

## Role of *Diadegma semiclausum* (Hymenoptera: Ichneumonidae) in Controlling *Plutella xylostella* (Lepidoptera: Plutellidae): Cage Exclusion Experiments and Direct Observation

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*We evaluated the role of the larval parasitoid, Diadegma semiclausum Hellén (Hymenoptera: Ichneumonidae), in controlling Plutella xylostella (L.) (Lepidoptera: Plutellidae) by cage exclusion experiments and direct field observation during the winter season in southern Queensland, Australia. The cage exclusion experiment involved uncaged, open cage and closed cage treatments. A higher percentage (54–83%) of P. xylostella larvae on sentinel plants were lost in the uncaged treatment than the closed (4–9%) or open cage treatments (11–29%). Of the larvae that remained in the uncaged treatment, 72–94% were parasitized by D. semiclausum, much higher than that in the open cage treatment (8–37% in first trial, and 38–63% in second trial). Direct observations showed a significant aggregation response of the field D. semiclausum populations to high host density plants in an experimental plot and to high host density plots that were artificially set-up near to the parasitoid source fields. The degree of aggregation varied in response to habitat quality of the parasitoid source field and scales of the manipulated host patches. As a result, density-dependence in the pattern of parasitism may depend on the relative degree of aggregation of the parasitoid population at a particular scale. A high degree of aggregation seems to be necessary to generate density-dependent parasitism by D. semiclausum. Integration of the cage exclusion experiment and direct observation demonstrated the active and dominant role of this parasitoid in controlling P. xylostella in the winter season. A biologically based IPM strategy, which incorporates the use of D. semiclausum with Bt, is suggested for the management of P. xylostella in seasons or regions with a mild temperature.*

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## INTRODUCTION

The diamondback moth, *Plutella xylostella* (L.) (Lepidoptera: Plutellidae), is the most destructive pest of cruciferous crops worldwide, particularly in the tropics and subtropics where it has increasingly developed resistance to almost all major classes of insecticides (Cheng, 1988; Tabashnik *et al.*, 1990; Talekar & Shelton, 1993). The resultant crisis in management has led to increasing interest in the development of biologically based integrated management systems for *P. xylostella*, in which the role of parasitoids is maximized to reduce reliance on insecticides (Ooi, 1992; Talekar & Shelton, 1993; Verkerk & Wright, 1996; Liu & Sun, 1998; Saucke *et al.*, 2000).

*Diadegma semiclausum* (Hellén) (Hymenoptera: Ichneumonidae) is one of the most important larval parasitoids of *P. xylostella* (Waterhouse & Norris, 1987; Talekar & Shelton, 1993). The parasitoid was introduced from Europe into New Zealand and Australia in the 1940s and later to many other Asia-Pacific regions (Waterhouse & Norris, 1987; Talekar & Shelton, 1993). It is now the dominating parasitoid of *P. xylostella* in Australia and New Zealand (Yarrow, 1970; Goodwin, 1979; Waterhouse & Norris, 1987), as well as in the relatively cooler highlands of other Asia-Pacific regions (Talekar & Shelton, 1993; Saucke *et al.*, 2000). However, in the hotter lowlands of many Asia-Pacific regions, *Cotesia plutellae* Kurdjumov (Hymenoptera: Braconidae) is the dominant larval parasitoid of *P. xylostella* (Talekar & Yang, 1991; Talekar & Shelton, 1993; Liu *et al.*, 2000; Saucke *et al.*, 2000). Quantitative comparison of host-searching behaviour between these two larval parasitoids showed that *D. semiclausum* is better adapted to its host's defensive behaviour, and is more effective at detecting and parasitising *P. xylostella* larvae than is *C. plutellae* under temperature conditions suitable for both species (Wang & Keller, 2002). Because *D. semiclausum* prefers a relatively cooler temperature range of 15–25°C (Talekar & Yang, 1991), its lack of tolerance to high temperature conditions may have limited its distribution in hotter climates (Talekar & Yang, 1991; Amend & Basedow, 1997; Saucke *et al.*, 2000).

In order to reduce pest pressure in hot climates, cruciferous vegetables are typically cultivated in the relatively cool seasons or cooler highlands of many tropical regions (Amend & Basedow, 1997; Saucke *et al.*, 2000). However, *P. xylostella* is still a serious pest that requires suppression even in these relatively cool habitats due to its extremely wide tolerance to temperature (Talekar & Shelton, 1993; Liu *et al.*, 2002). Because *D. semiclausum* also favours the relatively cool habitats, this study evaluated the role of natural enemies, particularly *D. semiclausum*, in controlling *P. xylostella* in winter broccoli in southern Queensland, using cage exclusion experiments and direct field observation on the foraging efficiency of the parasitoid.

Cage exclusion is a relatively rapid means of demonstrating the impact of natural enemies on pest population and has been widely used in biological control programs (DeBach *et al.*, 1976; Luck *et al.*, 1988). The principle underlying this approach is that pest populations on sentinel plants from which natural enemies have been excluded, suffer lower predator-induced mortality or parasitism than populations on plants to which natural enemies are allowed access. This information can be used to illustrate the effectiveness or shortcomings of existing natural enemies (DeBach *et al.*, 1976).

*Diadegma semiclausum* is a specialist on *P. xylostella* (Wang & Keller, 2002). In southern Queensland both *D. semiclausum* and *P. xylostella* dominate in crucifer crop fields during winter when other crucifer pests and parasitoids of *P. xylostella* occur at very low levels (Wang, 2001). Thus, the winter provides an ideal season for direct observation on the foraging behaviour of field *D. semiclausum* population in southern Queensland, when the possible interference effects of other insect species are minimized. An effective parasitoid

should concentrate search in areas of high host density to maximize the rate of host attack. Such a strategy results in an aggregated response of parasitoid foraging time on high host density patches (Waage, 1983; Wang & Keller, 2002). We directly observed the response of field *D. semiclausum* populations to plants experimentally infested with different densities of *P. xylostella* larvae in order to understand how the parasitoid exploits host resources during the winter season.

