

Selective flowers to attract and enhance *Telenomus laeviceps* (Hymenoptera: Scelionidae): a released biocontrol agent of *Mamestra brassicae* (Lepidoptera: Noctuidae)

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Abstract

The importance of the right food source for the survival and reproduction of certain insect species is well documented. In the case of biocontrol agents, this is even more important in order to reach a high predation or parasitism performance. The egg parasitoid *Telenomus laeviceps* (Förster, 1861) (Hymenoptera: Scelionidae) is a promising candidate for mass release as a biological control agent of the cabbage moth *Mamestra brassicae* (Linnaeus, 1758) (Lepidoptera: Noctuidae). However, adult *T. laeviceps* need a sugar-rich food source to increase their parasitism performance and produce a good amount of female offspring. Released biocontrol agents were shown to benefit from conservation biocontrol, which includes the provision of selected flowers as nectar resources for beneficial insects. In Switzerland, *Centaurea cyanus* L. (Asteraceae), *Fagopyrum esculentum* Moench (Polygonaceae) and *Vicia sativa* L. (Fabaceae) are successfully implemented in the field to attract and promote natural enemies of different cabbage pests. In this study, we investigated the potential of these selected flowers to attract and promote *T. laeviceps* under laboratory conditions. In Y-tube olfactometer experiments, we first tested whether the three nectar providing plant species are attractive to *T. laeviceps*. Furthermore, we assessed their effects on survival and parasitism performance of adult *T. laeviceps*. We found that flowers of *F. esculentum* and *C. cyanus* were attractive in contrast to *V. sativa*. Also fecundity and the number of female offspring produced were higher for females kept on *F. esculentum* and *C. cyanus* than on *V. sativa*. In contrast, survival was similar on all treatments. Our findings present a further key step towards the implementation of *T. laeviceps* as a biocontrol agent.

Keywords: biocontrol agent, fecundity, longevity, nectar, olfactometer, *Telenomus laeviceps*

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Introduction

In the life history of organisms, resources to invest in reproduction can be gained following two main strategies (Stephens *et al.*, 2009). Capital breeders are born with an accumulation of resources in their bodies that can be directly invested into reproduction, while income breeders acquire the

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needed resources immediately before reproduction (Stephens *et al.*, 2009). Among insects, these two strategies are often referred as pro-ovigeny and synovigeny, where the first one indicates females born with a complete stock of mature eggs, while in the second one this stock is absent or limited and eggs are produced throughout adult life (Jervis *et al.*, 2007, 2008). Most hymenopteran parasitoids are however between these two extremes and are considered to be moderately synovigenic, usually emerging with very limited larval reserves and die within few days without access to a suitable adult food source (Jervis *et al.*, 2001; Boivin, 2010). Thus, adults require suitable non-host food sources to satisfy their energy needs (Bianchi & Wäckers, 2008), enhancing their life expectancy, realized fecundity and dispersal capacity (Wäckers, 2004; Romeis *et al.*, 2005; Wäckers *et al.*, 2006, 2007; Bernstein & Jervis, 2008; Géneau *et al.*, 2012). Also small egg parasitoids from the genus *Trichogramma* benefit from sugar-rich food sources, as shown for *T. pretiosum* Riley (Hymenoptera: Trichogrammatidae) and *T. platneri* Nagarkatti (Hymenoptera: Trichogrammatidae) (Ashley & Gonzalez, 1974; McDougall & Mills, 1997). *Trichogramma* spp. are widely used in agriculture as biocontrol agents (Parra, 2010), but because *Trichogramma* spp. are clearly synovigenic, it can be challenging to apply them efficiently for pest control and keep the maintenance costs as low as possible (Mills *et al.*, 2000). Indeed, studies in agricultural fields conducted on *Trichogramma* spp. demonstrated the importance of a constant food provision throughout the parasitoid life and not just before their release in the field to ensure high parasitism rates (Ashley & Gonzalez, 1974; Leatemia *et al.*, 1995; Díaz *et al.*, 2012). Under field conditions, *Trichogramma* spp. were observed to consume nectar of different flowers near crop fields, like e.g. red clover (*Trifolium pratense* L.), buckwheat (*Fagopyrum esculentum* Moench), mustard (*Brassica juncea* L.), dill (*Anethum graveolens* L.) or avocado flowers (*Persea americana* Mill.) (Wellinga & Wysoki, 1989; Begum *et al.*, 2004, 2006; Witting-Bissinger *et al.*, 2008; Díaz *et al.*, 2012). Therefore, herbs in flower strips, planted next to the crop plants threatened by pest insects, could serve as nectar sources providing sugar and other nutrients (Witting-Bissinger *et al.*, 2008; Balzan & Wäckers, 2013; Balzan *et al.*, 2016). However, apart from nectar quality and quantity, nectar accessibility, determined by flower morphology and the presence of extra-floral nectar, is decisive for small-bodied parasitoids. This emphasizes the importance to select the right plant species for flower strips depending on the parasitoid of interest (Patt *et al.*, 1997; Tooker & Hanks, 2000; Vattala *et al.*, 2006).

