Do ‘food sprays’ improve natural enemy performance?

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Summary

- Many insect natural enemies are omnivorous, in that they include extra-floral nectar, floral nectar, honeydew and pollen, as well their insect prey in their diet for at least part of their life. Access to these plant-based foods can reduce pest pressure by increasing both the individual ‘performance’ and local density of natural enemies. Commercial ‘food sprays’ that are rich in carbohydrates and proteins, such as Amino-Feed® and Envirofeast®, can be applied to cotton crops to act as artificial plant-based foods.
- We examine the extent to which Amino-Feed and Amino-Feed UV benefit a predator, the Pacific damsel bug, and a pupal parasitoid, the banded caterpillar parasite. These insects are important natural enemies of heliothis.
- The provision of either wet or dry residues of Amino-Feed UV had no discernable effect on Pacific damsel bug survival and development rates.
- In contrast, the provision of artificial nectar, cotton extrafloral nectar and Amino-Feed (plus extrafloral nectar) had a marked effect on banded caterpillar parasite longevity. Whilst fed female parasitoids lived longer, the total number of eggs they laid was unaffected by diet. This is because females exhausted their maximum egg supply around the same time across all treatments, and death occurred shortly thereafter in the water control treatment but after considerably longer in the other diets. Because females of the banded caterpillar parasite were also able to feed on small amounts of host body fluid, this alternative source of nutrition best explains why there was no effect of diet on fecundity (i.e., the number of eggs laid).
- It is likely that providing natural enemies with a supplementary source of food will be more important during periods of prey/host scarcity or absence. Thus, from the perspective of integrated pest management, artificial and natural plant-based foods are worthy of further investigation because they have potential to improve the ecosystem service of biological pest control.

Background information

Many insect natural enemies of pests are omnivorous, in that they include extra-floral nectar, floral nectar, honeydew and pollen, as well as prey in their diet for at least part of their life (Coll & Guershon 2002). There are two mechanisms by which plant-based foods can reduce pest pressure. First, improved nutrition may enhance one or more measures of natural enemy ‘performance’, such as longevity, fecundity or foraging behaviour. In this way, individual daily and/or lifetime attack rates may increase through expanded searching for location of suitable hosts/prey, and indirectly, through increasing rates of egg maturation and/or daily and lifetime reproductive output (Heimpel & Jervis 2005). Second, natural enemies may be attracted to, and aggregate around, patches containing these foods, resulting in increased parasitism or predation by virtue of their greater localised density, rather than increased individual attack rates.

Various habitat manipulation tactics have been proposed to boost the provision of plant-based foods to natural enemies in crops (Landis et al. 2000). Although flowering non-crop ‘insectary’ plants situated within or surrounding crop areas can provide natural enemies with plant-based foods (e.g., Pontin et al. 2006), there are a several drawbacks associated with this approach. These include lost income on land devoted to the non-crop, disruption to standard agronomic practices, establishment and maintenance costs (particularly if the plant becomes invasive) and critically, only brief periods of food provisioning are generally possible with this approach. As most plants only flower for a brief period, at best they provide a sudden abundance of food for a short period, which is generally preceded and followed by scarcity of food. A potentially more refined and controlled habitat manipulation tactic that has been proposed involves the application of high carbohydrate and/or protein solutions to crops to act as artificial supplemental food sources for natural enemies (e.g., Ben Saad & Bishop 1976; Mensah & Singleton 2003). Typically the underlying rationale is to improve the synchrony of herbivore and natural enemy populations through time and space. However, the influence of artificial-plant based foods on the performance of key species of natural enemies has seldom been investigated under controlled laboratory or glasshouse conditions (e.g., McEwen et al. 1993, 1996).

