Within-field floral resources have the potential to increase parasitism rates in winter oilseed rape pests more than resources at field margins

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Agriculture, Ecosystems and Environment 344 (2023) 108288

https://doi.org/10.1016/j.agee.2022.108288.

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Abstract

Pollen beetles (*Brassicogethes spp.)* and stem flea beetles (*Psylliodes chrysocephala*) are among the main pests of oilseed rape crops in Europe, causing high yield losses. Increasing insecticide resistance and environmental concerns have highlighted the need for alternatives to chemical control, such as conservation biological control. Conservation biological control can be enhanced by providing resources to beneficial organisms in and around crop fields. Flower strips are a promising tool to provide nectar resources for parasitoids. In addition, the planting of such resources in field margins should be reinforced by the absence of insecticide application. However, the presence and the amount of weeds within the field could mitigate the effect of flower strips and needs to be explored.

We surveyed 25 oilseed rape fields in 2019 and 2020. Ten of these fields were adjacent to a sown wildflower strip. We compared an eco-friendly strategy (wildflower strip without insecticide in spring) to a conventional approach (no wildflower strip and chemical control of pests). We assessed the parasitism of stem flea beetle and pollen beetle larvae in the spring, at distances of 5 m and 30 m from the wildflower strip or the natural field margin. We performed botanical surveys in wildflower strips and natural field margins and within the field, to assess the availability of accessible nectar resources to parasitoids.

Stem flea beetle parasitism rates were not affected by the strategy used, but the eco-friendly strategy had a weak positive effect on pollen beetle parasitism. Unexpectedly, the amount of accessible nectar did not differ significantly between wildflower strips in the eco-friendly strategy and natural herbaceous field margins in the conventional strategy. The amount of accessible nectar provided by weeds in the field was positively related only to pollen beetle parasitism rates. By contrast, the amount of accessible nectar for both spontaneous field margin and flower strips had no effect on the rates of parasitism of either species. Parasitism rate decreased significantly with decreasing distance to the field edge, for both species.

Our results suggest that further studies are required: (i) to improve the composition of wildflower strips, so as to provide more accessible nectar resources at the start of spring and (ii) to explore more management strategy options, notably on in-field resources management such as unsown patches or intercropping.

Keywords: Pollen beetle, stem flea beetle, conservation biological control, accessible nectar, parasitism

1. Introduction

Winter oilseed rape is a crop of considerable economic and agronomic value that is grown on 8.5 % of all European arable land (*FAO 2016, in* Ruser et al., 2017). However, it is also subject to high pest pressure from insects such as pollen beetle (*Brassicogethes aeneus*, Coleoptera: Nitidulidae, Fabricius, 1775), stem flea beetle (*Psylliodes chrysocephala,* Coleoptera: Chrysomelidae, Linnaeus, 1758) and stem weevils (*Ceutorhynchus spp*., Coleoptera: Curculionidae) (Williams, 2006). The management of these pests has become more difficult with the move towards organic cropping systems and other types of cropping system without insecticides (Valantin-Morison and Meynard, 2008). For instance, the abundance of stem flea beetle populations has increased in Sweden following the introduction of a neonicotinoid ban (Lundin, 2021). Oilseed rape pests have also become a key issue in conventional cropping systems. Indeed, stem flea beetle and pollen beetle are increasingly resistant to pyrethroids, one of the last families of chemical substances authorized for their control since the ban on neonicotinoids in the European Union (Robert et al., 2017; Slater et al., 2011; Willis et al., 2020). In addition, oilseed rape is a mass-flowering crop very attractive to pollinators, where insecticide applications caused deleterious effects particularly when they are applied during flowering to control inflorescence insects such as pollen beetle (Mänd et al., 2010). It is therefore necessary to provide efficient and biodiversity-friendly crop protection strategies against insects considered as pests, such as through conservation biological control.

Most oilseed rape pests can be regulated by generalist predators (from insects to birds) or more specialist parasitoids (Gagnon, 2017; Riggi et al., 2017). Parasitoids are important biocontrol agents that target oilseed rape pests (Ulber et al., 2010). In Europe, the main parasitoid of pollen beetle is *Tersilochus heterocerus* (Hymenoptera: Ichneumonidae, Thomson, 1889) and the main parasitoid of stem flea beetle is *Tersilochus microgaster* (Hymenoptera: Ichneumonidae, Szépligeti, 1899 ; Ulber et al., 2010). These parasitoids are sufficiently widespread and abundant across Europe to be of potential importance for the conservation biological control of these target pests (Ulber et al., 2010).

Across Europe, the larval parasitism rates of pollen beetle generally lie within the 25-50% range in unsprayed crops, but they may occasionally exceed 80% (Ulber et al., 2010), highlighting the potential of parasitoids for the effective biological control of oilseed rape pests. However, stem flea beetle parasitism rates vary considerably between years and geographic contexts (Ulber and Wedemeyer, 2004; Ferguson et al., 2006).