The present study is focused on *Telenomus laeviceps* (Förster, 1861) (Hymenoptera: Scelionidae), an egg parasitoid distributed across Europe, able to parasitize eggs of different insect pests belonging to the Noctuidae, Geometridae and Nolidae (Mexia *et al.*, 2004; Klemola *et al.*, 2009; Bayle, 2012; Petrov, 2012). This parasitoid can be used in brassica fields as a biocontrol agent against the cabbage moth *Mamestra brassicae* L. (Lepidoptera: Noctuidae). We conducted studies on its biology and the results clearly showed that *T. laeviceps* emerges with limited larval reserves. This implies that, in order to reach a maximum parasitism performance and proportion of female offspring, sugar-rich food sources are needed directly after adult emergence. Since *T. laeviceps* is released via field delivery systems as parasitized eggs, similar to the one commercially used for different *Trichogramma* species, adult wasps emerge directly in the field, benefiting from an easily exploitable food source near

the release point. A possible solution would be the addition of honey to the field delivery systems. However, preliminary field trials revealed that the provided honey indirectly increased predation of the exposed parasitized eggs, reducing the effective number of released parasitoids. Furthermore, honey should be provided only shortly before the exposition in the field of the field delivery systems, causing additional efforts and costs for end users. Besides honey, a further solution can be the provision of nectar sources near the crop field, for example, as sown flower strips, as already implemented for some *Trichogramma* spp. that are used as biocontrol agents (Wellinga & Wysoki, 1989; Begum *et al.*, 2004, 2006; Díaz *et al.*, 2012).

Here, we focused on the promotion of released *T. laeviceps* through the provision of nectar sources. Studies were already conducted to promote natural enemies of different cabbage pests, such as *M. brassicae* or *Plutella xylostella* (Linnaeus, 1758) (Lepidoptera: Plutellidae) (Géneau *et al.*, 2012, 2013; Balmer *et al.*, 2013, 2014; Belz *et al.*, 2013). Based on these studies, *Centaurea cyanus* L. (Asteraceae); buckwheat, *F. esculentum* Moench (Polygonaceae) and common vetch, *Vicia sativa* L. (Fabaceae) were selected as the main components of a tailored flower strip for the promotion of beneficial insects of cabbage pests. These flowers, besides being part of the tailored flower strip for the cabbage production, were already implemented to promote other parasitoid species, like *Microplitis mediator* (Haliday, 1834) (Hymenoptera: Braconidae), *Dolichogenidea tasmanica* (Cameron) (Hymenoptera: Braconidae), *Trichogramma* spp. or *Anagyrus pseudococci* (Girault) (Hymenoptera: Encyrtidae) (Berndt *et al.*, 2002; Witting-Bissinger *et al.*, 2008; Géneau *et al.*, 2012, 2013; Irvin & Hoddle, 2015). Furthermore, these flowers did not increase the survival nor the fecundity of *M. brassicae*, the most important host of *T. laeviceps* (Géneau *et al.*, 2012). In Switzerland, to increase the pest control achieved by the tailored flower strip, *T. laeviceps* can be released as biocontrol agent. Under laboratory conditions we investigated the suitability of *C. cyanus*, *F. esculentum* and *V. sativa* to attract released *T. laeviceps* and increase their parasitism performance. The olfactory attractiveness can help reducing the time spent searching for food and thus increase the *per capita* host searching efficiency (Wäckers & Swaans, 1993; Hegazi *et al.*, 2000; Jervis & Heimpel, 2007; Jervis *et al.*, 2008). However, attractiveness alone is not enough to reach the desired pest control. In fact, flowers should also promote the performance of the target beneficial. To this end, we conducted (i) olfactometer trials to determine the attractiveness potential of the selected flowers for *T. laeviceps* and (ii) laboratory experiments testing their influence on survival and parasitism performance of *T. laeviceps*.

Material and methods

Parasitoid

Rearing of the egg parasitoid *T. laeviceps* started in 2012 at the Research Institute of Organic Agriculture (FiBL), Switzerland, from individuals collected with trap eggs (cabbage moth *M. brassicae*) from organic cabbage fields in the Swiss Plateau (47th parallel north). *T. laeviceps* was reared in glass tubes (14.5 cm, \varnothing 3 cm) on cabbage moth eggs in a climatic chamber at $22 \pm 2^\circ\text{C}$, 16:8 (L:D) photoperiod and $55 \pm 5\%$ relative humidity (RH). To ensure the supply of a sufficient number of adult wasps for the experiments, three new rearing units were started weekly. A rearing unit consisted of 1500–2000

cabbage moth eggs (<24 h old) and approximately 100 10-day-old wasps (70% females and 30% males). Females were allowed to parasitize the provided batch of eggs during 7 days. Afterwards, the parasitized eggs were placed in an empty rearing unit until wasp emergence. Parasitoids were fed with honey–gelatine *ad libitum* (200 g flower honey (Switzerland), 100 ml demineralized water and 3 g gelatine (Dr Oetker, Germany)), provided on a piece of white paper placed in each rearing unit. With these rearing conditions, following generations of parasitoids emerge 14 days after parasitisation started.