## MATERIALS AND METHODS

### Cage Exclusion Experiment

The experiment was conducted in a broccoli (Pacific) field (50 × 20 m) in 1999 at the Gatton Research Station (27°37'S, 153°18'E) in southern Queensland, Australia. The field was divided into three similar-sized plots subjected to different pest management practices: integrated pest management (IPM), conventional insecticides spray schedule and an unsprayed control. During the experiment, the plots were monitored weekly for pest levels by randomly sampling 10 plants per plot. When a control threshold of four to six small *P. xylostella* larvae per 10 plants was reached, the IPM plot was sprayed with *Bacillus thuringiensis* Berliner (Xentari, 750 mL/ha, Valent BioSciences) with a wetting agent (Agral, Syngenta) while the conventional insecticide spray plot was sprayed with either Phosdrin (mevinphos, 65 mL/ha, Rotam Chemical Co.) or Secure (chlorfenapyr, 400 mL/ha, BASF Chemical Co.). No insecticides were applied to the unsprayed control plot. Standard agronomic practices were used to grow the broccoli.

The experiment was conducted in each of three pest management systems twice in succession during the winter. The first trial started on 4 July, soon after transplanting, and the second trial started on 5 August and finished with the harvest of the broccoli in September. Each trial consisted of four treatments of sentinel plants: (a) closed cage; (b) open cage; (c) open cage with a sticky barrier (selective cage); and (d) no cage. The cylindrical cage (40 × 40 cm) was constructed of fine wire net (approximately one cell per cm) and covered by a fine nylon mesh sleeve. The mesh size (25 cells/cm) was sufficiently small to exclude all natural enemies except some tiny *P. xylostella* egg parasitoids such as *Trichogramma*. Each cage was held in place by three bamboo stakes and the bottom edge of the cage was buried approximately 10 cm in the soil. In the closed cage treatment, the sleeve was tied at the top to allow access only for sampling. In either the open or selective cage treatments, the cage was left open at the top to allow access of natural enemies, yet still maintaining an environment similar to that in the closed cage treatment. Additionally, with the selective cage treatment, a sticky barrier was applied to the bottom of the cage (a piece of 4-cm wide black sticky table was first placed around the outside surface of the cage, the table was then covered by water-proof sticky TAC-GEL, Rentokil Pty. Ltd., Australia), to selectively exclude entry of walking natural enemies directly from the ground.

The sentinel broccoli plants were grown in 14-cm diameter pots in a greenhouse until each plant had six to eight fully expanded leaves. The potted plants were exposed to *P. xylostella* adults in a laboratory cage (40 × 40 × 40 cm) until each plant contained enough *P. xylostella* eggs for the trials. The initial *P. xylostella* egg density per plant ranged between 15 and 16 for the first trial and 21 and 25 for the second trial. These densities were necessary for the detection of any treatment effects, but were low enough to ensure that no plants became severely defoliated during the experiments. All the experimental plants were carefully checked for the number of *P. xylostella* eggs and any extra eggs removed. Prior to hatching of the *P. xylostella* eggs, the sentinel plants were moved to the field and were randomly allocated to different treatments and plots. Each cage treatment was replicated four to eight times and any two adjacent treatments were 7 m apart.

Immigration of wild *P. xylostella* adults onto the sentinel plants would be inevitable in the open and uncaged treatments. Therefore, the plants in those treatments were checked at 3–5-day intervals to remove any newly laid *P. xylostella* eggs. When most of *P. xylostella* larvae

on the sentinel plants had nearly completed their development, the trial was terminated, and the numbers of *P. xylostella* larvae and pupae, and *D. semiclausum* cocoons on the sentinel plants recorded. All the *P. xylostella* larvae and pupae were collected and individually maintained in vials (3 × 5 cm) under laboratory conditions (25 ± 2°C, 50–70% RH) until either *P. xylostella* or parasitoid adults emerged.

In order to investigate the possible activities of egg parasitoids of *P. xylostella* in the field, 10 potted broccoli plants each containing 16–23 freshly laid *P. xylostella* eggs (collected within 12 h in the laboratory) were placed 5–7 m apart into the unsprayed plot four times during the course of the two trials. Each potted plant grown in 14-cm diameter pot had six to eight fully expanded leaves, and stood at about the same height as the field plants in the field. The positions of these *P. xylostella* eggs on the leaves were marked nearby with blank ink. After a 3-day exposure in the field, the plants were collected and the number of eggs remaining recorded. The eggs were reared in the laboratory to determine the levels of egg parasitism.

During the weekly monitoring of the pest population level in the field, the number of *D. semiclausum* cocoons was recorded. Although comparison of parasitoid cocoon and *P. xylostella* larval density does not give an accurate assessment of parasitism, such a measure could indicate the trends of both *P. xylostella* and the parasitoid populations in the field.

The percentage losses and parasitism of *P. xylostella* cohorts on the sentinel plants were calculated to provide an estimate of the overall impact of natural mortality and particularly the role of *D. semiclausum*, and to compare the differences among different pest management practices and treatments. All proportional data were transformed by arcsin square root before an analysis of variance (ANOVA, LSD test, Statistix for Windows 4.1, Microsoft, 1998 Analytical software), and were back-transformed to proportions for presentation. Results of each trial were first compared among different treatments within the same pest management practice and among different pest management practices within the same treatment, respectively. If a significant effect of pest management practice was detected, the data were subjected to further tests on the possible interaction effect between the different pest management practice and cage treatment.

### Direct Observation

Direct observation on the response of field *D. semiclausum* populations to broccoli plants deliberately infested with different densities of *P. xylostella* larvae were carried out adjacent to a cabbage field as well as the above broccoli field during the winter. The cabbage field was about 200 m away from the broccoli field and there was no other brassica crop field within several kilometres around the observation sites. The cabbage field plants, transplanted in April 1999, had been heavily infested by *P. xylostella* as no control practices were applied. Sampling in August showed that 95.6 ± 5.6% (mean ± SE) ( $n = 20$  plants) of the *P. xylostella* larvae and pupae were parasitized by *D. semiclausum*. In contrast, in the broccoli field density of *P. xylostella* larvae gradually increased in July but decreased in August with an increasing number of *D. semiclausum* present (see results of above experiment).

