Artificial shelters enhance the establishment of the aphidophagous predator *Scymnus interruptus* on sweet pepper plants

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HIGHLIGHTS

- The aphidophagous predator *Scymnus interruptus* strongly preferred to use a shelter device as an oviposition site rather than sweet pepper leaves infested with its prey, *Myzus persicae*.
- The oviposition rate of *S. interruptus* was double when shelters were provided.
- *Scymnus interruptus* offspring population increased by twofold when shelters were provided under greenhouse conditions.

GRAPHICAL ABSTRACT
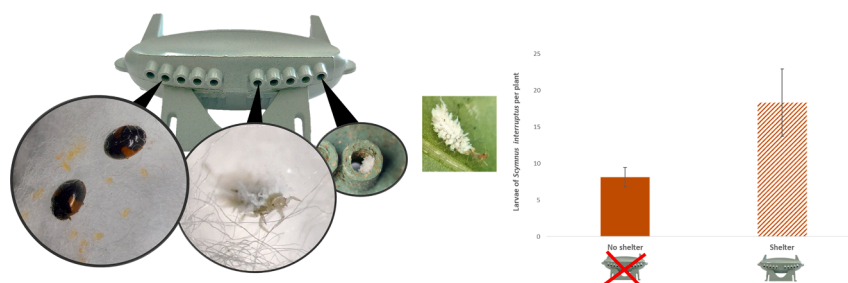
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ABSTRACT

The success of biological control programs often depends on the establishment of natural enemies in or around the crop. An emerging strategy that aims to improve establishment is through the provision of niches that act as shelters. These shelters can enhance the natural enemies' effectiveness by providing: a suitable microclimate; protection against other predators or pesticides; and/or alternative food. Generally, shelters are natural habitats such as flower strips, intercropped areas, hedgerows, or banker plants. The use of artificial devices as shelters remains underexplored. In this study, we assessed the functionality of an artificial shelter for the aphidophagous predator, *Scymnus interruptus* Goeze (Coleoptera: Coccinellidae). A tailor-made artificial shelter for oviposition was developed and tested in both laboratory and greenhouse conditions. Our results showed that *S. interruptus* females strongly preferred to use the shelters as an oviposition site compared with sweet pepper (*Capsicum annuum* L.) leaf discs supporting the aphid, *Myzus persicae* (Sulzer) (Hemiptera: Aphididae), its primary prey. Furthermore, *S. interruptus* oviposition rate was doubled when shelters were supplied. In a greenhouse experiment, population densities of *S. interruptus* larvae increased twofold when shelters were provided. Additionally, providing artificial shelters in a sweet pepper crop has the potential to enhance aphid control and could also be a valuable method for monitoring predator densities in the crop.

1. Introduction

Habitat management, also referred to as 'ecological engineering' is an emerging subdiscipline in the development of arthropod pest management programmes (Jonsson et al., 2008; Gurr et al., 2017a, b). It involves manipulation of an agroecosystem's structure with the aim of

suppressing pest densities by enhancing the impact of natural enemies (Landis et al., 2000; Landis et al., 2005; Bianchi et al., 2006; Gurr et al., 2017a, b). This manipulation generally includes: floral intercropping (Bianchi and Wäckers, 2008; Ramsden et al., 2015; Tschumi et al., 2015; Mei et al., 2021); field margin vegetation (Sengonca et al., 2002; Bischoff et al., 2016; Santos et al., 2018); and banker plants which provide shelter, nectar, alternative food (prey/hosts) and/or pollen for biological control agents (Huang et al., 2011; Lu et al., 2014; Karamouna et al., 2019; Snyder, 2019). Amongst all these resources, the provision of shelter remains underexplored and could be particularly beneficial in monocultures or annual crops, where resources are limited or unstable for the establishment of natural enemies (Landis et al., 2000; Thorbek and Bilde, 2004).