Plants

The flowering plants used in these experiments were grown in climatic chambers (GroBanks (CLF Plant Climatics, Germany)) at 24°C (day) and 18°C (night), 55 ± 5% RH and with a 16:8 (L:D) photoperiod. To ensure a constant supply of flowers, weekly 14 seeds per flower species were sown in 4 × 4 cm pressed soil blocks (Schwarz AG, Villigen, Switzerland). After 3 weeks, seedlings were transplanted to 12 cm diameter pots (10 cm height) in soil (Einheitserde Classic, Gebrüder Patzer GmbH & Co.KG, Germany) fertilized with slow-release formulation fertilizer (3 g l⁻¹ of Tardit 3 M (Haubert HBG Dünger AG, Switzerland)). Plants were regularly checked and watered as needed.

Olfactory attractiveness of different flowers for *T. laeviceps*

The attractiveness of the flowering plants was tested in a Y-tube olfactometer as described by Belz *et al.* (2013). The experiments were conducted in a dark room, during the period of main parasitoid activity, between 10 and 12 am. An

artificial light source (20 W) was placed 28 cm above the Y-tube glass. The humidified charcoal-filtered air passed at a speed of 757 ml min⁻¹ through two glass containers, each containing one odour. A visual barrier was placed between the Y-tube and the two odour containers (fig. 1).

Newly hatched (<24 h old) and unfed *T. laeviceps* females were used for the experiments. Females were inserted at the base of the Y-tube and the assessment started when they crossed the start line (fig. 1). They had 5 min time to take a decision by crossing one of the two finish lines (fig. 1). If they did not chose within the given time, they were discarded from the experiment.

We tested the attractiveness of the three flower species *C. cyanus*, *V. sativa* and *F. esculentum* against ambient air only. *C. cyanus* and *F. esculentum* were, in addition, tested one against the other. During the experiments, *V. sativa* flowers were absent, therefore only the attractiveness of plants displaying extra-floral nectar was tested. Thirty parasitoids were tested per comparison. Flower buds for *V. sativa* or inflorescences for *C. cyanus* and *F. esculentum* were freshly cut and placed in the odour containers. For *V. sativa*, the presence of extra-floral nectar was confirmed with the help of a binocular. After six tested wasps, the odour sources were renewed. The position of the odour sources was switched after three wasps had been tested to avoid biases due to positional effects.

Survival and parasitisation performance of *T. laeviceps* in the presence of different nectar resources

We tested the influence of nectar availability of *C. cyanus*, *F. esculentum*, *V. sativa* and a water control on the survival and parasitisation performance of *T. laeviceps*. During the

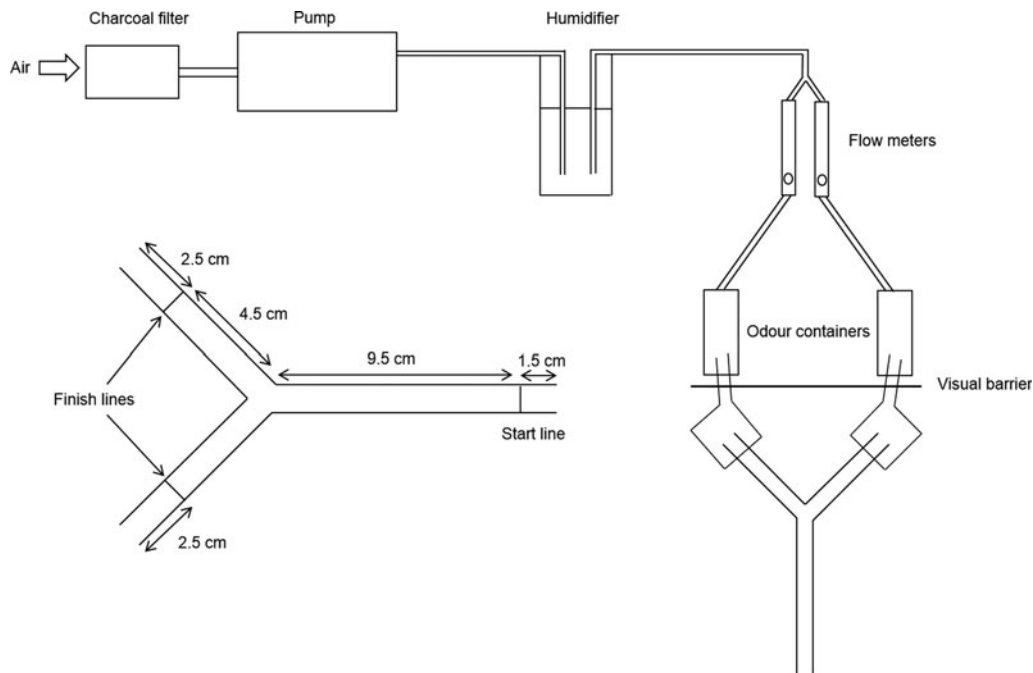


Fig. 1. Set up of the Y-tube olfactometer. Air passes through two odour containers and enters the Y-tube. Parasitoids were inserted at the base of the Y-tube and the assessments were started when the wasps crossed the start line.

experiments, flowers of *V. sativa* were absent, therefore only plants presenting extra-floral nectar were used.