Once *Tersilochus spp.* larvae have completed their development, they pupate and overwinter in the soil. The adults then emerge in early spring and disperse to colonize oilseed rape crops. At the landscape scale, higher rates of parasitism by *T. heterocerus* are often associated with more complex landscapes, with a low proportion of arable land (Scheid et al., 2011) or a high proportion of semi-natural habitats (Schmidt, Thies, and Tscharntke, 2004; Rusch et al. 2012). Semi-natural habitats adjacent or close to agricultural fields could provide nectar and pollen resources for parasitoids depending on their plant composition and vegetation structure (Rusch et al., 2010). Indeed, in most parasitoid species, adult survival is dependent on the availability of plant-derived sugar-rich foods, such as nectar (Vollhardt et al., 2010). These resources enhance longevity and fecundity, which can translate into higher reproductive success and parasitism rates (Gillespie et al., 2016; Tylianakis et al., 2004).

Like flower fields (Krimmer et al., 2021), flower strips may be a useful management option for providing trophic resources at the edge of the crop, to promote biological control by parasitoids. Büchi (2002) found significantly higher rates of pollen beetle parasitism in oilseed rape fields adjacent to wildflower strips than in an extensively managed meadow, a finding that may be explained by higher levels of nectar resources close to flower strips. Parasitism has been much less investigated in stem flea beetles than in pollen beetles. The parasitoids of stem flea beetles are active from February to March (Ulber et al., 2010), at least one month before those of pollen beetles, when nectar resources may be even more limited than later in the spring, due to the very small number of plants flowering during this period. In a recent field experiment, perennial wildflower strips increased the parasitism of *P. chrysocephala*, *B. aeneus* and of *Ceutorhynchus pallidactylus*, the magnitude of this effect depending on the amount of nectar available and accessible to parasitoids (Gardarin et al., 2021). However, the importance of the nectar resources provided by wildflower strips covering a small area at the edge of the field relative to those provided by weeds over the whole field area have not been investigated. Indeed, weeds growing spontaneously in fields may also be beneficial to parasitoids, through the provision of nectar and pollen resources (Norris & Kogan, 2000). These resources can act as a 'relay' in the field, as parasitism rates generally decline exponentially with increasing distance from floral patches (Tylianakis et al., 2004).

In this study, we investigated the extent to which a pest management strategy based on a wildflower strip with no spring insecticide treatment, as an alternative to conventional approaches based exclusively on insecticides, could promote the parasitism of two main pests of oilseed rape: stem flea beetle and pollen beetle. We compared parasitism rates in fields close to a wildflower strip without insecticide treatment (referred to hereafter as the 'eco-friendly strategy') to a reference situation consisting of fields with no flower strip that were treated with insecticide in the spring (hereafter referred to as the 'conventional strategy').

We expected (i) a positive effect of eco-friendly management on the rates of stem flea beetle and pollen beetle parasitism, with (ii) a decrease in efficacy with increasing distance from the flower strip. We also expected that (iii) the effect of wildflower strips would depend on the amount of accessible nectar available at the field margin and in the field (weeds), and that this effect might be strengthened when no insecticide is used.

2. Methods

2.1. Study sites

The northern half of France is dominated by the cultivation of arable crops (mostly cereals and oilseed rape) in open field landscapes. We followed 12 and 13 fields in 2019 and 2020 respectively, 10 of which were adjacent to a sown wildflower strip, the others being bordered by a natural, spontaneously growing herbaceous field margin. Each flower strip (approximately 4 m x 500 m) was sown in autumn 2018 with a mixture of 42 native, mostly perennial species (Table S1)
The plant species were selected to provide arthropods with physical habitats and diverse trophic resources throughout the year (Gardarin et al., 2018). We asked farmers to leave a 50 m buffer zone not treated with insecticide between the wildflower strip and the central part of the field, to increase the positive effects on biological control (Geiger et al. 2010). Removing insecticide treatments on the whole field was considered too risky by a majority of farmers. In fields treated in spring, farmers mainly applied insecticide once, sometimes twice. Treatments occurred in March and April at full dose with pyrethrinoids (e.g. Trebon 30 EC, Karate Zeon, Sentinel Pro) or less often with neonicotinoids (e.g. Mavrik Flo) or organophosphorous insecticides (e.g. Boravi WG).

In the years studied, over a network of 41 oilseed rape fields farmed conventionally and monitored by agricultural extension services within the study region (DRIAAF, 2020), the rate of oilseed rape plant infestation with stem flea beetle larvae ranged from 0 to over 30 individuals per plant, with a mean of four larvae per plant in 2019 and 2.5 larvae per plant in 2020. This level of infestation is among the highest recorded over the last decade. In the same network, pollen beetle pressure was also the highest in a decade, with a mean of 6 (2019) to 10 (2020) individuals per flowering stem at the sensitive stage, just before flowering.

