

# Suitability of pollen as an alternative food source for different developmental stages of *Amblyseius herbicolus* (Chant) (Acari: Phytoseiidae) to facilitate predation on whitefly eggs

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## Original research

### ABSTRACT

Alternative food sources could sustain predator populations by influencing their performances including predation, development, and reproduction in periods of shortage or scarcity of prey under laboratory or fields conditions. The aim of this study was to explore suitability of cattail pollen (*Typha orientalis*) as an alternative food source for different stages of *Amblyseius herbicolus* (Chant) (Acari: Phytoseiidae) on their predation capacity against the eggs of whitefly *Trialeurodes vaporariorum* (Westwood) (Hemiptera: Aleyrodidae), and the developmental and reproductive responses of the predators to different diet switching regimes under laboratory conditions (T1: nymphs and early adults fed pollen; T2: nymphs fed pollen but early adults fed whitefly eggs; T3: nymphs fed whitefly eggs but early adults fed pollen; T4: nymphs and early adults fed whitefly eggs). Results in the diet switching experiment revealed that although *A. herbicolus* could prey on whitefly eggs in all the treatment groups, the predatory mites obviously differed in their capacity to prey on the target pest whitefly eggs when fed on cattail pollen and whitefly eggs during different developmental stages. *A. herbicolus* in group T2 revealed the highest average predation rate, being 1.31, 1.17, and 1.49 times those in groups T1, T3, and T4, respectively. Our results indicated that food sources including cattail pollen and whitefly eggs were suitable for *A. herbicolus*, because it was able to develop from egg to adult females in all the treatment groups. The mixed diets (T2 and T3 groups) revealed positive effects on the developmental and reproductive parameters of *A. herbicolus* females, leading to higher oviposition rates and shorter developmental durations when compared with those of groups T1 and T4. *A. herbicolus* in group T2 revealed the highest average oviposition rates (1.32, 1.26, and 1.81 times those in groups T1, T3 and T4, respectively); however, no significant differences were observed between the T1 and T3 groups. All the eggs of *A. herbicolus* succeeded in reaching adulthood, but the duration of egg to adult developmental time of *A. herbicolus* was different among the four treatment groups. The longest and shortest developmental times were obtained in groups T4 and T3, respectively. All the results in the present study demonstrated that *A. herbicolus* adult females can prey on *T. vaporariorum* eggs, and cattail pollen was suitable as an alternative or additional food source with high nutritional value for this predatory mite to enhance the predation capacity towards whitefly eggs because *A. herbicolus* can successfully complete its immature development as well as reproduction.


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## Introduction

Whiteflies, thrips, aphids, spider mites, and leafminers include some of the most important pests in solanaceous greenhouse crops and can lead to serious yield losses on a global scale (Perdikis *et al.*, 2008). Among 1556 species of whiteflies in the world (Martin and Mound, 2007), the greenhouse whitefly, *Trialeurodes vaporariorum* (Westwood) and the tobacco whitefly, *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae) are the most important and destructive species in open field and greenhouse crops worldwide. Whiteflies suck and remove plant sap using mouthparts to reduce yield and weaken plant photosynthesis. Whitefly adults and nymphs feed on plant phloem and excrete a large amount of honeydew, which is a sugary substance for the development of saprophytic fungal mould, leading to lower fruit quality due to leaf/fruit blackening and reduced photosynthesis on covered leaves and fruits. In addition, whiteflies transmit numerous plant viruses (Perdikis *et al.*, 2008; Lee and Zhang, 2018; Spence *et al.*, 2020). *Trialeurodes vaporariorum* has a wide host range, attacking 859 species of plants from 469 genera and 121 families (CABI 2017). Additionally, Burnett (1949) reported the fecundity and longevity of females at temperatures ranging from 9 °C to 33 °C, and maximum longevity at 15 °C, with very short survival at 9 °C and temperatures over 27 °C and many generations every year. Moreover, *T. vaporariorum* was known to have high reproductive rates, reaching an average of 320 eggs per female throughout the life cycle (Burnett, 1949). Furthermore, whiteflies have rapidly developed resistance to many chemical pesticides because of the frequent application of synthetic insecticides to suppress whitefly populations (Spence *et al.*, 2020). All the biology and life history parameters of *T. vaporariorum* indicate that the effective management of this most economically important species has become a major challenge.

