Does the native natural predacious mite, *Neoseiulus womersleyi* Schicha (Acari: Phytoseiidae), overwinter on apple trees in northern Japan?

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Abstract

Neoseiulus womersleyi is a potentially important biological agent for controlling *Tetranychus urticae* in apple orchards. However, overwintering sites of *N. womersleyi* in apple orchards have not been identified; therefore, the population dynamics of *N. womersleyi* and *T. urticae* on apple trees were investigated in Akita Prefecture, northern Japan, from April 2007 to March 2008. The population of *T. urticae* peaked on the leaves in mid-September. In contrast, the population of *N. womersleyi* rapidly increased and peaked in early October, and many adults were present until early November. Eggs of the predacious mite were not observed at all in early November. Adult females of *N. womersleyi* were found under rough bark in early December. *N. womersleyi* was significantly more abundant under rough bark containing many diapausing *T. urticae* females than under bark where the prey were few; however, the mean number of *N. womersleyi* early in March 2008 was significantly lower than in early December. Both *N. womersleyi* and *T. urticae* were collected using Phyto traps attached to apple twigs. Neither was captured between April and September, but the numbers of both increased rapidly in October.

Key words: *Neoseiulus womersleyi*; overwintering site; apple orchard; *Tetranychus urticae*

INTRODUCTION

The predacious mite *Neoseiulus womersleyi* Schicha is a native species of Japan and a natural enemy of *Tetranychus* species on various crops in this country (Shinkaji and Adachi, 1978; Hamamura, 1986). *N. womersleyi* is distributed from northern Hokkaido to southern Okinawa (Amano et al., 2004), and has been observed in commercial apple orchards in Akita Prefecture in recent years (Hongo and Funayama, 2004); however, the density of predacious mites in fruit tree orchards generally increases only after the spider mite numbers reach damaging levels (Kishimoto, 2002). This time lag is considered the primary reason why predacious mites cannot effectively control spider mites, but their effectiveness as a biological control agent could be improved if larger populations could be maintained on trees before spider mite populations begin to grow (Kawashima and Amano, 2006). Jung et al. (2004) suggested that

the initial establishment and subsequent conservation of *N. womersleyi* are both important in the successful biological control of *T. urticae* in apple orchards.

Another native predacious mite, *Typhlodromus vulgaris* Ehara, has also been reported in apple orchards (Hongo and Funayama, 2004). This species overwinters in the cocoons of insects and in artificial microstructures (Phyto traps) attached to the twigs of woody plants (Kishi and Mori, 1979). Kawashima and Amano (2006) reported that *T. vulgaris* can also overwinter in Phyto traps (Koike et al., 2000) attached to the twigs, main branches, and trunks of Japanese pear until early spring. Such artificial microstructures attached to apple trees could possibly provide overwintering sites for *N. womersleyi*, which could in turn lead to an increase in *N. womersleyi* earlier in spring, coincident with the increase of the population of *T. urticae*. It is therefore necessary to clarify the hibernation habits of *N. womersleyi* on apple trees. Pre-

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viously, overwintering *N. womersleyi* have only been observed on herbaceous plants (Kishi and Mori, 1979; Takahashi and Mori, 1979). On the other hand, Kishi and Mori (1979) reported that they found overwintering female adults of *N. womersleyi* in the calyx end basin of mature pear fruits in late autumn, suggesting that woody hosts could also provide overwintering sites for *N. womersleyi*.

In the present study, the distributions and densities of predacious mites and spider mites were investigated on apple trees throughout the year. A possible method to improve the effectiveness of *N. womersleyi* in the control of *T. urticae* on apples is discussed from the viewpoint of overwintering ecology.

MATERIALS AND METHODS

Apple orchard. The investigation was conducted from April 2007 to March 2008 in an apple orchard of the Fruit-Tree Experiment Station of the Akita Prefecture Agriculture, Forestry and Fisheries Research Center (39.14°N, 140.32°E) in Yokote, Akita Prefecture. The orchard (15 a) contained about 20 apple trees (*Malus domestica* 'Fuji'), which were all older than 30 years and about 4 m height. The pest control record for the orchard is shown in Table 1.

