War of the Wasps: Is *Diadegma insulare* or *Microplitis plutellae* a More Effective Parasitoid of the Diamondback Moth, *Plutella xylostella*?

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Parasitism levels by *Diadegma insulare* (Muesebeck) (Hymenoptera: Ichneumonidae) and *Microplitis plutellae* (Haliday) (Hymenoptera: Braconidae) at various densities of their host, *Plutella xylostella* (L.) (Lepidoptera: Plutellidae), were assessed. Cages with densities of 10 hosts, 20 hosts, and 40 hosts were set up, with the cage volume (40 500 cm³) and number of wasps (2 females) remaining constant. The host populations were also exposed to the wasps for two different exposure times: 1 day and 3 days. The study showed that *D. insulare* was a better parasitoid overall, achieving a level of parasitism equal to or higher than *M. plutellae* at all densities. *Microplitis plutellae* performed best at a lower host density (76% \pm 9% of 10 hosts vs. 43% \pm 3% of 40 hosts). *Diadegma insulare* performed similarly at all densities tested (75% \pm 5% of 10 hosts, 83% \pm 4% of 20 hosts, and 79% \pm 6% of 40 hosts). This suggests that *D. insulare* may be the better parasitoid overall and should be applied in severe, large-scale infestations, while *M. plutellae* may be better for small-scale infestations.

Key Words: Diamondback Moth; Plutella xylostella; Microplitis plutellae; Diadegma insulare; parasitoids; biological control

Introduction

Invasive alien species are a large problem both for the environment and for agriculture. The Diamondback Moth (*Plutella xylostella* (L.) (Lepidoptera: Plutellidae)) (Figure 1) is one such species. It is invasive in North America and is thought to be native to Europe or Asia (Harcourt 1954). It is a major pest of crops in the plant family Brassicaceae (which includes canola, cabbage, and broccoli), causing an estimated one billion dollars U.S. in crop damage annually (Sarfaz *et al.* 2005a).



FIGURE 1. Adult Diamondback Moth, *Plutella xylostella*. Photo taken using microscope camera. CNC voucher Diptera 225229. Scale bar = 1 mm. Photo: Jeffery Skevington.

The Diamondback Moth is resistant to many pesticides (Harcourt *et al.* 1986). The scientific community is therefore exploring alternative ways of controlling it, including biological control. Biological control is a method of controlling a pest population (for example, an herbivore) by means of another organism (a parasitoid, predator, or pathogen). If applied properly, biological control can provide better control than pesticides. Not only does it avoid the introduction of dangerous chemicals into ecosystems, but it is also more effective because the target pest can never develop a resistance (the biological control agent evolves with the pest, countering pest adaptations with its own).

There are many parasitoid wasps that attack the Diamondback Moth, but two of the most studied are *Diadegma insulare* (Figure 2) and *Microplitis plutellae* (Figure 3). Both of these parasitoid wasps are native to Canada. The wasps attack the Diamondback Moth by implanting an egg into the larval host. The parasitoid inside matures with the host larva and eventually egresses to pupate, killing the host in the process (Sarfaz *et al.* 2005b). The two wasps differ slightly in life cycle: when the Diamondback host reaches the prepupal stage, the *D. insulare* larva consumes it and pupates within the host's already-spun cocoon. *Microplitis plutellae*, however, egresses from the fourth instar larva to find a dry location before pupating (Figure 4).

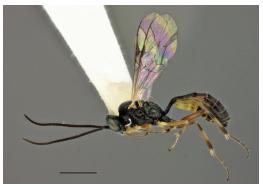


FIGURE 2. Adult male *Diadegma insulare*. CNC voucher Diptera 225228. Scale bar = 1 mm.



FIGURE 3. Adult female *Microplitis plutellae*. CNC voucher Diptera 225230. Scale bar = 1 mm.

