

Predation potential and prey-stage preference of two mirid bugs on different stages of *Plutella xylostella*

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Abstract

The predation rate and prey-stage preference of two mirid predatory bugs *Macrolophus pygmaeus* Rambur and *Nesidiocoris tenuis* (Reuter) (Hemiptera Miridae), were investigated on the diamondback moth (DBM) *Plutella xylostella* (L.) (Lepidoptera Plutellidae), a noxious pest of cruciferous plants. In particular, predation capacity of each predatory stages of both predators was studied on the first, second and third larval instars of DBM. There were no significant differences in predation between the predators on eggs, second and third instar larvae of DBM. Among the different life stages of both predators, females had the highest predation on eggs and all larval instars of DBM. Preference indices for both mirid species indicated that there was a preference toward eggs, first instar and second instar larvae versus first instar, second instar and third instar larvae, respectively. The data of the current study suggest that *M. pygmaeus* and *N. tenuis* are effective predators of DBM particularly against eggs and first instar larva. It would therefore be worth to investigate how these mirids can be incorporated in biological control programs of DBM.

Key words: generalist predators, voracity, diamondback moth (DBM), *Macrolophus pygmaeus*, *Nesidiocoris tenuis*, *Plutella xylostella*.

Introduction

The Brassicaceae (= Cruciferae) is a large and widespread family consisting of many economically important crops, including oilseed, vegetable, condiment, and fodder crops (Talekar and Shelton, 1993). The diamondback moth (DBM) *Plutella xylostella* (L.) (Lepidoptera Plutellidae) is a pest of a heterogeneous group of cruciferous plants throughout the world causing damage to cabbage, broccoli, canola, and other crucifers. The caterpillar destroys cruciferous plants by tunneling in the foliage and feeding on leaves and contaminates the crop by pupating on cabbage leaves (Talekar and Shelton, 1993; Zalucki *et al.*, 2012). Misuse of pesticides against DBM has increased resistance to nearly all conventional insecticides used against this pest (Talekar and Shelton, 1993). Tabashnik *et al.* (1990) demonstrated that despite the safety and efficacy of *Bacillus thuringiensis* subsp. *kurstaki*, DBM was the first lepidopteran pest that showed resistance to this biological control agent in the open field.

Nowadays, biological control is the principal method to reduce reliance on pesticides for controlling pest populations (Viñuela, 2005). Different natural enemies are commonly used in seasonal inoculative or inundative releases to reduce the key pest populations throughout the world (Malais and Ravensberg, 2004). There have been several studies on evaluation of a wide range of biocontrol agents, attack DBM (Tomiyama and Aoki, 1982; Lim, 1985; Talekar and Shelton, 1993; Philips *et al.*, 2014; Marchioro *et al.*, 2015; Li *et al.*, 2016).

The omnivorous hemipterans *Macrolophus pygmaeus* Rambur and *Nesidiocoris tenuis* (Reuter) (Hemiptera Miridae) have been used in various augmentative biological control programs (Calvo *et al.*, 2009; Urbaneja *et al.*, 2009). A number of studies have highlighted the preda-

tion capacity of these predatory mirids against the whiteflies *Trialeurodes vaporariorum* (Westwood) and *Bemisia* spp. (Hemiptera Aleyrodidae), aphids, thrips and other small arthropod pests in greenhouses and field crops. These predators as well as other dicyphine species have also been used for the biological control of *Tuta absoluta* (Meyrick) (Lepidoptera Gelechiidae) in several parts of the world (Malausá *et al.*, 1987; Malezieux *et al.*, 1995; Bueno and van Lenteren, 2012; Urbaneja *et al.*, 2012; De Backer *et al.*, 2014; Mollá *et al.*, 2014; Michaelides *et al.*, 2018; Ferracini *et al.*, 2019; Ingegno *et al.*, 2019). Recently, however, *N. tenuis* is considered a pest in the greenhouses of Asia and Northwestern Europe, due to its strong tendency to feed on and damage tomato plants (Pérez-Hedo and Urbaneja, 2016; Puentes *et al.*, 2018).