Seasonal fluctuations in mite numbers on leaves and rough bark of apple trees. Ninety apple leaves were sampled from three trees (i.e., 30 leaves per tree) at intervals of 10 days from June to October. Mites on sampled leaves were brushed

into a glass Petri dish (120 mm in diameter) spread with dilute detergent using a brushing machine (DKI-7200; Daiki Co. Inc., Tokyo, Japan), and adult female spider mites and eggs of predacious mites were counted under a binocular microscope. Predacious mites were mounted on Hoyer's medium, the species was identified under a microscope, and adult females of each species were counted.

Pieces of rough bark (about 2×3 cm) were sampled from the three apple trees by stripping the bark from the surface of primary scaffold branches in early December 2007 and early March 2008. Each piece of bark was classified into one of two groups based on the number of individual diapausing female *T. urticae*, either many (>30) or few (<4). Few pieces of rough bark contained intermediate numbers of *T. urticae* because the distribution of overwintering individuals on apple trees is clumped (Narita and Takahashi, 1981). Moreover, rough bark samples containing few (<4) individuals of *T. urticae* were also sampled similarly in early December 2007. The numbers of predacious mites were compared for each species between the following conditions: (1) bark pieces containing many and few *T. urticae* in December 2007, and (2) December 2007 and March 2008 for pieces with many *T. urticae* present. Mann-Whitney's *U*-test was used for the comparison.

Test of Phyto traps as an overwintering site. Phyto traps were constructed following the procedure of Koike et al. (2000), although the structure was modified slightly: a 2×1 cm piece of Velcro

Table 1. Pest control record for the apple orchard in 2007

Time of application	Insecticide (%AC, formulation)	Concentration (ppm)	Acaricide (%AC, formulation)	Concentration (ppm)	Fungicide (% AC, formulation)	Concentration (ppm)
Mid-May	flufenoxuron (10, EC)	33			dithianon (40, F)	200
Late May					hexaconazole (2, F)	20
					ziram (25,F)	500
					thiuram (15, F)	300
Mid-June	thiacloprid (30, WP)	75			manzeb (75, WP)	1,500
Late June					manzeb (75, WP)	1,500
Mid-July	clothianidin (16, SP)	40			fosetyl (40,WP)	500
					captan (40, WP)	500
Late July			etoxazole (10, F)	50	fosetyl (40,WP)	500
					captan (40, WP)	500
Early Aug.			chlorfenapyr (10, F)	50	kresoxim-methyl (41.5, F)	208
Late Aug.			milbemectin (1, EC)	10	kresoxim-methyl (41.5, F)	208

AC: active ingredient, WP: wettable powder, EC: emulsifiable concentrate, F: flowable, SP: water-soluble powder.

(hooked surface) was stapled to a piece of transparent vinyl tape (10×1.5 cm), and wool yarn was attached to the Velcro. Each Phyto trap was then attached to an apple twig using a double-arm clip such that the Velcro was in contact with the twig. The traps were washable and could be reused many times.

Within the orchard, 10 Phyto traps were attached to previous-year branches of three apple trees from April to October. The traps were collected at 7-d intervals and replaced with new traps. Collected traps were observed under a binocular microscope, and the numbers of spider mites and predacious mites in the traps were counted and identified to the species level.

RESULTS AND DISCUSSION

Seasonal fluctuations in predacious mites and spider mites on leaves in the apple orchard in 2007 are shown in Fig. 1. *T. urticae* was observed from late July and rapidly increased to a peak in mid-September. Diapausing females of *T. urticae* were observed from early October. *Panonychus ulmi* Koch were observed from late June to early August, though their numbers were low (fewer than 20 individuals per 90 leaves). *N. womersleyi* was observed from early September; the population density rose rapidly to a peak in early October, and the adults were still observed in early November. *T.*

vulgaris was also observed from early September; the population density rose rapidly and peaked in early October, and the adults were observed until late October. *N. womersleyi* on the leaves of apple trees have been reported to increase when *T. urticae* populations grow (e.g., Hongo and Funayama, 2004; Jung et al., 2004), although the increase was not observed until August in the present study (Fig. 1). Regarding the pesticides sprayed in this orchard, an insect growth regulator (flufenoxuron) and neonicotinoids (thiacloprid and clothianidin) have been shown to be harmless for *N. womersleyi* (Funayama, unpublished data), as has chlorfenapyr (Amano et al., 2004); however, the acaricides etoxazole and milbemectin have been found to be slightly or moderately harmful to *N. womersleyi* (Amano et al., 2004). Therefore, the occurrence of *N. womersleyi* on apple leaves may have been suppressed by spraying these acaricides from late July to late August.