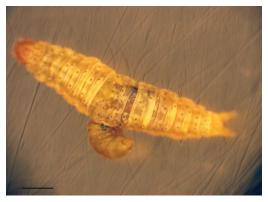


FIGURE 4. *Microplitis plutellae* larva emerging from its host, a fourth instar larva of the Diamondback Moth, *Plutella xylostella*. Scale bar = 1 mm.

Despite the large amount of information collected about these two parasitoid wasps, there are very few data on the effect of host density on the level of parasitism, or the functional response, of the wasps. Some studies (Bolter and Laing 1984) have indicated that there is a difference in functional response, but a comparison of effectiveness of both wasps is lacking.

An important way of evaluating the effect of host density on a parasitoid's level of parasitism is by looking at its functional response curve. Most parasitoids have a type II functional response curve, although those with a type III functional response curve tend to be more effective parasitoids (Fernandez-Arhex and Corley 2002). The functional response curves of *D. insulare* and *M. plutellae* are currently unknown.

The objective of this study was to evaluate the functional response of *M. plutellae* and *D. insulare* to their *P. xylostella* host population, to determine which would be better suited as a biological control agent for Diamondback Moth infestations.

Methods

The experiment tested three levels of density treatments (10, 20, and 40 hosts per cage) crossed with two levels of exposure time treatments (1 day and 3 days). Parasitism by female *Diadegma insulare* and *Microplitis plutellae* wasps was measured separately (see Table 1).

TABLE 1. Number of cages used to produce data for each host density and exposure time treatment, for both *D. insulare* and *M. plutellae*.

Wasp	Exposure time	Host density (No. of hosts per cage)	No. of cages
Microplitis plutellae	1 day	10	5
		20	6
		40	5
	3 days	10	5
	2	20	6
		40	4
Diadegma insulare	1 day	10	4
	2	20	4
		40	4
	3 days	10	5
		20	7
		40	4

Parasitoid pupae were collected from a locallyreared culture and placed in a 19 cm \times 14 cm \times 19 cm cage (fabricated onsite) with a vial containing a 10% solution of sucrose in water. This cage was checked daily so that the approximate age of each of the adult parasitoid wasps to be used in experiments was controlled. The range of acceptable age for wasps was 2– 5 days post-egression, to ensure that the wasps were young enough and had been able to mate before being introduced to the host population.

Diamondback Moth larvae (Figure 5) were taken from a culture and placed in a 30 cm \times 30 cm \times 45 cm cage with a single canola plant. Larvae were placed in each cage at different host densities (in this experiment, host density is specifically the number of hosts, either 10, 20 or 40, per cage), and 10% sucrose vials were placed inside as a source of food for the wasps. After setup, the cages were left in a climate-controlled room (set at 22°C ± 1 Celsius degree and 70% humidity, with 16 hours of light provided by two Sylvania F48T12D/ VHO bulbs, 115 W and 6500 K, and two Sylvania Cool Whites F48T12/CW/VHO bulbs, 115 W and 4200 K). This was the beginning of the wasp exposure time,



FIGURE 5. Third instar larva of the Diamondback Moth, *Plutella xylostella*. CNC voucher Diptera 225231. Scale bar = 1 mm.

which lasted for 1 or 3 days, depending on the treatment.

After the wasp exposure time was complete, the parasitoids were removed and the cage was left inside the climate-controlled room for a further 14 days to allow hosts and parasitoids to pupate. The parasitoid and host pupae were removed from the cage and counted. For each replicate, the level of parasitism (based on the number of parasitoid pupae recovered) was recorded as a percentage of the original host population.

Specimens are deposited in the Canadian National Collection of Insects, Arachnids, and Nematodes at Agriculture and Agri-Food Canada in Ottawa, Ontario (CNC) (collection acronyms follow the Registry of Biological Repositories, http://www.biorepositories .org).