In open field conditions, shelters are green semi-permanent areas with vegetation that provide natural enemies with overwintering and/or reproduction habitats; protection against unsuitable climatic conditions through a better microclimate; and a reduced risk of intraguild predation (MacLeod et al., 2004; Griffiths et al., 2008; Sánchez et al., 2010; Snyder, 2019). Moreover, these habitats may act as dispersal corridors for beneficial insects that connect species-rich ecosystems with low diversity farmland (Hossain et al., 2002; Nilsson et al., 2016; Martin et al 2019). In addition, the presence of shelters within the crop reduces natural enemy mortality caused by tillage, harvesting processes and chemical control treatments (De Snoo, 1999; Marshall 2002). In contrast with open field crops, the use of green shelters in greenhouse crops is still an uncommon practice because sacrificing valuable cropping areas for this purpose is not usually feasible (Messelink et al., 2014). However, banker plants, which are used in greenhouse systems, have the potential to be used (to some extent) as green shelters. For instance, certain plants that possess acarodomatia can be particularly beneficial as refuges for predatory mites (Walter, 1996). Acarodomatia are minute pubescent areas located in the vein axils on the underside of leaves; the presence of these cavities increases predatory mite densities by boosting oviposition, acting as nurseries, or decreasing intraguild predation (Grostal and O'Dowd, 1994; Agrawal et al., 2000; Roda et al., 2000, 2001; Parolin et al., 2013). By improving establishment, predatory mites are able to colonize the crop more effectively and achieve early pest control (Parolin et al., 2013). Banker plants can also provide refuge from pesticides if they are removed temporarily from the greenhouse during necessary pesticide applications (Frank, 2010).

In addition to natural shelters formed from a variety of plant species, artificial shelters can be constructed and used as a valuable tool for supporting biological control strategies (Iuliano and Gratton, 2020). During the last twenty years, artificial shelters have been used in open agricultural landscapes for monitoring natural enemy population densities or as overwintering refuges (Horton 2004, Nicholas et al., 2005; Horton et al., 2006; Kawashima and Jung, 2010). However, to the best of our knowledge, they have never been used in greenhouse crops, where they have the potential to be particularly useful for supporting the establishment and reproduction of natural enemies in cost-intensive crops. The tendency towards a monoculture in greenhouses means that habitat complexity is limited; this can result in a lack of suitable oviposition sites for natural enemies (Messelink et al., 2014), and may also increase negative interactions such as intraguild predation, compared with more diversified cropping systems (Synder, 2019). Under these circumstances, it would seem likely that the provision of artificial shelters could increase habitat complexity and enhance biological control efficacy (Gontijo, 2019). Some previous studies have attempted to develop artificial acarodomatia as oviposition sites for predatory mites (Bresch et al., 2019; Loughner et al., 2019), but this has not been applied in practice yet.

In greenhouse crops, aphids are amongst the most destructive primary pests (Rabasse and Steenis, 1999; Blackman and Eastop, 2000; Blümel, 2004; van Emden and Harrington, 2017). Biological control strategies are mainly based on frequent releases of specialized aphid parasitoids and the predatory midge *Aphidoletes aphidimyza* (Rondani)

(Ramakers, 1989; Blümel, 2004); and additionally, chrysopid, syrphid or coccinellid predators when aphid densities are high. However, aphid populations remain difficult to suppress due to their high growth rates, the presence of hyperpredation, hyperparasitism or lack of natural enemy establishment when prey is scarce (van Schelt and Mulder, 2000; Messelink et al., 2011; Prado et al., 2015; Tougeron and Tena, 2019; Postic et al., 2020). Biological control of aphids might be greatly improved if natural enemies are able to establish in the crop prior to aphid infestation, which could enable a faster response to new aphid infestations thereby preventing aphid outbreaks.

Recent studies have identified the potential value that micro-coccinellids have in preventing early season aphid outbreaks (Singh and Mishra, 2016; Arruda, 2021), particularly species belonging to the genus *Scymnus* (Bouvet et al., 2019a,b; Rosagro et al., 2020; Bouvet et al., 2021; Borges et al., 2022). *Scymnus* predators are one of the most abundant aphidophagous species which appear early in the spring season, highly associated with the appearance of the first aphid colonies in the crop (Woin et al., 2006; Aroun et al., 2015; Bouvet et al., 2019a). This early presence makes *Scymnus* species key for preventing or limiting aphid outbreaks (Agarwala and Yasuda, 2001; Bouvet et al., 2021). *Scymnus* adult measure only about ~ 2 mm in length, significantly smaller than other key macrococcinelids. The reduced body size of *Scymnus* entails low predation rates, and the capability to survive and complete their entire life cycle when prey densities are low (Wilson, 1927; Slipiński et al., 2012). In addition, *Scymnus* adults can survive long periods of time when prey is absent (Buntin and Tamaki, 1980, Naranjo et al., 1990). These traits could make *Scymnus* a valuable preventive biological control agent for aphid suppression (Bouvet et al., 2021). Although *Scymnus* species seem to be promising candidates for early aphid control, previous studies in which *Scymnus* individuals were released did not succeed in probing their establishment (Askar, 2021).

