 3900–3911.
- Doutt, R.L., Viggiani, G., 1968. The classification of the Trichogrammatidae (Hymenoptera: Chalcidoidea). *Proc. Calif. Acad. Sci.* 355, 477–586.
- Evans, E.W., England, S., 1996. Indirect interactions in biological control of insects: pests and natural enemies in alfalfa. *Ecological Applications* 6 (3), 920–930.
- Fadamiro, H.Y., Heimpel, G.E., 2001. Effects of partial sugar deprivation on lifespan and carbohydrate mobilization in the parasitoid *Macrocentrus grandii* (Hymenoptera: Braconidae). *Ann. Entomol. Soc. Am.* 94, 909–916.
- Fiedler, A.K., Landis, D.A., Wratten, S.D., 2008. Maximizing ecosystem services from conservation biological control: the role of habitat management. *Biol. Control* 45, 254–271.
- Géneau, C.E., Wäckers, F.L., Luka, H., Daniel, C., Balmer, O., 2012. Selective flowers to enhance biological control of cabbage pests by parasitoids. *Basic Appl. Ecol.* 13, 85–93.
- Goulet, H., Huber, J.T., 1993. Hymenoptera of the World: An Identification Guide to Families. Research Branch, Agriculture Canada.
- Grissell, E., Schauf, M., Gibson, G., Huber, J., Woolley, J., 1997. Annotated Keys to the Genera of Nearctic Chalcidoidea (Hymenoptera). National Research Council Research Press, Ottawa, Ontario, Canada, pp. 709–725.
- Halhoul, M.N., Kleinberg, I., 1972. Differential determination of glucose and fructose, and glucose-and fructose-yielding substances with anthrone. *Anal. Biochem.* 50, 337–343.
- Hardy, I., Griffiths, N., Godfray, H., 1992. Clutch size in a parasitoid wasp: a manipulation experiment. *J. Anim. Ecol.* 121–129.
- Harlan, J.R., deWet, J.M., 1965. Some thoughts about weeds. *Econ. Bot.* 19, 16–24.
- Hayat, M., 1983. The genera of Aphelinidae (Hymenoptera) of the world. *Syst. Entomol.* 8, 63–102.
- Heimpel, G.E., 2019. Linking parasitoid nectar feeding and dispersal in conservation biological control. *Biol. Control* 132, 36–41.
- Heimpel, G.E., Jervis, M.A., 2005. Does Floral Nectar Improve Biological Control by Parasitoids? Plant-Provided Food for Carnivorous Insects: a Protective Mutualism and its Applications. Cambridge University Press, Cambridge, pp. 267–304.
- Heimpel, G., Lee, J., Wu, Z., Weiser, L., Wäckers, F., Jervis, M., 2004. Gut sugar analysis in field-caught parasitoids: adapting methods originally developed for biting flies. *Int. J. Pest Manage.* 50, 193–198.
- Huber, J.T., Viggiani, G., Jesu, R., 2009. Order Hymenoptera, family Mymaridae. *Arthropod Fauna UAE* 2, 270–297.
- Jervis, M., Kidd, N., Fitton, M., Huddleston, T., Dawah, H., 1993. Flower-visiting by hymenopterian parasitoids. *J. Nat. Hist.* 27, 67–105.
- Kishinevsky, M., Cohen, N., Chiel, E., Wajnberg, E., Keasar, T., 2018. Sugar feeding of parasitoids in an agroecosystem: effects of community composition, habitat and vegetation. *Insect Conserv. Divers.* 11, 50–57.
- Landis, D.A., Wratten, S.D., Gurr, G.M., 2000. Habitat management to conserve natural enemies of arthropod pests in agriculture. *Annu. Rev. Entomol.* 45, 175–201.
- Lavandero, B., Wratten, S., Shishebor, P., Worner, S., 2005. Enhancing the effectiveness of the parasitoid *Diadegma semiclausum* (Helen): movement after use of nectar in the field. *Biol. Control* 34, 152–158.
- Lee, J.C., Heimpel, G.E., 2008. Floral resources impact longevity and oviposition rate of a parasitoid in the field. *J. Anim. Ecol.* 77, 565–572.
- Lee, J.C., Heimpel, G.E., Leibe, G.L., 2004. Comparing floral nectar and aphid honeydew diets on the longevity and nutrient levels of a parasitoid wasp. *Entomol. Exp. Appl.* 111, 189–199.
- Lee, J.C., Andow, D.A., Heimpel, G.E., 2006. Influence of floral resources on sugar feeding and nutrient dynamics of a parasitoid in the field. *Ecol. Entomol.* 31, 470–480.
- Masner, L., 1976. Revisionary notes and keys to world genera of Scelionidae (Hymenoptera: Proctotrupoidea). *Mem. Entomol. Soc. Can.* 108, 1–87.
- Masner, L., 1980. Key to genera of Scelionidae of the Holarctic region, with descriptions of new genera and species (Hymenoptera: Proctotrupoidea). *Mem. Entomol. Soc. Can.* 112, 1–54.
- McDougall, S., Mills, N., 1997. The influence of hosts, temperature and food sources on the longevity of *Trichogramma platneri*. *Entomol. Exp. Appl.* 83, 195–203.
