Intraguild Predation and Cannibalism between Two Phytoseiid Mites, Neoseiulus californicus and Amblyseius andersoni

H. Rahmani¹,²*, and N. Afshari¹

ABSTRACT

Intraguild predation and cannibalism (conspecific predation) of two generalist phytoseiid mites were determined in this study. Adult females and protonymphs of Neoseiulus californicus (McGregor) and Amblyseius andersoni Chant were considered as intraguild and cannibalistic predators, provided with eggs, eggs+larvae and protonymphs as intraguild or conspecific prey. A side from predator species and prey type (IG prey versus conspecific prey), females exhibited lowest predation rates on eggs. Not including prey type, the highest predation rate was recorded in A. andersoni and N. californicus when provided with eggs+ larvae. Female predators were not interested in the first nymphal stage of prey and there was no significant difference in consumption rates and oviposition when provided with conspecific or intraguild prey. Feeding on intraguild larval prey did not only increase mortality of A. andersoni immatures, but also decreased their consumption rates. For N. californicus immatures, predation on IG larval prey was higher than on conspecific larvae. Therefore, it seems that the predator species are able to survive on both con- and heterospecific prey.

Keywords: Acari, Cannibalistic predators, Conspecific prey, Interspecific predation.

INTRODUCTION

The two-spotted spider mite, Tetranychus urticae is known as one of the economically most important pests in greenhouse production and field crops (Zhang, 2003). It has the highest pesticide resistance among arthropods (Van Leeuwen et al., 2010). Repetitive applications of chemicals to control insect and mite pests lead to environmental pollution and serious health problems for consumers (Çalmaşur et al., 2006). Therefore, biological agents and IPM Management (Integrated Pest Management) are taken into account to help or even to substitute the synthetic insecticides and acaricides.

Competition, cannibalism, and IntraGUILD Predation (IGP) may influence the success of natural enemies including phytoseiid species in management methods, since they may co-occur in the environment or may be released simultaneously in greenhouse crops (Schausberger and Walzer, 2001). Knowledge on the strength and direction of competition and intraguild predation and their consequences at the population level is important to evaluate the success of pest control (Walzer and Schausberger, 1999a; Schausberger and Walzer, 2001; Walzer et al., 2001). Some studies have indicated positive effects of combined release of phytoseiid mites in suppression of pests (Mori et al., 1990; Schausberger and Croft, 2000 a,b;
Schausberger and Walzer, 2001; Walzer et al., 2001; Onzo et al., 2003; Hatherly et al., 2005; Cakmak et al., 2006, 2009; Walzer et al., 2009), while others have shown that intraguild predation (IGP) may reduce the control of the target pests (Polis et al. 1989, Rosenheim et al. 1995; Holt and Polis, 1997; Arim and Marquet, 2004; Walzer et al. 2001; Hatherly et al. 2005; Negloh et al. 2012).

Neoseiulus californicus (McGregor) and Amblyseius andersoni Chant are predators of phytoseid family, which are used commercially as biocontrol agents. Neoseiulus californicus, which is classified in type II group of Phytoseiidae (McMurtry et al., 2013), can feed on spider mites, tarsonemid mites, small arthropods, pollen (Castagnoli et al., 1999, Khamanani et al., 2017), fungi (McMurtry et al., 2013) and Thrips tabaci (Rahmani et al., 2009). It is distributed around the world and considered as a cosmopolitan species (Gotoh et al., 2004; Canlas et al., 2006; McMurtry et al., 2013; Barbosa and de Moraes, 2015). Efficient functional and numerical response to spider mites (Song et al., 2016) is the other trait of this predator.

Amblyseius andersoni is a generalist predator; belongs to subtype III-b phytoseid family, that have the ability to feed on fungi. In addition, it is abundant as a predator of spider mites on grape varieties (McMurtry et al., 2013). The red spider mite, Panonychus ulmi (Koch) was controlled effectively by this species (Koveos and Broutas, 2000). It is cosmopolitan but widespread in Europe (Tixier et al., 2016).