Results

Parasitism by *Diadegma insulare* remained relatively consistent for both exposure times, sitting at $62\% \pm 7\%$ for the 1-day exposure time and $75\% \pm 5\%$ for the 3-day exposure (Figure 6).

Parasitism by *Microplitis plutellae* showed some variation among densities. In the 1-day exposure treatments, the level of parasitism was very low $(16\% \pm 8\%)$

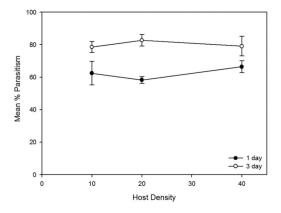


FIGURE 6. Mean (± SE) parasitism of the Diamondback Moth, *Plutella xylostella*, by the larval parasitoid *Diadegma insulare* at different host densities and exposure times.

to $21\% \pm 3\%$); the most likely explanation is that the wasps did not have adequate time to parasitize. In the 3-day exposure treatments (where the wasps had more time for parasitism), *M. plutellae* performed optimally at the lowest density, parasitizing 76% ± 9% of the 10 hosts (Figure 7).

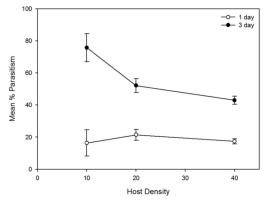


FIGURE 7. Mean (± SE) parasitism of the Diamondback Moth, *Plutella xylostella*, by the larval parasitoid *Microplitis plutellae* at different host densities and exposure times.

In the 1-day exposure treatment, *Diadegma insulare* achieved a much higher level of parasitism than *Microplitis plutellae* at all densities $(58\% \pm 2\% \text{ to } 67\% \pm 4\%$ for *D. insulare* versus $16\% \pm 8\%$ to $21\% \pm 3\%$ for *M. plutellae*) (Figure 8). In the 3-day exposure treatment, *M. plutellae* performed comparably to *D. insulare* at the lowest density, with both wasps achieving similar levels of parasitism $(76\% \pm 9\% \text{ and } 75\% \pm 5\%, \text{ respectively})$. However, as density increased, the parasitism rate for *M. plutellae* dropped to $52\% \pm 4\%$ (of 20 hosts), and finally to $43\% \pm 3\%$ (of 40 hosts). The parasitism rate for *D. insulare* remained at approximately the same level (Figure 9).

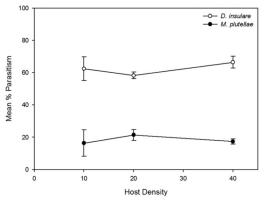


FIGURE 8. Mean (± SE) parasitism of the Diamondback Moth, Plutella xylostella, by the larval parasitoids Diadegma insulare and Microplitis plutellae at the 1-day exposure time at different host densities.

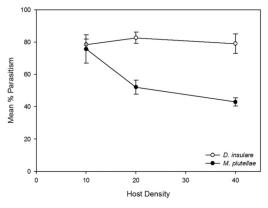


FIGURE 9. Mean (± SE) parasitism of the Diamondback Moth, Plutella xylostella, by the larval parasitoids Diadegma insulare and Microplitis plutellae at the 3-day exposure time at different host densities.

When looking at the number of hosts parasitized (as opposed to the percentage of the total host population), a different trend emerges. Both *D. insulare* and *M. plutellae* parasitized a larger number of hosts as host density increased. However, the number of hosts parasitized by *D. insulare* increased at a constant rate, while the number of hosts parasitized by *M. plutellae* increased at a constantly decreasing rate.

Discussion

From a purely functional response standpoint, *Diadegma insulare* seems to be a more effective parasitoid of the Diamondback Moth than *Microplitis plutellae*. *Diadegma insulare* maintained a level of parasitism equal to or higher than that of *M. plutellae* at all densities.