The present study examines the extent to which the commercial artificial plant-based foods Amino-Feed® and Amino-Feed UV® benefit a predator, the Pacific damsel bug, *Nabis kinbergii*, and a pupal parasitoid, the banded caterpillar parasite, *Ichneumon promissorius*. Both insects are important natural enemies of various species of heliothis, *Helicoverpa* spp. (Lepidoptera: Noctuidae), which are major worldwide pests of arable crops such as cotton, soybean and sorghum (Johnson et al. 2000). Specifically, we examined how Amino-Feed and Amino-Feed UV affects: (i) rates of survival and development of the predator in the laboratory; and (ii) the longevity and fecundity of the parasitoid in the glasshouse. Additionally, two high carbohydrate-low protein diets were tested for the parasitoid as a comparison with Amino-Feed, which is high in both carbohydrates and proteins.

Procedures

The artificial plant-based food used in Trial 1 was Amino-Feed UV. This commercially available liquid formulation has increased stability to ultra-violet light compared to standard Amino-Feed and fewer mixing and application problems compared with wettable powder and granule formulations. Nonetheless, standard Amino-Feed was used in Trial 2 because this was the only formulation readily available at the time. The two formulations vary only in the addition of 10 % UV protectant, with

Amino-Feed containing 24.2 % sugars, 21.3 % crude protein and amino acids and 1.8 % nitrogen (product label data; Agrichem Manufacturing Industries, Loganholme, Queensland).

Trial 1 – Survival and development of the Pacific damsel bug

Insects and Amino-Feed UV application

Pacific damsel bug nymphs were supplied with excess heliothis eggs as prey and a water-soaked cotton dental roll for moisture both prior to and during the trial, but not during Amino-Feed UV application. Newly emerged first-instar nymphs were provided with either wet and thereafter dried residues (hereafter labelled 'wet residue') or dry residues only (hereafter referred labelled to as 'dry residue' treatment) of Amino-Feed UV. Wet residue and dry residue water controls were included. Each treatment was replicated 10 times. A total spray volume of 141.7 l per ha was used in order to convert the Amino-Feed UV dilution rate from 3 l per hectare to 21.2 ml per litre. The sprayer used was a 500 ml plastic hand held atomiser. The inner surfaces of 10 polystyrene enclosures were sprayed with each treatment on 29 October 2001. In the wet residue treatments, 10 healthy one-day old nymphs were placed in each enclosure prior to spraying. Conversely, in the dry residue treatments, nymphs were introduced one-hour after spraying, with the spray having dried after 5 minutes. Predators were then transferred to a controlled environment room.

Assessments and analysis

The number of predators that remained alive was initially assessed after two days exposure to determine the immediate effects of treatment exposure. To test for longer-term effects, individuals were transferred to ventilated enclosures with prey and a moist dental roll, but were free of potential Amino-Feed UV residues (to limit feeding time, as might happen in the field). In order to maintain a density of 10 predators per enclosure (few had died during the initial exposure period), individuals from each treatment were assigned to new replicate cages with ca. 10 predators per enclosure. The number of predators alive and their life-stage was recorded daily over the next five days. The rate of insect development was calculated based on the number of days elapsed since the start of the trial until at least fifty percent of live predators in each enclosure had reached the third-instar. The number of predators that remained alive when more than 50% of predators in an enclosure had reached the third-instar was also recorded. It was predicted that rates of predator survival and development would be enhanced in the Amino-Feed UV treatments, particularly in the wet residue treatment where predators had access to the product before it dried as well as afterwards. It is likely that insects with sucking mouthparts such as Pacific damsel bugs may have difficulty feeding on dry residues. A series of generalized linear models in the program SAS were used to statistically analyse the data.