We digitized the land cover types of cropped and non-cropped areas within a 500 m radius around each field (the landscape buffer), based on an intensive land survey, with QGIS software (version 3.12; www.qgis.org). We calculated the proportion of semi-natural habitats (including woods, fallow, hedges, and wildflower strips) within this radius. The proportion of semi-natural habitats within a radius of 500 m ranged from 0.18 to 63.72% (median=7.48%) of the landscape buffer areas. The proportion of oilseed rape ranged from 4.75 to 37.79% (median=18.68%). Our sampled fields were mostly located in open landscapes and the proportions of semi-natural habitats and oilseed rape tested in the first model did not significantly explain the rates of parasitism in the two species (Table S2). We did not, therefore, introduce landscape variables into the statistical models, to prevent overparametrisation.

2.2. Assessment of parasitism rates in pollen beetle and stem flea beetle larvae

Stem flea beetle

In April 2019 and 2020, we collected 100 last-instar larvae from the petioles of basal leaves of oilseed rape. The larvae were samples at random along a transect of 50 m, at 5 m and 30 m from the wildflower strip, or from the natural herbaceous field margin. We excluded one and four fields from the analysis of stem flea beetle parasitism in 2019 and 2020, respectively, due to the presence of too few stem flea beetle larvae in the plants. We reared the larvae to the adult stage for the determination of parasitism rates. The larvae were placed in hermetic plastic boxes (9.5 cm in diameter, 7 cm high), filled to two thirds of their total volume with moist sieved soil. The soil used consisted of a sand and soil mixture (50:50), which was sterilized before use.

The rearing boxes were placed at room temperature (about 20°C) for six months. We recorded the number of the emerging adults of *P. chrysocephala* (developing from non-parasitized larvae) during the first two months after collection in the field. Rearing boxes were then stored at 4°C for a further three months, to simulate winter temperatures, and were then returned to room temperature. Parasitoids emerged after the release from low temperatures and were recorded over a period of two months, until no further emergence occurred. The parasitoids species was morphologically similar to *Tersilochus microgaster* and *T. obscurator* (Hymenoptera: Ichneumonidae, Aubert, 1959) according to published identification tools (Barari et al., 2005), and could not be assigned to either species with certainty. For simplicity, we named them *Tersilochus sp*. for the purposes of this study.

Parasitism rate was calculated as the number of emerging parasitoids divided by the sum of the number of emerging stem flea beetles and the number of emerging parasitoids, expressed as a percentage.

Pollen beetle

At the end of blooming periods (BBCH stage 66: Lancashire et al., 1991; Weber & Bleiholder, 1990; i.e. in early May 2019 and late April 2020), we collected a minimum of 100 last-instar larvae of more than 3 mm in length from oilseed rape flowers. The larvae were sampled randomly along a transect of 50 m parallel to the field edge, at 5 m and 30 m from the wildflower strip and from the natural herbaceous field margin. The collected larvae were stored in 70° alcohol. Under a binocular microscope, we recorded the presence of eggs in the larvae. They were identified as eggs of *T. heterocerus* due to their black colour (Osborne, 1960). This species is the principal parasitoid of *B. aeneus* in the area (Rusch et al., 2011). Parasitism rates were calculated as the number of larvae containing at least one parasitoid egg divided by the total number of hosts, expressed as a percentage.

2.3. Nectar resources at the field margin and within the field

Standardized botanical surveys were carried out during the sampling of each oilseed rape pest, to assess the nectar resources provided to parasitoids by sown wildflower strips, by the natural vegetation growing spontaneously at the field margin (always herbaceous) and within fields.

We performed three transects: at the field edge, and at distances of 5 m and 30 m from the edge of the field. Every 2 m, over a distance of 40 m (20 plots of 1 m^2), we attributed a cover class to each dicotyledonous species present (as a putative nectar-producing resource) according to its percent cover (Class 1: 1% or less; Class 2: 2-10%; Class 3: 11-30%: Class 4: 31-75%; Class 5: 76-100%). We noted the phenological stage of each species (vegetative, flowering, fruiting). We then calculated the cover of plants (%) providing floral and extrafloral nectar for parasitoids as the sum of the plant species cover producing available and accessible nectar (Supplementary Materials Appendix A). Nectar was considered to be available if it was produced during the period of parasitoid activity. Floral nectar accessibility depends on the morphological match between the plant and parasitoid, and was determined by measuring traits for each parasitoid and plant within the assemblage (Gardarin et al., 2021). For the calculation of floral nectar accessibility, we adapted the geometric model proposed by van Rijn and Wäckers (2016). We considered the ability of the insect to penetrate the flower and its ability to reach the nectar, which depended on corolla shape, nectar depth, insect head size and the size of the mouthparts.

2.4. Statistical analyses

We ran generalized linear mixed models (GLMM), assuming a binomial distribution, with the 'glmer' function (nlme package, Pinheiro et al., 2017). We used the 'cbind' function with the number of parasitized larvae as 'successes' and the number of larvae without parasitism as 'failures'.

We generated two models for each species, for (i) the type of strategy (eco-friendly *vs.* conventional) and (ii) for the gradient of accessible nectar resources at each distance. In the first type of model, we introduced *strategy*, *distance* to the field margin (5 or 30 m) and *year* as explanatory fixed-effects variables, together with the first-order interaction between *strategy* and *distance*. In the second type of model, we introduced *strategy*, *distance*, *year*, and *mean cover of plants providing accessible nectar* at the edge of the field (natural vegetation or wildflower strip) and in the field (weeds) as explanatory fixed-effect variables. We tested the first-order interactions between the *cover of plants providing accessible nectar in the fields* and (a) *strategy* and (b) *distance*.

