Bulletin of Insectology ***72*** *(2): 273-280, 2019*
ISSN 1721-8861 eISSN 2283-0332

Predation rate of competing *Chrysoperla carnea* and *Hippodamia variegata* on *Aphis fabae* at various prey densities and arena complexities

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Abstract

We measured predation rate of several developmental stages of two aphidophagous predators - larvae of lacewing *Chrysoperla carnea* (Stephens) (Neuroptera Chrysopidae) and larvae and adults of ladybird *Hippodamia variegata* (Goeze) (Coleoptera Coccinellidae) combined together in the same experimental arena. Two arenas with different complexity but similar surface were compared, simple with aphids on a leaf of host plant, and microcosm with potted plant *Vicia faba* L. The initial number of prey, larvae of aphid *Aphis fabae* Scopoli (Rhynchota Aphididae), was set up to 50, 150 and 400 individuals. The first day of experiment, after previous starvation, the pair of predators exhibited type I functional response. The second and third days, after feeding ad libitum, the predators exhibited type II functional response. Consumption rate was lower on the second day (130 aphids of 400) than on the first day (180 aphids) in the simple arena, probably due to an effect of starvation vs. feeding ad libitum. Consumption increased day after day in the microcosm arena (1st day: 30 of 400 aphids, 2nd day: satiation limit 174 aphids / 3rd day: 261 aphids), probably due to the habituation to the complex environment. Overall predation rate and control mortality of aphids without predators at the microcosm arena were lower than in the simple arena.

Key words: variegated lady beetle, green lacewings, voracity, feeding rate.

Introduction

Predation rate or voracity is an important parameter characterizing relationship between a predator and its prey, useful for prediction of pest control in the field and prey consumption in artificial rearings of biocontrol agents. Because the increase of killed and consumed prey individuals is seldom linear with the increase of prey available, reaction norms called functional response I, II, III were established by Holling (1959). Estimation of the type and the equation describing functional response of concrete predator and prey relationship requires experimental set up with several prey densities. The type II functional response is the most frequently reported type in insects including coccinellids (Hodek and Honěk, 1996; Dixon, 2000).

Although the initial number of prey individuals is usually considered and controlled for in experiments quantifying functional response (e.g. Jiang *et al*., 2019), the prey density per surface area (Reynolds, 2012) or volume water (Mondal *et al*., 2017) can also be considered. Closed artificial laboratory and semi-field arenas as well as real plants can differ in their architecture or complexity, even when surface area is kept identical. It is considered more difficult to find prey in more complex (which often means more realistic) arena, so that the estimated predation rate should be lower (Reynolds, 2012). Influence of predator foraging by physical plant characteristics was examined e.g. in females of *Stethorus gilvifrons* (Mulsant) foraging for *Tetranychus urticae* Koch on castor bean, common bean, and cucumber leaves (Bayoumy *et al*., 2014). The two response parameters - attack rate and handling time - varied independently.

Starvation for the period of 24 hours prior the experiment is often used to motivate the predators to high consumption rate (Jalali *et al*., 2010; Jiang *et al*., 2019). However, such situation would not be natural if continuous feeding is available either in laboratory or in the field, and ad libitum feeding prior to the experiment can be recommended (Papanikolau *et al*., 2016).

If predators are abundant and prey rare, they compete for the prey. Combined predation rate of several predators can vary with prey density due to the competition between conspecifics or functionally similar species but also due to resource partitioning between functionally distinct species. Per capita searching efficiency and killing power decreased with increasing number of females of *S. gilvifrons* preying on *T. urticae* (Bayoumy *et al*., 2014). Combining distinct predators of *Leptinotarsa decemlineata* Say increased predation only when prey densities were low (Werling *et al.*, 2012). When aphidophagous insect larvae are considered, that have limited mobility and tend to aggregate at prey patches, they are exposed to each other. Frequent encounters may affect their foraging success: the feeding rate was almost independent of predator density at high prey density, but dependence in the functional response was observed at the combination of low prey and high predator density (Papanikolaou *et al.*, 2016).

Predation sometimes combines with competition into intraguild predation (IGP) which occurs when potential competitors of a shared prey (the extraguild = EG prey) attack each other (Polis *et al.*, 1989). Before we start to

observe IGP, as done by Tavoosi Ajvad *et al.* (2014) regarding the combination of ladybird *Hippodamia variegata* (Goeze) and hoverfly *Episyrphus balteatus* (De Geer) the evaluation of EGP by multiple predators combined in a common arena should be considered (Chang, 1996; Devee *et al*., 2018). Effect of diverse prey density on IGP of aphidophagous predators must be evaluated (Burgio *et al*., 2002; 2005; Rondoni *et al*., 2014).