A total of nine observation experiments were undertaken on sunny and calm days from 30 July to 2 September, with the first four observations beside the cabbage field and the last five beside the broccoli field. On each observation date, two plots each consisting of 16 potted broccoli plants infested with three different densities of *P. xylostella* larvae were set up on the bare soil 3 m upwind of the parasitoid source fields to intercept naturally occurring and dispersing *D. semiclausum* adults (Figure 1). The host density (number of plants per density treatment) was 0 (8), 2 (4) and 4 (4) in the low host density plot, and 0 (8), 4 (4) and 16 (4) larvae in the high host density plot. In order to determine if the location of an observation site would influence the response of parasitoids to the set-up host plant plots, three

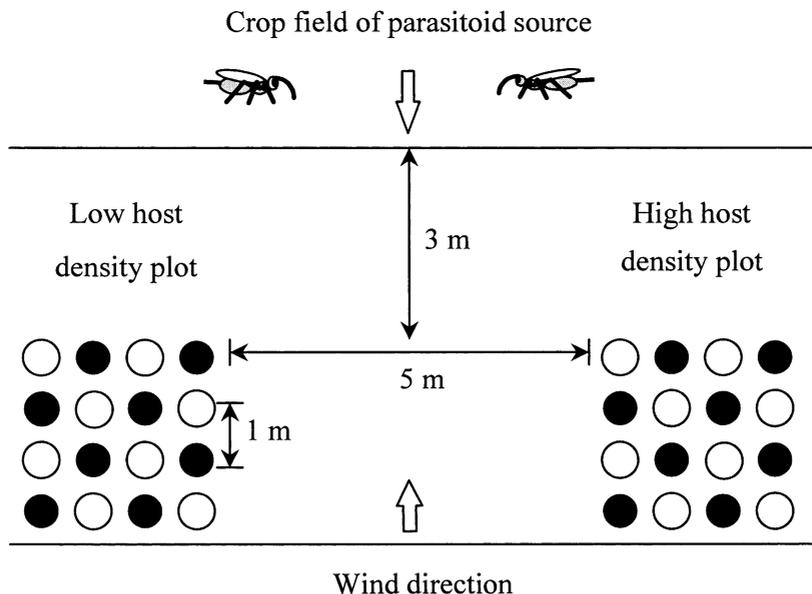


FIGURE 1. The layout of field observation: two different host density plots each consisting of 16 potted broccoli plants either infested (black circles) with *P. xylostella* larvae or uninfested (white circles) were set up next to resource fields of naturally occurring *D. semiclausum* population. The host density of per plant (number of plants per density treatment) was 0 (8), 2 (4) and 4 (4) larvae in the low host density plot, and 0 (8), 4 (4) and 16 (4) larvae in the high host density plot. The plants were assigned by a random block design to 4 × 4 grids, i.e., each row or column contains two different host density plants, respectively.

observations with two using only the high host density plot arrangement were simultaneously carried out on 30 July at three different sites 3 m upwind of the cabbage field.

The potted broccoli plants were grown in a glasshouse until four to five fully expanded leaves were present. The plants were infested in the laboratory by second and third instar *P. xylostella* at the densities required 24 h prior to the observations, and moved into the field on the morning of each observational date. Before setting up the experimental plots, all infested plants were checked to ensure that the exact number of host larvae were present. A yellow mesh cloth was spread over the bare soil of each plot to enhance the observation. A 4 × 4 1-m grid was marked on the cloth, and the plants were assigned in a random block design to the 4 × 4 grids, i.e., each row or column having four plants with three levels of different host density (Figure 1). A tray was placed under each pot in an attempt to catch any larvae dropping following attacks by the parasitoid. In this way we were able to recover the host larvae after the observations. A 1.5-m high mesh fence was set-up 5 m upwind of the experimental arena to protect the observational arena from occasional wind gusts.

Observations started from 08:30 to 10:30 and finished from 15:30 to 16:30 (1 h break from 12:30 to 13:30). The start time of each observation depended on the field conditions, with the exception of the last three observations, which ended at 12:30. There was no apparent wasp activity in the field when the plant surface was still wet early in the morning, or when the temperature started dropping in the late afternoon. During the winter, daily temperatures, recorded at a weather station located about 500 m from the experimental field, varied from 5 to 25°C. However, during the day time the temperature varied from ca. 15 to 25°C (Wang, 2001).

At 10-min intervals, observers walked around the plots and checked the plants from all sides, and recorded all *D. semiclausum* wasps on each experimental plant. With practice it was possible to census the plants quickly and to record virtually all the wasps on the experimental plants. In most cases one to four wasps were observed during a recording interval. If a wasp was observed, the observer then walked close to the plant, and checked the sex of the parasitoid with minimal disturbance. At the end of each observation, all larvae were collected and dissected to determine the presence of parasitoid eggs. Mean percentage recovery of the experimental larvae was 84–96%. Parasitism was calculated based on the recovered hosts.

Waage (1983) pioneered the study of parasitoid foraging behaviour in the field. We used a similar method as he used to estimate the relative amount of time spent by searching wasps on different plants based on the number of wasps observed per plant per unit time. In order to test if the parasitoid population may respond differently at different scales to local variation in host density and distribution, the data were analyzed at two levels: (1) parasitoid response to variation in host density to individual plants; and (2) parasitoid response to variation in host density to a group of plants (plot).

We used an aggregative index ( $A$ ) to compare the degree of the parasitoid's aggregation response relative to the host density index ( $D$ ) at the two spatial levels across the observational dates. At the plant level,  $A_{\text{plant}}$  = number of wasps observed on the high host density plants/total number of wasps observed on the whole plot, and  $D_{\text{plant}}$  = number of hosts present on the high host density plants/total number of hosts present in the whole plot (which were 0.67 and 0.80 for the low and high host density plot, respectively). At the plot level,  $A_{\text{plot}}$  = number of wasps observed on the high host density plot/total number of wasps observed on the two plots, and  $D_{\text{plot}}$  = number of hosts present in the high host density plot/total number of hosts present in the two plots (which was 0.77). If the parasitoids distribute themselves in proportion to hosts or aggregate more on patches with high host density, i.e.,  $A \geq D$ , the positive aggregation response by parasitoids should generate a direct density-dependent parasitism (Hassell & May, 1974; Hassell, 1982). The resultant pattern of percent parasitism was compared in relation to the relative degree of aggregation response across the observational dates using Student  $t$ -test with the adjusted Sequential Bonferroni Method (Rice, 1989).

## RESULTS

### Cage Exclusion Experiment

*Diadegma semiclausum* was the dominant parasitoid of *P. xylostella* from the collections of *P. xylostella* larvae and pupae on the sentinel broccoli plants during the winter in southern Queensland, accounting for 95% of all parasitoids collected. Occasionally, two pupal parasitoids, *Diadromus collaris* (Gravenhorst) and *Brachymeria* sp., and one fungal agent, *Zoophthora* sp. were recorded. Predators (mainly spiders and Coccinellids) were occasionally observed in the field.