At high prey densities, there is no problem about simultaneous use of predators, but predation may take place between different predatory mite species among various life stages within a guild (IGP) and/or within each species (cannibalism) in the absence of the main food. Consequence of these interactions is vital for having sustainable control (Walzer et al., 2001).

There are many studies about interactions of N. californicus with other phytoseiids (Walzer and Schausberger, 1999a,b; Schausberger and Croft, 2000a; Schausberger and Walzer, 2001; Walzer et al., 2004; Cakmak et al., 2006, 2009; Rhodes et al., 2006; Mendel and Schausberger, 2011; Farazmand et al., 2015; Kazak et al., 2015; Rahmani et al., 2015). Some studies also assessed the interaction between A. andersoni and the other members of this family (Duso, 1989; Zhang and Croft, 1995; Schausberger, and Croft; 2000a,b; Ahmad et al., 2015).

Both A. andersoni and N. californicus are efficient biocotrol agents for spider mite species (Escudero and Ferragut, 2005; Jolly, 2000; Gotoh et al., 2006; Amano and Chant, 1977; Duso, 1992) when used separately, but no study has been conducted on interaction between them at the simultaneous use. These two predators may co-exist in an environment, or may be deployed at the same time for better pest management at greenhouses. Therefore, the objective of this study was to compare predation of adult females and protonymphs of the two predator species when offered conspecific or IntraGuild (IG) prey. The hypothesis tested was predators consume more heterospecific than conspecific prey in a closed cage with no choice situation.

**MATERIALS AND METHODS**

**Origin of Predatory Mites, Rearing, and General Methods**

Neoseiulus californicus was obtained from a commercial producer (Koppert). Amblyseius andersoni was taken from the laboratory population in the University of Natural Resources and Life Sciences, Vienna, Austria. The colonies of each species were separately held on artificial arenas (half-filled plastic boxes containing water-saturated foam cubes with plastic tiles) and fed with mixed stages of T. urticae, by adding infested common bean leaves (Phaseolus vulgaris L.) on the arenas (Walzer and Schausberger 1999a, b).

Eggs, larvae, and protonymphs of the two predatory mite species were used as conspecific and heterospecific prey. For this purpose, even-aged cohorts were obtained by transferring gravid females from the rearing
units to 6 cm Petri-dishes containing detached bean leaves and fed with spider mites. After 24 h, newly laid eggs were collected and used for experiments or transferred to the separate arenas for having other required stages.

Closed cages were used as experimental units. Each cage consisted of rectangular pieces of an acrylic glass (6×3×0.6 cm) with circular cavities (Ø= 2.0 cm) closed at the bottom by gauze and covered with a microscope slide at the upper side. The microscope slide was fixed with a rubber band (Schausberger, 1997). Rearing and experimental units were maintained in a climate chamber at 25±2°C, 70±5% RH and 16:8 L:D hours photoperiod.

Experimental Procedures

Gravid females of both species were chosen randomly from rearing arenas and, after 24 hours starvation, used as IG predators. Each single female was placed into the experimental cage (one female predator, \(A. \text{ andersoni} \) or \(N. \text{ californicus} \) in each experimental cage) and provided with: (a) 6 eggs of < 24 hours age, (b) 4 eggs of 24 to 48 hours age+4 newly hatched larvae, and (c) 4 protonymphs of con- and/or heterospecifics at separate treatments (in each treatment just one type of mentioned prey was offered to a female predator). Every 24 hours, experimental units were observed and some parameters such as predation rates, survival and oviposition were recorded for 10 consecutive days. The eggs laid by the female predator were removed and predator was transferred to the new experimental cage with fresh food on daily basis. Replication was 16-20 times for each experiment and species.

