Predatory mite Amblyseius orientalis prefers egg stage and low density of prey Carpoglyphus lactis

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Abstract

*Amblyseius orientalis* (Ehara) (Acari: Phytoseiidae) is an effective predatory mite for spider mites control on fruit trees in China. In recent decade, it can be produced commercially and intensively via *Carpoglyphus lactis* (Linnaeus) in natural enemy factory. During the practical predator production, ratio of predatory mites to their prey was found critical for the population increase of *A. orientalis* in large-scale rearing. In this study, we investigated the predatory capacity of *A. orientalis* on different developmental stages of the prey *C. lactis* and the effect of prey numbers on predator reproductions. The maximum predation number of *A. orientalis* adults on *C. lactis* adults was 2.21 at the lowest density of 5 and on *C. lactis* eggs was 45.07 at the highest density of 60. The preference index $C_i$ of *A. orientalis* on *C. lactis* eggs and adults was 0.4312 and $-0.9249$, respectively, suggesting that *A. orientalis* preferred eggs to adults of *C. lactis*. *A. orientalis* could reproduce when it preyed either eggs or deutonymphs of *C. lactis*. However, the fecundity of the predatory mites is not proportional to the provided prey density in a week. Higher density of prey deutonymphs resulted in lower fecundity, while more prey eggs can bring higher fecundity of *A. orientalis*. Therefore, our study indicated that suitable density and developmental stage of prey is the basis for *A. orientalis* production in a large scale.

Introduction

*Amblyseius orientalis* (Ehara) (Acari: Phytoseiidae) is a widely distributed and effective predatory mite for spider mite control in fruit productions in China (Sheng et al. 2014; Zheng et al. 2008). It was once only used for spider mite control (Zhang et al. 1992; Zheng et al. 2008), then it was found to be effective against thrips and whiteflies in recent decade (Sheng et al. 2014; Yang et al. 2018). Thus, the mass breeding and large-scale production of this native predatory mite is gradually attracting attention. Early studies of *A. orientalis* breeding involved in host plants and spider mite. However, it was not practical for large-scale production of *A. orientalis* in natural enemy factory for the long and costly process (Zhang et al. 2015). Some studies have attempted to rear predatory mites by pollen, whereas the cost was too high without satisfactory results (Liao & Zhu 1985).

*Carpoglyphus lactis* (Linnaeus) is a fast-reproducing, adaptable storage pest mite widely used as an alternative prey (Knapp et al. 2018; Ramachandran et al. 2021; Wang et al. 2008; Wu et al. 2009). It can be used for successfully rearing many commercial predatory mites, such as *Amblyseius swirskii* (Athias-Henriot) in large scale (Nguyen et al. 2013; Zhang et al. 2021). Our lab has reported that *A. orientalis* feeding on *C. lactis* can complete development, mating, and reproduction (Sheng et al. 2014). Compared with diets of *Bemisia tabaci*, *Panonychus citri* and castor pollen, *A. orientalis* feeding on *C. lactis* had a shorter preoviposition period and faster reproduction rate (Liao & Zhu 1985; Sheng et al. 2014; Zhang et al. 2015; Zhang 1990). Due to diet compatibility with the alternative prey *C. lactis*, it's promising of *A. orientalis* in large-scale production and field applications.

Instead of spider mites, few studies have clarified how predatory capacity of *A. orientalis* on *C. lactis* or whether there is a preference on prey developmental stage. During both laboratory observation and
factory production, we noticed that the density of prey to *A. orientalis* was strikingly stringent in predator population increase. Some studies have already found the critical effect of prey on predation and female fecundity of the predators (Werling et al. 2013; Osman et al. 2016; Rasmy & Abou-Elella 2002), whereas mass rearing was also considered with no negative impact on predation and development of predatory mite (Saemi et al. 2017). When ratio of predatory mites to pest mites was too small, the inhibitory effect could occur in the expansion of *A. orientalis* populations instead.

In this study, we investigated the predatory capacity of *A. orientalis* on different developmental stages of the prey *C. lactis*, the stage preference and the effect of prey numbers on predator reproductions. The aim of this study was to uncover the potential reason of low population growth of *A. orientalis* when using *C. lactis* as an alternative prey in large-scale production.