In this study we evaluated whether provision of tailor-made oviposition shelters enhanced establishment of the predatory micro-coccinellid *Scymnus interruptus* (Goeze) (Coleoptera: Coccinellidae). *Scymnus interruptus* is predominantly aphidophagous but can also predate scale insects, mealybugs and spider mites (Tawfik et al., 1973; Bouvet et al., 2019a). We determined whether artificial shelter provisioning i) was preferred as an ovipositing site for *S. interruptus* ii) affected the preoviposition time and iii) oviposition rate under laboratory conditions. Subsequently, we assessed the efficacy of shelter provisioning under greenhouse conditions, specifically its effect on *S. interruptus* population density in the presence of an excess of aphids.

2. Material and methods

2.1. Insect culture

The micro-coccinellid *S. interruptus* was field collected in Limburg province, Netherlands and was subsequently reared at Wageningen University & Research for more than 20 generations. Insects were kept in climate cabinets (Snijders Micro Clima-series™) under constant conditions (25 °C, 70 % RH and a 16L:8D light:dark regime) and were fed *ad libitum* with the green peach aphid *Myzus persicae* (red phenotype). Aphids were reared for more than 20 generations on 50 cm high sweet pepper plants (*Capsicum annuum* L., cv. Maranello). All aphid stock colonies were kept under greenhouse conditions at 25 ± 2 °C, 65 ± 10 % RH and an ambient photoperiod.

2.2. Description of shelter devices

Shelter devices, commercialised as Bioclips©, were provided by Global-Biodesign (Belgium). The device consisted of a closed capsule of 10 × 3 × 3 cm, comprised of two halves: a cover and a concave structure located below the cover (see graphical abstract). The cover protected the interior from wind, rain, solar and UV radiation, and had ten × 2.4 mm diameter holes. These holes facilitated entry and exit of small insects and

the size was just sufficient for entry of *S. interruptus* adults (2 mm wide). The perimeter of each hole was surrounded by a protuberance that prevented entry of water. The other concave half of the device had 31 lateral holes each of 2.4 mm diameter facing down and 30 holes of 1 mm diameter on base. The two halves of the device were joined at two points via a hinge mechanism that enabled the cover to be opened. Opposite the two hinge joints there was a small platform that was intended to help insects enter through the holes. Following the European standard EN 17,033 and EN 13432, the device was entirely made of biodegradable materials (confidential composition) and was pale green in colour to mimic the environment of green plants.

2.3. Laboratory trial

2.3.1. Experimental arena

Arenas consisted of a closed plastic container (30 × 20 × 10 cm) Curver Grand Chef ©. Ventilation was provided via two 15 cm diameter holes covered with fine mesh. Inside each container, one small plastic cup (6 cm diameter, 6 cm depth) with a layer of 2 cm water agar (15 g/L) and a 6 cm diameter sweet pepper leaf on it was added. Sweet pepper leaf was immersed in the agar with its abaxial side exposed and infested with ca. 200 *M. persicae* individuals of mixed age. All sweet pepper leaves used in the experiments were of similar age (3 months old) with well-formed domatia at the axils of the leaf veins, as this location is preferred for *S. interruptus* oviposition (Pérez-Rodríguez personal observation). To determine whether the presence of shelters affected the preoviposition time, oviposition rate and fertility of *S. interruptus,* two different treatments were established: containers with one sweet pepper leaf disc and containers with one artificial shelter and one sweet pepper leaf disc. Shelters used were as described above and included a natural cotton wool ball inside as a known oviposition substrate (Pérez-Rodríguez personal observation). Ten replicates per treatment were conducted.

2.3.1.1. Effect of shelter on preoviposition time, oviposition rate and fertility of Scymnus interruptus. To assess the effect of artificial shelters on *S. interruptus* reproduction, a pair of adults was released into each container. Pairs consisted of one newly emerged female (<24 h) and one 10 days old male to ensure that egg fertility did not depend on males. Oviposition was recorded daily in each container for 22 days, both in leaf discs and shelters, and these were replaced every day in each container. To determine egg fertility, eggs were kept in climate cabinets (Snijders Micro Clima-series™) under constant conditions (25 °C, 70 % RH and a 16L:8D light:dark regime) with a supply of aphids and were checked daily until hatching was observed.

2.4. Greenhouse trial: Effect of shelters on Scymnus interruptus densities

The greenhouse trial was conducted in a 140 m² greenhouse compartment to determine the effect of shelters on *S. interruptus* density. The average temperature and relative humidity were 20 ± 3 °C and 74 %, respectively, and there was a 14L:10D light:dark regime. Both temperature and relative humidity were recorded every 5 min throughout the experiment using a climate recorder (Hoogendoorn Growth Management, Vlaardingen, The Netherlands).