In most phytoseiid mites, larvae reach the protonymphal stage without consumption, and protonymph is usually the first developmental stage starting to prey and act as IG predator (Walzer et al., 2015) or cannibal. Six conspecific and/or six heterospecific larvae were regarded as prey (each prey offered to one protonymph in the experimental cage of each treatment). Dorsal shield of the predators (protonymphs) were marked with a tiny watercolor point in order to make them distinguishable from larva prey, which may enter the next stage. Predation, survival, and development of the predators were checked twice daily in intervals of 10 and 14 hours until the predator reached adulthood or died. Prey larvae were replenished daily. Each treatment (con- or heterospecific prey) was replicated 20 times for each experiment and species.

Statistical Analysis

All data were analyzed using SPSS ver. 18.0.1. The number of preys consumed and number of laid eggs by adult females provided with either con- or heterospecific prey during 10 days were compared by one-way ANOVA for each predator, separately. Differences between treatments were compared using Tukey’s multiple range tests. \(t\) tests were used to compare the predation rate and oviposition of female predators between conspecific and IG prey. The same method was used to analyze effects of con- or heterospecific larvae on developmental time of immatures and the juvenile predation rates. Mortality of immatures and adult females was analyzed for each species separately using Pearson Chi-square tests.

RESULTS

Adult Females Feeding on Con- and Heterospecific Eggs

Predation on eggs regardless of prey was very low for both predators (Tables 1 and 3). \(Amblyseius \text{ andersoni} \) females consumed no conspecific eggs during 10 days, whereas
Table 1. Predation and oviposition (Mean±SE) of adult *A. andersoni* and *N. californicus* females caged singly and provided with conspecific or heterospecific prey for 10 consecutive days. *a*

| Prey species  | Stage     | Predation |  | Oviposition |
|---------------|-----------|-----------|  | ------------|
|               |           | *A. andersoni* |  | *N. californicus* |  | *A. andersoni* |  | *N. californicus* |
| *A. andersoni* | Egg       | 0.00 ±0    |  | 0.05 ±0.050  |  | 0.3 ±0.128  |  | 0.5 ±0.170  |
| *N. californicus* | Egg  | 0.15±0.0820 |  | 0.45±0.153  |  | 0.7 ±0.147  |  | 0.4 ±0.134  |
| *A. andersoni* | Egg+Larva | 3.5±0.448  |  | 5.75±0.479  |  | 1.16 ±0.245 |  | 1.44 ±0.223 |
| *N. californicus* | Egg+Larva | 7.8±0.890  |  | 6.80±0.565  |  | 1.4 ±0.254  |  | 0.10 ±0.069 |
| *A. andersoni* | Protonymph | 5.2±0.592  |  | 6.45±0.673  |  | 0.05 ±0.050 |  | 0.23 ±0.113 |
| *N. californicus* | Protonymph | 5±0.465    |  | 4.48±0.506  |  | 0.1 ±0.069  |  | 0.22 ±0.108 |

*a* Means followed by different letters are significantly different for each predator, between two types of prey, in each stage (P < 0.05, t test).

*N. californicus* females consumed more conspecific eggs than heterospecific (t tests for independent samples: *N. californicus*: T= -2.4, df= 38, P= 0.04) (Table 1). There was no significant difference in oviposition of *N. californicus* females fed on con- or heterospecific eggs (t tests for independent samples: T= 0.69, df= 38, P= 0.5) (Table 1).

Oviposition was negligible for both predators and could be due to primary feeding (Tables 1 and 3). *Amblyseius andersoni* females laid more eggs when fed on heterospecific eggs during 10 days (t tests for independent samples; T= -2.05, df= 38, P= 0.04) (Table 1). There was no significant difference in oviposition of *N. californicus* females fed on con- or heterospecific eggs (t tests for independent samples: T= 0.69, df= 38, P= 0.5) (Table 1).