It is possible to identify the functional response curves for D. insulare and M. plutellae based on the results of this experiment. It appears that D. insulare has a type I functional response curve: as the number of available hosts increased, D. insulare parasitized a larger number of hosts while maintaining a constant percentage parasitism. It is a somewhat unexpected result, as only one other parasitoid biological control agent (Eretmocerus eremicus (Hymenoptera: Aphelinidae)) is known to exhibit a type I functional response (Fernandez-Arhex and Corley 2002). It is important to note that the host densities tested in this experiment were relatively low compared to other experiments (Putnam 1968), and it is likely that the percentage parasitism by D. insulare would decrease when the number of available hosts reached a critical mass (as is evident with other type I functional response organisms) (van Alphen and Jervis 1996).

Microplitis plutellae seems to have a type II functional response curve. The number of hosts parasitized by *M. plutellae* rose at a constantly decreasing rate. Thus, as host density increased, a smaller percentage of hosts was parasitized. These are characteristics of type II and type IV functional responses (van Alphen and Jervis 1996). A type IV response has never been observed in a parasitoid before (Fernandez-Arhex and Corley 2002), so it is more likely that *M. plutellae* has a type II response. Further experimentation (using higher host densities) could provide data to support or refute the inferences made in this paper about the functional response curves of *D. insulare* and *M. plutellae*.

The characterization of the functional responses of *D. insulare* and *M. plutellae* also reveals information about their behaviour. A type I functional response, such as *D. insulare*'s, occurs when a parasitoid's handling time is negligible and egg supply is the only factor that limits parasitism. *M. plutellae* has a type II functional response, suggesting that it takes a relatively long time to find and implant an egg into a host.

Past experiments seem to agree with the results of this experiment. Microplitis plutellae has been shown to be more egg-limited than D. insulare (Bolter and Laing 1984), producing only 40 eggs per day compared to D. insulare's 50 eggs per day. The studies by Bolter and Laing (1984) have also revealed that the behaviour of the wasps may, in some cases, have a greater impact on their level of parasitism than the number of eggs. In their experiment, M. plutellae wasps were able to lay only half of their daily quota of eggs, whereas D. insulare wasps were able to lay all of them (when a large enough host population was provided) (Bolter and Laing 1984). This further supports the classification of M. plutellae as a parasitoid with a type II functional response-it may be unable to use all of the eggs it produces in a day because it spends too much time interacting with the host.

It is possible that the inferior percentage parasitism of *M. plutellae* is related to superparasitism, a form of parasitism in which the host is attacked more than once by a single species of parasitoid. The percentages superparasitism of *M. plutellae* and *D. insulare* have been compared in previous experiments. Bolter and Laing (1984) suggested that *M. plutellae* is unable to distinguish between a parasitized and a non-parasitized host. However, the functional response experiments of Bolter and Laing (1984) indicated that *D. insulare* and *M. plutellae* maintain an equally low level of superparasitism within a host population. Further research is required to determine the impact that superparasitism may have on the functional response of *M. plutellae*.

There are multiple factors to take into consideration when determining the optimal biological control agent for a pest. In addition to functional response, a parasitoid's adaptability to climate and its impact on nontarget species must also be evaluated. Both *D. insulare* and *M. plutellae* are widespread in North America (Krombein *et al.* 1979a,b). *Microplitis plutellae* is found as far north as Ontario and Saskatchewan, as far west as California, and as far east as Virginia. *Diadegma insulare* has an even wider North American distribution. While both parasitoids are considered native species to Canada and the U.S., the Diamondback Moth is widely believed to have originated in Europe. *Diadegma insulare* is known to attack two other European moths, *Plutella armoraciae* (Busck) (Lepidoptera: Plutellidae) and *Hellula undalis* (Fabricius) (Lepidoptera: Crambidae). Similarly, *M. plutellae* can attack *Trichoplusia ni* (Hübner) (Lepidoptera: Noctuidae), another European pest of Brassicaceae (Krombein *et al.* 1979a). It is likely that these two parasitoids have non-target host species that are native to North America, but these host species are currently unknown.

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