The predation rate of a predator is an important indicator of its effectiveness to control pest populations in biological control projects. Predators may normally encounter a range of prey species in the environment (Pearre, 1982). Understanding the prey preferences of predators toward a certain developmental stage of the target pest or among different pest species is crucial. The prediction of the predation potential of a predator could be more accurate when different prey types or different life-stages of prey species are simultaneously present (Carrillo and Pena, 2012; Rahman *et al.*, 2012; Xiao *et al.*, 2013; Ganjisaffar and Perring, 2015).

The feeding preference of a predator exposed to different types of prey is mostly controlled by the energetic costs of searching and consuming of the prey. These energetic costs are closely related to prey size and prey behaviour (Krebs and McCleery, 1984). Despite the acceptance of a wide range of prey by generalist predators, they often show preferences for particular prey species or different life-stages within prey species (Provost *et al.*,

2006; Reitz *et al.*, 2006; Soleymani *et al.*, 2016). However, little attention has been paid to mechanisms of prey selection by generalist predators. Generalist predators require different prey to balance their nutritional needs (Stephens and Krebs, 1986). As pointed out by Chesson (1989), the presence of alternative prey may reduce predation pressure of generalist predators on a population of a pest, but it may also enhance survival and reproduction of the predators and in this manner increase predation pressure on other species, including the target pest (Leman *et al.*, 2020).

In order to strengthen insight into the potential predation of *M. pygmaeus* and *N. tenuis*, the present study evaluated the predation rates of both predatory bugs on egg and larval stages of DBM. Furthermore, the prey preference of both predators was investigated when different life stages of DBM were presented together. To our knowledge, this is the first report on predation and prey preference of *M. pygmaeus* and *N. tenuis* on the immature DBM stages.

Materials and methods

Insect rearing

P. xylostella

A laboratory colony of DBM was started in 2012 with pupae originating from a cabbage greenhouse in Rafsanjan University (Kerman Province, Iran). The pupae were transferred to a growth chamber maintained at 25 ± 1 °C, $65 \pm 10\%$ RH, and a photoperiod of 16:8 (L:D). Upon emergence, adults were transferred to net cages ($60 \times 40 \times 40$ cm) and a piece of cotton wool soaked with 20% honey solution was supplied as food. Chinese cabbages (Hybride Hero of *Brassica campestris* L., Takh & Co. Ltd., Kyoto, Japan) were 20-25 cm tall and grown in plastic pots (16 cm in diameter) with soil mix of peat moss, coco peat and perlite for 2:1:1 v/v and watered as needed. After adult oviposition on Chinese cabbage and larval emergence, larvae were fed by adding fresh cabbage pots daily. The colony was yearly renewed by adding wild pupae to prevent inbreeding depression.

M. pygmaeus and *N. tenuis*

The laboratory colonies of *M. pygmaeus* and *N. tenuis* were set up from commercial strains (Biobest N.V., Westerlo, Belgium). The insects were reared in Plexiglas cylinders (9 cm diameter, 20 cm high) on fresh stems of tomato plants (*Solanum lycopersicum*), as an oviposition substrate and source of moisture. The cut end of the stems was covered with cotton and placed in a 10 ml Eppendorf tubes with water to keep the tomato stems fresh for a few days. The food of the stock colony of the predators primarily consisted of frozen and irradiated eggs (Biobest N.V.) of the Mediterranean flour moth *Ephestia kuehniella* Zeller (Lepidoptera Pyralidae) three times a week. A piece of moist cotton wool soaked with 20% of honey solution served as a source of water for the predators. The insects were reared in a growth chamber at 25 ± 1 °C, $65 \pm 10\%$ RH, and a photoperiod of 16:8 (L:D).

Predation by different life stages of *M. pygmaeus* and *N. tenuis* on different life stages of *P. xylostella*

Predation capacity of the different predatory stages of both predators was studied on eggs and first, second and DBM third larval instars. Predation during a 24-h period was observed for each juvenile instar and for male and female of the predators.