*A. andersoni* had higher survival on IG prey. Only 10 percent of *A. andersoni* females could not survive the experimental period when supplied with heterospecific eggs as prey (P < 0.05) (Table 2). Percentage of

Table 2. Mortality of adult *A. andersoni* and *N. californicus* females caged singly and provided with conspecific or heterospecific prey for 10 consecutive days. *a*

<table>
<thead>
<tr>
<th>Prey species</th>
<th>Stage</th>
<th>Mortality (percent)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td><em>A. andersoni</em></td>
</tr>
<tr>
<td><em>A. andersoni</em></td>
<td>Egg</td>
<td>25 ±0</td>
</tr>
<tr>
<td><em>N. californicus</em></td>
<td>Egg</td>
<td>10 ±0</td>
</tr>
<tr>
<td><em>A. andersoni</em></td>
<td>Egg+Larva</td>
<td>27 ±0</td>
</tr>
<tr>
<td><em>N. californicus</em></td>
<td>Egg+Larva</td>
<td>14 ±0</td>
</tr>
<tr>
<td><em>A. andersoni</em></td>
<td>Protonymph</td>
<td>25 ±0</td>
</tr>
<tr>
<td><em>N. californicus</em></td>
<td>Protonymph</td>
<td>30 ±0</td>
</tr>
</tbody>
</table>

*a* Means in columns (two prey for one predator in each stage) followed by the different letters are significantly different (Chi square test).

Table 3. Predation and oviposition (Mean±SE) of adult *A. andersoni* and *N. californicus* females caged singly and provided with conspecific or heterospecific prey for 10 consecutive days. *a*

| Prey species  | Stage     | Predation |  | Oviposition |
|---------------|-----------|-----------|  | ------------|
|               |           | *A. andersoni* |  | *N. californicus* |  | *A. andersoni* |  | *N. californicus* |
| *A. andersoni* | Egg       | 0 ±0      |  | 0.05 ±0.050  |  | 0.3 ±0.127  |  | 0.55 ±0.169 |
| *N. californicus* | Egg  | 0.15±0.081 |  | 0.45±0.153  |  | 0.7 ±0.146  |  | 0.4 ±0.134  |
| *A. andersoni* | Egg+Larva | 3.5±0.448  |  | 5.75±0.479  |  | 1.16 ±0.244 |  | 1.44 ±0.223 |
| *N. californicus* | Egg+Larva | 7.8±0.890  |  | 6.80±0.565  |  | 1.4 ±0.254  |  | 0.10 ±0.069 |
| *A. andersoni* | Protonymph | 5.2±0.592  |  | 6.45±0.673  |  | 0.05 ±0.050 |  | 0.23 ±0.112 |
| *N. californicus* | Protonymph | 5±0.464   |  | 4.48±0.506  |  | 0.1 ±0.068  |  | 0.22 ±0.108 |

*a* Means in columns followed by different letters are significantly different at P < 0.05 (ANOVA, Tukey).
mortality was similar for *N. californicus* females in both treatments (Table 2).

### Adult Females Feeding on Con- and Heterospecific Eggs+ Larvae

Mean predation rate of the two predators increased by adding larval stage of the prey (Tables 1 and 3). Consumption of IG eggs+larvae was greater in *A. andersoni* than cannibalism (t tests for independent samples: T= -4.5, df= 32, P= 0.00) (Table 1), but oviposition rate was similar between the two types of prey (t tests for independent samples: T= -0.67, df= 32, P= 0.5) (Table 1). Predation rate of con- and heterospecific eggs+larvae was similar for adult females of *N. californicus* (t tests for independent samples: T= -1.37, df= 34, P= 0.17) (Table 1). These females laid more eggs while feeding on IG prey (t tests for independent samples: T= 6.27, df= 34, P= 0.00) (Table 1).

Comparison of all treatments showed that adult *A. andersoni* and *N. californicus* consumed IG eggs+larvae and conspecific eggs+larvae, respectively, more than other foods during 10 days (Table 3).

Mortality percentage was higher in *A. andersoni* when fed on conspecific prey, but was similar to *N. californicus* females for the two types of prey (conspecific and IG prey) (Table 2).

### Adult Females Feeding on Con- and Heterospecific Protonymphs

*Amblyseius andersoni* consumed the same amount of both protonymphs species (conspecific and IG prey) (t tests for independent samples: T= 0.26, df= 38, P= 0.7) (Table 1). For *N.californicus*, mean predation of heterospecific protonymphs was higher than conspecific one (t tests for independent samples: T= 2.3, df= 43, P= 0.02) (Table 1).