A total of 16 six-week-old sweet pepper plants (*C. annuum* cv. Maranello) were placed individually in insect cages (60 × 60 × 90 cm, 650 μm mesh aperture, Vermandel, Hulst, The Netherlands). Plants were grown in rockwool slabs and were drip irrigated with a mix of water and a nutrient solution specific for sweet pepper crops (Sonneveld and Voogt, 2009). Two treatments were assigned at random: plants with no shelter or plants with shelter provision. In the shelter treatment, one shelter was installed per plant. Shelters used were as described above and included a natural cotton wool ball inside as a known oviposition substrate (Pérez-Rodríguez personal observation) Two 25 cm high

wooden sticks were fixed to the extreme edge of each shelter device and then inserted in the rockwool block. There were eight replicates per treatment.

To determine the effect of shelters on *S. interruptus* densities, plants were infested with high densities of *M. persicae,* to ensure that aphid densities were not the limiting factor for oviposition rates of *S. interruptus*. Aphid infestation was conducted by placing a sweet pepper leaf harbouring 30 mixed instar aphids (from 2nd to 4th) at the top of each plant. 24 h later, when aphids were settled in the plant, three pairs (one male, one female) of *S. interruptus* were then released in each cage. Based on our laboratory trials, and due to *S. interruptus* have a preoviposition period of around 6 days (Tawfik et al., 1973) all *S. interruptus* adults were 15 days old and selected randomly. To prevent sex determination mistakes, *S. interruptus* were paired in Petri dishes and only when mating was observed were couples selected. One week after *S. interruptus* release and once per week for four consecutive weeks, the number of aphids on the whole plant was recorded by visual counting. In the shelter treatment, the number of eggs of *S. interruptus* inside the shelter was counted once per week under the binocular microscope; during the experiment eggs on the plant were not counted in either treatment as they were hard to observe using a hand lens without destructive sampling. Four weeks after the introduction of *S. interruptus*, plants in both treatments were destructively sampled. In each cage, plants were cut, enclosed individually in plastic bags, transported to the lab and all stages of *S. interruptus* (eggs, larvae, adults) recorded under a binocular stereoscope. In the shelter treatment, mature and immature *S. interruptus* that remained in the shelters were also counted.

2.5. Data analysis

Preoviposition time (days before the first egg was laid) and total oviposition (eggs laid per female) were analysed using a generalized linear model (GLM) with a Poisson error distribution. Daily oviposition rate (eggs per female per day) was analysed using a generalized linear mixed effect model (GLMM) with a Poisson error distribution. Treatment was the fixed effect while female identity and time were the random effects. Egg fertility was calculated as the percentage of eggs hatched per ovipositing day and female and was analysed using a GLM with binomial error distribution and probit link. To assess the preference of *S. interruptus* females for ovipositing in the shelter or in the leaf when shelter was provided, a GLM with binomial error distribution and probit link was used.

In the greenhouse trial, aphid densities were analysed using a GLMM with a Poisson error distribution. Treatment was the fixed effect and time the random factor. Differences in *S. interruptus* densities between treatments were analysed using a GLM with Poisson error distribution. To deal with under- or overdispersion in the GLMs, we switched error distributions to quasipoisson for count data and quasibinomial for binary data (McCullagh and Nelder 1989). We used the glht function from the multcomp package for Tukey HSD tests/ *post-hoc* pairwise comparisons (Hothorn et al., 2008). All analyses were done using the statistical software R Version 4.0.2 (R Core Team 2021).

3. Results

3.1. Laboratory trial: Effect of shelter on preoviposition time, oviposition rate and fertility of Scymnus interruptus

The preoviposition period was not significantly different between treatments with or without shelters (GLM quasiPoisson: $F_{1, 16} = 16.29$; $P = 0.40$). In the no shelter treatment, *S. interruptus* females began to lay eggs 10 days after adult emergence and in the shelter treatment around nine days after adult emergence. During the whole laboratory trial, the daily oviposition rate of *S. interruptus* remained higher in the treatment where artificial shelters were provided (GLMM Poisson: $\chi^2 = 4.75$, df = 1, 368; $P < 0.05$) and achieved a maximum number of ten eggs laid

per day. In contrast, in the treatment without shelters, six was the maximum mean number of eggs laid per female (Fig. 1). Total oviposition (mean number of *S. interruptus* eggs per female during the trial) was significantly higher when shelters where provided (GLM quasiPoisson: $F_{1, 368} = 5.03$; $P < 0.001$). When shelters were absent, females laid around 50 eggs in total but when shelters were present, this increased by more than twofold (110 eggs) (Fig. 2). *Scymnus interruptus* showed a clear preference for egg-laying in shelters, in all occasions inside the cotton surface. During the oviposition time, when shelters were provided, they were in use on 99 % of the days, meanwhile leaf discs were only used on 32 % of the days (GLM quasibinomial: F $_{1, 205} = 5.24$; $P < 0.001$). Fertility was similar in both treatments. When shelters were provided, the hatch rate was 84 % and in the absence of shelters it was 86 % (GLM quasibinomial: $F_{1, 368} = 0.38$; $P > 0.05$).