We accounted for the spatial dependence of observations between distances in the same field, by introducing *field name* as a random-effect variable. All variables were standardized (i.e. by subtracting the mean and dividing by the standard deviation). Correlations between covariates in full models were assessed with variance inflation factors (all VIF < 5; James et al. 2013). Diagnostic residual plots were generated with DHARMa (Hartig and Hartig, 2017) and confirmed a good fit for all models. Spatial correlation in the residuals was evaluated with the 'bubble' function from the 'sp' package and with Correlogram (package 'ncf'); no spatial correlation was detected. All analyses were performed with a significance threshold of 5% in R statistical software version 3.5.2 (2018-12-20).

3. Results

3.1. Availability of floral resources

3.1.1. Availability of field margin resources during stem flea beetle sampling

During stem flea beetle sampling, 29 dicotyledonous plant species were found in the natural field margins (18 species with nectar accessible to stem flea beetle parasitoids) and 18 were found in wildflower strips (11 species with accessible nectar, Fig. S2). A complete list of the plant species providing accessible nectar for each parasitoid studied is provided in Table S4. There was some overlap (61%) in flower species composition with accessible nectar between natural field margins and wildflower strips at the start of the season.

In spontaneous field margins, *Veronica persica*, *Taraxacum officinale* and *Sinapis arvensis* were the main species providing accessible nectar (Fig. S2). In the wildflower strips, nectar was mainly provided by *Vicia sativa*, *Lamium purpureum*, *L. hybridum*, *and Veronica hederifolia* (Fig. S2)

During stem flea beetle sampling, the average percentage of plant coverage providing accessible nectar at field margins was 9.8 % (min-max = 0.1-60.5 %). The mean cover of plants at the field edge providing nectar accessible to parasitoid of stem flea beetle did not significantly differ between the two management strategies: conventional *vs.* eco-friendly (Fig. 1).

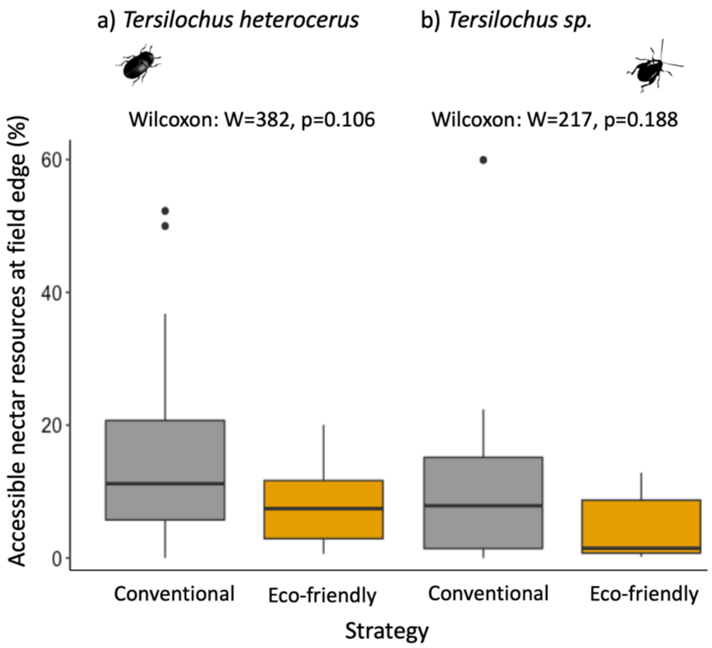

Figure 1. Percent cover of nectar resources accessible to a) *Tersilochus heterocerus* (parasitoids of pollen beetle) and b) *Tersilochus sp.* (parasitoids of stem flea beetle) in field margins, according to management strategy. Conventional = natural herbaceous field margin plus insecticides. Eco-friendly = wildflower strip without insecticides in spring. Boxplots show medians, and the 1st and 3rd quartiles.

3.1.2. Availability of weeds resources during stem flea beetle sampling

During stem flea beetle sampling, 12 dicotyledonous weed plant species with accessible nectar to parasitoids were found in oilseed rape field (Table S5). *Stellaria media*, *Vicia sativa*, *Lamium hybridum* and *Veronica persica* were the main weed species providing accessible nectar over the two distances (Table S5).

The average percentage of weed cover within the field providing nectar accessible to parasitoids of stem flea beetle was 6.2 % (min-max = 0.2-14.2 %) and 2.1 % (min-Mmax = 0.1-12.9 %) in the eco-friendly or in the conventional strategy, respectively.

There was a 50% overlap in weed species communities found in conventional and eco-friendly strategy fields (50% of the species were found in both strategies; Fig. S4).

We detected no difference in weeds cover between the two management strategies (Wilcoxon test: W=172, *P*=1).