Our study uses a unique experimental set up for quantification of predation rate and functional response of pairs of predators - diverse developmental stages of two commonly occurring aphid predators - lacewing *Chrysoperla carnea* (Stephens) and ladybird *H. variegata* in diverse EG prey densities and two arenas with different complexity but similar surface, and both initial starvation and later feeding ad libitum. The widespread and common aphid, *Aphis fabae* Scopoli, accepted by both predators as essential food (Hodek and Evans, 2012), was used as their prey.

Materials and methods

Prey

Initial colony of the aphid *A. fabae* was collected in the broad bean (*Vicia faba* L.) fields in Hamedan province, Iran. We established laboratory culture on potted broad bean (c.v. Barekat) grown in a greenhouse at 25 ± 5 °C, 50 ± 10% RH and 16:8 L:D.

Predators

Adults of ladybird *H. variegata* and lacewing *C. carnea* were collected in alfalfa fields in Hamedan Province, Iran. We established rearings in a climate controlled chamber at 25 ± 1 °C , 60 ± 10% RH and 16:8 L:D. The ladybirds were kept in ventilated Petri dishes (9 cm diameter), lacewings were placed in oviposition containers (6.5 cm diameter, 11.5 cm height). The larvae of *H. variegata* and *C. carnea* were fed daily and ad libitum with *A. fabae* and eggs of Mediterranean flour moth, *Ephestia kuehniella* Zeller respectively. Adult lacewing were also supplied with a mixture of yeast, water and honey and ladybirds were offered with honey solution (10%) as supplement food. Both predators were reared for five generations prior to experiments. Predators laid eggs on the provided oviposition substrate (pieces of corrugated paper for ladybirds and black cardboard for lacewings). Eggs were collected every 8 hours (ladybirds) or daily (lacewings) and transferred to separate Petri dishes (3 cm diameter, 1 cm height). Dishes were checked for egg hatching every 12 hours and larvae were transferred to individual Petri dishes where they were fed with *A. fabae* ad libitum until they reached the required stage. All larvae were starved for six hours prior the experiment to increase their incentive to search for prey.

Experimental design

To assess the influence of arena complexity, two experimental set-ups were used: 1) Simple arena: ventilated Plexiglas containers (12 × 9 cm, 4 cm height) with black bean aphids feeding on two broad bean leaves placed upside down on a moistened cotton wool and petioles covered with cellophane; the two predators were placed each on its own leaf; 2) Microcosm arena: two transparent plastic containers (drink cups), each 12 cm height, 7 cm diameter; bottom one with soil and a potted broad bean seedling (about 10 cm high with 4 to 6 leaves); top one ventilated, enclosing the seedling with black bean aphids evenly distributed on leaves; the predators were placed randomly on the plants.

According to preliminary assays, we used three initial aphid densities: 50, 150 and 400 3rd instar nymphs of *A. fabae* in both arenas. Immature aphids were used to avoid any reproduction during the experiments (Papanikolau *et al*., 2016). Two predators were competing for this prey: all six combinations of one larva of second (2nd) or third (3rd) instar of *C. carnea* and one larva of third (3rd) or fourth (4th) instar or adult female of *H. variegata* were used for all prey densities and both arenas. The females *H. variegata* were three days old and mated. The first and second instar ladybeetle larvae and the first instar lacewing larvae were excluded from the experiments due to their relatively low predation rate. In addition, control without any predator was set up to assess inherent aphid mortality. All experimental units were checked non-destructively after three 24-h intervals, i.e. after 24, 48 and 72 hours. The number of alive aphids was recorded, the number of dead aphids calculated. Prey was not replenished during the experimental period, so that the initial aphid density for the second and third days varied. Ten replications per each combination of predator stages and aphid density were used at both arenas. Experiments took place in a climate controlled chamber at 25 ± 1 °C, 60 ± 10% RH and 16:8 L:D.

Statistical analysis

The number of aphids killed by the predators was corrected for mortality in the control: Corrected mortality = Observed mortality in treatment – Initial density in treatment × Mortality in control / Initial density in control. If the corrected predation rate was negative in a particular replication, it was set to zero. Mann-Whitney U Test was employed to compare control mortality between the simple arena and microcosm.