Weekly monitoring showed that *P. xylostella* population density increased quickly soon after the broccoli plants were established but decreased with increasing *D. semiclausum* population in the unsprayed plot (Figure 2A). Overall, both *P. xylostella* and *D. semiclausum* population density were higher in the unsprayed control plot than in the conventional insecticide spray schedule or IPM plot (Figure 2A,B). The conventional insecticide spray schedule plot was sprayed using mevinphos or chlorfenapyr and the IPM plot was sprayed using *Bt*, each on three occasions, during the whole growing season of the crop (Figure 2A).

Within each cage treatment, there was no significant difference in the mean percentage of *P. xylostella* recovered, including those that were parasitized, among the three different pest management practices in both trials (Table 1). Within each pest management practice, both trials showed that the *P. xylostella* populations suffered the greatest loss in the uncaged

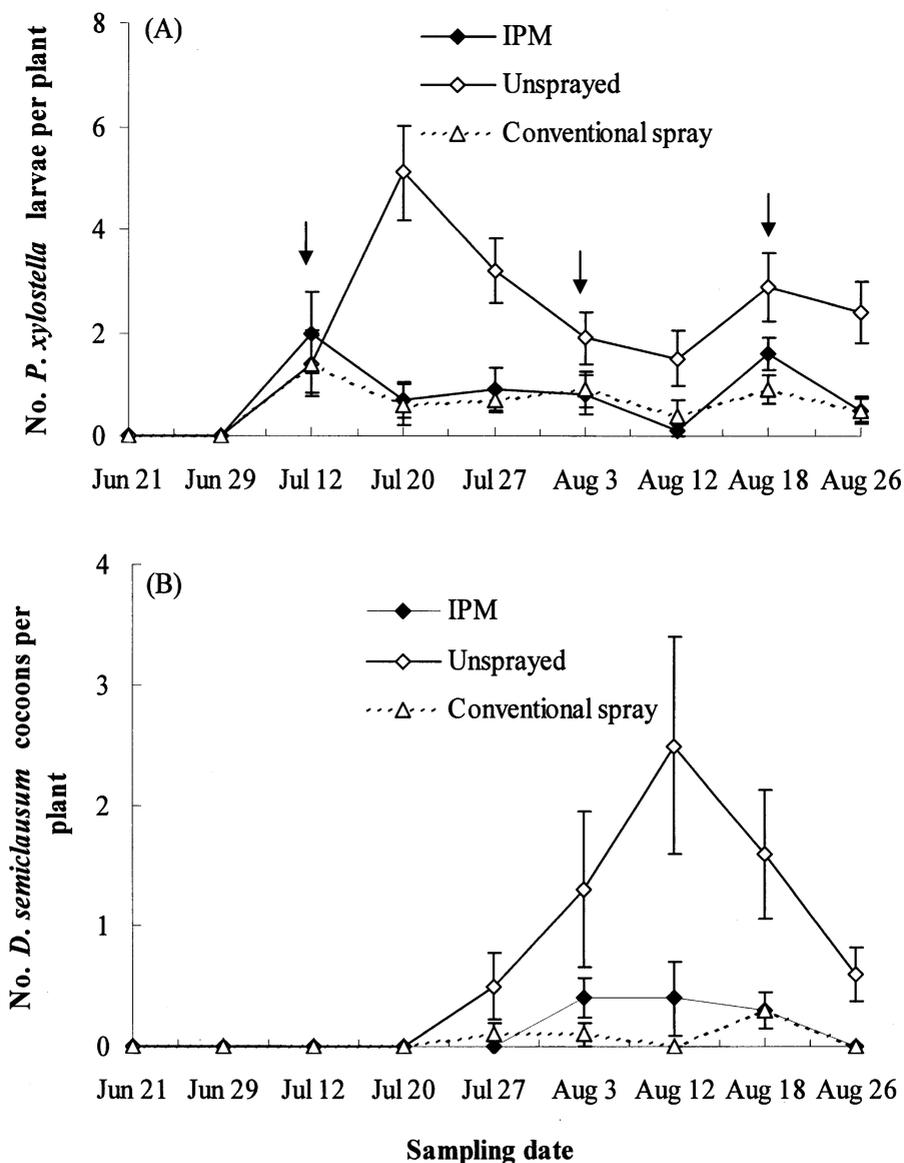


FIGURE 2. Seasonal abundance (Mean  $\pm$  SE) of *P. xylostella* larvae (A) and *D. semiclausum* cocoons (B) in plots with different pest management practices. Arrows indicate spraying dates in both IPM and conventional insecticide spray schedule plot.

treatments and there was no significant difference in the mean percentage of *P. xylostella* recovered, between the three pest management treatments (Table 1).

There was no significant difference in the overall percent parasitism of *P. xylostella* among different pest management practices for each cage treatment (Table 2). However, within each pest management practice, 72–94% of the larvae that remained were parasitized by *D. semiclausum* in the uncaged treatments, much more than in the open or selectively

TABLE 1. Percentage (mean  $\pm$  SE) of recovered *P. xylostella* at the end of each experiment from different treatments in plots of different pest management practices

| Treatment      | Replicates | Initial egg density | % Population recovered from different plots |                  |                  |
|----------------|------------|---------------------|---|------------------|------------------|
|                |            |                     | IPM   | Unsprayed        | Conventional     |
| First trial    |            |                     |   |                  |                  |
| Closed cage    | 4          | 15–16               | 91.6 $\pm$ 4.2Aa                            | 91.7 $\pm$ 1.7Aa | 93.3 $\pm$ 7.3Aa |
| Open cage      | 4          | 15–16               | 73.3 $\pm$ 8.6Aa                            | 70.9 $\pm$ 6.9Aa | 81.9 $\pm$ 4.2Aa |
| Selective cage | 4          | 15–16               | 72.3 $\pm$ 5.3Aa                            | 72.3 $\pm$ 6.5Aa | 75.9 $\pm$ 9.8Aa |
| No cage        | 8          | 15–16               | 36.7 $\pm$ 7.0Ab                            | 26.3 $\pm$ 8.6Ab | 45.7 $\pm$ 5.9Ab |
| Second trial   |            |                     |   |                  |                  |
| Closed cage    | 5          | 21–24               | 95.6 $\pm$ 3.2Aa                            | 93.2 $\pm$ 5.1Aa | 94.7 $\pm$ 3.0Aa |
| Open cage      | 5          | 23–24               | 86.3 $\pm$ 3.2Aa                            | 86.2 $\pm$ 5.1Aa | 85.5 $\pm$ 7.3Aa |
| Selective cage | 5          | 22–25               | 89.5 $\pm$ 9.2Aa                            | 87.2 $\pm$ 9.3Aa | 83.4 $\pm$ 5.7Aa |
| No cage        | 5          | 23–24               | 31.7 $\pm$ 15.8Ab                           | 20.2 $\pm$ 8.6Ab | 16.7 $\pm$ 5.4Ab |