Trial 2 – Longevity and fecundity of the banded caterpillar parasite

Insects, plants and Amino-Feed application

Potted cotton plants (cv. Delta Emerald) produced copious amounts of extrafloral nectar from numerous foliar and bracteal extrafloral nectaries throughout the trial. Newly emerged adult banded caterpillar parasites were provided with one of the following four treatments: (1) cotton extrafloral nectar plus Amino-Feed UV, (2) cotton extrafloral nectar, (3) artificial nectar, or (4) a water only control. Treatments were replicated five times, with two newly emerged male-female pairs of the parasitoid in each, and arranged randomly in the glasshouse. The cotton plants used in both extrafloral nectar treatments were individually enclosed by cylindrical covers made from fine white polyester gauze, supported by wooden rods inserted into the soil. Enclosures used in the artificial nectar and water only treatments were constructed from aluminium frame and covered with gauze. Amino-Feed mixed with water at the rate of 10ml per litre was sprayed (equipment described earlier) onto the upper-leaf surface and squares and flowers of cotton plants at 0, 9, 23, 37 and 51 days after the trial commenced on 13 February 2001. The wasps were temporarily removed from the enclosures during spraying to prevent them from escaping and returned before the spray had fully dried. 1-2 ml of honey contained in a Petri dish lid was added to the artificial nectar treatment enclosures. Honey was used as an artificial nectar source because it contains fructose, glucose and sucrose as well as trace amounts of amino acids, which are the main components of floral and extrafloral nectar from most plants (Baker & Baker 1983). Water was provided in all treatments by way of a filled glass vial with a cotton wick protruding through a hole in the cap.

Assessments and analysis

Parasitoid survival under the different treatments was assessed daily until all had died. Starting on day 1, parasitoid fecundity was assessed every other day by placing two pupae contained in a Petri dish lid in each enclosure for the proceeding 48 h. Assessments ceased once both females in each enclosure had died or 69 days after spraying (DAS). Although it is possible for individual female wasps to each attack more than two hosts in 48 hours, only two host pupae were provided during this period because of the low numbers available. Hence, rather than analyse the number of hosts attacked, realised fecundity was indicated by the total number of eggs deposited in a few hosts (individual female parasitoids will readily attack the same host more than once). Potentially parasitised pupae were examined under a microscope to record the number of discernible blackened scars or ‘sting spots’ present on the exoskeleton. The number of sting spots indicates deliberate acts of non-destructive feeding on host body fluid, which reflects in part on the nutritional status of parasitoids in the various treatments. Pupae were then dissected to record the number of parasitoid eggs. The numbers of sting spots and eggs deposited in both hosts was adjusted for the number of female parasitoids alive at the start of each two-day census period to produce measures of per capita reproductive output. A series of survival analyses and ANOVA tests were used to analyse the data.

Findings

Trial 1 – Survival and development of the Pacific damsel bug

Immediate and longer-term survival

There was no immediate effect of treatments on survival of the Pacific damsel bug at 2 DAS ($P > 0.05$) by product type, exposure manner or the interaction of product type and exposure manner. Of the survivors that were transferred to residue free enclosures at 2 DAS, their longer-term survival under the various treatments approximately 4.5 DAS was not significantly affected by product type, exposure manner or the interaction of product type and exposure manner. This corresponded to when more than 50% of predators that remained alive had reached the third-instar (Table 1).

Development time

The time elapsed from the start of the trial until the majority of surviving predators had reached the third-instar under the various treatments was not significantly affected by product type or the interaction of product type and exposure manner. However, the rate of immature development was significantly affected by exposure manner. In the wet residue treatment (pooled across product types) predators required 0.57 days or 14% longer to complete development of the first- and second-instars compared with the dry residue treatment (i.e., 4.7 ± 0.1 vs. 4.1 ± 0.1 days, respectively) (Table 1).

Table 1. Immediate and longer-term survival and immature development rates of Pacific damsel bug nymphs under various treatments

Product type	Exposure manner	Mean ± SE number alive 2 DAS†	Mean ± SE number alive when ≥ 50% had reached third-instar‡	Mean ± SE DAS until ≥ 50% had reached third-instar
Amino-Feed UV	Wet residue	8.5 ± 0.4	8.9 ± 0.6	4.8 ± 0.1
Amino-Feed UV	Dry residue	8.4 ± 0.7	9.3 ± 0.5	4.1 ± 0.1
Water control	Wet residue	8.1 ± 0.6	8.1 ± 0.7	4.6 ± 0.2
Water control	Dry residue	8.8 ± 0.8	9.1 ± 0.2	4.1 ± 0.1

† 10 insects were placed in the enclosures at the start of the trial
‡ Surviving insects were transferred in groups of 10 to residue free enclosures 2 DAS (days after spraying). This explains why the number alive in the enclosures at the first survival assessment at 2 DAS was lower than the number alive at the second assessment approximately 4.5 DAS.