Parasitisation performance and survival experiments were conducted in plastic cages (fig. 2) in a laboratory at $23 \pm 2^\circ\text{C}$ and $90 \pm 9\%$ RH in the presence of flowers and at $23 \pm 1^\circ\text{C}$ and $46 \pm 6\%$ RH in the negative control. Temperature and RH were measured inside the cages with small data loggers (DS1923 Hygrochron, Thermodata). In contrast to temperature, RH in the plastic cages differed between the control and the three flowers. To exclude biases in the results due to this difference, a small trial was conducted in a climatic chamber with higher RH values compared with the laboratory ($55 \pm 5\%$). Here, we compared the survival of females when provided with water only. Under higher RH, the parasitoids

died within 1 or 2 days, as in the control with lower RH (data not shown). We thus concluded that this difference in the RH between flowers and control should not influence the outcome of the trials. Plastic cages were designed to allow air circulation and at the same time to prevent the small parasitoids from escaping. One and half litre plastic bottles opened at the bottom and closed with a sponge cut in the middle were used as cages. This opening allowed the insertion of the flowers in the cages (fig. 2). On the top, bottles were closed with wet cotton, which was daily watered to ensure water provision during the whole experiment. Plants in pots were used for the trials. To assess the survival of *T. laeviceps*, one female and one male (both <24 h old) were placed in a plastic cage in the presence of one of the four treatments. For

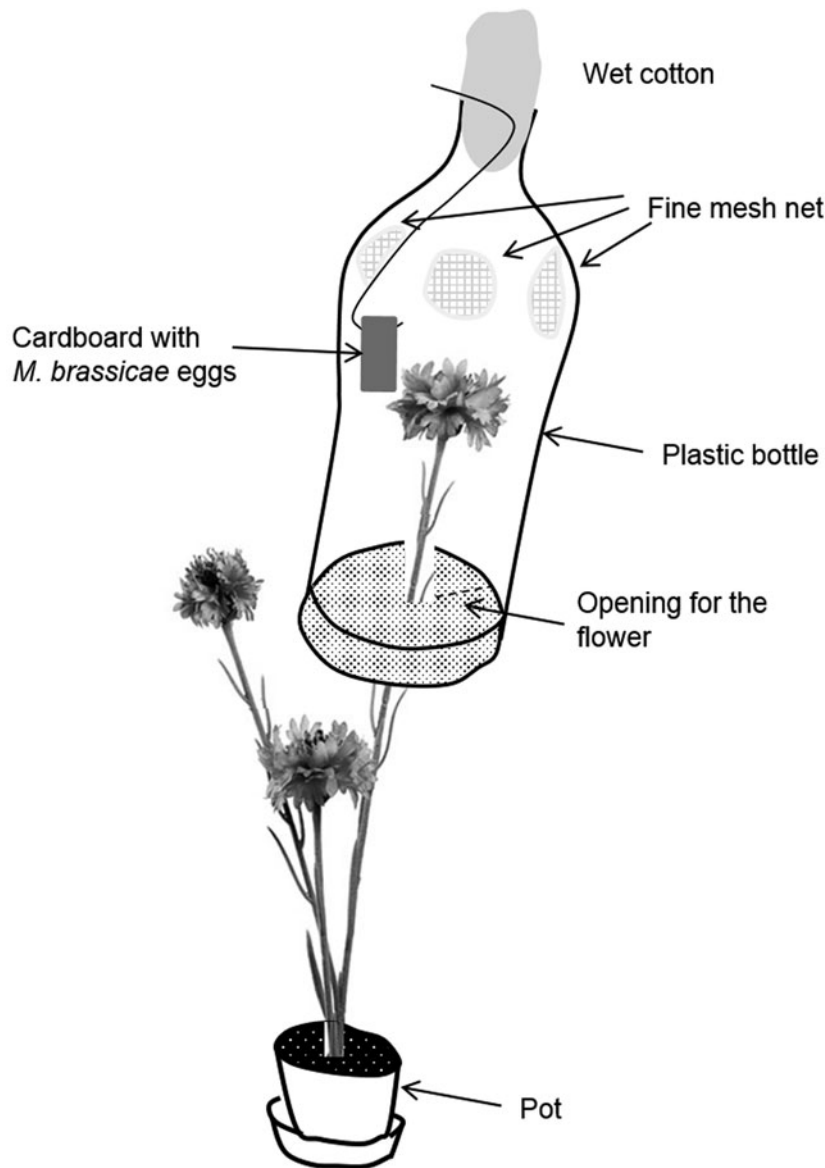


Fig. 2. Set up of the cage used for the fecundity and survival experiments. A plastic bottle was opened at the bottom and closed with a sponge, cut in the middle to allow the positioning of the flower. For the fecundity experiments, additional to the flower, *Mamestra brassicae* eggs were provided.

the fecundity experiment, cabbage moth eggs were additionally provided at the top of the bottle, on a daily basis, until female death (fig. 2). In both trials, mortality was assessed daily at 9.30 am. For both trials, ten replicates per treatment were tested. One replicate per treatment was started weekly. The parasitized eggs were counted, as well as the number of emerging parasitoids. The sex of the offspring was also determined.