Percentage of *P. xylostella* recovered was calculated as [(live fourth instar larvae + live pupae + diseased fourth instar larvae + *D. semiclausum* cocoons)/initial egg density]  $\times$  100. Within each trial, means in the same row followed by the same capital letter, and means in the same column followed by the same lower case letter are not significantly different, respectively ( $P > 0.05$ , LSD test, ANOVA).

cage treatments (8–37% first trial, and 38–63% second trial, Table 2). Overall, percent parasitism of *P. xylostella* by *D. semiclausum* in the open and selective cage treatments was higher in the second trial than in the first trial (Table 2). There was no significant difference in the percent parasitism of *P. xylostella* by *D. semiclausum* between the open and selective cage treatments within each pest management practice (Table 2).

On average, 41–56% of the *P. xylostella* eggs were lost during the 3-day exposures in the field but no egg parasitoid was reared from the recovered *P. xylostella* eggs.

### Direct Observation

*Diadegma semiclausum* wasps became active around 08:30 in the morning and soon were observed to emigrate from the source field into the experimental plots (Figure 3). In the cabbage field experiments, the number of wasps observed per hour increased over time during the day, and this trend was consistent at the three different sites on 30 July (Figure

TABLE 2. Percent parasitism (mean  $\pm$  SE) of the recovered *P. xylostella* larvae by *D. semiclausum* at the end of each trial from different treatments in plots of three different pest management practices

| Treatment      | Replicates | % Parasitism      |                   |                   |
|----------------|------------|-------------------|-------------------|-------------------|
|                |            | IPM               | Unsprayed         | Conventional      |
| First trial    |            |                   |                   |                   |
| Closed cage    | 4          | 0                 | 0                 | 0                 |
| Open cage      | 4          | 8.5 $\pm$ 3.2Aa   | 36.7 $\pm$ 17.3Aa | 25.5 $\pm$ 13.2Aa |
| Selective cage | 4          | 8.3 $\pm$ 3.2Aa   | 8.6 $\pm$ 3.4Aa   | 4.8 $\pm$ 4.1Aa   |
| No cage        | 8          | 93.6 $\pm$ 3.2Ab  | 84.6 $\pm$ 6.8Ab  | 92.6 $\pm$ 2.3Ab  |
| Second trial   |            |                   |                   |                   |
| Closed cage    | 5          | 0                 | 0                 | 0                 |
| Open cage      | 5          | 38.2 $\pm$ 6.2Aa  | 55.3 $\pm$ 5.6ABa | 63.2 $\pm$ 5.8Ba  |
| Selective cage | 5          | 49.8 $\pm$ 9.6Aab | 44.9 $\pm$ 3.8Aab | 43.9 $\pm$ 9.9Aa  |
| No cage        | 5          | 75.9 $\pm$ 21.8Ab | 71.7 $\pm$ 9.0Ab  | 88.5 $\pm$ 15.7Ab |

Within each trial, means in the same row followed by the same capital letter, and means in the same column followed by the same lowercase letter are not significantly different, respectively ( $P > 0.05$ , LSD test, ANOVA).

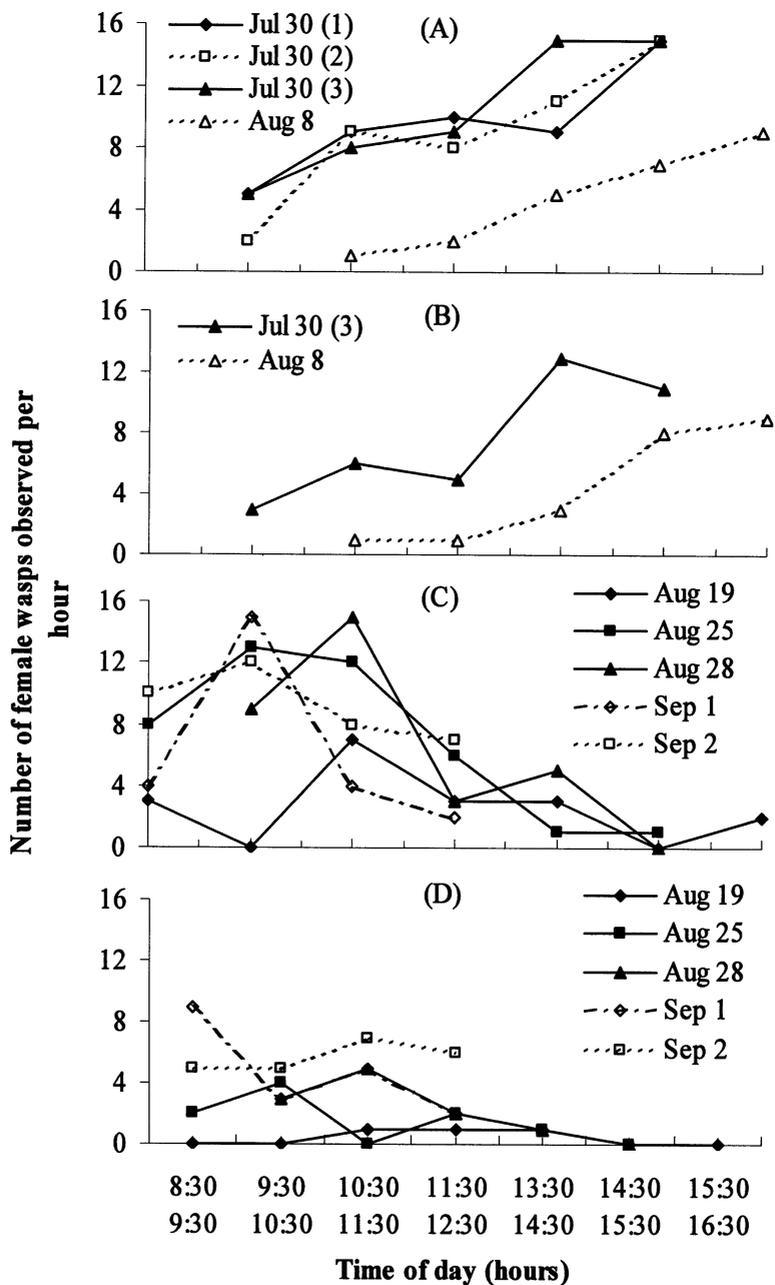


FIGURE 3. Number of *D. semiclausum* female wasps observed per hour per plot in two fields. Cabbage field (four observations): (A) high host density plot; (B) low host density plot (no low host density treatment for the first two observations). Broccoli field (five observations): (C) high host density plot; (D) low host density plot.