Trial 2 – Longevity and fecundity of the banded caterpillar parasite

Longevity

Diet had a significant effect on both male and female parasitoids. The mean (± SE) longevity of male parasitoids was the shortest when provided with water only (9.8 ± 0.3 days) compared with the other diets (artificial nectar, 27.8 ± 5.0; cotton extrafloral nectar, 31.8 ± 7.7; Amino-Feed plus extrafloral nectar, 29.2 ± 5.2 days) (Figure 1a). Female parasitoids lived longer than males and produced a similar response in that longevity was shortest when provided with water only (27.6 ± 4.0 days). Females lived the longest in the diet of Amino-Feed plus extrafloral nectar (65.5 ± 6.1 days), artificial nectar (51.6 ± 4.6), and then cotton extrafloral nectar (43.1 ± 8.4) (Figure 1b).

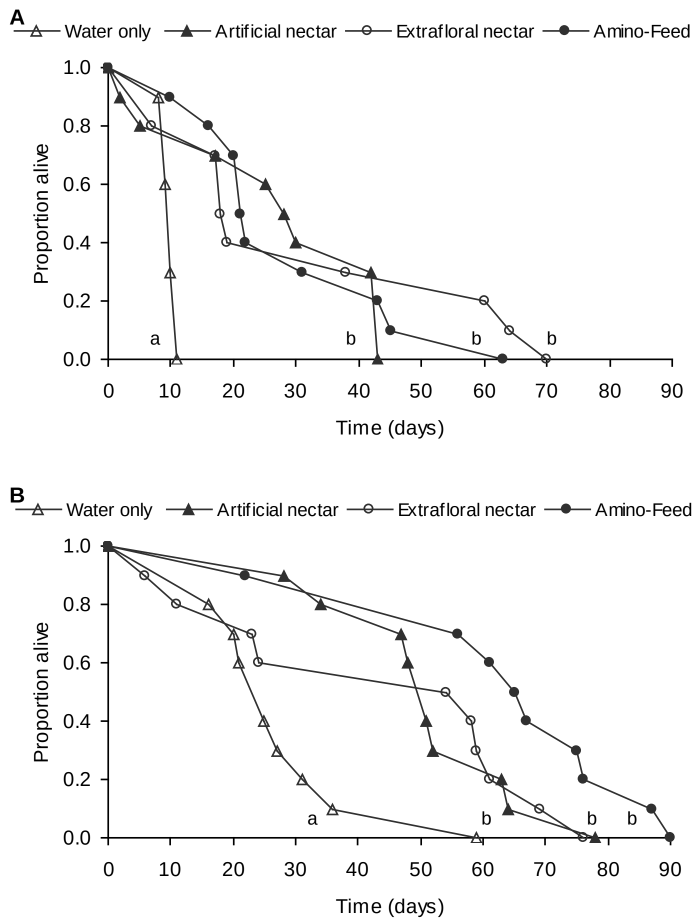


Figure 1. Survivorship curves of male (A) and female (B) banded caterpillar parasites under various treatment diets. Different letters denote a significantly different survivorship based on pair wise survival analysis comparisons.

Lifetime fecundity

Whilst the longevity of female parasitoids in the water only treatment was significantly reduced, the lifetime fecundity of an average female parasitoid was not significantly affected by diet (Figure 2). Similarly, the total number of sting spots was not significantly affected by diet (Figure 2).