Biological control is the most prominent and sustainable method worldwide in integrated pest management (IPM) in agriculture, especially in greenhouse crops. It is necessary because crops are harvested frequently over short intervals, and thus the intensive use of chemicals becomes questioned due to the possible contamination of products with chemical residues (Perdikis *et al.*, 2008). Biological control against insect and mite pests has taken environmentally friendly advantages over the conventional chemical control, including neither resistance development, nor risk in subsequent pesticide residues in food and environment. Numerous studies revealed that biological control of whitefly populations based on using a wide variety of parasitoids and predators has proved very successful on several crops (Perdikis *et al.*, 2008; Lee and Zhang, 2018; Medd and GreatRex, 2014). The data indicated that 24 predator and 21 parasitoid species were reported to control *T. vaporariorum*, and 48 predators and 33 parasitoids were known for controlling *B. tabaci* (CABI, 2017). Ladybird beetles are prominent natural enemies of whiteflies and known to prey successfully on *B. tabaci* and *T. vaporariorum*. The results of prey consumption preferences demonstrated that larvae and adults of *Serangium parcesetosum* Sicard (Coleoptera: Coccinellidae) could prey on all whitefly immature stages, and all *S. parcesetosum* stages could also effectively prey on *B. tabaci* and *T. vaporariorum* at both low and high temperatures (18 °C and 30 °C) (Al-Zyoud and Sengonca, 2004). Greenberg *et al.* (2002) reported that two parasitoid species, *Eretmocerus eremicus* Rose & Zolnerowich and *Eretmocerus mundus* Mercet (Hymenoptera: Aphelinidae), could efficiently attack both whitefly species, *Bemisia argentifolii* Bellows & Perring and *T. vaporariorum* (Westwood) on tomato. Apart from these insect predators, recent studies have revealed that predatory mites of the Phytoseiidae family were widely used to control infestations of whiteflies. Among them, *Amblyseius swirskii* Athias-Henriot has been reported as an effective natural enemy for suppressing whitefly populations due to its high intrinsic rate of increase and its ability to utilize alternative food sources such as pollen and prey-produced honeydew (Nomikou *et al.*, 2003). Provision of pollen as supplementary food could enhance population build up and predation efficiency over either a suitable or a marginal suitable prey species (Pappas *et al.*, 2013; Samaras *et al.*, 2019, 2021). *Amblydromalus limonicus* has been reported as a crucial biological control agent against *T. vaporariorum* in the greenhouse environment (Knapp *et al.*, 2013; Lee and

Zhang, 2018). *Amblyseius swirskii* is also an important predator of the whitefly, *Aleuroclava jasmini* (Takahashi) on paper mulberry in Iran (Shahbaz *et al.*, 2019). Three predatory mite species — *A. swirskii*, *Typhlodromips montdorensis* (Schicha), and *Amblydromalus limonicus* — could suppress greenhouse whitefly (*T. vaporariorum*) populations (Medd and GreatRex, 2014). *Amblyseius tsugawai* Ehara, and *Amblyseius orientalis* (Ehara) could also prey on whitefly eggs (Yang *et al.*, 2019; Zhang *et al.*, 2015). Recently, the predatory mite *Neoseiulus bicaudus* (Wainstein) was shown to be a promising biocontrol agent of the whitefly *B. tabaci* (Han *et al.* 2020).