For each treatment, a newly moulted individual per each juvenile stage (2nd, 3rd, 4th and 5th instars) and a mated adult per each sex (circa 6 days old) of *M. pygmaeus* and *N. tenuis* were randomly taken from coetaneous cohorts from laboratory cultures and placed individually into a Petri dish arena (10 cm in diameter and 4 cm in height). Each Petri dish was ventilated by a 4-cm-diameter circular screened hole in the lid covered with a fine mesh. A leaf disk of cabbage (4.5 cm in diameter) was placed in each arena with its adaxial surface facing upwards on moistened cotton. Both predator nymphs (< 12 hours old) and adults were starved for 24 hours before onset of the experiments. A moist paper plug fitted into a small cup (2.5 cm in diameter) supplied water during starvation. 100 eggs or 20 larvae of each DBM instar, were uniformly distributed on the cabbage leaflet and offered to a single starved individual of the different tested stages of *M. pygmaeus* or *N. tenuis* per arena per 24 hours. Predation by individuals of *M. pygmaeus* and *N. tenuis* was recorded by counting preyed units at the stereo microscope. Each treatment was replicated 10 times simultaneously and controls (consisting of arenas without a predator) were set per each prey stage. In this experiment, survival of eggs and different DBM larval stages was high (> 99%) in the controls. Therefore, control mortality was considered negligible and we decided to don't use control data. All experiments were conducted in a climate chamber at 25 ± 1 °C, $65 \pm 10\%$ RH and a photoperiod of 16:8 (L:D).

Preference of *M. pygmaeus* and *N. tenuis* to different stages of *P. xylostella*

Prey stage preference of the fifth nymphal instar and female of both predator species was studied using eggs and larvae of different instars [first instar (L1), second instar (L2) and third instar (L3)] of DBM as prey. Different stages of DBM were exposed in the following ratios: 50-10 (Egg-L1), 10-10 (L1-L2) and 10-10 (L2-L3). Ten replicates were done for each predator stage, species and prey ratio combination.

All treatment combinations and controls (consisting of arenas without a predator) were tested in Petri dishes (14 cm in diameter and 2 cm in height) divided into two parts with Ant Escape Prevention Lacquer (Ameisen Ausbruchsschutz Lack, Germany). Two discs of cabbage plants leaves (4.5 cm in diameter) with their adaxial surface upwards were placed on moistened cotton in each section of the Petri dishes and each tested stage of the prey was placed on each of these leaf discs (figure 1). A single starved (24 hours) individual of each predator was released in the centre of the Petri dishes. This setup allowed the predator to access to the prey but prevented the DBM larvae from escaping from their side of the Petri dish. After 24 hours exposition, the total number of live

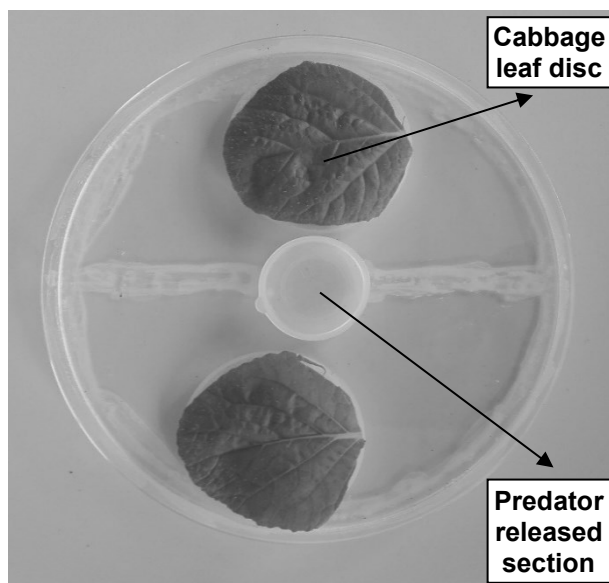


Figure 1. A Petri dish (14 cm diameter, 2 cm high) used in the prey preference experiments containing two leaf discs and painted with Ant Escape Prevention Lacquer.

and dead prey was counted. In the absence of predators, eggs and different larval stages of DBM had high survival rates (> 99%). Therefore, there was no need to correct mortality data from the preference test.