3.1.3. Availability of field margin resources during pollen beetle sampling

During pollen beetle sampling, 44 dicotyledonous plant species were found in the natural field margins (21 species with accessible nectar) and 33 were found in wildflower strips (24 species with accessible nectar, Fig. S2). A complete list of the plant species providing accessible nectar for each parasitoid studied is provided in Table S4.
There was a 52% overlap in the flower species composition with accessible nectar between the natural field margin and wildflower strips. The differences in composition between the natural field margin and flower strips were more marked later in the season, i.e. during pollen beetle sampling than during stem flea beetle (Fig. S3).
In spontaneous field margins, *Veronica persica*, *Geranium spp.* and *Taraxacum officinale* were the main species providing accessible nectar (Fig. S2). In the wildflower strips, nectar was mainly provided by *Vicia sativa*, *Leucanthemum vulgare, Lamium purpureum*, and *Barbarea vulgaris* (Fig. S2).
During pollen beetle sampling, the average percentage of plant coverage providing accessible nectar at field margins was 13.4 % (min-max = 0.0-52.2 %). The mean cover of plants at the field edge providing nectar accessible to parasitoid of pollen beetle did not significantly differ between the two management strategies: conventional *vs.* eco-friendly (Fig. 1).

3.1.4. Availability of weeds resources during pollen beetle sampling

During pollen beetle sampling, 19 dicotyledonous weed plant species with accessible nectar to parasitoids were found in oilseed rape field (Table S5). *Vicia sativa*, *Stellaria media*, *Lamium hybridum* and *Veronica persica* were the main weed species providing accessible nectar over the two distances (Table. S5).
The average percent of weed cover within the field providing nectar accessible to parasitoids of pollen beetle) was 5.4 % (min-max = 0.1-12.3 %) and 3.7 % (min-max = 0.1-52.5 %) in the eco-friendly or in the conventional strategy, respectively.
There was a 60% overlap in the weed species found in conventional and eco-friendly strategy fields (60% of the species were found in both strategies; Fig. S4).
We detected no difference in weeds cover between the two management strategies (Wilcoxon test: W=359, *P*=0.244).

3.2. Effect of strategy and of distance to the edge of the field on parasitism rate

The mean parasitism rates for stem flea beetle (mean=29.55% ± 21.78) and pollen beetle (mean=13.12% ± 18.59) varied considerably between fields, ranging from 0 to 90% and 0 to 43.75%, respectively. Study year had no effect on parasitism rate for either of the species (Table 1, Fig. 2).

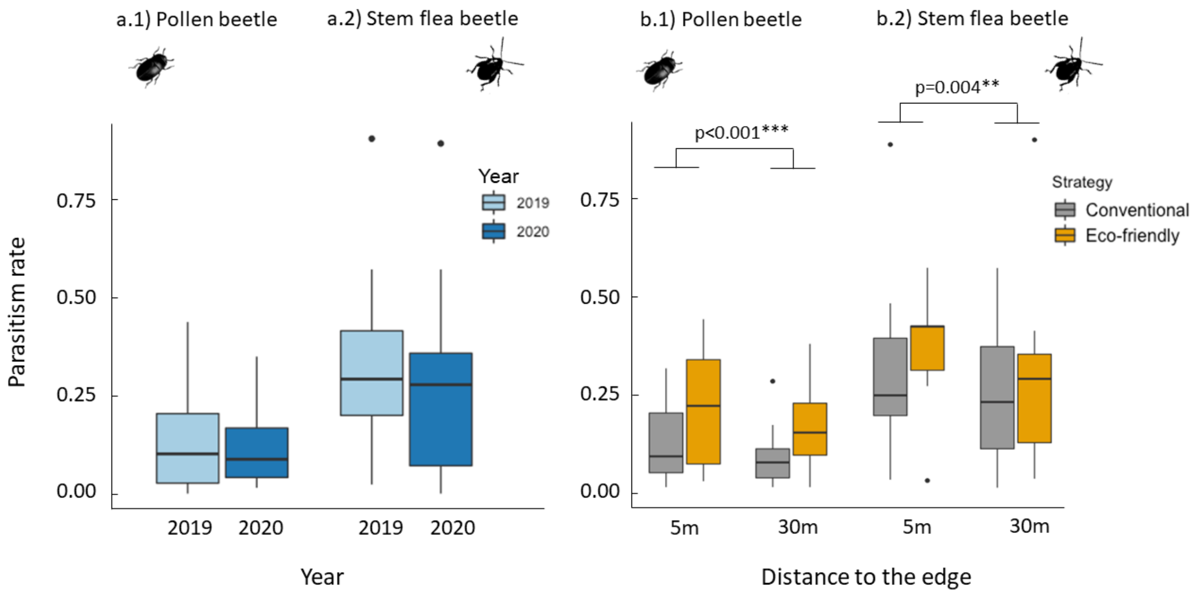

Figure 2. Mean parasitism level of pollen beetle and stem flea beetle according to a) sampling year and b) distance to the edge of the field and management strategy. Boxplots show medians, and the 1st and 3rd quartiles. Conventional = the natural spontaneously growing herbaceous field margin plus insecticide treatment. Eco-friendly = wildflower strip without insecticide application in spring.