3.2. Greenhouse trial: Effect of shelters on Scymnus interruptus densities and aphid population levels

Throughout the greenhouse trial, *S. interruptus* eggs were recorded in the shelters with a clear peak of 36 eggs per shelter in the third week (Fig. 3). Four weeks after *S. interruptus* releases, and in the destructive sampling, an average of two eggs were recorded per shelter. In contrast, no eggs were found on plants where shelters were installed and less than an egg per plant was recorded on plants with no shelters (Fig. 4). *Scymnus interruptus* larval density in cages where shelters were provided was twice that in the cages with no shelter (GLM quasiPoisson: $F_{1, 14} = 2.62$; $P > 0.01$). On average, 18 larvae were recorded per cage in the shelter treatment compared with eight in the plants without shelters. When the number of adults recovered was counted, no significant differences were found (GLM quasiPoisson: $F_{1, 14} = 1.11$; $P > 0.05$); in both treatments around five adults were recorded (Fig. 5). Aphid density did not differ throughout the trial between treatments and a mean of >500 aphids were recorded per plant (GLM quasiPoisson: F $_{1, 14} = 5.08$; $P > 0.05$ (Fig. 6).

4. Discussion

Our results show the potential for shelter provision to support natural enemies in greenhouse crops. To our knowledge, this is the first study demonstrating that the use of artificial shelters is a valuable tool to enhance establishment of biological control agents in augmentative programmes. The predatory ladybird *S. interruptus* showed a clear preference for ovipositing in the shelter rather than on sweet pepper leaves infested with aphids. Previous studies have reported that suitable oviposition substrates are selected by coccinellids based on visual, tactile and/or architectural cues (Gautam, 1990). For instance, *Adalia bipunctata* laid more eggs (90 % of the total) on filter paper rather than on plant tissue (10 %) when both surfaces were provided (Timms and Leather, 2007). *Coleomegilla maculata* (Coleoptera: Coccinellidae) and *Scymnus louisianae* (Coleoptera: Coccinellidae) showed an oviposition preference for leaves with trichomes, where eggs could be nested together (Griffin and Yeargan, 2002; Brown et al., 2003). One plausible explanation for selection of pubescence habitats such as domatia or cottony surfaces as oviposition sites, is that these areas are protected from other foraging predators that could be intraguild predators. Roda et al., 2000 and 2001 showed that female *Phytoseiulus persimilis* mites deposited more eggs when plant pubescence increased and that mite egg predation by thrips *Frankliniella occidentalis* (Thysanoptera: Thripidae) was significantly reduced when mite eggs were laid under webbing. In addition to protection against potential predators, a cottony substrate inside a shelter offers protection against abiotic stresses such as low humidity, wind or rain, and maintains a stable microclimate. Cottony surfaces could also be preferred for egg laying because of particular physical properties; some coccinellid species such as *Cryptolaemus montrouzieri* (Coleoptera: Coccinellidae) show positive thigmotaxis towards cotton for oviposition (Maes et al., 2014) and probing the substrate with the ovipositor is the final step prior to egg laying (Merlin et al., 1996).