Age-specific fecundity

The numbers of eggs deposited or sting spots by an average female parasitoid in two hosts in 48 hours was not significantly affected by the provision of the various diets (Figure 3). However, the numbers of eggs deposited and sting spots by an average female parasitoid varied significantly between the 19 census dates. In general, the numbers of eggs deposited and sting spots rapidly increased in the first nine days, then remained fairly constant for the next 30 days, and was followed by a gradual decline between days 39 and 67. The interaction of diet and census date was not significant for either the numbers of eggs deposited or sting spots (Figure 3).

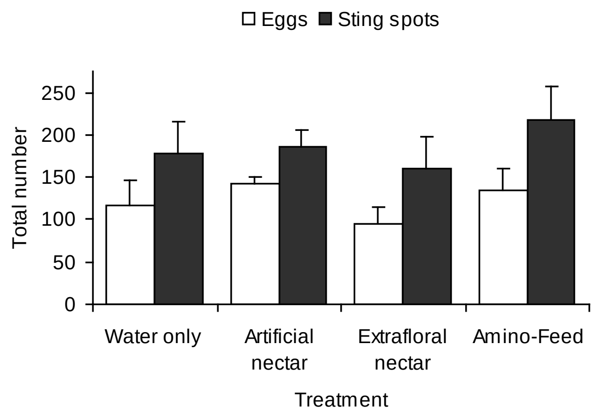
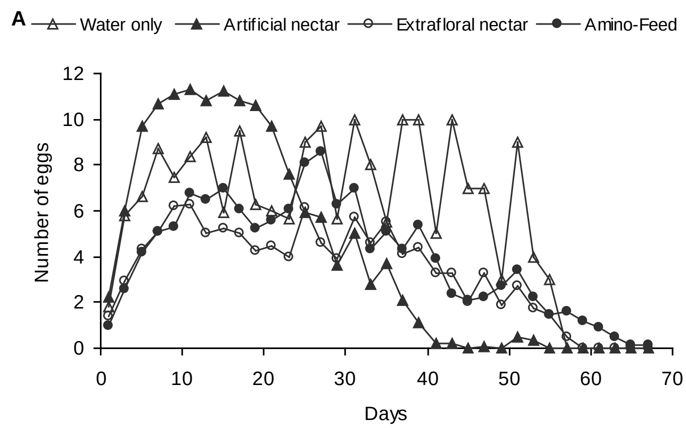


Figure 2. Mean (+ SE) numbers of eggs laid and sting spots in hosts per female banded caterpillar parasite over its lifespan under various treatment diets. The numbers of eggs laid and sting spots were not significantly affected by diet.



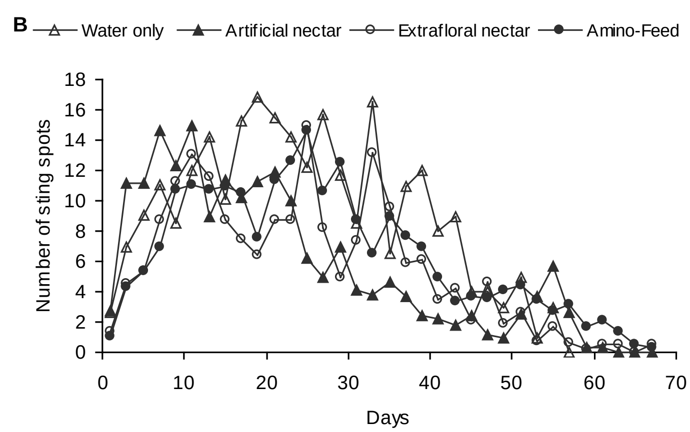


Figure 3. Mean numbers of eggs laid (A) and sting spots (B) in hosts per female banded caterpillar parasite in 48 hour periods over its lifespan under various treatment diets. The numbers of eggs laid and sting spots were significantly affected by census date, but not by diet and the interaction of diet and census date.