- Melander, B., Rasmussen, I.A., Bärberi, P., 2005. Integrating physical and cultural methods of weed control—Examples from European research. *Weed Sci.* 53, 369–381.
- Moller, G., 2019. Effect of Weed Management on Parasitoid Diversity in Vineyards of Israel. MSc thesis. Ben Gurion University, Israel.
- Nafziger Jr., T.D., Fadamiro, H.Y., 2011. Suitability of some farmscaping plants as nectar sources for the parasitoid wasp, *Microplitis croceipes* (Hymenoptera: Braconidae): effects on longevity and body nutrients. *Biol. Control* 56, 225–229.
- Navasse, Y., Derocles, S.A.P., Plantegenest, M., Le Ralec, A., 2018. Ecological specialization in *Diaeretiella rapae* (Hymenoptera: Braconidae: Aphidiinae) on aphid species from wild and cultivated plants. *Bull. Entomol. Res.* 108, 175–184.
- Noyes, J.S., 2003. Universal Chalcidoidea Database. Access date: 2018. <http://www.nhm.ac.uk/research-curation/research/projects/chalcidooids/>.
- Oksanen, J., Kindt, R., Legendre, P., O'Hara, B., Simpson, G.L., Solymos, P., Stevens, M.H.H., Wagner, H., 2008. The vegan package. *Community Ecol. Package* 10, 631–637. <http://r-forge.r-project.org/projects/vegan/>.
- Parolin, P., Bresch, C., Desneux, N., Brun, R., Bout, A., Boll, R., Poncet, C., 2012. Secondary plants used in biological control: a review. *Int. J. Pest Manage.* 58, 91–100.
- Phillips, C., Hyszczynska-Sawicka, E., Iline, I., Novoselov, M., Jiao, J., Richards, N., Hardwick, S., 2018. A modified enzymatic method for measuring insect sugars and the effect of storing samples in ethanol on subsequent trehalose measurements. *Biol. Control* 126, 127–135.
- Pinto, J.D., 2006. A review of the new world genera of Trichogrammatidae. *Int. Soc. Hymenopterists* 15, 38–163.
- Pricop, E., 2013. Identification key to European genera of the Mymaridae (Hymenoptera: Chalcidoidea), with additional notes. *ELBA Bioflux* 5, 69–81.
- R Core Team, 2013. R: a Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria.
- Rusch, A., Bommarco, R., Ekbom, B., 2017. Conservation biological control in agricultural landscapes. *Anonymous Advances in Botanical Research*. Elsevier, pp. 333–360.
- Segoli, M., Rosenheim, J.A., 2013. Spatial and temporal variation in sugar availability for insect parasitoids in agricultural fields and consequences for reproductive success. *Biol. Control* 67, 163–169.
- Shaw, M., Huddleston, T., 1991. Classification and biology of braconid wasps. In: *Handbooks for the Identification of British Insects*, 7, p. 7.
- Steppuhn, A., Wäckers, F.L., 2004. HPLC sugar analysis reveals the nutritional state and the feeding history of parasitoids. *Funct. Ecol.* 18, 812–819.
- Tena, A., Pekas, A., Wäckers, F.L., Urbaneja, A., 2013. Energy reserves of parasitoids depend on honeydew from non-hosts. *Ecol. Entomol.* 38, 278–289.
- Tena, A., Pekas, A., Cano, D., Wäckers, F.L., Urbaneja, A., 2015. Sugar provisioning maximizes the biocontrol service of parasitoids. *J. Appl. Ecol.* 52, 795–804.
- Tena, A., Wäckers, F.L., Heimpel, G.E., Urbaneja, A., Pekas, A., 2016. Parasitoid nutritional ecology in a community context: the importance of honeydew and implications for biological control. *Curr. Opin. Insect Sci.* 14, 100–104.
- Tylianakis, J.M., Didham, R.K., Wratten, S.D., 2004. Improved fitness of aphid parasitoids receiving resource subsidies. *Ecology* 85, 658–666.
- Ulrich, W., 2006. Body weight distributions of European Hymenoptera. *Oikos* 114, 518–528.
- Van Handel, E., 1984. Metabolism of Nutrients in the Adult Mosquito. *Mosq. News*, pp. 573–579.
- Van Handel, E., 1985. Rapid determination of glycogen and sugars in mosquitoes. *J. Am. Mosq. Control Assoc.* 1, 299–301.
- Williams III, L., Roane, T.M., 2007. Nutritional ecology of a parasitic wasp: food source affects gustatory response, metabolic utilization, and survivorship. *J. Insect Physiol.* 53, 1262–1275.
- Woolley, J.B., 1997. Aphelinidae. Annotated Keys to the Genera of Nearctic Chalcidoidea (Hymenoptera), 794. NRC Research Press, Ottawa, Canada, pp. 134–150.
- Wratten, S., Lavandero, B., Tylianakis, J., Vattala, D., Cilgi, T., Sedcole, R., 2003. Effects of flowers on parasitoid longevity and fecundity. *N. Z. Plant Prot.* 56, 239–245.
- Wyckhuys, K.A., Strange-George, J.E., Kulhanek, C.A., Wäckers, F.L., Heimpel, G.E., 2008. Sugar feeding by the aphid parasitoid *Binodoxys communis*: how does honeydew compare with other sugar sources? *J. Insect Physiol.* 54, 481–491.