Statistical analysis

Data analyses were conducted with R version 3.3.0 (R Core Team, 2016). The count data from the olfactometer experiment were analysed with a Pearson's χ^2 test by comparing the observed frequencies against 0.5 (expected frequency of the zero hypotheses: no preference).

Data from the survival experiment were interval censored and therefore plotted following the non-parametric maximum likelihood estimate for the distribution of interval censored data. The overall influence of treatments on survival was tested by an asymptotic log-rank k -sample test. Since this kind of analysis does not allow a *post-hoc* test for the pairwise comparison of the different treatments, single analyses were conducted through the asymptotic log-rank two-sample test. The resulting P -values were adjusted with the Bonferroni correction for multiple comparisons. The survival data were fitted to the model with the *icfit* and *icest* functions included in the *interval* package.

The number of parasitized eggs and the number of females produced were analysed with generalized linear models with Poisson errors (*glmer* function from the package *lme4*) and the fixed factor treatment (four levels: water and the three flower species), corrected for overdispersion.

Results

Olfactory attractiveness of different flowers for *T. laeviceps*

Out of the three flowers tested, only *C. cyanus* and *F. esculentum* were significantly attractive for *T. laeviceps* females compared with the control (fig. 3a, b). We found no significant difference between *V. sativa* (extra-floral nectar only) and the control (fig. 3a, b). *C. cyanus* and *F. esculentum* were found to be equally attractive (fig. 3a, b).

Survival and parasitism performance of *T. laeviceps* in the presence of different nectar resources

The presence of eggs during the survival experiment did not influence the survival of both females and males (asymptotic log-rank two-sample test, all $P < 0.3$). Therefore, we pooled the data for each treatment. We found a significantly higher survival rate of *T. laeviceps* females for all three flowers tested compared with the control (asymptotic log-rank two-sample test, $N = 20$, all $P < 0.0001$) (fig. 4a). In the presence of *V. sativa* (extra-floral nectar only), *F. esculentum* and *C. cyanus* (floral and extra-floral nectar), females survived significantly longer compared with water (fig. 4a). No difference was found in the survival of females between the three flowers (asymptotic log-rank two-sample test, $N = 20$, all $P > 0.8$). Similar results were found for males (fig. 4b).

Similar to survival, the number of parasitized eggs in the water control (19.1 ± 11.2 eggs) was significantly lower than in the *F. esculentum* (204.7 ± 42 eggs; generalized linear model, $z = 3.542$, $P < 0.0001$) and *C. cyanus* (202.5 ± 42.8 eggs; generalized linear model, $z = 3.531$, $P < 0.0001$) treatment.

No significant difference was found between the water control and *V. sativa* (144.8 ± 67.91) (generalized linear