We used the Hartley (F), Cochran (C) and Bartlett (χ^2) tests for homogeneity of variances to judge whether General Linear Models in Statistica 13 (TIBCO, 2017) are appropriate for computation of significance of the categorical factors. Initial prey density, developmental stage of *C. carnea* and developmental stage of *H. variegata* were analysed for explaining predation rate on the initial day (24 hours) separately for each arena.

Polynomial logistic regression equations and associated parameters are used for estimation of functional response (Mondal *et al*., 2017). For the second day of experiment in both arenas and the third day in microcosm, we calculated functional response curve of the type II: $y = (a \times x) / (1 + a \times h \times x)$, where a stands for attack rate and h for handling time. These parameters with standard errors were calculated using Nonlinear Estimation utility in Statistica 13 (TIBCO, 2017). On the first day, the functional response did not fit to the type II,

resembled rather type I with no possibility to calculate satiation level. For the third day in simple arena, very low initial aphid density remained that did not allow expression of real predation capacity of the predators nor comparison of parameters with the other treatments.

Results

Simple arena, 24 hours

Average control mortality of aphids after the first 24-h period of the experiment in simple arena was 23.6 ± 5.3%. Predation rate in the experimental treatments was corrected by this constant.

For the prey consumption rate after the first 24-h period of the experiment, tests for homogeneity of variances was not significant ($F = 1.03$, $C = 0.51$, $\chi^2 = 0.016$, $p = 0.90$). GLM revealed the effects of initial prey density and stages of *H. variegata* figured below, and non-significant effect of *C. carnea* stages on prey consumption rate.

Predation rate of all six combinations of predators observed during the first 24 hours of the experiment was strongly gradually and linearly increasing ($F_{2,174} = 786$, $p < 10^{-6}$) with the initial number of aphids (figure 1). However, the percentage of aphids killed decreased from 61 ± 15% at the lowest initial prey density (50 aphids), to 45 ± 13% and 45 ± 8% at the medium and high densities (150 and 400 aphids).

The combined prey consumption rate during initial 24 hours did not differ significantly between 2nd and 3rd instars of the lacewing *C. carnea* ($F_{1,174} = 2.29$, $p = 0.132$). Combined with *H. variegata*, 2nd instar of *C. carnea* consumed in average 29 ± 8 of 50, 65 ± 20 of 150 and 177 ± 30 of 400 initial aphids, while the 3rd instar of *C. carnea* combined with ladybird consumed 33 ± 6 of 50, 70 ± 19 of 150 and 183 ± 35 of 400 initial aphids.

The combined prey consumption rate during initial 24 hours differed significantly among the stages of the ladybird *H. variegata* ($F_{2,174} = 7.66$, $p = 0.00065$). Consumption of the third instar larvae was smaller than that of the 4th instar (Tukey HSD test, $p = 0.00035$), while consumption of adult females was intermediate, not different from the other two stages. Combined with *C. carnea*, 3rd instar of *H. variegata* consumed 25 ± 8 of 50, 57 ± 17 of 150 and 173 ± 32 of 400 initial aphids, 4th instar of *H. variegata* consumed 33 ± 6 of 50, 78 ± 15 of 150 and 189 ± 32 of 400 initial aphids, and adult females of *H. variegata* consumed 34 ± 6 of 50, 68 ± 21 of 150 and 178 ± 33 of 400 initial aphids.

Estimation of separate predation rates for individual predators is 10.2, 25.6, 83.6 for 2nd instar of *C. carnea*, 14.2, 31.0, 88.8 for 3rd instar of *C. carnea*, 13.3, 28.3, 86.2 for 3rd instar of *H. variegata*, 21.4, 49.6, 102.8 for 4th instar of *H. variegata*, and 21.5, 39.3, 91.9 for adult of *H. variegata* for the three initial prey densities (50, 150, 400 aphids).

Simple arena, 48 hours

For the prey consumption rate during the second day of the experiment, we calculated the initial number of aphids as those left alive after the first 24-h period (after predation and natural mortality) and then observed how many aphids were eaten, considering mortality in control, during the second 24-h period. Large proportion of the prey items was eaten, decreasing at high densities, resulting in a functional response type II curve with attack rate $a = 0.842 \pm 0.054$ and handling time $h = 0.00480 \pm 0.00064$ (figure 2). The predicted number of aphids eaten when 50 were offered was 35, 79 eaten for initial 150 and 129 for the initial 400 aphids. Satiation limit (curve plateau) was 208 aphids.