Stem flea beetle parasitism rates decreased significantly with increasing distance from the field edge and were not influenced by the type of strategy tested (Table 1, Fig. 2). Parasitism rates also decreased with increasing distance from the field edge in pollen beetle. Parasitism rates were significantly higher at 5 m than at 30 m from the edge of the field, in both stem flea beetles and pollen beetles (Table 1, Fig. 2). In pollen beetle, a trend towards a positive effect of the eco-friendly strategy was observed (Table 1, Fig. 2).

Table 1. Effect of management strategy on the rates of parasitism of stem flea beetle and pollen beetle in oilseed rape fields managed with an 'eco-friendly' (wildflower strip without insecticides in spring) or a conventional (natural herbaceous field margin plus insecticides) strategy. Reference levels for factors: Distance = 30 m; Strategy = conventional; Year = 2019. Significant results are indicated in bold. *P < 0.05, **P < 0.01, ***P < 0.001.

Response variable	Fixed effects	Estimate	Standard error	*Z* value	*P*
Stem flea beetle parasitism rate	**Intercept**	**-1.30**	**0.50**	**-2.63**	**0.009****
	Year 2020	-0.46	0.61	-0.76	0.445
	Distance 5 m	**0.39**	**0.14**	**2.85**	**0.004****
	Strategy eco-friendly	0.19	0.62	0.30	0.765
	Distance x strategy	-0.24	0.20	-1.21	0.226
Pollen beetle parasitism rate	**Intercept**	**-2.94**	**0.38**	**-7.85**	**$<10^{-4***}$**
	Year 2020	0.11	0.46	0.23	0.82
	Distance 5 m	**0.45**	**0.10**	**4.48**	**$<10^{-4***}$**
	Strategy eco-friendly	0.84	0.47	1.77	0.077.
	Distance x strategy	-0.09	0.14	-0.65	0.515

3.3. Effect of accessible nectar on parasitism rates

The nectar resources at the margin of the field did not influence the parasitism rates of stem flea beetles and pollen beetles (Table 2). However, within-field resources enhanced parasitism rates, regardless of distance from the field margin for pollen beetles, and particularly at 5 m from the field margin for stem flea beetle (Table 2; Fig. 3). An interaction between management strategy and within-field nectar resources was observed only for pollen beetle. For this species, the nectar resources provided by weeds had a stronger positive effect in fields managed with the conventional strategy than in fields managed with the eco-friendly strategy (Table 2; Fig. 3). The interaction between the percentage of semi-natural habitat and the strategy was not significant for both species (Table S2).

Table 2. Effect of oilseed rape management strategy (eco-friendly *vs.* conventional), distance to the edge of the field and of nectar resources accessible to parasitoids on rates of parasitism in stem flea beetle and pollen beetle larvae. Reference levels for factors: Distance = 30 m; Strategy = conventional; Year= 2019.

Response variable	Fixed effects	Estimate	Standard error	*Z* value	*P*
Stem flea beetle parasitism rate	**Intercept**	**-1.41**	**0.54**	**-2.62**	**0.009***
	Year 2020	-0.49	0.65	-0.75	0.452
	Distance 5 m	**0.37**	**0.11**	**3.38**	**<0.001*****
	Strategy Eco-friendly	0.10	0.64	0.16	0.873
	Accessible nectar at field edge	0.10	0.35	0.29	0.772
	Accessible nectar in the field (weeds)	-0.66	0.60	-1.10	0.273
	Distance × Accessible nectar in the field (weeds)	**0.37**	**0.18**	**2.02**	**0.043***
	Strategy × Accessible nectar in the field (weeds)	1.04	0.68	1.54	0.123
Pollen beetle parasitism rate	**Intercept**	**-2.81**	**0.42**	**-6.69**	**<0.001*****
	Year 2020	-0.07	0.52	-0.13	0.899
	Distance 5 m	**0.34**	**0.07**	**4.70**	**<0.001*****
	Strategy Eco-friendly	0.68	0.53	1.27	0.203
	Accessible nectar at field edge	-0.27	0.28	-0.99	0.32
	Accessible nectar in field (weeds)	**0.51**	**0.21**	**2.43**	**0.015***
	Distance × Accessible nectar in the field (weeds)	0.01	0.08	0.17	0.862
	Strategy × Accessible nectar in the field (weeds)	**-0.86**	**0.28**	**-3.14**	**0.002***

Significant results are shown in bold: *$p < 0.05$, **$p < 0.01$, ***$p < 0.001$.