Statistical analysis

Statistical analyses were performed using the SPSS Statistics for Windows, version 22.0 (IBM Corp, Armonk, NY, USA). The normality of data was checked before analysing. A generalized linear model (GLM) procedure with factorial design based on Completely Randomized Design and Tukey's Studentized Range (HSD) ($p = 0.05$) were used to compare predation rates of the different life stages of *M. pygmaeus* and *N. tenuis* among the different DBM stages offered.

Prey preferences of *M. pygmaeus* and *N. tenuis* were analysed using Manly's preference index (Manly, 1974):

$$\beta_A = (\log A/a) / (\log A/a + \log B/b)$$

where A is the number of individuals of a given prey type A available for predation, B is the number of individuals

of a given prey type B available for predation, a is the number of prey type A that was not preyed upon and b is the number of prey type B that was not preyed upon.

When testing the preference of *M. pygmaeus* and *N. tenuis* between eggs (prey type A) and first instar larvae (prey type B) of DBM, for instance, Manly's preference index β_A gives the probability that the next prey will be eggs of DBM (prey type A). The preference index β_A was calculated for the three tested ratios of different DBM stages. The calculated value of β_A is situated between 0 and 1 and $\beta_A + \beta_B = 1$. This value is compared with the expected β_A -value, (i.e. the ratio of the total number of offered prey type A and the total number of offered prey type A+B). When the calculated β_A is higher than the expected β_A , this indicates a preference for prey type A. For each prey combination both calculated and expected preference indices were determined. We compare the calculated indices between *M. pygmaeus* and *N. tenuis* and between the fifth nymphal instar and female of each predator in each prey combination. Since the calculated indices for the tested prey combinations were not normally distributed, they were compared by Wilcoxon rank sum test (i.e. Mann Whitney U test).

Results

Predation by different life stages of *M. pygmaeus* and *N. tenuis* on different life stages of *P. xylostella*

All tested nymphal stages and adults of both predators preyed upon the DBM eggs (table 1). Results indicated that there was no significant difference in predation between the predator species on DBM eggs ($F = 0.52$, $df = 1, 126$, $P = 0.471$). While the predation among the different stages of each predator on the eggs did differ statistically ($F = 517.58$, $df = 6, 126$, $P < 0.0001$). Among the different life stages of both predators, adult females of *M. pygmaeus* and *N. tenuis* had the highest predation on DBM eggs. Predation on eggs by the first instars of both predators and second instar of *N. tenuis* was significantly lower than that by the other stages of both predator stages (table 1).

There was a significant difference in the predation rates of the two predators on DBM first instar larvae ($F = 11.01$, $df = 1, 126$, $P = 0.001$). The female adult predation of *N. tenuis* on first instar larvae was significantly higher

Table 1. Mean numbers (\pm SE) of DBM eggs, first, second and third instar larvae killed in 24 hours by different developmental stages of *M. pygmaeus* and *N. tenuis* [first (N1), second (N2), third (N3), fourth (N4) and fifth (N5) nymphal instars, and male and female adults].