Implications

For Pacific damsel bugs

The provision of either wet or dry residues of Amino-Feed had no discernable effect on Pacific damsel bug survival and development. While no formal observations were made of the feeding behaviour by Pacific damsel bugs during the trial, on several occasions bugs were seen with mouthparts extended, apparently feeding on both wet and dry Amino-Feed residues. This indicates that predators were indeed using the sprayed Amino-Feed, but were not nutritionally limited in the absence of an artificial plant-based food. Thus, adequate nutrition was provided by the surfeit of heliothis eggs presented to predators in all treatments, regardless of whether Amino-Feed was provided as wet or dry residues or not at all.

Whilst the addition of Amino-Feed had no discernable impact on Pacific damsel bugs when excess prey were made available, the provision of food sprays may be more valuable during periods of prey scarcity or absence. This could buffer them against an unpredictable supply of food resources. Food resources can be quite variable for predators in ephemeral agricultural systems, such as during periods following planting, harvesting or spraying of insecticides (Coll & Guershon 2002). It is possible that Amino-Feed may be more important when only nutritionally inferior prey species are available, such as pea aphids, compared with nutritionally superior prey such as heliothis (Eubanks & Denno 2000). In addition, life stages of Pacific damsel bugs other than first-instar nymphs, such as adult females, may benefit more from Amino-Feed due to slight differences in their nutritional requirements. Indeed, these differences may be more pronounced in insect species that undergo complete metamorphosis (e.g., parasitoid wasps such as the banded caterpillar parasite); a process that allows the immature and adult stages to consume different food sources.

For banded caterpillar parasites

In contrast with Pacific damsel bugs, the provision of Amino-Feed (plus extrafloral nectar) increased the longevity of males and females of the banded caterpillar parasite by 3.0 and 2.4 fold, respectively, compared with the water only diet. This provides clear evidence of carbohydrate-limitation in banded caterpillar parasites, whilst the similar result achieved in the Amino-Feed (plus extrafloral nectar) diet, which is high in both carbohydrate and protein, and the extrafloral nectar only diet that is high in carbohydrate only, indicates that parasitoids were either not protein limited or the Amino-Feed component of the former treatment was not well utilised. While no formal observations were made of the feeding behaviour by banded caterpillar parasites during the trial, wasps were seen lapping at the leaf surface, apparently feeding on dry Amino-Feed residues on several occasions, as well as at extrafloral nectaries and the cotton wick containing artificial nectar. Indeed, the ability to feed on dry sugar residues has been documented in numerous species of natural enemies, including parasitoid wasps (Bartlett 1962). Hence, it is likely that this parasitoid was able to imbibe the different foods, but there was no additive effect of Amino-Feed plus extrafloral nectar compared with extrafloral nectar only.

While female-banded caterpillar parasites that received supplementary food lived an additional 15.5-37.9 days on average, their age-specific and lifetime fecundity were remarkably no different to those in the water control diet. The difference between the two measures can be explained by female wasps having exhausted their egg supply in all treatments around 60 DAS, which was before death. Hence, in the water control treatment females died shortly thereafter, while in the three high carbohydrate diets they stayed alive for considerably longer, so extending this post-exhaustion period would confer no reproductive gain.

The most plausible explanation for diet not having a significant affect on parasitoid fecundity is that body fluid from feeding on heliothis pupae provided females in all treatments with adequate nutrition for egg development (a portion of eggs mature gradually over their lifetime) (Godfray 1994). Informal observations revealed that female wasps regularly fed upon host body fluid that exuded from the site where the ovipositor was previously inserted. The explanation that host feeding alone could provide females with adequate nutrition is consistent with the similar number of sting spots having been recorded across the treatments; host feeding was predicted to be more frequent in the water control, but no evidence supported this hypothesis. Thus, the present result for the banded caterpillar parasite provides evidence to support the view that the addition of artificial and natural plant-based foods to host-based foods provides no additional resources used for reproduction, and thus no resultant improvement in fecundity. Indeed, the most limiting factor on reproductive success in this parasitoid is probably host-based food.