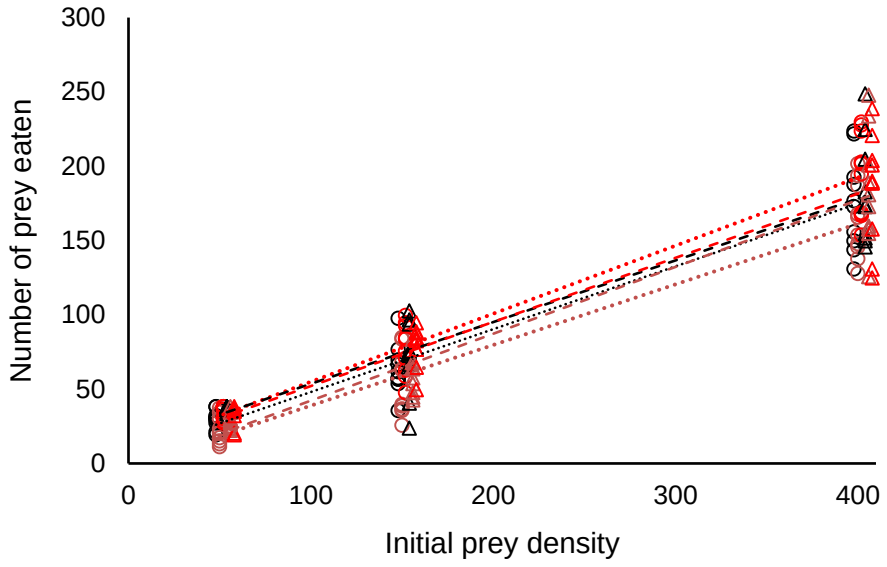

Figure 1. Functional response of six combinations of developmental stages of *C. carnea* and *H. variegata* to three initial prey densities of *A. fabae* in simple arena. Circles and dotted lines: *C. carnea* 2nd instar; triangles and dashed lines: *C. carnea* 3rd instar; black: *H. variegata* adult female; orange: *H. variegata* 3rd instar; red: *H. variegata* 4th instar. Linear trend lines.

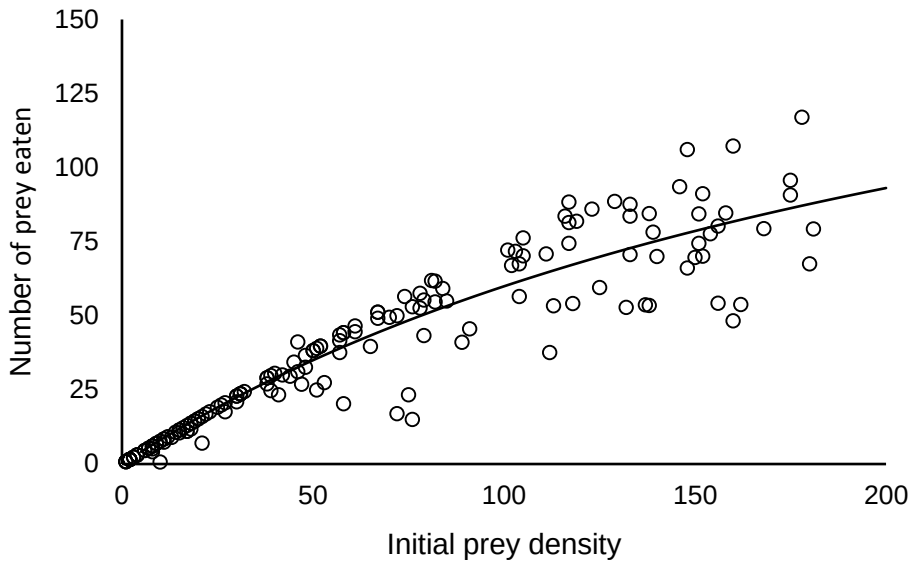

Figure 2. Functional response of merged combinations of developmental stages of *C. carnea* and *H. variegata* to the prey densities of *A. fabae* left after the first 24 hours of the experiment eaten during the next 24 hours in simple arena. Functional response type II curve: $y = (0.842 \times x) / (1 + 0.842 \times 0.0048 \times x)$, $R^2 = 0.882$.