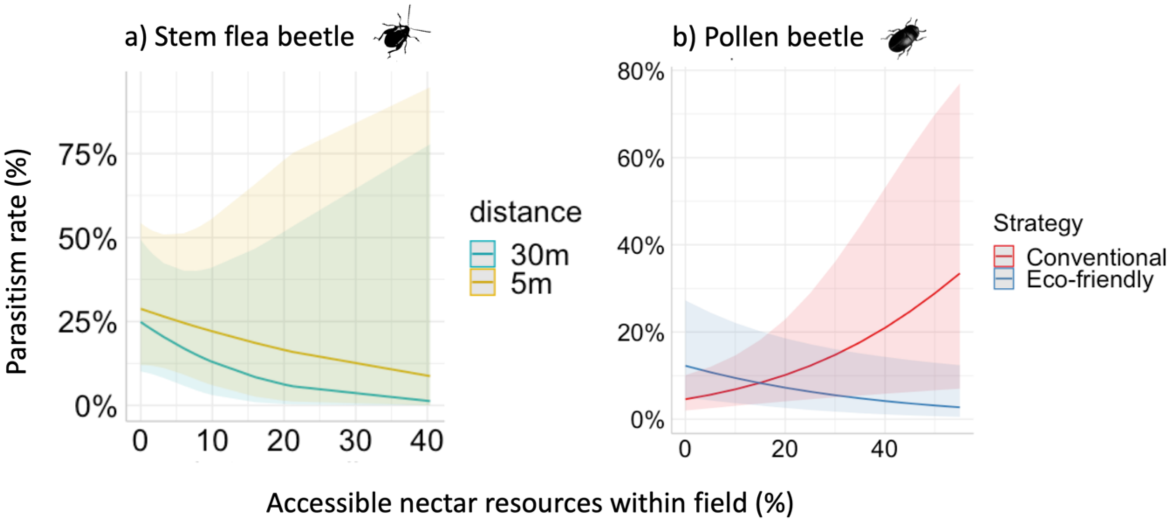

Figure 3. Rates of parasitism of (a) stem flea beetle and (b) pollen beetle as a function of the percentage of accessible nectar resources within the field in interaction with the distance to the field edge for (a) stem flea beetle, or according to management strategy for (b) pollen beetle. Conventional = natural, spontaneously growing herbaceous field margin plus insecticides. Eco-friendly = wildflower strip without insecticide application in spring. The figures show the predictions of the models, with confidence intervals.

4. *Discussion*

4.1. Effect of eco-friendly oilseed rape management on parasitism rates

We compared the effects of perennial wildflower strips without spring insecticide spraying of the crop (eco-friendly management) with a conventional strategy (no flower strips plus insecticide) on the parasitism rates of two major pests of the oilseed rape: stem flea beetle and pollen beetle. The eco-friendly strategy did not significantly increase the rate of stem flea beetle parasitism, but tended to increase the rate of pollen beetle parasitism. For both species, parasitism rates were higher 5 m from the field margin than at 30 m from the field margin, but the eco-friendly strategy did not have the expected strengthening effect on this relationship.

In our study, most farmers who managed their fields conventionally sprayed insecticides between mid-March and mid-April, essentially after the period during which the adults of *T. microgaster* would be flying around in the crop (during February to March). This may account for the very small differences, if any in parasitism rates for stem flea beetle observed between the two crop management strategies. Parasitoids, such as *T. heterocerus,* are active later in the season (from late March to late April, according to our observations) and prefer to oviposit in L2-stage pollen beetle larvae (Ulber et al., 2010). According to the treatment calendar of conventional farmers, the trend of a positive effect of the eco-friendly strategy on pollen beetle parasitism may be linked to the absence of insecticide treatment in spring. Indeed, the trend of a positive effect of flower strips on parasitism rates is known to be greater when no pesticide is applied (Krimmer et al., 2021), highlighting the need to combine a reduction of pesticide use with flower strips to maintain parasitism rates in oilseed rape pests. In addition, the amount of available nectar in wildflower strips and natural field margins was similar, but the difference in species composition in May (Figure S3) could explain this trend.

4.2. Effect of nectar resources and management on parasitism rates

We expected that the effect of wildflower strips on pest parasitism, relative to that of natural spontaneously growing herbaceous field margins, would depend on the trophic resources provided to parasitoids. In a previous factorial field experiment, the percent cover of plants providing accessible nectar in wildflower strips to parasitoids was positively related to stem flea beetle and pollen beetle parasitism (Gardarin et al. 2021). Here, parasitism rates were highly variable but, contrary to our hypothesis, did not seem to be related to the amounts of accessible nectar available at the edge of the fields. The trend towards a weak positive effect of the eco-friendly strategy on pollen beetle parasitism rates was, therefore probably more closely linked to the absence of insecticide treatment than to flower strip resources. However, the absence of wildflower strip was confounded with the absence of treatment in our study, making a more detailed interpretation of the mechanisms difficult. In general, very few plants in the flower strips were flowering in March and April in our study. The amount of available nectar provided by the flower strips was essentially similar to that of natural herbaceous margins, but they exhibit different species composition in May, as shown in figure S3. Differences in their effect could be explained by traits not taken into account here, such as the amount and quality of nectar, flower attractivity toward parasitoids as well as possible preferences toward specific plant species. In addition, despite a choice of species to ensure the provision of food resources throughout the entire season, the nectar of sown species, such as *Bellis perennis,* was not accessible to parasitoids (as shown by morphometric models) whereas other widespread wild species, such as *Taraxacum officinale* or *Veronica spp*., provided accessible resources. These wild species grew spontaneously in the natural field margins, and their presence therefore decreased the expected difference in the efficacy of flower strips relative to natural field margins. Although the nectar of the majority of species was accessible to stem flea beetle and pollen beetle parasitoids, we estimated the resource by the ground cover of these plants, without counting the flowers or taking into account the amount of nectar produced (see for example Baude et al., 2016). The high presence of low nectar-producing species could explain the lack of relationship between the amount of resources observed (in terms of cover of nectar-producing species) at the field margins and parasitism rates of oilseed rape pests.