Oviposition rate of both female predators was not significantly different between conspecific and IG protonymphs (t tests for independent samples: *A. andersoni*: T= -0.58, df= 38, P= 0.56; *N. californicus*: T= -0.63, df= 43, P= 0.9) (Table 1). Feeding protonymphal stage of the intraguild preys had negative effect on survival, as the number of both predators not completing the experimental period was higher compared to other types of prey. Almost 30 percent of female predators died when fed on either con- or heterospecific protonymphs (Table 2).

### Immatures Feeding on Con- and Heterospecific Larvae

Feeding on heterospecific larva not only increased the mortality percentage of *A. andersoni* immatures, but also decreased the consumption rates (Table 4). Almost 50 percent of immatures died before reaching adulthood, which was significantly higher than other prey species (Table 4). Protonymph (t tests for independent samples: T= 0.46, df= 38, P= 0.6), deutonymph (t tests for independent samples: T= -0.25, df= 25, P= 0.7) and total developmental time of *A. andersoni* (t tests

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**Table 4.** Predation and mortality of immature *A. andersoni* and *N. californicus* caged singly and provided with conspecific or heterospecific larvae over 10 days.

<table>
<thead>
<tr>
<th>Predator species</th>
<th>Prey</th>
<th>Predation</th>
<th>Mortality (Percent)</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>A. andersoni</em></td>
<td><em>A. andersoni</em></td>
<td>0.6±0.17</td>
<td>20$^b$</td>
</tr>
<tr>
<td><em>N. californicus</em></td>
<td><em>N. californicus</em></td>
<td>0.25±0.09</td>
<td>55$^a$</td>
</tr>
<tr>
<td><em>N. californicus</em></td>
<td><em>A. andersoni</em></td>
<td>1.3±1.3</td>
<td>35$^a$</td>
</tr>
<tr>
<td><em>N. californicus</em></td>
<td><em>N. californicus</em></td>
<td>0.8±0.7</td>
<td>20$^b$</td>
</tr>
</tbody>
</table>

$^a$ Means followed by different letters are significantly different for each predator, between two types of prey.

$^b$ (Predation: P< 0.05, t test), $^c$ (Mortality: Chi-square test).
Table 5. Developmental times (days) (Mean±SE) of A. andersoni and N. californicus caged singly and provided with conspecific or heterospecific larvae.

<table>
<thead>
<tr>
<th>Predators</th>
<th>A. andersoni</th>
<th>N. californicus</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protonymph</td>
<td>3.6±0.24</td>
<td>2.9±0.15</td>
</tr>
<tr>
<td>Deutonymph</td>
<td>3.4±0.02</td>
<td>4±0.16</td>
</tr>
<tr>
<td>Totala</td>
<td>6.57±0.05</td>
<td>5.52±0.51</td>
</tr>
<tr>
<td>Totalb</td>
<td>5.55±0.6</td>
<td>7.75±0.56</td>
</tr>
</tbody>
</table>

a Means followed by different letters are significantly different for each stage, between two types of prey (P< 0.05, t test). b (Protonymph+Deutonymph).
phytoseiid mite group (MCMurtry et al., 2013) and the same behavior is expected. In no choice situation, adult females of A. andersoni had more predation on other species egg and egg+larva, but predation on protonymphs was higher on conspecific (predation was slightly greater on conspecific prey and there was no significant difference between consumption of con- or heterospecific protonymphs for this predator).

Both females of phytoseiid species had the lowest amount of predation on IG eggs. This observation agrees with Farazmand et al. (2015) who determined the predation rate of N. californicus and T. bagdasarjani on IG prey eggs, larvae, and protonymphs on cucumber leaf discs. The same result was reported for A. swirskii and N. cucumeris (Buitenhus et al., 2010), however, P. persimilis and T. bagdasarjani preferred egg stage and consumed more heterospecific eggs when placed singly on cucumber leaf discs in both absence and presence of extraguild prey, T. urticae (Moghadasi and Allahyari, 2017).