Numerous recent studies, focusing on evaluating diet suitability of alternative/supplementary food sources for predatory mites in biological control of pest species may become the emerging novel topics in nutritional ecology. These non-prey food sources not only allowed the predators to sustain the population in the event of prey scarcity, but also were valuable for rearing predators in laboratories or mass rearing for biological control (Khanamani *et al.*, 2017). A previous study indicated that different types of alternative food sources, including pollens (cattail, maize, and apple) and eggs of the Mediterranean flour moth *Ephestia kuehniella*, were all suitable for *Amblyseius swirskii* and *Neoseiulus cucumeris*, and both predatory species were able to develop and reproduce when fed with these pollens as well as moth eggs (Delisle *et al.*, 2015). Bouvet *et al.* (2019) evaluated the suitability of the two most abundant aphid species in citrus agroecosystems, *Aphid gossypii* and *A. spiraecola*, for their principal natural enemies, the coccinellid predators *Scymnus subvillosus* and *S. interruptus*. Results indicated that the fitness of both predators when offered a mixed diet of both aphids was higher than that fed only a single aphid species. A previous study suggested that non-prey food sources such as cattail pollen and whitefly-produced honeydew were suitable for the survival, development, and reproduction of two phytoseiid species, *Euseius scutalis* (Athias-Henriot) and *Typhlodromips swirskii* (Athias-Henriot) (Nomikou *et al.*, 2003). The laboratory trials performed by Pirayeshfar *et al.* (2020) revealed that immature stages (mixed living, frozen eggs or frozen larvae) of astigmatid mites (*Tyrophagous putrescentiae*) as a supplementary food source strongly affected the diet suitability for *A. swirskii* by influencing the predatory mite's developmental times and oviposition rates. Interestingly, the result of Soltaniyan *et al.* (2018) revealed that pistachio pollen (*Pistacia vera*) could be a more suitable alternative food source for the development and reproduction of *Neoseiulus californicus* than its natural prey *T. urticae* because of shorter preoviposition periods and higher fecundity.

The predatory mite, *Amblyseius herbicolus* Chant (Acari: Phytoseiidae) is widely established throughout New Zealand (Ma *et al.*, 2019) as well as being commonly found in many countries (Argov *et al.*, 2002; Cavalcante *et al.*, 2015; Döker *et al.*, 2014; Hajizadeh and Nazari, 2012). According to the classification of lifestyles of phytoseiid mites, it is considered a generalist facultative predator (type III) that can feed on many species of prey (mites and small insects) as well as a variety of plant-derived foods (McMurtry *et al.*, 2013). It is an important predator of multiple pest mite species such as the broad mite *Polyphagotarsonemus latus* (Banks) (Duarte *et al.*, 2015; Rodríguez-Cruz *et al.*, 2013), false spider mite *Brevipalpus phoenicis* (Geijskes) (Reis *et al.*, 2007), citrus rust mite *Phyllocotruta oleivora* (Ashmead) (Argov *et al.*, 2002), and spider mites *Tetranychus urticae* Koch (Moghadam *et al.*, 2010). It has been reported to attack small insects such as thrips (*Pseudodendrothrips mori* (Niwa) and *Sericothrips staphylinus* Haliday) (Lam *et al.*, 2019) and whitefly *B. tabaci* (Gennadius) (Cavalcante *et al.*, 2015). It can also feed on many non-prey food sources, including honeydew, nectar, and pollen grains from different plant species (Hoogerbrugge *et al.*, 2011; Marcossi *et al.* 2020; Moghadam *et al.*, 2010). The wide distribution and range of suitable food sources for this species suggest it could well adapt to extensive natural environments, including greenhouses and fields. Accordingly, the aim of the present study was to explore the suitability of cattail pollen (*Typha orientalis*) as an alternative food source for different stages of *A. herbicolus* in their predation capacity against whitefly eggs *T. vaporariorum* and the developmental and reproductive responses of the predators to different diet-switching regimes under laboratory conditions.

## Materials and methods

### Plants

The seedlings of *Solanum mauritianum* and *Solanum nigrum* were respectively grown from seeds in small plastic pots (30 cm height, 20 cm diameter) filled with potting soil. These plants were placed inside a mesh cage (52 x 52 x 72 cm), watered twice per week, and kept at approximately 25 °C, 60 ±10% relative humidity and a natural photoperiod in the greenhouse at Landcare Research, Auckland, New Zealand. None of the plants were ever exposed to any pesticides.

### Stock colonies of whitefly and predatory mite

*Trialeurodes vaporariorum* colony was reared on the seedlings of *S. mauritianum* in a greenhouse at Landcare Research. These whitefly-infested solanaceous plants were enclosed inside mesh cages. The whitefly rearing was continuous and required a fresh batch of whitefly-infested *S. mauritianum* plants once every two weeks.