Both our laboratory and greenhouse trials demonstrated that *S. interruptus* doubled its oviposition rate and larval densities when shelters were provided. This strong population increase might reflect the fact that *S. interruptus* females lay their maximum egg load per day when the oviposition substrate is optimal. As with other ladybird species, *S. interruptus* has a maximum and limited number of eggs that it can produce during its life, and it can refrain from oviposition when conditions are unsuitable (Tawfik et al., 1973; Pervez, 2004; Laubertie et al., 2006; Soroushmehr et al., 2008). In this context, their egg laying/oviposition rate can vary depending on various factors including prey availability, food quality, abiotic parameters or chemical cues (Hodek et al., 2012). Our results showed that, in addition to these parameters, the quality of the oviposition site can also encourage or discourage *S. interruptus* egg laying. Our results are even more striking knowing that
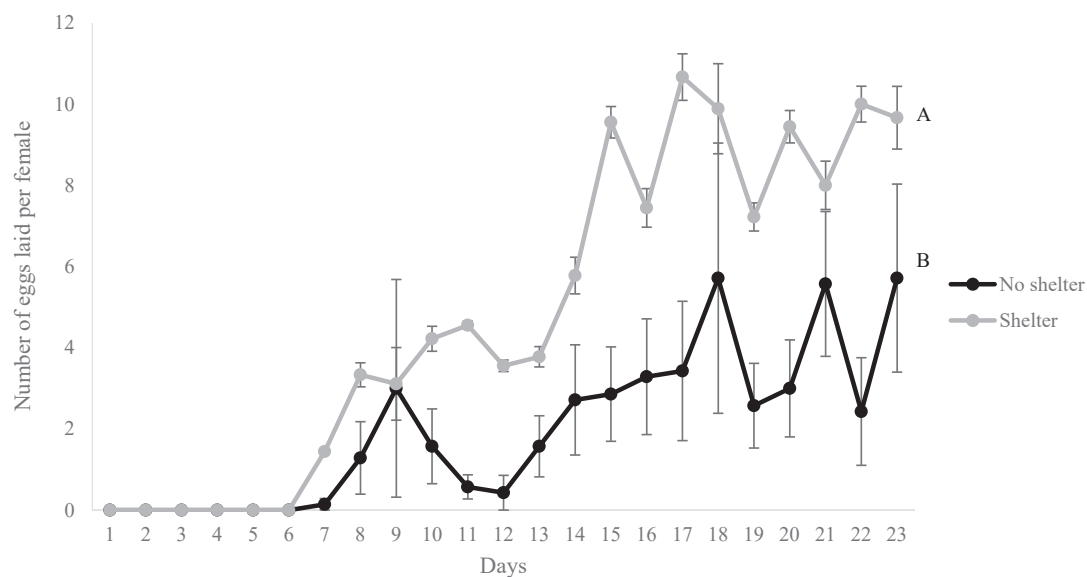

Fig. 1. Mean daily oviposition rate ($\pm$SE) of *Scymnus interruptus* females after adult emergence. Different letters indicate a significant difference between treatments ($P < 0.05$).
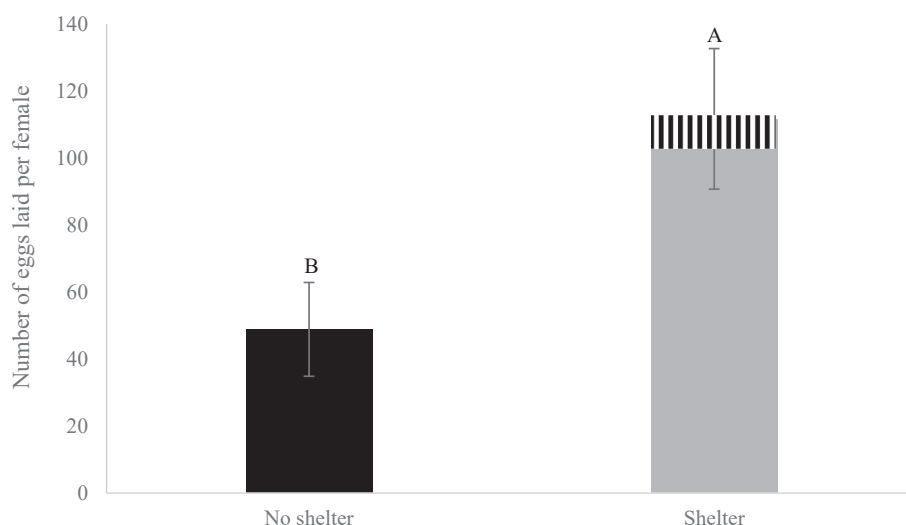
Fig. 2. Mean number of *Scymnus interruptus* eggs (±SE) laid per female over 22 days. The different letters on the bars indicates a significant difference between treatments ($P < 0.001$). The area of the bar with a striped pattern represents the mean number of eggs per female on leaf discs in the shelter treatment.
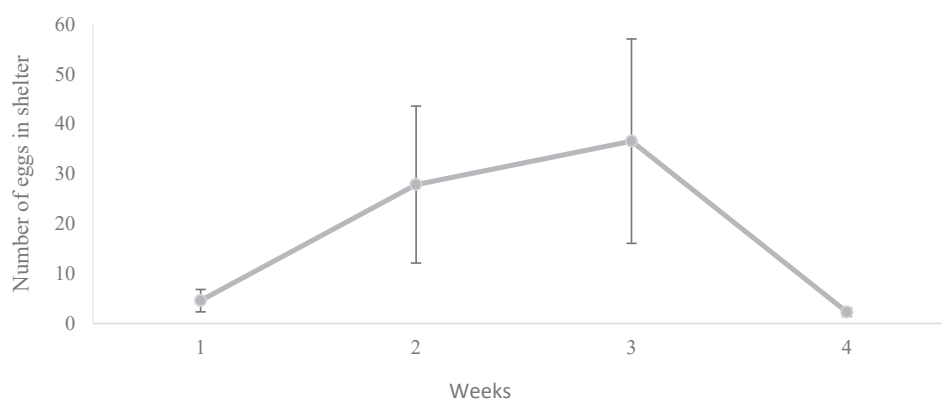