Prey stage	Predator	N1	N2	N3	N4	N5	Male	Female
Egg	<i>M. pygmaeus</i>	3.7 \pm 0.33 ^e	6.3 \pm 0.36 ^{de}	9.4 \pm 0.56 ^{dce}	14.2 \pm 0.89 ^c	23.8 \pm 1.7 ^b	12.1 \pm 1.06 ^{dc}	70.4 \pm 2.71 ^a
	<i>N. tenuis</i>	4.2 \pm 0.41 ^e	4 \pm 0.33 ^e	9.1 \pm 0.67 ^{dce}	14.4 \pm 0.63 ^c	27.5 \pm 1.36 ^b	9 \pm 1.01 ^{dce}	75.8 \pm 3.92 ^a
1 st instar larvae	<i>M. pygmaeus</i>	2.7 \pm 0.30 ^g	3.3 \pm 0.30 ^{gf}	5 \pm 0.25 ^{def}	5.9 \pm 0.34 ^{dc}	6.5 \pm 0.34 ^{dc}	2.3 \pm 0.30 ^g	14 \pm 0.59 ^b
	<i>N. tenuis</i>	2.8 \pm 0.24 ^g	3.7 \pm 0.26 ^{gef}	5.1 \pm 0.31 ^{de}	5.9 \pm 0.34 ^{dc}	7.3 \pm 0.33 ^c	2.7 \pm 0.21 ^g	16.8 \pm 0.69 ^a
2 nd instar larvae	<i>M. pygmaeus</i>	2.8 \pm 0.24 ^{fe}	2.8 \pm 0.24 ^{fe}	4.3 \pm 0.26 ^{dce}	4.4 \pm 0.33 ^{dce}	5.3 \pm 0.3 ^c	2 \pm 0.25 ^f	11.6 \pm 0.52 ^b
	<i>N. tenuis</i>	2.1 \pm 0.23 ^f	2.7 \pm 0.3 ^{fe}	3.5 \pm 0.34 ^{dfe}	4.9 \pm 0.31 ^{dc}	5.6 \pm 0.49 ^c	2.4 \pm 0.3 ^f	13.6 \pm 0.61 ^a
3 rd instar larvae	<i>M. pygmaeus</i>	1.9 \pm 0.27 ^{cd}	2 \pm 0.21 ^{cd}	3.1 \pm 0.31 ^{ebcd}	3.4 \pm 0.33 ^{cbd}	4.4 \pm 0.26 ^b	1.8 \pm 0.29 ^c	8.9 \pm 0.48 ^a
	<i>N. tenuis</i>	1.7 \pm 0.26 ^e	2.3 \pm 0.3 ^{cd}	2.8 \pm 0.32 ^{ced}	4 \pm 0.25 ^{cb}	4.1 \pm 0.34 ^{cb}	1.8 \pm 0.24 ^c	9.3 \pm 0.39 ^a

Means with the same letter in each prey stage are not significantly different according to Tukey's studentized range test ($P > 0.05$).

Table 2. Expected and calculated Manly's preference-index (β_A) for fifth instars (N5) and female adults of *M. pygmaeus* and *N. tenuis* in each of the three tested prey combinations. Calculated β_A values indicate the probability of the next prey.

Predator	Prey combination	Prey ratio	Expected β_A	*Calculated β_A (Means \pm SE)	
				N5	Female
<i>M. pygmaeus</i>	Egg-L1	50-10	0.833	0.901 \pm 0.031 ^{aA}	0.857 \pm 0.036 ^{aA}
<i>N. tenuis</i>	Egg-L1	50-10	0.833	0.847 \pm 0.039 ^{aA}	0.853 \pm 0.029 ^{aA}
<i>M. pygmaeus</i>	L1-L2	10-10	0.5	0.745 \pm 0.089 ^{aA}	0.646 \pm 0.069 ^{aA}
<i>N. tenuis</i>	L1-L2	10-10	0.5	0.7008 \pm 0.065 ^{aA}	0.601 \pm 0.079 ^{aA}
<i>M. pygmaeus</i>	L2-L3	10-10	0.5	0.826 \pm 0.055 ^{aA}	0.653 \pm 0.069 ^{aA}
<i>N. tenuis</i>	L2-L3	10-10	0.5	0.721 \pm 0.073 ^{aA}	0.679 \pm 0.071 ^{aA}

* Calculated β_A values indicate the probability that the next prey will be the first one mentioned in the combination. Values within the same column in each section between *M. pygmaeus* and *N. tenuis* followed by the same lowercase letter indicate no significant difference ($P > 0.05$; Wilcoxon rank sum test). Values within same row followed by the same uppercase letter indicate no significant difference ($P > 0.05$; Wilcoxon rank sum test).

than that of *M. pygmaeus* (table 1). The predation of different stages of both predators on the first instars was statistically different ($F = 289.25$, $df = 6$, 126 , $P < 0.0001$). Predation by male adults and first instar of both predators was significantly lower than that by the other stages except second nymph of *M. pygmaeus* (table 1).