3A,B). Because the observed patterns of wasp activity were consistent at the three different sites, the data from the three observations were pooled for the analysis hereafter. In contrast, in the broccoli field the number of wasps observed per unit time increased in the morning but gradually declined in the afternoon (Figure 3C,D).

The density of observed parasitoid populations per unit time on both high and low host density plots decreased from 30 July to 8 August in the cabbage field but gradually increased from 19 August to 2 September in the broccoli field (Figure 4A). The proportion of male

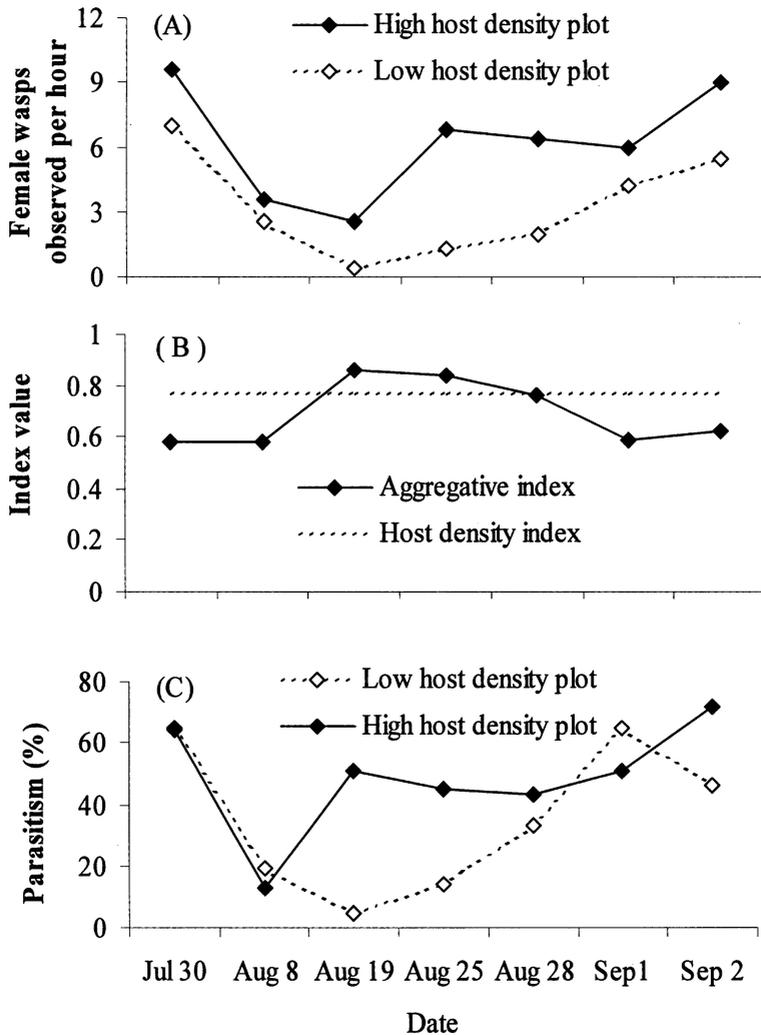


FIGURE 4. Aggregation responses of field *D. semiclausum* populations to two host plant plots infested with different densities of *P. xylostella* larvae and the resultant pattern of parasitism at the plot level. The first two observations were beside a cabbage field while the last five observations were beside a broccoli field. (A) Number of *D. semiclausum* female wasps observed per hour per plot. (B) Aggregative index ( $A_{\text{plot}} = \text{number of wasps observed in the high host density plot} / \text{total number of wasps observed in the two plots}$ ), and host density index ( $D_{\text{plot}} = \text{number of hosts present in the high host density plot} / \text{total number of hosts present in the two plots} = 0.77$ ). (C) The pattern of percent parasitism.

wasps observed were generally lower than 10%, except on 8 August when a high proportion of males (72%) was observed in the cabbage field.

At the plot level, more wasps were observed on the high host density plot than the low host density plot in both fields (Figure 4A). However, the degree of aggregation response varied across all the observation dates (Figure 4B). When  $A_{\text{plat}}$  was low relative to  $D_{\text{plot}}$  in both the cabbage and broccoli fields, parasitism did not increase with host density levels (Figure 4B,C). When  $A_{\text{plat}}$  was high relative to  $D_{\text{plot}}$  in the broccoli field, parasitism increased at higher density levels (Figure 4B,C), suggesting that a density-dependent relation may exist between the parasitoid and host.

At the plant level, the parasitoids also showed a positive aggregation response to high host density plants in both high and low host density plots as percentage of wasp observations increased with host density (Figure 5). However, at the plant level,  $A_{\text{plant}}$  was generally low