The predatory mite, *Amblyseius herbicolus* (about 60 adult females) was initially collected from *Pittosporum crassifolium* (Karo) leaves on the campus of Landcare Research in St John, Auckland. The stock population was maintained in a climate controlled cabinet at approximately 25 ± 1 °C, 70 ±5% RH and 16:8 h photoperiod (Light: Dark) at Landcare Research, Auckland. The population was supplied with *T. orientalis* pollen as the food source. Part of their progeny was used for morphological species identification (Ma *et al.*, 2019) and the remainder of the progeny was maintained to establish the laboratory colony for use in the present study. Briefly, the phytoseiid stock culture of *A. herbicolus* was reared on a black PVC sheet (approximately 15 cm x 10 cm, with 12 holes of 0.5-mm diameter) placed on black tissue-papers wrapped over the edge of the water-saturated sponge in a plastic tray filled with water according to the protocols described by Patel & Zhang (2017). A few cotton threads were added underneath a white plastic sheet (about 1–2 cm<sup>2</sup>), functioning as shelter and substrate for oviposition for the predatory mites. *T. orientalis* pollen was dusted on these sheets with a brush twice every week.

### Preparation of developmentally synchronized *T. vaporariorum* eggs

The experiments were conducted on detached *S. nigrum* leaves cut from the first leaves of 2-week-old plants. Before the experiment, leaves were checked for the presence of any arthropods under a stereomicroscope and only the leaves without arthropods were used in the following experiment. The cohorts of whitefly eggs were used in the laboratory experiments. In order to prepare whitefly eggs in synchronized development, many experimental arenas were created, according to Li & Zhang (2016): each experimental arena contained two plexiglass slides clipped together at the sides: the top slide was 38 mm wide, 38 mm long and 3 mm thick, with a 16-mm-diameter hole in the middle, and the bottom slide was covered with a piece of freshly detached *S. nigrum* leaf that was placed upside-down on the black water-soaked tissue-paper. Approximately 20 adult whiteflies were collected with a fine hairbrush from *S. mauritianum* leaves in the greenhouse, subsequently transferred into each experimental arena, covered with a coverslip, and then incubated at 25 °C and 16:8 LD for 24 h. During that time, the adult female whiteflies laid eggs on the *S. nigrum* leaf discs. After one day, the adult females were removed from the leaf disc with a brush without damaging the eggs left on the leaf disc. The eggs laid by *T. vaporariorum* adults were used as a food source for nymphs or adult mites of *A. herbicolus* in the different experimental treatments. The number of the whitefly eggs was adjusted to twenty per leaf disc with a thin entomological needle and each leaf disc was used as an experimental arena. Our preliminary predation tests suggested that the amounts of whitefly eggs were sufficient and could meet the maximum prey consumption of the predators.

## Preparation of the predatory mites synchronized in development

*Amblyseius herbicolus* displays thelytokous parthenogenesis in its reproduction (De Moraes & Mesa, 1988), so adult female mites were used in this laboratory experiment. To produce the large cohorts of newly molted adult female mites that were synchronized in their development, 40 adult female mites were transferred from the stock rearing colony to the new plastic arenas (approximately 15 cm x 10 cm) placed on black tissue-papers and sponges in a plastic tray containing water, and the adult females were allowed to lay eggs at 25 °C for 24 h before removal. These developmentally synchronized eggs of *A. herbicolus* were maintained with *T. orientalis* pollen until they reached protonymphs, which were used in the following experimental treatments.

## Experimental design

To evaluate the effects of pollen and *T. vaporariorum* eggs on the performance of *A. herbicolus*, the following treatments were conducted:

- T1: Pollen (protonymph and deutonymph) + pollen (adult female).
- T2: Pollen (protonymph and deutonymph) + whitefly egg (adult female).
- T3: Whitefly egg (protonymph and deutonymph) + pollen (adult female).
- T4: Whitefly egg (protonymph and deutonymph) + whitefly egg (adult female).

Developmentally synchronized eggs of *A. herbicolus* prepared in the last section were used in the experiments and were transferred individually to each rearing arena of each treatment. A total of 24 eggs of *A. herbicolus* were used per treatment. In two treatments (T1 and T4), either pollen or whitefly eggs alone were given to them as a food source during immature stages and adults throughout the experiments. In T3 and T4 treatments, there was diet switching: when the eggs of predatory mites developed to protonymphs and deutonymphs, whitefly eggs (T3) or pollen (T2) were given to them as a food source; but when the deutonymphs molted and developed to adult mites, their food was switched to pollen or whitefly eggs, respectively. In the present study, the whitefly eggs were renewed daily to keep them fresh and at a density of 20 eggs per leaf disc during the experiment.