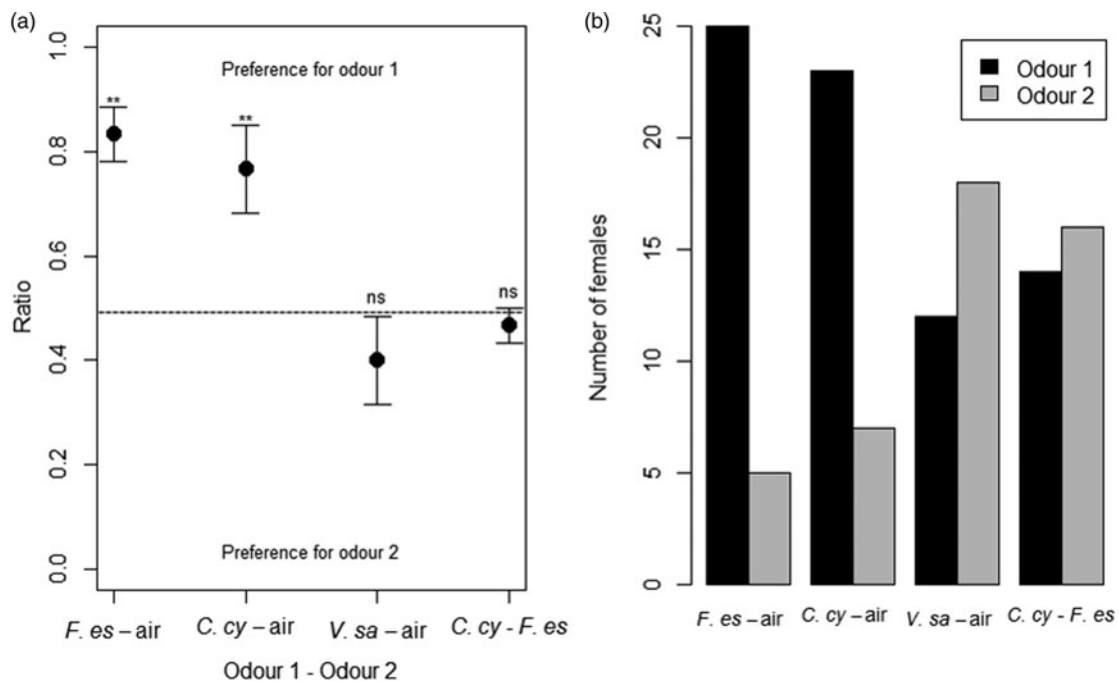


Fig. 3. (a) Proportion of females choosing odour 1. Values above the dotted line (expected frequency = 0.5) indicate a preference for odour 1 and below for odour 2. (b) Number of females choosing either odour 1 or odour 2. *F. es*, *Fagopyrum esculentum*; *C. cy*, *Centaurea cyanus*; *V. sa*, *Vicia sativa*. Pearson's χ^2 test, ** $P < 0.01$; ns: not significant ($P > 0.05$), $N = 30$ per treatment.

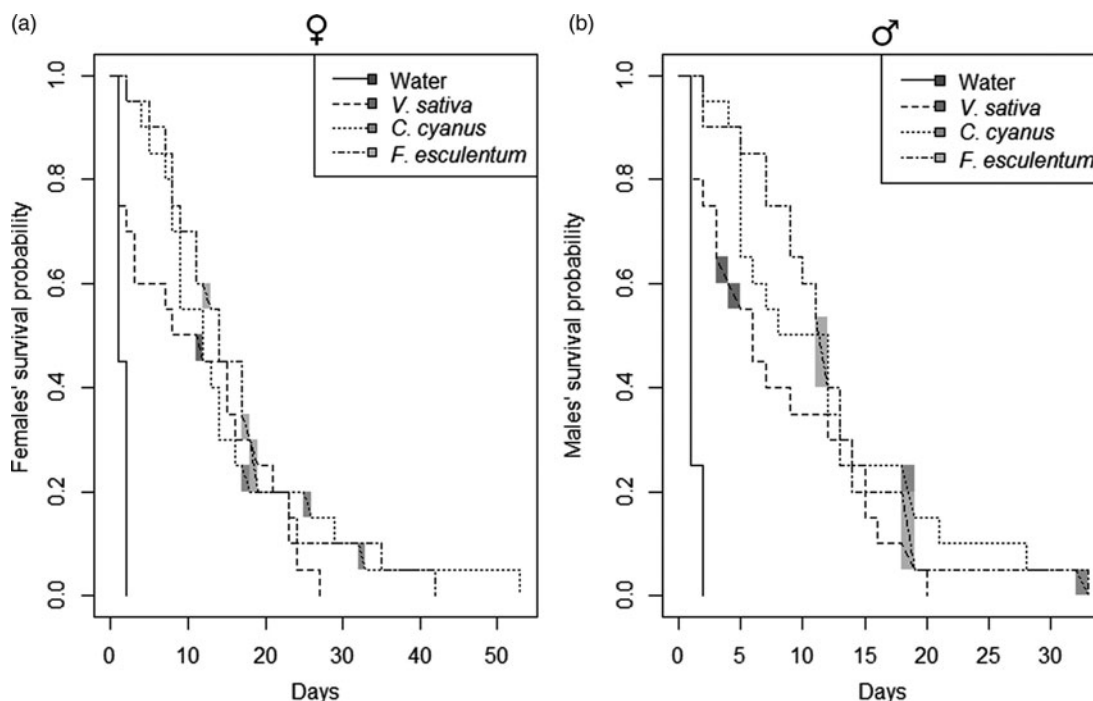


Fig. 4. (a) Survival of *Telenomus laeviceps* females in the presence of *Vicia sativa*, *Centaurea cyanus*, *Fagopyrum esculentum* and water. The three flowers significantly increased their longevity (asymptotic log-rank k -sample test, $N = 20$ per treatment, $\chi^2 = 17.87$, $P = 0.0005$). (b) Survival of *T. laeviceps* males in the presence of *V. sativa*, *C. cyanus*, *F. esculentum* and water. The three flowers significantly increased their longevity (asymptotic log-rank k -sample test, $N = 20$ per treatment, $\chi^2 = 20.517$, $P = 0.0001$).

model, $z = 1.666$, $P = 0.09$). *C. cyanus* and *F. esculentum* equally enhanced the fecundity of *T. laeviceps* females (generalized linear model, $z = -0.013$, $P = 0.989$), but significantly differed with *V. sativa* (generalized linear model, both $P < 0.03$) (fig. 5a).

The number of female offspring produced depended also significantly on the specific food source. Compared with *V. sativa* (25.5 ± 13.72 female offspring), parasitizing females produced significantly more female offspring in the presence of *C. cyanus* (generalized linear model, $z = -2.4$, $P = 0.016$) and *F. esculentum* (generalized linear model, $z = -2.42$, $P = 0.016$), respectively, 57.2 ± 13.94 and 55.2 ± 15.22 female offspring. The number of female offspring did not significantly differ between *C. cyanus* and *F. esculentum* (generalized linear model, $z = 0.024$, $P = 0.98$). No females were produced in the water control (fig. 5b).