Microcosm, 24 hours

Average control mortality of aphids after the first 24-h period of the experiment was 4.8 ± 2.2%. Test revealed very significant difference from the mortality in the control of simple arena ($Z = 3.74$, $p = 0.00018$).

For the prey consumption rate after the first 24-h period of the experiment, tests for homogeneity of variances was not significant ($F_{1,178} = 2.59$, $p = 0.109$). GLM revealed the effects of initial prey density and stages of *C. carnea* figured below, and non-significant effect of *H. variegata* stages on prey consumption rate.

Predation rate of all six combinations of predators observed during the first 24 hours of the experiment was strongly gradually and linearly increasing ($F_{2,174} = 45$, $p < 10^{-6}$) with the initial number of aphids (figure 3). However, the percentage of aphids killed fluctuated from 16.7 ± 17.2% at the lowest initial prey density (50 aphids), trough 20.0 ± 11.2% at the medium density (150 aphids) to 9.9 ± 6.7% at the high density (400 aphids). Within-treatment variability of consumption rate in microcosm was enormous (see the SD values compared to means).

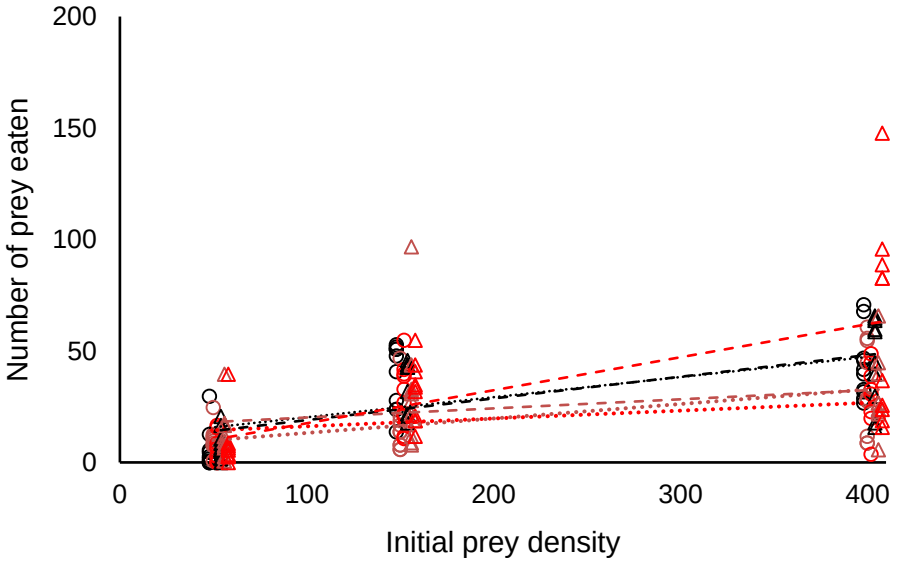

Figure 3. Functional response of six combinations of developmental stages of *C. carnea* and *H. variegata* to three initial prey densities of *A. fabae* in microcosm arena. Circles and dotted lines: *C. carnea* 2nd instar; triangles and dashed lines: *C. carnea* 3rd instar; black: *H. variegata* adult females; orange: *H. variegata* 3rd instar; red: *H. variegata* 4th instar. Linear trend lines.

The combined prey consumption rate during initial 24 hours differed significantly between 2nd and 3rd instars of *C. carnea* ($F_{1,174}$ = 5.02, p = 0.026). Combined with *H. variegata*, 2nd instar of *C. carnea* consumed in average 7.4 ± 7.4 of 50, 28.0 ± 16.3 of 150 and 33.2 ± 19.5 of 400 initial aphids, while the 3rd instar of *C. carnea* combined with *H. variegata* consumed more: 9.3 ± 9.7 of 50, 31.8 ± 17.5 of 150 and 46.0 ± 31.3 of 400 initial aphids.

The prey consumption rate during initial 24 hours of the 3rd instar larvae combined with ladybirds was the least, that of the fourth instar intermediate, and the consumption of adult females was highest. However, due to the high variability, the average consumption did not differ significantly among the stages of *H. variegata* ($F_{2,174}$ =2.20, p = 0.113). Combined with *C. carnea*, 3rd instar of *H. variegata* consumed 11.2 ± 8.7 of 50, 23.6 ± 21.2 of 150 and 31.5 ± 17.8 of 400 initial aphids, 4th instar of *H. variegata* consumed 6.9 ± 9.0 of 50, 30.5 ± 13.4 of 150 and 42.6 ± 38.5 of 400 initial aphids, and adult females of *H. variegata* consumed 6.9 ± 7.8 of 50, 35.8 ± 13.2 of 150 and 44.6 ± 17.1 of 400 initial aphids.