## Predation, survival, and reproduction of the predatory mite *A. herbicolus*

All the developmentally synchronized adult female mites that were treated according to above experimental treatments (T1, T2, T3 and T4) were individually starved for 24 hours before being used in this experiment. Each *A. herbicolus* adult female was transferred to an experimental arena containing twenty whitefly eggs, and the top slide of each experimental arena was then covered with a piece of transparent cling film punctured with an insect pin to allow ventilation and the maintenance of humidity. All the experiments were conducted in a room at 25±1 °C, 60 ±10% RH and 12:12 h (L:D) photoperiod. After 24 hours, twenty whitefly eggs of each experimental arena in every treatment were renewed to ensure sufficiently fresh prey supply. The number of whitefly eggs consumed and the number of eggs laid by each predatory mite within 24 and 48 hours were recorded, respectively. All the predatory mites were removed with the brush. Each egg laid by each female mite was then individually transferred to a new separate rearing arena and supplied with *T. orientalis* pollen as a food source every other day until the mites reached adulthood. The survival and the developmental stages of the offsprings' generation of experimental females were checked under the stereoscopic microscope and recorded daily.



## Statistical analysis

The predation rates, oviposition rates, and developmental durations of immature stages of *A. herbicolus* were analyzed using one-way analysis of variance (ANOVA); all mean comparisons with Duncan's test at  $P < 0.05$  were considered to have statistically significant differences. The statistical analyses were conducted using the package software SPSS v.20.0 (Chicago, IL, USA).

## Results

### Predation rates of *A. herbicolus* adult females on *T. vaporariorum* eggs

The predation rates of *A. herbicolus* adult females on *T. vaporariorum* eggs were statistically different among the four groups ( $P < 0.05$ ; Table 1). The highest and lowest predation rates of *A. herbicolus* were exhibited in groups T2 (young adults exposed to whitefly eggs) and T4 (both immature and adults exposed to whitefly eggs), respectively. *A. herbicolus* in group T2 at day 1 consumed 1.31, 1.16, and 1.50 times those in groups T1, T3 and T4, respectively. In addition, at day 2, the highest and lowest predation rates of *A. herbicolus* were exhibited in groups T2 and T4, respectively. However, no significant differences in predation rates of *A. herbicolus* were found between groups T1 and T3. *A. herbicolus* in the experimental group T2 at day 2 had the highest predation rate, which was 1.31, 1.21, and 1.60 times that in groups T1, T3, and T4, respectively. As for the average predation rate of *A. herbicolus* adult female on *T. vaporariorum* egg within two days, predation rates among the four groups revealed statistically different significances ( $P < 0.05$ ): the highest and lowest predation rates of *A. herbicolus* were recorded in groups T2 and T4, respectively. *A. herbicolus* in experimental group T2 revealed the highest average predation rate: 1.31, 1.17, and 1.49 times that in groups T1, T3 and T4, respectively.

### Development and reproduction of *A. herbicolus* fed on *Typha orientalis* pollen or *T. vaporariorum* egg

Although there were no statistically significant differences in the developmental durations of egg and larvae of *A. herbicolus* within the four groups ( $P > 0.05$ ), the developmental times of the protonymph and deutonymph stages of *A. herbicolus* revealed obvious differences (Table 2). The longest developmental durations for *A. herbicolus* protonymphs and deutonymphs were observed in groups T2 and T4, respectively. The developmental time of *A. herbicolus* protonymphs in group T2 was 1.15, 1.11, and 1.03 times those in groups T1, T3 and T4, respectively. Similarly, the developmental time of *A. herbicolus* deutonymphs in group T4 was 1.09, 1.18, and 1.20 times those in groups T1, T2, and T3, respectively. Taken together, all the eggs succeeded in reaching adulthood, but the duration of egg to adult developmental time of *A. herbicolus* was obviously different among the four treatment groups. The longest and shortest

**Table 1** Predation rates by adult female of *Amblyseius herbicolus* on *Trialeurodes vaporariorum* egg and cattail pollen (*Typha orientalis*) after 24 and 48 h ( $25 \pm 1$  °C,  $60 \pm 10\%$  RH and 12:12 h (L:D) photoperiod).