**Materials And Methods**

**Mite rearing**

*A. orientalis* was collected from soybean fields in Changli Research Institute of Pomology, Hebei Academy of Agricultural and Forestry Sciences (119°09'E, 39°43'N). The colony had been maintained on *C. lactis* for 10 years in the Laboratory of Predatory Mites, Institute of Plant Protection, Chinese Academy of Agricultural Sciences (Yan et al. 2022). *C. lactis* feeding on yeast were provided by Shoubonong Biotechnology Company (Beijing, China). All mites were reared at 25 ± 1°C, 70 ± 5% RH and L14:D10 photoperiod in an incubator (RXZ, Ningbo, China).

The predatory capacity of *A. orientalis* on *C. lactis*

To investigate the predatory capacity of *A. orientalis* on *lactis* at different densities, we selected approximate 1050 female *C. lactis* adults and deutonymphs into small arenas (Yan et al. 2022). Briefly, the arena was a tiny device with two tightly clipped layers: a transparent acrylic board (30×20×3 mm³) with a 10 mm diameter hole in the middle sealed by nylon mesh with glue, one piece of rectangular glass (30×20×1 mm³) at top. The prey densities were set for 5, 10 and 20 adults or deutonymphs per arena, respectively. *A. orientalis* female adults were starved for 24 hours in a plastic box before moving to each arena with *C. lactis* adults separately. All treatment was repeated by 30 *A. orientalis* adults. The consumed number of both stages of *C. lactis* was recorded within 24 hours.

Similarly, we then investigated the predatory capacity of *A. orientalis* adults and larvae on eggs of *C. lactis* at different densities. We collected sufficient *C. lactis* eggs to set three densities of 30, 45 and 60 per arena. *A. orientalis* females were starved for 24 hours and then separated into the arena with different densities of prey eggs. The same process was repeated, with one newly hatched *A. orientalis* larva per arena, and three prey densities of *C. lactis* was set at 10, 20 and 30 eggs per arena.

Stage preference of *A. orientalis* to prey *C. lactis*
We studied the developmental stage preference when *A. orientalis* was exposed to egg, nymph, and adult in the same arena. We collected one starved *A. orientalis* female adult and transferred it to the arena with 10 eggs, 10 deutonymphs and 10 adults of *C. lactis*. The predation number of *A. orientalis* on each developmental stages of *C. lactis* was recorded upon 6, 12 and 24 hours, respectively. The experiment was replicated 30 times.

**Effect of developmental stage and density of *C. lactis* on fecundity of *A. orientalis***

We moved one gravid *A. orientalis* adult into the arena with the densities of 30, 45, and 60 eggs or 5, 10 and 20 deutonymphs of *C. lactis*. Each density was repeated 30 times. These arenas were changed daily with the same number of eggs or deutonymphs of *C. lactis*, respectively. We then recorded the number of consumed eggs or deutonymphs of *C. lactis* daily and the oviposition of each *A. orientalis* in 7 consecutive days.

**Statistical analysis**

Kruskal-Wallis test with pairwise comparisons were used to examine the effects of different *C. lactis* densities on predatory capacity, prey preference and fecundity of *A. orientalis*. All mean comparisons with *p* < 0.05 were considered to have statistically significant differences. All analyses were run in SPSS 25.0 and results were visualized in Graphpad 8.02.

In the preference experiment, the prey preference index (C<sub>i</sub>) of predators to various stages could be expressed as (Eq. 1) (Ivlev et al. 1962; Zhou *et al*. 1987), where F<sub>i</sub> was the proportion of the *i*<sup>th</sup> prey type in the environment, and Q<sub>i</sub> was the proportion of predation of the *i*<sup>th</sup> prey type by the predator.

\[
C_i = \frac{(Q_i - F_i)}{(Q_i + F_i)}
\] (1)

When C<sub>i</sub> = 0, it meant that the predator had no preference for the *i*<sup>th</sup> prey. 0 < C<sub>i</sub> < 1, it meant that the predator had a positive preference for the *i*<sup>th</sup> prey. -1 < C<sub>i</sub> < 0, it meant that the predator had a negative preference for the *i*<sup>th</sup> prey.