Oviposition rate of the two predatory mite species was higher when feeding on egg+larva (both con- and heterospecific). It seems that piercing eggs and overwhelming protonymphs is difficult for adult females and predation on them was only for survival and not producing offspring. (Schausberger and Croft, 2000b; Meszaros et al., 2007). Momen and Abdel-Khalek (2009) indicated that cannibalizing Amblyseius swirskii females did not lay eggs, whereas in our study both female predators sustained oviposition on conspecific prey.

Developmental duration of immature could be affected by consumption. Total developmental period of immature N. californicus when fed on T. urticae was reported almost 3 (Uddin et al., 2016) and 6 days (Escudero and Ferragut, 2005) at 25°C, but cannibalism and intraguild predation increased developmental period and adults emerged after 10 and 7 days, respectively. Moreover, in the presence of preferred prey, 93% of N. californicus could survive to adulthood (Uddin et al., 2016; Escudero and Ferragut, 2005), whereas in the present study, 80 and 65% of N. californicus immatures could reach adulthood by feeding conspecific prey and IG prey, respectively. Amano and Chant (1977) stated that adult A. andersoni reached adulthood after 7 days. It was longer in our study (9 and 8 days) for nymphs that consumed con- and heterospecific prey. Overall, short pre-adult period, higher predation, and mortality were the consequences of IGP for N. californicus. It is similar to what we have found for A. andersoni, although mean predation rates of immatures were higher on conspecific prey.

Some phytoseiid mites could lay eggs and survive when fed with heterospecific prey. For example, Farazmand et al. (2015), Ghasemloo et al. (2016), and Moghadasi and Allahyari (2017) reported oviposition by T. bagdasarjani on intraguild prey stages. Also, adult females of K. aberrans, T. pyri, A. andersoni, and Phytoseius finitimus were able to survive and oviposit when fed with heterospecific larvae (Ahmad et al., 2015). It seems that generalist phytoseiids are stronger predators than specialists on intraguild predation and can outcompete them (Schausberger, 1997; Walzer and Schausberger, 1999a; Hatherly et al. 2005; Meszaros et al. 2007; Momen and Abdel-Khalek, 2009; Moghadasi and Allahyari, 2017).

In this study, two phytoseiid species could survive and feed on conspecific prey as well as on IG prey. Since they were given no choice, this may affect their ability to distinguish between preys and avoid cannibalism. Moreover, predators might have different behavior in the small experimental units compared to situations in greenhouses or the field. As a result, additional choice experiments are needed in natural condition and at the population level.

ACKNOWLEDGEMENTS

The authors thank the vice presidency for research of the University of Zanjan (Iran) for financial support of this project.
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*Neoseiulus* درون گونه‌ای و هم‌واری بین دو کن شکارگر فیتوژید.

*Amblyseius andersoni* و *californicus*

ج. حمایی و ن. افتخاری

چکیده

در این مطالعه به شکارگری درون گونه‌ای و هم‌واری (تغذیه از هم گونه) در دو کن شکارگر نظیر *Neoseiulus* و *Amblyseius andersoni* و *californicus* ترکیبی از نظر غذایی در نظر گرفته شد. نتایج نشان داد که میزان تغذیه از هم گونه بین نواحی مختلف کن شکارگر نظیر *Neoseiulus* و *Amblyseius andersoni* و *californicus* متفاوت است.

راهنمای و نکات

*Neoseiulus* درون گونه‌ای و هم‌واری (تغذیه از هم گونه) در دو کن شکارگر نظیر *Neoseiulus* و *Amblyseius andersoni* و *californicus* ترکیبی از نظر غذایی در نظر گرفته شد. نتایج نشان داد که میزان تغذیه از هم گونه بین نواحی مختلف کن شکارگر نظیر *Neoseiulus* و *Amblyseius andersoni* و *californicus* متفاوت است.

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