Treatment	Day 1	Day 2	Total
T1	$11.08 \pm 0.11c$	$2.54 \pm 0.15bc$	$13.63 \pm 0.07c$
T2	$14.54 \pm 0.11a$	$3.33 \pm 0.15a$	$17.79 \pm 0.25a$
T3	$12.50 \pm 0.19b$	$2.75 \pm 0.22b$	$15.25 \pm 0.31b$
T4	$9.67 \pm 0.11d$	$2.08 \pm 0.11c$	$11.92 \pm 0.11d$

Notes: Means  $\pm$  SE within each column followed by the same lowercase letter are not significantly different (Duncan's test,  $P > 0.05$ ).

**Table 2** Mean developmental duration of immature stages of *Amblyseius herbicolus* after predation on *Trialeurodes vaporariorum* egg and cattail pollen (*Typha orientalis*) (25±1 °C, 60 ±10% RH and 12:12 h (L:D) photoperiod).

Treatment	Developmental duration (days)				
	Egg	Larva	Protonymph	Deutonymph	Egg to Adult
T1	1.13±0.00a	1.04±0.02a	1.08±0.04b	1.15±0.02b	4.35±0.08b
T2	1.06±0.01a	1.04±0.02a	1.25±0.00a	1.06±0.00c	4.44±0.00b
T3	1.08±0.02a	1.02±0.02a	1.13±0.06ab	1.04±0.02c	4.16±0.08c
T4	1.15±0.06a	1.06±0.00a	1.21±0.02ab	1.25±0.04a	4.77±0.06a

Notes: Means ± SE within each column followed by the same lowercase letter are not significantly different (Duncan's test,  $P > 0.05$ ).

developmental times of egg to adult were obtained in groups T4 and T3, respectively (Table 2). Although no significant differences were observed between groups T1 and T2, there were statistically significant differences among groups T1 (or T2), T3, and T4.

The oviposition rates of *A. herbicolus* adult females at day 1 were highest in group T2, which was 1.54, 1.54, and 2.15 times those in groups T1, T3, and T4, respectively (Table 3). However, there were no significant differences among groups T1, T3, and T4 ( $P < 0.05$ ). *A. herbicolus* adult females in groups T2 and T3 at day 2 revealed the highest oviposition rates, which were 1.09 and 1.52 times those in groups T1 and T4, respectively; however, no significant differences in oviposition rates of *A. herbicolus* were found among groups T1, T2, and T3. Last, *A. herbicolus* adult females in group T2 within two days revealed the highest average oviposition rates, which were 1.32, 1.26, and 1.81 times those in groups T1, T3, and T4, respectively. The average oviposition rate of *A. herbicolus* adult females in group T1 was not significantly different from that of group T3 ( $P < 0.05$ ) (Table 3).

## Discussion

Some phytophagous mites (e.g. spider mites) and other small arthropods (e.g. whiteflies and thrips) are the most prominent targets of predatory mites in agroecosystems. Under moderate environmental conditions such as temperature and moisture, these target pests can concurrently breakout in greenhouses and fields. Therefore, to survive, develop, and reproduce, the biocontrol agents should have the capacity to prey on suitable natural target preys. *Amblyseius swirskii* successfully preyed on eggs and/or immature stages of whiteflies and thrips, as well as broad mites, simultaneously (Buitenhuis *et al.*, 2015). The eggs of the Mediterranean flour moth *Ephesia kuehniella* were a suitable food source for *A. swirskii* and *N. cucumeris* because

**Table 3** Oviposition rates of *Amblyseius herbicolus* after predation on *Trialeurodes vaporariorum* egg and cattail pollen (*Typha orientalis*) after 24 and 48 h.