The very small effect of wildflower strips may also be due to their young age (established for less than two years) and their relatively small size (<1 ha). Krimmer et al. (2021) recently showed that newly established flowering fields (sown the previous year) did not greatly improve pollen beetle parasitism rates, whereas older flower fields (5-6 years old) and calcareous grasslands did. They also showed that flower fields with an area greater than 1.5 ha promoted parasitism more effectively than smaller flower fields.

The presence of weeds offering floral resources within the field influenced the parasitism rates. A greater abundance of weed resources at 5 m resulted in higher stem flea beetle parasitism rates. Pollen beetle parasitism rates also increased with the abundance of weed resources. This effect was greater in conventionally managed fields than in fields under eco-friendly management, indicating that the field resources were more effective without wildflower strip and with insecticide application. Parasitism rates declines exponentially with increasing distance from floral patches (Albrecht et al., 2020; Tylianakis et al., 2004). Within-field resources, close to the hosts, may, therefore, increase parasitism rates at field scale more effectively than resources at the margin of the field.

Also, considering the 48% difference in species field margin composition between the ecofriendly and conventional strategies, the effect of weeds in the conventional strategy could originate from traits and processes not taken into account to relate plant species composition and parasitoid activity. At similar levels of available and accessible nectar, all plant species

may not have similar effects. For instance, the chemical characteristics of nectar which can affect their attractivity, nutritional quality and preferences toward parasitoids (Belz et al., 2013). Maintaining desirable target levels of weeds in the crop is difficult for farmers, particularly in terms of preventing competition with the crop, but our results highlight the importance of managing the resources available to natural enemies at whole-field level, rather than just at the edge of the field. One recent study highlighted increases in species richness and parasitoid abundances in the presence of within-field unsown patches in oilseed rape fields (González et al., 2022). Further studies of the effects of unsown patches on parasitism rates could lead to the formulation of guidelines for farmers.
Otherwise, intercropping or undersowing are also two agricultural practices that could decrease pest pressure and enhance within field resources for parasitoids (e.g. extra-floral nectar secreted by leguminous plant) while allowing to reduce herbicide applications by competition with undesirable weeds for farmers. In oilseed rape, intercropped service plants modify the architecture of the crop canopy by increasing its structural heterogeneity, and they make it more difficult for pests to find their hosts (Finch and Collier, 2000), leading to a decrease of pest oviposition rates (Âsman et al., 2001). This resulted in a reduction of the abundance and damage of several oilseed rape pests (Breintenmoser et al., 2022; Cadoux et al., 2015). Although promising, some studies are required to assess their effect on parasitism rate.

5. *Conclusion*

This study shows that a pest management strategy based on a wildflower strip combined with an absence of spring insecticide treatment, as an alternative to conventional methods based on insecticides, tended to promote the parasitism of pollen beetle, but not that of stem flea beetle. The eco-friendly strategy alone is therefore not enough sufficient to significantly improve parasitism rates. The effect of the flower strip did not depend on distance or on the amount of resources provided. Conversely, the presence of weeds within the field seemed to have a good potential for promoting parasitism.

These results open up new perspectives for promoting the conservation biological control of oilseed rape pests. Further studies are required to improve the composition of wildflower strips, to provide more accessible nectar resources at the start of spring.

The implementation of this strategy should also be accompanied by a reflection on the management of other factors influencing the quality of crop habitats, including weed management, in particular. We would argue for the exploration of more management strategy options, trying to achieve a balance between competitive ability and the ability to sustain conservation biological control, for example. Furthermore, it will only be possible to do without insecticides if other technical levers, such as those related to bottom-up control via autumn intercropping with legumes to deal with stem flea beetles or with early flowering varieties to deal with pollen beetles, are implemented.

Author Contributions

LS, AB, MVM, FC and AG conceived the ideas and designed the methodology; LS, AG and AB collected the data; LS analyzed the data and wrote the first draft of the manuscript. All authors contributed critically to the drafts and approved the final version for publication.

Acknowledgements

This work was funded by INRAE (metaprogram EcoServ), the François Sommer Foundation, AgroParisTech, the *Office Français de la Biodiversité*, the *Fondation pour la Recherche sur la Biodiversité*, and Labex BASC.

We would like to thank Eleonore Courteau, Arnaud Butier, Alexandre Morisseau, Dominique Le Floch, Emmanuel Frankenreiter and Solene Morelle for technical assistance in the collection of field data and larval dissection. We thank all the farmers involved in the study.

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