Overall, there were no significant differences in total predation between *M. pygmaeus* and *N. tenuis* on DBM second instar larvae ($F = 1.41$, $df = 1$, 126 , $P = 0.237$). However, the predation on second instars by different stages of each predator did differ ($F = 201.43$, $df = 6$, 126 , $P < 0.0001$). The predation by female adults of *N. tenuis* on this stage was significantly higher than that by *M. pygmaeus*, whereas the predation rate of male adults of both predators and first instar of *N. tenuis* was significantly lower than that of the other stages (table 1).

ANOVA revealed no significant differences between the two predators when fed on DBM third instar larvae ($F = 0.18$, $df = 1$, 126 , $P = 0.672$). Results indicated that the different stages of each predator differed significantly in their predation on third instars of the pest ($F = 132.67$, $df = 6$, 126 , $P < 0.0001$). The highest predation rate on this stage was observed for female adults, whereas the lowest was found for male adults and first instar of *N. tenuis* (table 1).

Prey preference of *M. pygmaeus* and *N. tenuis* for different stages of *P. xylostella*

The relationships between expected and calculated Manly's preference-index (β_A) for fifth instars and female adults of *M. pygmaeus* and *N. tenuis* in various combinations of different DBM stages are shown in table 2. There was no significant difference in prey preferences of neither fifth instar nymphs nor female adults between the mirid predators in all tested combinations of DBM stages [fifth instar nymphs: Egg-L1 ($S = 93.5$, $df = 18$, $P = 0.399$), L1-L2 ($S = 96.5$, $df = 18$, $P = 0.537$) and L2-L3 ($S = 91.5$, $df = 18$, $P = 0.314$); female adults: Egg-L1 ($S = 98.5$, $df = 18$, $P = 0.649$), L1-L2 ($S = 99.5$, $df = 18$, $P = 0.701$) and L2-L3 ($S = 109.5$, $df = 18$, $P = 0.759$)]. The prey preference between the fifth instar nymphs and female adults of *N. tenuis* did not differ in

the several tested combinations: Egg-L1 ($S = 108$, $df = 18$, $P = 0.850$), L1-L2 ($S = 116.5$, $df = 18$, $P = 0.400$) and L2-L3 ($S = 109.5$, $df = 18$, $P = 0.759$). Likewise, no significant differences in prey preference between the fifth instar nymphs and female adults of *M. pygmaeus* were detected [Egg-L1 ($S = 116.5$, $df = 18$, $P = 0.399$), L1-L2 ($S = 118$, $df = 18$, $P = 0.337$) and L2-L3 ($S = 129.5$, $df = 18$, $P = 0.064$)] (table 2).

Both fifth instar nymphs and female adults of either predator had a distinct preference to first instar larvae of the pest in the L1-L2 prey combination. A clear preference for second instar larvae was observed in nymphs and adults of both predators, when exposed to the L2-L3 combination of DBM.

Discussion and conclusion

Many ecological, physiological and behavioural processes in insects are linked to their nutritional context. The alimentary regime may have an impact on the life cycle of predators, because better fed individuals reproduce more and benefit in terms of growth, survival, and longevity (Lemos *et al.*, 2003; Urbaneja-Bernat *et al.*, 2013). Therefore, a good understanding of the alimentary preferences and nutritional needs of a natural enemy is elementary for its implementation in biological control strategies. The result of this study indicated that female adults and fifth instars of both predators were the most voracious stages on DBM regardless of exposure stage. This suggests that larger nymphs and female adults of two mirid bugs extract more nutrients from different DBM stage at an appropriate rate to fully support their growth and reproduction. The data of the current study suggest that *M. pygmaeus* and *N. tenuis* are effective predators of DBM particularly against eggs. This might be due to the fact that nymphs and adults of both predators fed in clusters which make them relatively easy to handle them and DBM eggs have smaller size and they are immobile as well. Within the three DBM larval instars tested, for each stage of both predators, it was seen that higher is the larval instar, lower is the consumption. This is to be expected as it is more difficult for predator to handle larger mobile larvae.