**Results**

**The predatory capacity of *A. orientalis* on *C. lactis***

Prey densities significantly affected predator’s predation number but in different trends (Fig. 1). The maximum predation number of *A. orientalis* adults on *C. lactis* adults was 2.21 at the lowest density of 5 mites per arena (*χ^2 (2) = 12.436, p = 0.002, Fig. 1A*). In contrast, only 0.80 of predation quantity was observed in the density of 20 *C. lactis* adults per arena. The opposite tendency was observed when *A. orientalis* feeding on deutonymphs. The maximum predation number at the highest density of 20 mites per arena was 6.44, which was significantly more than 4.07 in the group of 5 prey mites per arena.
When testing *A. orientalis* adults and larvae on *C. lactis* eggs, the consumption number increased with prey densities. The maximum predation number of *A. orientalis* adults was 45.07 at the highest density of 60 eggs per arena, in contrast to the predation on 30 eggs per arena at 25.30 (Fig. 1C, $\chi^2(2) = 44.869, p < 0.001$). Similarly, the maximum predation number of *A. orientalis* larvae was 13.08 at the highest density of 30 eggs per arena that were more than 7.62 in an arena with 10 prey eggs (Fig. 1D, $\chi^2(2) = 24.536, p < 0.001$). In a total, taking *C. lactis* eggs and deutonymphs as preys, the predation of *A. orientalis* adults tended to increase with prey density, while the predation number tended to decrease when feeding on high density of *C. lactis* adults (Fig. 2, $\chi^2(1) = 26.336, p < 0.001$).

**Stage preference of *A. orientalis* for *C. lactis***

*Amblyseius orientalis* significantly preferred eggs to deutonymphs and adults of *C. lactis* (Table 1). *C. lactis* eggs was consumed in the largest proportion by 81.84%. The preference index $C_i$ of 0.4312 indicated a positive preference of *A. orientalis* to *C. lactis* eggs ($0 < C_i < 1$). On contrary, *C. lactis* adults were the least chosen with only 1.3%. $C_i$ of -0.9249 suggested that *A. orientalis* have a strong negative preference to adult prey ($-1 < C_i < 0$).

<table>
<thead>
<tr>
<th>Stages of <em>C. lactis</em></th>
<th>Density</th>
<th>Consumed number</th>
<th>Proportion (%)</th>
<th>$C_i$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Eggs</td>
<td>10</td>
<td>8.43 ± 0.54a</td>
<td>81.84</td>
<td>0.4212</td>
</tr>
<tr>
<td>Deutonymphs</td>
<td>10</td>
<td>1.74 ± 0.27b</td>
<td>16.89</td>
<td>-0.3272</td>
</tr>
<tr>
<td>Adults</td>
<td>10</td>
<td>0.13 ± 0.072c</td>
<td>1.3</td>
<td>-0.9249</td>
</tr>
</tbody>
</table>

In the 24-hour preference experiment, the consumed number of *C. lactis* eggs in the three time periods was 3.87, 5.87, 8.43, which were significantly higher adult numbers of 0, 0.04, 0.09 (Fig. 3). It showed that *A. orientalis* preying on eggs of *C. lactis* was continues at each time period.

**Effect of developmental stage and density of *C. lactis* on *A. orientalis* fecundity**

The preference experiment revealed different options of *A. orientalis* to eggs and deutonymphs of *C. lactis*, so we further tested the effects of eggs and deutonymphs at different densities on *A. orientalis* fecundity. The maximum predation was 4.15 at the highest density of 20 deutonymphs per day (Fig. 4A, $\chi^2(2) = 8.107, p = 0.017$). However, the maximum number of eggs reproduction with 2.13 per day occurred in the density of 5 deutonymphs preys (Fig. 4C, $\chi^2(2) = 21.487, p < 0.001$). When preyed on eggs, 38.62 eggs were consumed at the highest density of 60 eggs per arena, significantly greater than 31.50 and 26.94 eggs from the other two density groups (Fig. 4B, $\chi^2(2) = 32.833 p < 0.001$). The corresponding
maximum number of egg reproduction was 1.90 in the density of 45 preys (Fig. 4D, \( \chi^2(2) = 14.869, p = 0.001 \)).

The predatory number and reproductive quantity of *A. orientalis* were only correlated when prey density was high (Fig. 5). When prey number was lower, although not correlated, it did not decrease reproduction, suggesting the unstrict nutrition demand of *A. orientalis* for reproduction.

**Discussion**

The results of this study showed that *A. orientalis* female adults had strong predatory capacities on *C. lactis* eggs and deutonymphs, but not adults. In the preference experiment, \( C_i \) value of the preference to *C. lactis* eggs was 0.4212, showing a positive preference, and a strong negative preference to adults with \( C_i \) of -0.9249. It is consistent with what we observed during practical breeding. Some studies also revealed predatory mites preferred eggs as prey (Furuichi et al. 2005; Ganjisaffar & Perring 2015; Moghadasi et al. 2013; Naeem et al. 2017; Song et al. 1995). We consider that physical barrier (ie. body size) could be one of the determinative factors in developmental preference.