Fig. 3. Mean number of eggs (±SE) laid by *Scymnus interruptus* in the shelter over the 4-week trial.
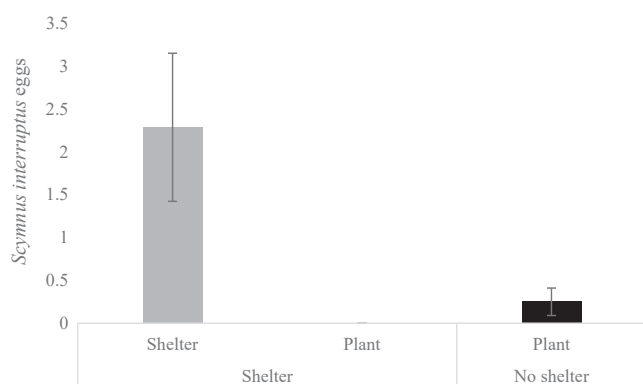

Fig. 4. Mean number of eggs (±SE) laid by *Scymnus interruptus* in the shelter device and on the plant in treatments with shelter and no shelters at the end of the trial.

sweet pepper leaves have tufts of dense hairs near the vein axils (Faraji et al 2002), which are also suitable oviposition sites. In fact, all eggs of *Scymnus* that were found on leaves were always found in these domatia, but apparently, the artificial shelter was still much more attractive for oviposition. A possible drawback of high oviposition rates is that they are negatively correlated with female longevity. Tawfik et al. (1973)

reported that when mated female *S. interruptus* were compared with unmated females (lower oviposition rate and unfertilised eggs), mated coccinellids had a shorter lifespan. Providing shelters might thus decrease female lifespan, although this will probably be largely compensated by the much stronger population increase.

The hatching rate of *S. interruptus* eggs was similar in the shelter and no shelter treatments; approximately 85 % of eggs hatched. This is in accordance with previous studies on other species in the genus *Scymnus*. For example, *Scymnus subvillosus* and *Scymnus apetzi* showed a hatch rate of circa 80 % (Atlihan and Kaydan, 2002), *Scymnus nubilus* 93 % (Borges et al., 2013) and *S. interruptus* circa 85 % (Bouvet et al., 2019b). Therefore, our results demonstrate that despite the increase in egg production, fertility of *S. interruptus* eggs remained similar throughout the trial in both treatments. Furthermore, it is worth mentioning that, when eggs hatched, first instar larvae showed positive phototropism and were able to colonise the entire plant immediately (personal observation). This behaviour, together with the low cannibalism described for *Scymnus* species (Völkl and Vohland, 1996) is an advantage for the practical implementation of artificial egg shelters in the crop.