In conclusion, the findings of the present study suggest it is possible to alter the survival, but not reproduction of the banded caterpillar parasite, by providing a source of artificial or natural plant-based foods. Indeed, the dichotomous response for the species tested emphasises that we cannot assume that one plant-based food suits all. In particular, the lack of a positive response to Amino-Feed (plus extrafloral nectar and hosts) for the banded caterpillar parasite or to Amino-Feed UV (plus prey) for the Pacific damsel bug casts doubt over the suitability of this specific strategy of applying artificial plant-based food to crops. Nonetheless, in the context of integrated pest management, artificial and natural plant-based foods have potential to improve the ecosystem service of biological pest control in targeted crops by providing natural enemies with an alternative source of food (e.g., Ben Saad & Bishop 1976; McEwen et al. 1996; Mensah & Singleton 2003). Artificial plant-based foods could also be used in combination with classical, inoculation and inundation biological control methods to potentially improve the success of the release strategies in an approach dubbed ‘integrated biological control’ (Gurr & Wratten 1999).

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References

Baker HG, Baker I, 1983. A brief historical review of the chemistry of floral nectar. In: Bentley B, Elias T (Eds.), The Biology of Nectaries. Columbia University Press, New York, pp. 126-152.

Bartlett BR, 1962. The ingestion of dry sugars by adult entomophagous insects and the use of this feeding habit for measuring the moisture needs of parasites. J. Econ. Entomol. 55, 749-753.

Ben Saad AA, Bishop GW, 1976. Effect of artificial honeydews on insect communities in potato fields. Environ. Entomol. 5, 453-457.

Coll M, Guershon M, 2002. Omnivory in terrestrial arthropods: mixing plant and prey diets. Ann. Rev. Entomol 47, 267-297.

Eubanks MD, Denno RF, 2000. Healthy food versus fast food: the effects of prey quality and mobility on prey selection by a generalist predator and indirect interactions among prey species. Ecol. Entomol. 25, 140-146.

Godfray HCJ, 1994. Parasitoids: Behavioral and Evolutionary Ecology. Princeton University Press, Princeton, New Jersey, USA.

Gurr GM, Wratten SD, 1999. 'Integrated biological control': a proposal for enhancing success in biological control. Int. J. Pest Manag. 45, 81-84.

Heimpel GE, Jervis MA, 2005. Does floral nectar improve biological control by parasitoids? In: Wäckers FL, van Rijn PCJ, Bruin J (Eds.), Plant-Provided Food for Carnivorous Insects: A Protective Mutualism and its Applications. Cambridge University Press, UK, pp. 267-304.

Johnson M-L, Pearce S, Wade M, Davies A, Silberbauer L, Gregg P, Zalucki M, 2000. Review of beneficials in cotton farming systems. Report to CRDC, Narrabri, NSW (86 pp).

Landis DA, Wratten SD, Gurr GM, 2000. Habitat management to conserve natural enemies of arthropod pests in agriculture. Annu. Rev. Entomol. 45, 175-202.

McEwen PK, Jervis MA, Kidd NAC, 1993. Influence of artificial honeydew on larval development and survival in *Chrysoperla carnea* (Neur., Chrysopidae). Entomophaga 38, 241-244.

McEwen PK, Jervis MA, Kidd NAC, 1996. The influence of an artificial food supplement on larval and adult performance in the green lacewing *Chrysoperla carnea* (Stephens). Int. J. Pest Manag. 42, 25-27.

Mensah RK, Singleton A, 2003. Optimum timing and placement of a supplementary food spray Envirofeast® for the establishment of predatory insects of *Helicoverpa* spp. in cotton systems in Australia. Int. J. Pest Manag. 49, 163-168.

Pontin DR, Wade MR, Kehrli P, Wratten SD, 2006. Attractiveness of single and multiple species flower patches to beneficial arthropods in agroecosystems. Ann. Appl. Biol. 148, 39-47.