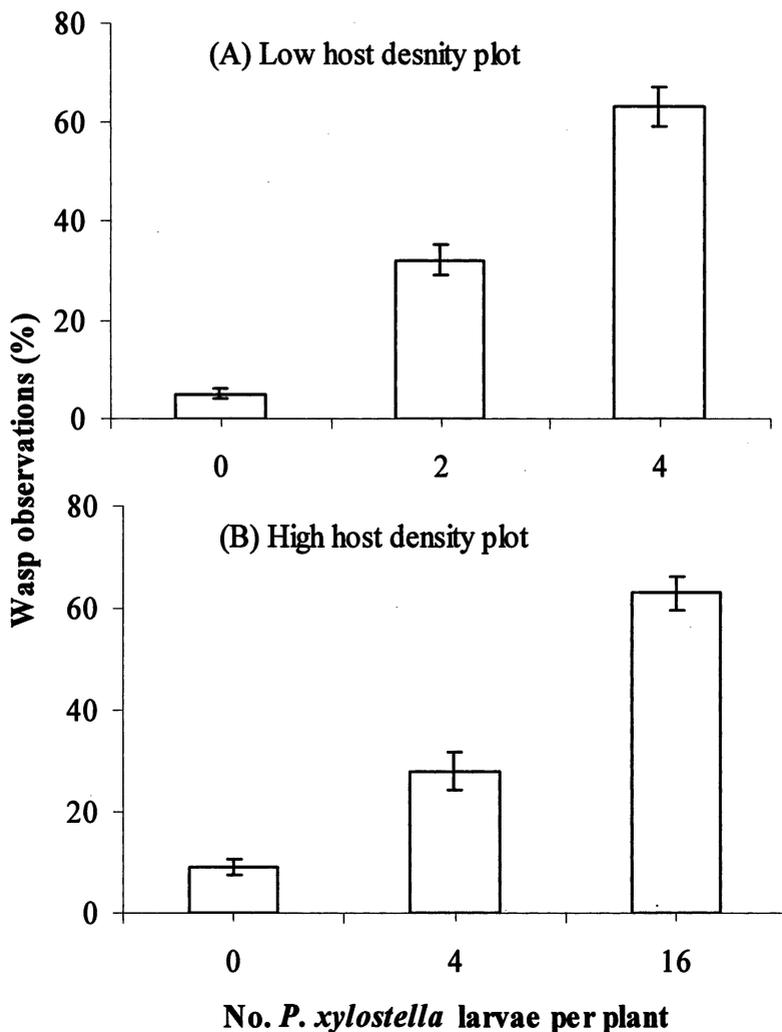


FIGURE 5. Mean ( $\pm$ SE) percentage of *D. semiclausum* female wasps observed on different host density plants in each plot. (A) Low host density plot ( $n = 7$ ); (B) high host density plot ( $n = 9$ ).

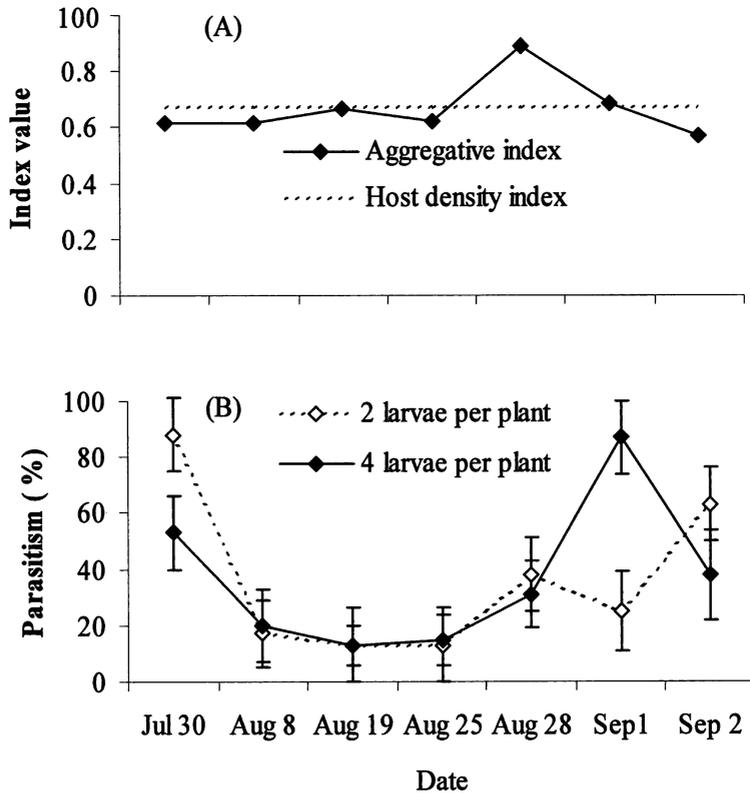


FIGURE 6. Aggregation responses of field *D. semiclausum* populations to individual plants infested with varying densities of *P. xylostella* larvae and the resultant patterns of parasitism at the low host density plot. The first two observations were beside a cabbage field while the last five observations were beside a broccoli field. (A) Aggregative index ( $A_{\text{plant}}$  = number of wasps observed on the high host density plants/total number of wasps observed in the whole plot), and host density index ( $D_{\text{plant}}$  = number of hosts present on the high host density plants/total number of hosts present in the whole plot, which is 0.67). (B) The pattern of mean ( $\pm$ SE) parasitism. There was no significant difference in the parasitism between low and high host density plants except on 30 July and 1 September (Student *t*-test with adjusted sequential Bonferroni method,  $P > 0.05$ ).

relative to  $D_{\text{plant}}$  (except on 28 August in the low host density plot) (Figures 6 and 7), the overall pattern of parasitism was independent of host density (Figures 6 and 7). There was no significant difference in the percent parasitism between low and high host density plants except on 30 July and 1 September in the low host density plot (Figure 6).

## DISCUSSION

The cage exclusion experiment showed that *D. semiclausum* played a dominant role in controlling *P. xylostella* during the winter season in southern Queensland. Parasitism of *P. xylostella* by other parasitoids, mainly the two pupal parasitoids, *D. collaris* and *Brachymeria* spp., was low (<10%) although it was likely underestimated because the experiments were terminated before all larvae had pupated. The lack of egg parasitism during our study agrees with the results of a longer term assessment of *P. xylostella* egg parasitoids in the same locality, which showed that *P. xylostella* egg parasitism by *Trichogramma* spp. occurs frequently in spring, summer, and autumn but is usually absent