Treatment	Day 1	Day 2	Average
T1	0.46±0.03b	0.46±0.04a	0.46±0.02b
T2	0.71±0.04a	0.50±0.00a	0.60±0.02a
T3	0.46±0.03b	0.50±0.01a	0.48±0.02b
T4	0.33±0.04b	0.33±0.04b	0.33±0.04c

Notes: Means ± SE within each column followed by the same lowercase letter are not significantly different (Duncan's test,  $P > 0.05$ ).

both phytoseiid species could develop from egg to adult (Delisle *et al.*, 2015). In addition, a couple of studies confirmed *A. herbicolus* could predate and reproduce on the immature stages of *S. staphylinus* in New Zealand (Lam *et al.*, 2019) and on the eggs of *Bemisia tabaci* (Gennadius) in Brazil (Cavalcante *et al.*, 2015). *Amblyseius herbicolus* females were found to have the lowest mean duration of immature stages ( $4.86 \pm 0.19$  day) and the highest fecundity ( $48.2 \pm 1.85$  eggs/female) when feeding on *Pseudodendrothrips mori* compared with *T. urticae* and the date palm pollen (Moghadam *et al.*, 2010). In our study, *A. herbicolus* could feed and develop to adults, and reproduce when fed with *T. vaporariorum* eggs, either as nymphs or adult females. Nevertheless, the immature developmental duration of *A. herbicolus* was significantly longer on *T. vaporariorum* eggs alone than on *Typha orientalis* pollen alone or the mixed diets, revealing the *T. vaporariorum* eggs alone were of inferior quality. We predicted the delayed immature developmental duration of *A. herbicolus* feeding and reduced oviposition rates on whitefly eggs would ultimately result in slower population growth in greenhouses and under field conditions. However, some studies suggested the low performance of phytoseiid mites such as *A. swirskii* fed on whiteflies or thrips or spider mites alone, indicated that although these predators can complete their immature development when fed this diet, they potentially cannot reproduce (Delisle *et al.*, 2015; Janseen and Sabelis, 2015). In the present study, when comparing with other groups, the average predation rates and oviposition rates of *A. herbicolus* adult females in group T4 were lowest, and development time was longest. The explanation for these differences in development and reproduction of these phytoseiid predators may be due to the deficiency of nutrients in the target prey alone (Muñoz-Cárdenas *et al.*, 2014; Bouvet *et al.*, 2019) and/or the difference of the nutritional requirements of different developmental stages of predators (Schultz *et al.*, 2019).

In general, generalist predators are widely used as prominent biological control agents worldwide because they can prey on phytophagous mites as well as on other small arthropods such as whiteflies and thrips. Indeed, they can also feed and reproduce on different types of pollen (Nomikou *et al.*, 2003; Hoogerbrugge *et al.*, 2011; Kolokytha *et al.* 2011; Delisle *et al.*, 2015; Liu & Zhang, 2017; Tsolakis *et al.*, 2019; Nemati *et al.* 2020; Pascua *et al.* 2020; Yazdanpanah *et al.* 2021). When prey are rare or even absent, or when prey quality is low, pollen functions as supplementary food for the predators (insects and mites) (Tuovinen and Lindqvist, 2010). Moreover, pollen also serves as a favorite food source of immature stages of several phytoseiid mites (Vantornhout *et al.*, 2004). Other studies also indicated the higher reproductive potential of the predatory mite *N. californicus* and *Typhlodromus bagdasarjani* on pollen compared with their natural prey (Riahi *et al.*, 2016; Khanamani *et al.*, 2017). The provision of pollen as supplementary food source on greenhouse-grown strawberries increased thrips and whitefly control of *A. limonicus* and *A. swirskii*. (Hoogerbrugge *et al.*, 2011). The predacious mites *A. swirskii* and *N. cucumeris* could develop and reproduce when fed on pollens (maize, cattail, and apple) (Delisle *et al.*, 2015). Our result suggested that *A. herbicolus* can prey on *T. vaporariorum* eggs when fed with *T. orientalis* pollen at the different developmental stages of protonymph/deutonymph or adult female. Furthermore, this study also found that *A. herbicolus* can develop and reproduce when fed with *T. orientalis* pollen either at protonymph/deutonymph or adult stage during the life cycle. A similar result was reported by Kolokytha *et al.* (2011), in which nine different plant pollens were all suitable for the immature stages, development, and reproduction of the predacious mite *Typhlodromus athenas* Swirski and Ragusa. A possible explanation for this could be due to the role played by the nutritional values of different pollens. In fact, previous studies have indicated that cattail pollen was considered a low-cost alternative food source with high nutritional value for some phytoseiid species such as *Euseius scutalis* and *A. swirskii* (Nomikou *et al.*, 2003), *A. limonicus* (Liu & Zhang, 2017; Lee & Zhang, 2018; Lam *et al.*, 2019), and *Typhlodromus athenas* (Kolokytha *et al.*, 2011). In addition, Moghadam *et al.* (2010) reported that date palm pollen may variously be used as an alternative food of *A. herbicolus* in the absence of primary food. Therefore, we consider that predatory mites utilize pollens when prey is scarce to increase survival and maintain their population in the crops. Indeed, under greenhouse and field conditions, many