The findings of the current study are consistent with previous studies reporting a preference for eggs to larvae of lepidopterans in both third instars and female adults of *M. pygmaeus* and *N. tenuis* (Arnó *et al.*, 2009; Urbaneja *et al.*, 2009; Chailleux *et al.*, 2013). Arnó *et al.* (2009) stated that adults and nymphs of *M. pygmaeus* and *N. tenuis* were able to prey on eggs and larvae of the tomato leaf miner *T. absoluta*. The nymphs of *N. tenuis* consumed more eggs than those of *M. pygmaeus*, while the adults of both mirid species had higher predation rates than nymphs. The overall predation by females of both mirid species upon first and second instar larvae of *T. absoluta* was very low. In the current study the observed preference for DBM eggs over larvae may be explained by their immobility and the size of the prey. The numbers of DBM eggs that were consumed by the different stages of *M. pygmaeus* and *N. tenuis* were higher as compared with the other stages of DBM. It can therefore be assumed that these two predators prey more on the immobile stages of their lepidopteran prey. This finding corroborates the reports of Chailleux *et al.* (2013) and Jaworski *et al.* (2013) who indicated that the predation rate of these predators on larvae of *T. absoluta* was low. Our study also showed that the female adults of *M. pygmaeus* and *N. tenuis* killed more DBM larval instars as compared to the nymphal instars of the mirids. These differences can be due to the defence reactions of the larvae. The larger size of the DBM later instars may be associated with stronger defence displays against attacks by different stages of both predators, even of the later stages. The first instars of both predators displayed the lowest predation rates on the different stages of DBM. Hillert *et al.* (2002) speculated that this could have resulted from the small size of the first instar nymphs of both predators, their searching behaviour and prey location abilities.

Although our study suggests that *M. pygmaeus* and *N. tenuis* may have limited effectiveness on DBM larval stages but they could be a source of mortality of prey larvae in the field, especially first-instar larvae. However, the importance of this mortality should be explored in further studies in a more realistic setting. A combination of different biological control agents with different mechanisms of action may therefore be useful in the suppression of this key pest of crucifers. Previous reports have shown that releases of *M. pygmaeus* together with the egg parasitoid *Trichogramma achaeae* Nagaraja et Nagarkatti (Hymenoptera Trichogrammatidae), or of *N. tenuis* combined with *Bacillus thuringiensis* Berliner against the early instars of *T. absoluta* were more reliable strategies for control of this pest (Mollá *et al.*, 2011; Trottin-Caudal *et al.*, 2012).

The findings of the current study indicate that the DBM eggs were more suitable prey to the mirid than the larvae of DBM that in accordance with Ebrahimi *et al.* (2019). Lykouressis *et al.* (2009) found that consumption rate by fifth instars of *M. pygmaeus* on eggs of *T. vaporariorum* was significantly higher than that on different nymphal stages of the whitefly. They showed that with the increase of age of the whitefly nymphs, consumption rate was reduced (Lykouressis *et al.*, 2009). A possible explanation

for this reduction in consumption rate might be that the larger prey requires longer time to handle (i.e. to attack and feed on).

Factors such as prey species, predator hunger level, mechanisms of prey defence, and/or the nutritional quality of prey individuals can affect the prey-stage preference of predators (Badii *et al.*, 2004; Carrillo and Pena, 2012). It is worth noting that in prey preference experiments, a relatively high number of prey individuals should be provided. A too limited number of prey can give rise to the fact that predation of a certain prey type occurred out of hunger instead of preference. Jaworski *et al.* (2013) noted that the preference of *M. pygmaeus* for *Bemisia tabaci* (Gennadius) over *T. absoluta* was related to the ratio of the offered prey. These authors showed that the experimental arena may affect the observed prey preference of predators. Furthermore, Maselou *et al.* (2014) indicated plant resources may affect prey preference of predators and thus their potential for biological control of target pests.

The findings of our laboratory study indicated that all stages of *M. pygmaeus* and *N. tenuis* can significantly contribute to the control of the eggs of *P. xylostella* but only to a limited extent to that of the larvae. However, lower predation rates and preference indices recorded for these two predators towards the larval stages of *P. xylostella* may be an obstacle for their use to suppress larval stages of this and other lepidopteran defoliators. It would therefore be imperative to further develop and evaluate strategies for incorporating these mirids in biological control programs against DBM in cabbage fields and greenhouses.

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