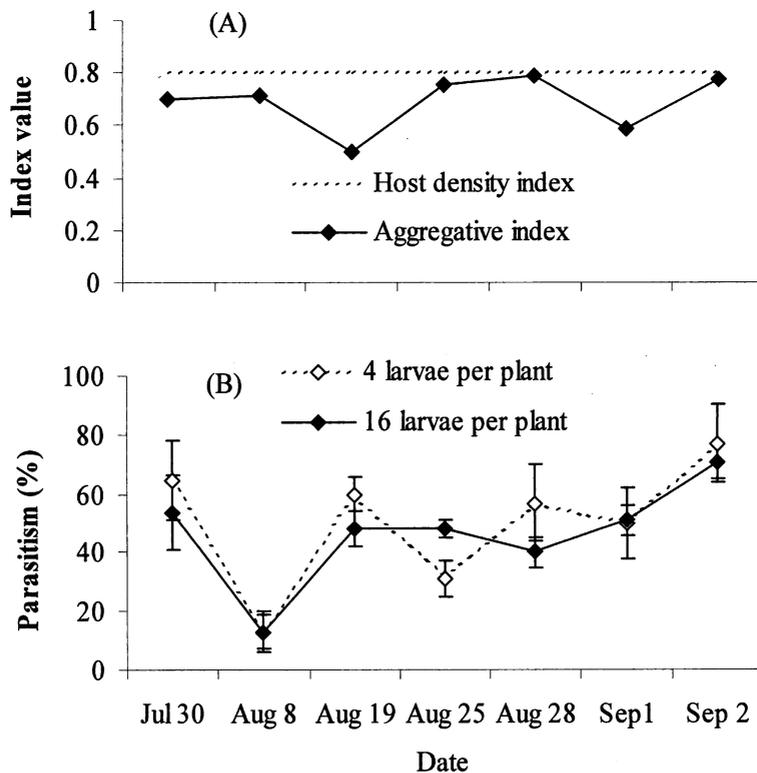


FIGURE 7. Aggregation responses of field *D. semiclausum* populations to individual plants infested with varying densities of larval *P. xylostella* and the resultant patterns of parasitism at the high host density plot. The first two observations were beside a cabbage field while the last five observations were beside a broccoli field. (A) Aggregative index ( $A_{\text{plant}}$  = number of wasps observed on the high host density plants/total number of wasps observed in the whole plot), and host density index ( $D_{\text{plant}}$  = number of hosts present on the high host density plants/total number of hosts present in the whole plot, which is 0.80). (B) The pattern of mean ( $\pm$ SE) percent parasitism. There was no significant difference in percent parasitism between low and high host density plants (Student *t*-test with adjusted sequential Bonferroni method,  $P > 0.05$ ).

in winter months (Liu *et al.*, 2004). Activities of predators were probably minimal during the winter as there was no significant difference in the percentage of *P. xylostella* recovered between the open cage and selective cage treatments. Thus, the high loss of eggs in the field was possibly due to abiotic factors such as rain and wind.

Parasitism of *P. xylostella* larvae by *D. semiclausum* was higher in the uncaged than the open cage treatment. There are two possible explanations. Firstly, the cage with an open on the top may have partly obstructed access of the parasitoid to hosts. Furlong *et al.* (2004) showed that parasitism of *P. xylostella* larvae on uncaged plants was similar to those on plants in cages with horizontal access. Secondly, the surviving host larval densities on the sentinel plants were much lower in the uncaged treatment than were in the open cage treatment. With the build up of the field parasitoid population, *P. xylostella* parasitism by *D. semiclausum* in the open cage treatment was higher in the second than the first trial (Table 2). Thus, the low parasitism on the sentinel plants may be due to the lack of a high degree of parasitoid aggregation relative to on the high host densities. No obvious difference in the developmental rate of the *P. xylostella* larvae among the different treatments was observed. Also, there was no trend showing higher rates of disease development of the *P. xylostella*

larvae in the closed cage (2–7%) than in the open cage (1–6%) treatments. Thus, any effect resulting from microclimatic changes as a result of caging (temperature, humidity and light) on the observed pattern of parasitism may be minimal in this study.

Direct field observation showed that the response of *D. semiclausum* population from the resource fields to adjacent experimental plots varied with habitat quality of the parasitoid source field, parasitoid population density, and scales of the manipulated host patches. Because the only source of parasitoids we knew of was an adjacent field of cabbage or broccoli, we assumed changes in parasitoids on experimental plants were due to movement from these fields. When the hosts were seriously depleted in the cabbage field and the plants were at the late stage of growth, the parasitoid populations seemed to disperse consistently towards the nearby experimental plots. This pattern could reflect a combination of passive (dispersal) and active aggregation response, which resulted in a relatively low degree of wasp aggregation to the high host density plot. However, in the broccoli field when the plants were at the young stages of growth as the plants used in the experimental plots, and when host density in the broccoli field (0–10 larvae per plant, see Figure 1) seemed comparable to the experimental plots (0–4 larvae per plant in low host density plot and 0–16 larvae per plant in the high host density plot), the parasitoid populations moved between the resource field and the experimental plots, i.e., active aggregation response. When the observed *D. semiclausum* population density was relatively low in the broccoli field during the first three observation dates (Figure 4B), the parasitoids aggregated more on nearby plots with high host density than low host density. However, at the plant level, the parasitoid population always showed a strong aggregation response to high host density plants, suggesting that the parasitoid population might respond differentially to the two spatial scales.

Active aggregation in parasitoids was observed in many studies (Waage, 1983; Smith & Maelzer, 1986; Jones & Hassell, 1988; Sheehan & Shelton, 1989; Wang & Keller, 2002). The aggregation response of parasitoids has been identified as an important factor that contributes to host regulation (Hassell & May, 1974; Reeve & Murdoch, 1985; Murdoch *et al.*, 1987; Ives *et al.*, 1999). Positive aggregation response by parasitoids should generate a direct density-dependent parasitism (Comins & Hassell, 1979; Hassell, 1982; Waage, 1983; Lessells, 1985). However, empirical studies often fail to detect density-dependent parasitism (Waage, 1983; Smith & Maelzer, 1986), and reveal diverse patterns of parasitism (Lessells, 1985; Stiling, 1987; Walde & Murdoch, 1988). Theoretical explanations for non density-dependence in parasitism include behavioural or physiological limitations of foraging parasitoids, such as limited availability of eggs and handling time when foraging in high host density patches (Comins & Hassell, 1979; Hassell, 1982; Waage, 1983; Lessells, 1985), stochastic variation in foraging time allocation (Morrison, 1986), or other forms of aggregation response (Walde & Murdoch, 1988). Any conflicting or interacting mechanisms that influence the attack rate may outweigh the aggregation effect on parasitism (Wang, 2001). Our studies showed that, although *D. semiclausum* did show a strong aggregation response to high host density plants or plots, the density-dependence in the resultant pattern of parasitism depended on the degree of aggregation relative to host density at a particular spatial level. When the aggregative index was low relative to host density index, parasitism was approximately density-independent; when the relative aggregation index was high relative to host density index, parasitism was basically density-dependent. Thus, a high degree of aggregation was necessary to generate direct density-dependent parasitism by this parasitoid.

Based on the work presented here, it is suggested that *D. semiclausum* is active during the winter and plays an important role in suppressing *P. xylostella* population in southern Queensland. A final destructive sampling did not find significant differences in density of *P. xylostella* larvae and broccoli yield among the different plots (Wang, 2001). Therefore, a biologically based strategy by maximizing the role of *D. semiclausum* with the use of *Bt* or other microbial agents may deliver effective control of *P. xylostella* without the problems of

insecticide resistance and excessive use of chemical insecticide in the relatively cool seasons or regions where the weather conditions are favourable to *D. semiclausum* (Ooi, 1992; Talekar & Shelton, 1993; Amend & Basedow, 1997; Saucke *et al.*, 2000).

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