The daily fecundity of *T. laeviceps* females was approximately the same in all treatments. Females were able to parasitize right after hatching on average 9.6 ± 2.9 eggs, with a rapid increase up to 26.1 ± 8 eggs on the second day. Female offspring was produced from the third to the fourth experimental day, while the production of males started from the first day and stayed constant until death of the parasitizing female.

Discussion

The aim of the present study was to clarify if selected flowers can increase the longevity and parasitisation performance of the biocontrol agent *T. laeviceps*, as well as attract them through volatile cues. Results clearly showed that *C. cyanus*

and *F. esculentum* enhance the performance of *T. laeviceps* and further, successfully attract them.

The olfactory attractiveness of the selected flowers is important in some biological control programmes, like conservation biocontrol, where natural enemies have to be attracted into the crop field (Jervis & Heimpel, 2007; Balmer *et al.*, 2013, 2014). In an augmentative biological control programme, natural enemies are released into the crop field, reducing the need to select highly attractive food sources. On the other hand, flowering strips are usually sown at the field margin and in big crop fields several hundred metres should be covered by parasitoids to reach them. If the sown flowers are not only beneficial, but also attractive, the food searching time can be considerably reduced and the *per capita* host searching efficiency increased (Jervis & Heimpel, 2007). Two of the three tested flower species, cornflowers and buckwheat, equally attract *T. laeviceps* and can successfully be applied to decrease their food searching time. As already pointed out, we were able to test only the extra-floral nectar of the common vetch. The lack of attractiveness of the volatiles released by the extra-floral nectar is in line to what was shown by G eneau *et al.* (2013) and Rose *et al.* (2006), for respectively, cornflower and cotton. Further experiments should be conducted to assess the olfactory attractiveness of common vetch floral nectar.

Once the parasitoid located the food source, it should be able to take advantage of it. Carbohydrates represent an important source of energy for many adult parasitoids (Leatemia *et al.*, 1995; Steppuhn & W ackers, 2004). A minor source of carbohydrate is represented by host-feeding, which is found to take place in some egg parasitoids (Rivero & West, 2005; Ferracini *et al.*, 2006), but was never been described for

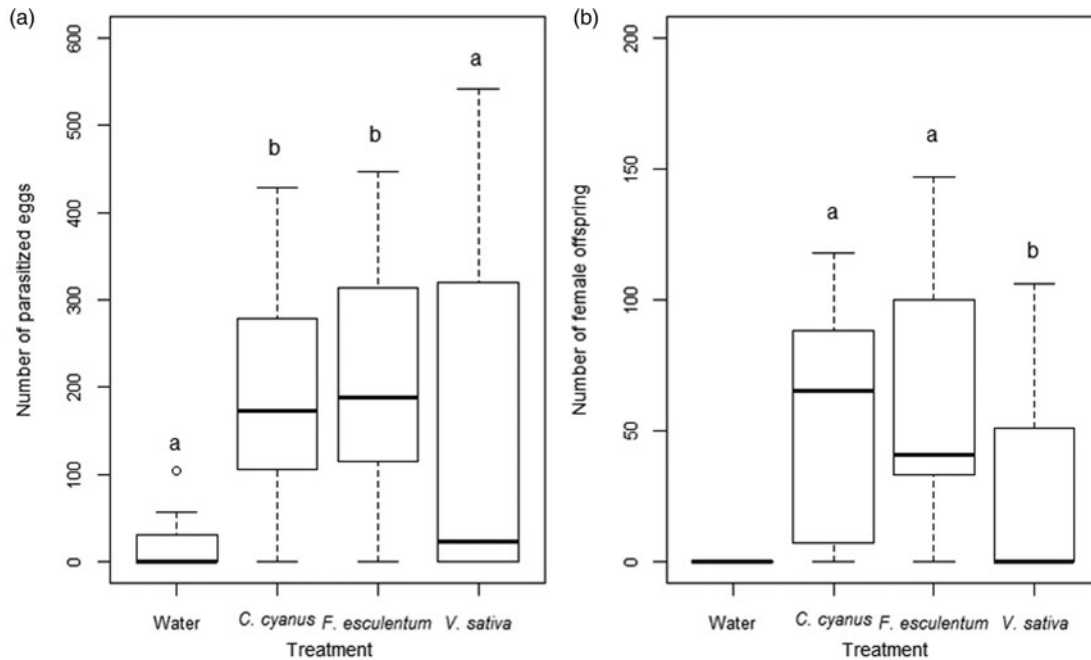


Fig. 5. (a) Number of parasitized eggs and (b) number of female offspring produced in the presence of the four treatments. Different letters indicate significant differences (generalized linear model, $P < 0.05$, $N = 10$ per treatment).