Estimation of separate predation rates for individual predators is 4.7, 9.9, 9.3 for 2nd instar of *C. carnea*, 6.6, 13.7, 22.1 for 3rd instar of *C. carnea*, 5.6, 11.8, 15.8 for 3rd instar of *H. variegata*, 1.3, 18.7, 26.9 for 4th instar of *H. variegata*, and 1.2, 24.0, 28.9 for adult of *H. variegata* for the three initial prey densities (50, 150, 400 aphids).

Microcosm, 48 hours

For the prey consumption rate during the second day of the experiment, we calculated the initial number of aphids as those left alive after the first 24-h period (after predation and natural mortality) and then observed how many aphids were eaten, considering mortality in control, during the second 24-h period. Small proportion of the prey items was eaten, decreasing at high densities, resulting in a functional response type II curve with attack rate a = 0.189 ± 0.032, and handling time h = 0.00576 ± 0.00290 (figure 4). The predicted number of aphids eaten when 50 were offered was 9, 24 eaten for initial 150 and 53 for the initial 400 aphids. Satiation limit (curve plateau) was 174 aphids.

Microcosm, 72 hours

For the prey consumption rate during the third day of the experiment, we calculated the initial number of aphids as those left alive after 48-h period (after predation and natural mortality) and then observed how many aphids were eaten, considering mortality in the control, during the third 24-h period. Higher proportion of the prey items than after 48-h was eaten, decreasing at high densities, resulting in a functional response type II curve with attack rate a = 0.926 ± 0.061, and handling time h = 0.00383 ± 0.00032 (figure 5). The predicted number of aphids eaten when 50 were offered was 39, 91 eaten for initial 150 and 153 for the initial 400 aphids. Satiation limit (curve plateau) was 261 aphids.

Discussion

Simple arena, 24 hours

As expected, the more aphids were offered, the more aphids were consumed by the two predators in both types of arenas and during each day of observation. However, the increase was surprisingly linear over a wide range of initial prey densities during the first day. This means that the functional response resembles most the type I and that the highest potential consumption rate was not achieved and even cannot be estimated. Number of aphids eaten at the highest offered initial

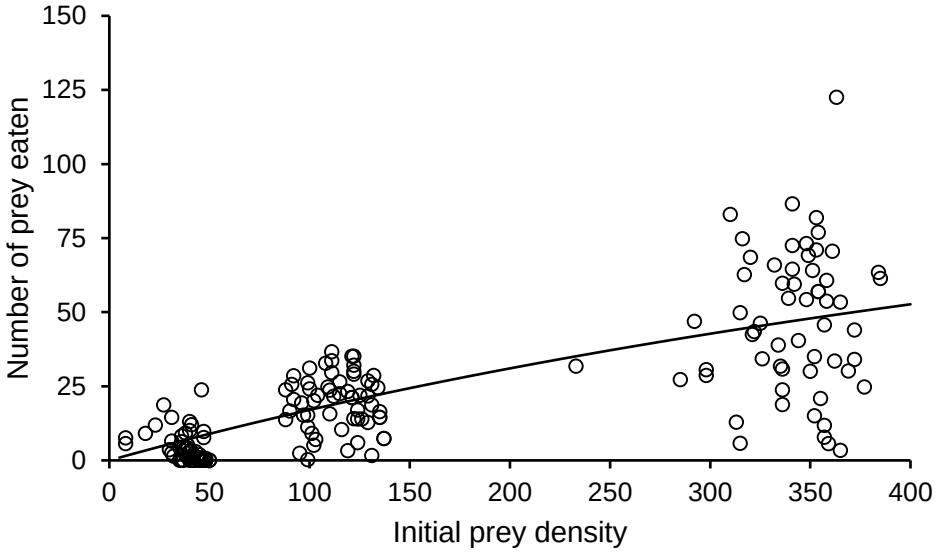

Figure 4. Functional response of merged combinations of developmental stages of *C. carnea* and *H. variegata* to the prey densities of *A. fabae* left after the first 24 hours of the experiment eaten during the next 24 hours in microcosm arena. Functional response type II curve: $y = (0.189 \times x) / (1 + 0.189 \times 0.00576 \times x)$, $R^2 = 0.523$.