Body size affects *A. orientalis* predation. In the experiment, the starved *A. orientalis* can prey on *C. lactis* adults and deutonymphs, both of which were similar in size with predator, however, the predation time was much longer than prey on eggs. It was observed that catching same size of *C. lactis* was difficult for *A. orientalis*, since it’s seen to attempt many times to grasp and prey *C. lactis*. Predators cannot efficiently utilize oversized prey because of physical constraints on feeding (Brose 2010). We considered that large body size, strong mobility and thick tegument of *C. lactis* result in predatory difficulty and risks. Compared with the *C. lactis* adults, thin egg shell is easy to be pierced by mouthparts of *A. orientalis*. When the body-mass ratio of predator-prey is larger, the predator would have a higher preference for the prey (Kalinkat et al. 2011).

The development and reproduction of predators require some crucial nutrition (Harvey et al. 2012; Williams & Roane 2007; Woods et al. 2020). Compared with high-protein prey, high-lipid prey had negative effects on the survival and reproduction of the predator (Wen 2020). On the other hand, adding carbohydrates (ie. glucose and trehalose) to the artificial diet could improve the fecundity of female *Harmonia axyridis* (Li et al. 2020). Predatory mite *Parasitus consanguineus* separately feeding on two prey species resulted in significant differences in survival, female longevity and fecundity (Szlendak & Lewandowski 2009). Predators are considered to less likely to exploit undersized prey, since they may not be able to meet the specific nutritional needs of predators for developmental maturity (Brose 2010). We thus consider prey eggs could provide sufficient nutrition for predator reproduction, in particularly, *A. orientalis* has striking bias to *C. lactis* egg.

Our result showed that *A. orientalis* preyed solely on eggs can not only lay eggs normally, but also reproduce as many eggs as *A. orientalis* feeding on *C. lactis* deutonymphs. It suggests that *C. lactis* eggs contain enough nutrients for *A. orientalis* reproduction. Our results are also consistent with previous study
that the eggs are sufficiently nutritious to support predatory mite to complete reproduction and egg prey preference does not pose a survival risk to predators (Moghadasi et al. 2013).

In our study, we found that when a high density of *C. lactis* was provided, predator reproduction was even lower than those in groups with low density prey, indicating that predation might consume far more energy than additional prey provision. Our research provided a case study for natural enemy production of this beneficial mite. In factory rearing, the ratio of predator to prey needs to be increased, that is, prey quantity and densities should be strictly controlled to ensure continued population growth of *A. orientalis*. Our study here not only introduced *A. orientalis* sensitivity to prey abundance, but also offered a reference for potential solution that other predatory mites may encounter in commercial large-scale production.

**Declarations**

**Acknowledgements**

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**References**


Figures
Figure 1

Predatory capacity of *A. orientalis* on *C. lactis* at different densities. (A) Predatory capacity of *A. orientalis* adults on *C. lactis* adults. (B) Predatory capacity of *A. orientalis* adults on *C. lactis* deutonymphs. (C) Predatory capacity of *A. orientalis* adults on *C. lactis* eggs. (D) Predatory capacity of *A. orientalis* larvae on *C. lactis* eggs. Each point represented an individual. Horizontal lines indicated the mean (± SD) of biological replicates. Different letters above the dots indicated significant differences between treatments (*p* < 0.05).
Figure 2

Predatory capacity of *A. orientalis* adults on *C. lactis* at different densities. Asterisks indicated differences between densities.
Figure 3

Developmental stage preference of *A. orientalis* adults. Each point represented an individual. Horizontal lines indicated the mean (± SD) of biological replicates.
Figure 4

Predatory capacity and fecundity of *A. orientalis* on eggs and deutonymphys of *C. lactis*. (A) Predatory capacity of *A. orientalis* on *C. lactis* deutonymphys. (B) Predatory capacity of *A. orientalis* on *C. lactis* eggs. (C) Fecundity of *A. orientalis* on *C. lactis* deutonymphys. (D) Fecundity of *A. orientalis* on *C. lactis* eggs. Each point represented an individual. Horizontal lines indicated the mean (± SD) of biological replicates. Different letters above the dots indicated significant differences between treatments (*p* < 0.05).
Figure 5

Linear relationship between predatory capacity and fecundity of *A. orientalis*. (A) Linear relationship between predatory capacity and fecundity of *A. orientalis* on *C. lactis* deutonymphs. (B) Linear relationship between predatory capacity and fecundity of *A. orientalis* on *C. lactis* eggs.