Although our study clearly showed a strong increase in *S. interruptus* densities on sweet pepper plants when artificial shelters were provided, this did not result in greater suppression of aphids. The set-up of our trial was probably not suitable for evaluating this effectively. Aphid populations are known to grow exponentially with extremely high rm values (Lykouressis, 1984; Jarošík et al., 2003; Bell et al., 2008; La Rossa et al.,
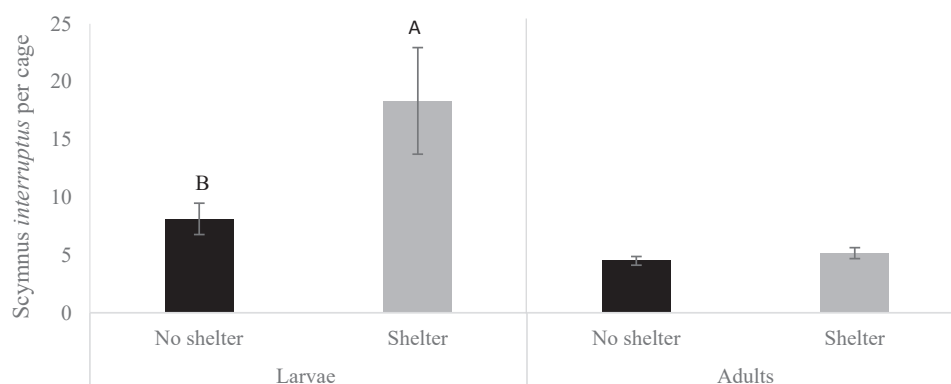
Fig. 5. Mean number (±SE) of *Scymnus interruptus* larvae and adults per cage. For each life stage, different letters on bars indicates a significant difference between treatments (P < 0.05).
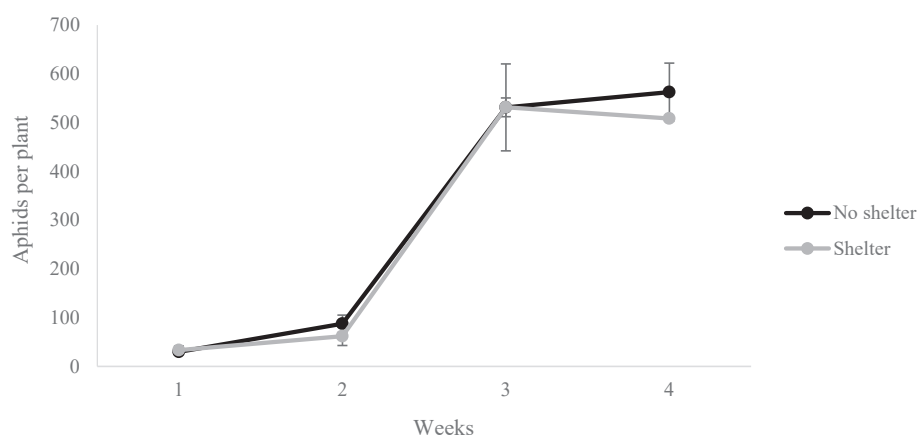

Fig. 6. Mean density (±SE) of green peach aphid *Myzus persicae* on sweet pepper plants with and without an artificial shelter over the 4-week trial.

2013) and the relatively low predator densities were probably insufficient in our study to significantly influence aphid population size, particularly because the micrococcinellids that belong to the genus *Scymnus* are small and have a relatively low predation capacity (Beltrà et al., 2017; Bouvet et al., 2019b; Bouvet et al., 2021). For instance, *Scymnus marginicolis* males predate around 2 *Myzus persicae* aphids per day and females around five (Buntin and Tamaki, 1980). Similarly, *S. nubes* adults predate less than four fourth instar aphids per day (Davidson, 1923). Beltrà et al. (2017) showed that *S. interruptus* females predate 27 *M. persicae* per day. However, these high values were obtained in small laboratory arenas and *Scymnus* individuals remained starved for 24 h. A similar trial with higher initial predator–prey ratios would then probably result in clearer effects on aphid suppression.

Preventive establishment of natural enemies, also known as a 'standing army approach', currently plays an important role in augmentative biological control programmes (Messelink et al., 2014; Pijnakker et al., 2017). It involves building a stable population of biological control agents while pests are still absent or in low densities. In our greenhouse trial, we showed that artificial shelters have potential to be helpful in this regard. *Scymnus interruptus* not only used shelters for oviposition but also as a resting and pupation site; in conjunction with low voracity, long longevity and the fact that *S. interruptus* can survive for long periods on pollen in the absence of prey (in preparation), this could be advantageous for preventive strategies aimed at aphid suppression. In addition, artificial shelters could facilitate monitoring or scouting tasks in greenhouse compartments. Nicholas et al., (2005) studied the phenology of predatory earwigs using cardboard bands in an apple orchard. Furthermore, Horton (2004) and Horton et al., (2006) were able to characterize the diapause and emergence phenology of

diverse predator species using cardboard bands as an overwintering habitat. Similarly, Kawashima and Jung, (2010) studied the overwintering ecology of different predatory mite species by providing artificial shelters in the ground. Our devices could be useful for recording overwintering periods, estimating population levels and assessing the establishment of *S. interruptus* over short periods of time.

It is worth mentioning that in our study, shelters were tailor-made based on the traits of *S. interruptus*. The entrance consisted of tiny little holes that restricted entry and use by larger coccinellids, such as *Propylea quatuordecimpunctata* (Coleoptera: Coccinellidae) or *Adalia bipunctata* (Coleoptera: Coccinellidae) adults; this also benefits *S. interruptus* establishment by providing refuge from intraguild predation by these larger coccinellids. Further development of tailor-made shelters for oviposition and survival of other natural enemy species might also have potential, particularly for species that are less well-adapted to crops with low trichome densities.

CRediT authorship contribution statement

JPR: Conceptualization, experimental design, data collection, data analysis and writing - original draft. **G.J.M:** experimental design, writing.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

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