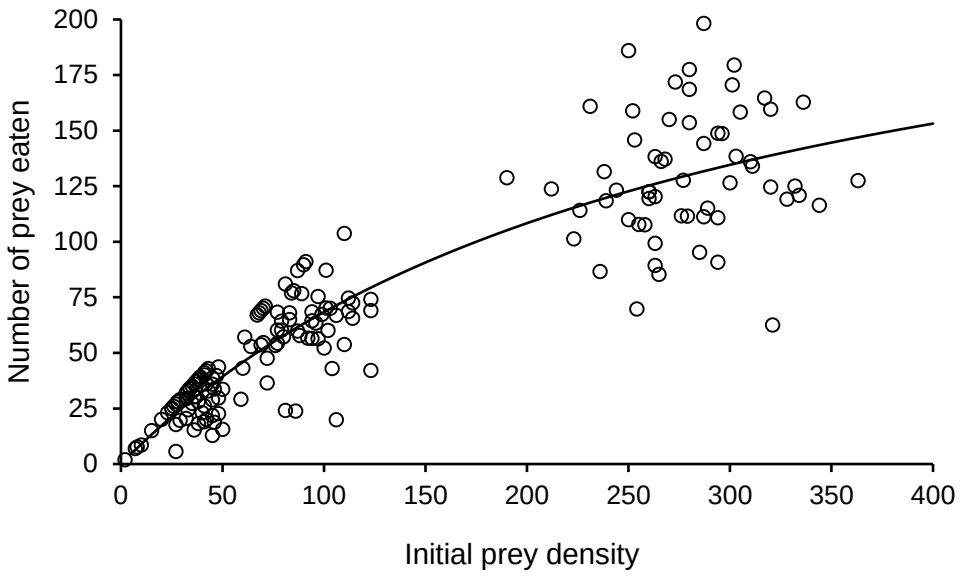

Figure 5. Functional response of merged combinations of developmental stages of *C. carnea* and *H. variegata* to the prey densities of *A. fabae* left after 48 hours of the experiment and eaten during the next 24 hours in microcosm arena. Functional response type II curve: $y = (0.926 \times x) / (1 + 0.926 \times 0.00383 \times x)$, $R^2 = 0.828$.

density was 180. The functional response of type I is rarely observed. It was, however, found when pupae of the beetle *Dinoderus porcellus* Lesne were offered simultaneously to two predators, assassin bug *Alloeocranum biannulipes* Montrouzier et Signoret and beetle *Teretrius nigrescens* Lewis (Loko *et al*., 2017). The immobility of pupae possibly contributed to the linear increase of the predation rate. Both the combination of two predators and low mobility of aphids also occur in our experiments leading to this type I response.

Although the predators ate only about 45% of 150 aphids offered, they consumed similar proportion of prey at the highest density, i.e. more than 150 individuals if 400 aphids were offered. It suggests a) high biocontrol abilities even at high aphid densities, b) the importance of high density of prey provided in laboratory rearings and experiments. Not all individuals were consumed whole, which would require a correction if energetic balance of the predation were considered, but which does not make any difference for biocontrol use. For comparison, *Harmonia axyridis* (Pallas) sucked out 26 to 58% aphid individuals without eating entire body (Šenkeříková and Nedvěd, 2013).

Feeding rate changes much during ladybird larval development. Thus, if a daily average number of prey eaten was calculated over the four instars of *H. variegata*, it showed much lower values (64 third and fourth instars of *Agonoscena pistaciae* Burckhardt et Lauterer, 69 of *Aphis gossypii* Glover; Jalali *et al*., 2018) than those measured in our study.

Simple arena, 48 hours

The number of aphids remaining alive after the first day of experiment became the initial number of prey for the second day of the experiment. We thus achieved almost continuous distribution of initial prey density that allowed for smooth estimation of the response curve (figure 2). Although the number of prey eaten was similar at the moderate prey density (about 70 eaten of 150 offered) in both days, consumption rate was lower during the second day (130 aphids) than during the first day (180 aphids) at the highest density, leading to the response type II. We attribute the decrease to the fact that the predators were starved before the first day, but they were fed on the second day. Estimation of consumption rate on the third day was impossible due to the low initial aphid number left.