T. laeviceps. Despite that, in Lepidopteran eggs, carbohydrates are present as glycogen and most parasitoids, lacking the specific debranching enzyme, could not utilize it (Leatemala *et al.*, 1995; Romeis *et al.*, 2005). An important sugar source is represented by floral nectar, but not every flower is equally suitable for a particular insect. In fact, factors determining nectar accessibility, like floral morphology, are of crucial importance (Jervis, 1998; Jervis & Heimpel, 2007). Small-bodied parasitoids, such as *T. laeviceps* or *Trichogramma* spp., have difficulties exploiting floral nectar, because petals and stamen filaments can act as barriers (Patt *et al.*, 1997). Therefore, the presence of easily accessible extra-floral nectar and other exposed nectaries can sensibly enhance the fitness of these small parasitoids (Patt *et al.*, 1997; Jervis, 1998).

Our results clearly showed that *T. laeviceps* longevity is significantly enhanced by *C. cyanus*, *F. esculentum* and *V. sativa*. Both *C. cyanus* and *V. sativa* display extra-floral nectar, explaining the increased longevity of *T. laeviceps*. Therefore, also if extra-floral nectar of *V. sativa* does not attract *T. laeviceps*, combined with highly attractive flowers, like *C. cyanus*, can be successfully used to enhance this parasitoid. On the other hand, *F. esculentum* did not present any extra-floral nectar, but its simple floral structure allows *T. laeviceps* to easily reach the nectar, as already demonstrated for *Trichogramma* spp. (Witting-Bissinger *et al.*, 2008). Besides floral structure, nectar composition also plays an important role determining the suitability of a nectar source for the target parasitoid. The two main components of nectar are sugars and amino acids (Gardener & Gillman, 2002), the first being important for somatic maintenance and locomotion, while the second for egg manufacture (Bernstein & Jervis, 2008). The presented results indicate that the three flowers tested display an exploitable sugar composition, allowing the processing into blood sugar and glycogen, both important fuel of somatic functions (Bernstein & Jervis, 2008). Results about fecundity

of *T. laeviceps* reveal something interesting. Two out of the three tested flowers, *C. cyanus* and *F. esculentum*, equally increased the parasitoid performance, but *V. sativa* did not, although it increased their survival. This suggests that the nectar of *V. sativa* lacks some kind of component important for egg manufacture. As a moderate synovigenic parasitoid, egg-limitation is a major constraint for *T. laeviceps*, which emerge with a limited stock of mature eggs and need a suitable food source throughout their life to continuously produce those. An important component of the dietary intake of many insects responsible for egg manufacture is represented by amino acids (Mevi-Schütz & Erhardt, 2005; Bernstein & Jervis, 2008). Nectar is the most relevant amino acid source for insects and can significantly vary between flower species, as well as within the same family (Gardener & Gillman, 2002). The nectar amino acid composition of *V. sativa* was analysed by Gardener & Gillman (2002) and revealed a total amount of amino acids of 4581 ± 1928.1 pmol μl^{-1} of nectar. The total amount of amino acids present in the nectar of *C. cyanus* is similar to the one of *V. sativa*, namely 5496 ± 1627 pmol μl^{-1} of nectar (Gardener, personal communication). Looking at the data more carefully shows that the major difference in the nectar amino acid composition of these two flowers lies in the absence of proline in *V. sativa*, against the 1937 ± 360 pmol μl^{-1} of nectar present in *C. cyanus*. Proline is a rapidly metabolized amino acid, resulting in high levels of adenosine triphosphate (Hajirajabi *et al.*, 2016). In the egg parasitoid *Trissolcus grandis* (Thomson, 1861) (Hymenoptera: Scelionidae), proline added to a normal sugar-rich diet was shown to enhance fecundity (Hajirajabi *et al.*, 2016). With these results, we additionally confirm the importance of proline for the egg manufacture in egg parasitoids. Amino acid analysis of *F. esculentum* nectar could further confirm this point, by potentially showing a similar amount of proline as in *C. cyanus*.

The back-up of a released biocontrol agent through food provision could downsize the necessary number of parasitoid releases and ultimately reduce the costs for the end user. Our results clearly showed that *T. laeviceps* longevity and fecundity are significantly increased by *C. cyanus* and *F. esculentum*. These two flower species, together with *V. sativa*, are the main components of an already existing tailored flowering strip for brassica crops (Balmer *et al.*, 2013, 2014). In Switzerland, this flowering strip is implemented by farmers as part of a conservation biocontrol programme to promote natural enemies of different cabbage pests, like the parasitoids *M. mediator* and *Diadegma fenestrata* (Hymenoptera: Ichneumonidae) or predators like carabid beetles or spiders (Balmer *et al.*, 2013, 2014; Ditner *et al.*, 2013). Furthermore, there is evidence that the presence of non-target habitats, like flowering strips, give the released parasitoids the chance to overwinter in the proximity of the field, being already present at the beginning of the next growing season (Babendreier *et al.*, 2003; Kuske *et al.*, 2003). With the present work, we showed that *T. laeviceps* can take advantage of nectar sources and therefore that the combination of augmentative biological control with habitat management could lead to an even more efficient pest control in brassica fields, potentially reducing, or even replacing, the use of insecticides applied against the cabbage moth.

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