kinds of pollens could be used as alternative/supplementary food sources for predaceous mites, not only to provide nutritional ingredients but also to sustain the population of predators before target preys become available (Lee & Zhang, 2018; Palevsky, 2017; Vangansbeke *et al.*, 2016; Warburg *et al.*, 2020).

The mixture of food sources has positive and/or negative effects on the developmental and reproductive performances of predatory insects and mites controlling the target pests, as well as on their population growth rates (Messelink *et al.*, 2008; Muñoz-Cárdenas *et al.*, 2014; Janseen & Sabelis, 2015; Samaras *et al.*, 2019, 2021). The quality of food source during the immature development is a key determinant of both fecundity and fertility in the mature stages of insects (Awmack & Leather, 2002). The suitability of the mixture of different food sources potentially showed positive effects on the performances of predators. For example, mixed diets were considered the most suitable for coccinellids development because the reproductive parameters of predators were significantly influenced by the diet consumed (Bouvet *et al.*, 2019). A significant increase of the intrinsic growth rate of a predator *Balaustium* population was found when predators fed on a mixture of *T. vaporariorum* and *T. urticae* eggs compared with other diets (Muñoz-Cárdenas *et al.*, 2014). Furthermore, a laboratory experiment by Messelink *et al.* (2008) with the predatory mites *A. swirskii* and *Euseius ovalis* demonstrated that the densities of predators were up to 15 times higher in the presence of both two major greenhouse pests *Frankliniella occidentalis* and *T. vaporariorum* than in the single-pest treatments because of a higher juvenile survival and developmental rate on a mixed diet. In the present study, results demonstrated that the average oviposition rates and developmental times from egg to adult of *A. herbicolus* in groups T2 and T3 were all higher and shorter, respectively, compared with groups T1 and T4. Additionally, the average oviposition rate in group T2 was the highest and the development time from egg to adult of *A. herbicolus* in group T3 was the shortest. The oviposition rates were affected by the previous food source of the adult predatory mites (Sabelis, 1990) and the peak oviposition rates of predatory mites were known to be a good stand-in parameter for population growth rates (Janssen and Sabelis, 1992). In our study, cattail pollen was added to the mixture of diets of different stages of *A. herbicolus*. Cattail pollen functioned as a supplementary ingredient in the mixed diet to elevate the nutritional values of the primary prey or in the early establishment of some phytoseiid species in crops (Nomikou *et al.*, 2003; Kolokytha *et al.*, 2011). We consider that the mixture of pollen and whitefly eggs were suitable for different developmental stages of *A. herbicolus* and indicated the positive effects on the development and reproduction of *A. herbicolus*. As a generalist facultative predator of type III, *A. herbicolus* can feed on a wide range of food types including thrips, whiteflies, phytophagous mites, and pollen from different plant species (Argov *et al.*, 2002; Cavalcante *et al.*, 2015; Hoogerbrugge *et al.*, 2011; Lam *et al.*, 2019; Marcossi *et al.*, 2020; Moghadam *et al.*, 2010). As an indigenous predatory mite in New Zealand, *A. herbicolus* can better adapt to the local conditions, which would ensure a greater chance of successful control of the target pests, including whiteflies. The suitability of cattail pollen and whitefly eggs as a diet for *A. herbicolus*, therefore, is an indication that mixed diets have the potential to support this predatory mite in controlling the target pest species in the agroecosystem. The ability of *A. herbicolus* to successfully complete development on whitefly eggs suggested it is a potential predator for controlling whitefly in biological control programs. Further comprehensive investigations under field conditions are necessary to test the potential suggested in this laboratory study.

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## Conflict of interest

The authors declare that they have no conflicts of interest with the contents of this article.

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