Microcosm, 24 hours

Much smaller control mortality in the microcosm arrangement (5%) than in the simple arena (24%) suggest better plant quality and microclimatic conditions (probably higher humidity) that can also affect performance of the predators, either directly, or indirectly through the number of prey items alive. Because it is not excluded that predators are also scavenging on some of the dead aphids (like in the ladybird *Cheilomenes lunata* F., Bayissa *et al*., 2016), the low control mortality in microcosm leads to better estimation of true consumption rate of the predators.

There was much higher individual variability in the feeding rate in the microcosm arena (figure 3) than in the simple arena (figure 1) and the average feeding rate was much lower in microcosm (about 30 aphids eaten of 150 offered) than in the simple arena (about 70 eaten of 150 offered). Both differences are attributable to the complexity of arena without much difference in its size. Two experimental arrangements similar to our two arenas were compared for *H. axyridis*, and green lacewing larvae, *C. carnea*, feeding on pea aphids, *Acyrthosiphon pisum* (Harris). In agreement with our observation, predators consumed significantly more aphids in homogeneous environment of Petri dishes than in heterogeneous environment (whole plants) of the same area (Reynolds, 2012).

Microcosm, 48 and 72 hours

The average number of prey eaten during the first day in microcosm when the highest initial number of aphids (400) was offered was only 30. Contrastingly, satiation limit (curve plateau of the response type II) of the predators during the second day was 174 aphids, and 261 aphids during the third day of experiment. This steady increase, occurring despite the fact that predators were starved before the first day and fed ad libitum afterwards, suggests habituation or learning process in the predators. They probably needed time to adapt to the complexity of microcosm arena, a phenomenon not mentioned for any insect predator of aphids. We thus recommend to use several subsequent days when measuring functional response of predators, even simple ones like insects, in complex microcosm and semi-field experiments.

General comparisons

Estimation of predator functional response in the field was modelled as extrapolation from laboratory observations made in *Delphastus catalinae* (Horn) feeding on *Bemisia tabaci* (Gennadius). It was predicted that type II observed on individual leaflets switches to the type III on whole plants (Rincon *et al*., 2017). The efficacy of predators as biological control agents showing type II functional response have been questioned. Predators showing type response of III are theoretically more capable of suppressing prey populations (Holling, 1965). Although the type of the functional response is an important factor, it is insufficient as a criterion to predict success or failure of a predator as a biocontrol agent (Jalali *et al*., 2010).

Our experimental predators are common species that can be found and used in the nature and in agricultural ecosystems. However, *C. carnea* forms a group of cryptic species that can be reliably determined by their courtship song. New species are being described recently, even from Iran (Henry *et al*., 2018), and thus we did not determine the exact identity of our experimental species. However, no important differences in predation rate are expected. Voracity of our predators was higher than the voracity of similar sized species. An individual *Adalia bipunctata* (L.) female, that has similar body size to our *H. variegata*, can kill a theoretical maximum number of 67 3rd-4th instars of *Myzus persicae* (Sulzer) per day at 23 °C (Jalali *et al*., 2010). On the other hand, *Coccinella septempunctata* L. females consumed about 130 of 150 offered aphids *Aphis craccivora* Koch during 24 hours and expressed type II of functional response (Jiang *et al*., 2019). Such consumption rate is higher than in our experiment, if similar body size of the two congeneric aphids is supposed. The difference is well explainable by the difference in body size between *C. septempunctata* and our predators.

All combinations of developmental stages of our two predatory species seemed to express additive foraging activity with no loss of ecological function due to competition. There was slightly lower consumption by the younger instars of the larvae, and high variability between pairs of predators in the combined predation rate during the first day in microcosm, when the predators were not habituated to the environment. IGP between the two predatory species at the diverse experimental set ups that we evaluated here will be quantified in a future study.

Conclusions

Overall predation rate and control mortality of aphids without predators at the microcosm arena were lower than in the simple arena. In a simple arena, where the prey (aphids) was simply accessible, consumption rate was higher the first day after previous starvation than the second day after previous feeding ad libitum. In more complex microcosm, the consumption rate increased from the first through the third day, probably due to the habituation to the environment. Variable prey numbers after one or two days spent in the individual arenas allows for more smooth estimation of functional response. Predation experiments lasting a few days are therefore recommended.

Acknowledgements

This research was financially supported by Bu-Ali Sina University, Hamedan, Iran.

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Received January 23, 2019. Accepted September 25, 2019.