Adult *N. womersleyi* diapause in November in Hiroshima Prefecture, southwestern Japan (Hamamura, 1982), and in mid-September in Sapporo city, Hokkaido Prefecture, northern Japan (Takahashi and Mori, 1979). The induction of diapause in adult predacious mites is reported to occur when they have completed egg deposition (Hamamura, 1982). In the present study, the number of eggs of predacious mites increased from early September and peaked in late September to early October;

Fig. 1 Seasonal fluctuations in the numbers of adult females and eggs of predacious mites and spider mites on the leaves of apple trees in the orchard from June to November 2007. Ninety apple leaves were sampled from three trees (i.e., 30 leaves per tree) at intervals of 10 d.

thereafter, the number decreased until late October and no eggs were observed in early November (Fig. 1). This observation indicates that most *N. womersleyi* and *T. vulgaris* entered diapause after early November in Akita Prefecture. The numbers of predacious mites and *T. urticae* found under pieces of rough bark of apple trees in early December 2007 are shown in Table 2. Adults of *N. womersleyi* and *T. vulgaris* were found under rough bark; all were females. This observation suggests that at least some adult females of the *N. womersleyi* population overwintered on apple trees in the orchard, as is the case for *T. vulgaris*.

The mean number of *N. womersleyi* was significantly larger under rough bark that hosted many (>30) *T. urticae* than under bark that hosted few (<4) *T. urticae* ($p<0.01$) (Table 2). There was no significant difference between the mean numbers of *T. vulgaris* observed under rough bark at the two densities of *T. urticae* ($p>0.01$) (Table 2). This ob-

servation suggests that the number of *N. womersleyi* respond to the aggregation size of *T. urticae* infesting rough bark, even in the selection of overwintering sites. On the other hand, the mean number of *N. womersleyi* observed under rough bark containing many diapausing female *T. urticae* in early March 2008 was significantly lower than that observed in early December 2007 ($p<0.01$) (Table 2). There was no significant difference between the mean numbers of *T. vulgaris* observed under rough bark in the same two periods ($p>0.01$) (Table 2). This observation suggests that the rough bark of apple trees may not provide a particularly suitable overwintering site for *N. womersleyi*; however, it remains unknown whether the survival rate of *N. womersleyi* declined during the winter. Thus, more detailed investigation will be necessary to clarify the selection of overwintering sites by *N. womersleyi* on apple trees.

Seasonal fluctuations in the numbers of adult fe-

Table 2. The numbers of predatory mites and *Tetranychus urticae* found in the bark of apple trees in the orchard in early December 2007 and early March 2008

Species	N	Number observed in old bark (Mean $\pm$ SE)			a	b
		Dec. 2007		Mar. 2008		
		With many (>30) <i>T. urticae</i>	With few (<4) <i>T. urticae</i>	With many (>30) <i>T. urticae</i>		
<i>Tetranychus urticae</i>	42	73.4 $\pm$ 6.6	0.5 $\pm$ 0.2	89.2 $\pm$ 9.1	**	
<i>Neoseiulus womersleyi</i>	42	1.4 $\pm$ 0.5	0.1 $\pm$ 0.1	0.3 $\pm$ 0.1	**	**
<i>Typhlodromus vulgaris</i>	42	0.2 $\pm$ 0.1	0.1 $\pm$ 0.0	0.3 $\pm$ 0.2		

Asterisks (**) indicate significant difference between (a) number of bark pieces containing many and few *T. urticae* in December 2007 within each species, and (b) number of *N. womersleyi* on bark with many *T. urticae* in December 2007 and March 2008 ($p<0.01$, Mann-Whitney *U*-test).

Fig. 2 Seasonal fluctuations in the numbers of adult female predacious mites and *Tetranychus urticae* in Phyto traps attached to apple twigs in the orchard from April to October, 2007. Ten Phyto traps were attached to each of three apple trees and were collected for observation and replaced at 7-d intervals.

male of predacious mites and *T. urticae* in Phyto traps attached to twigs of apple trees in the orchard from April to October 2007 are shown in Fig. 2. Adult female *T. vulgaris* were collected in Phyto traps from late April to early May and in October. This observation suggests that *T. vulgaris* used the Phyto traps not only as overwintering sites but also as a refuge. On the other hand, no *N. womersleyi* were collected in the traps from April to September (Fig. 2), but numbers increased through the first weeks of October, suggesting that *N. womersleyi* used the Phyto traps only for overwintering. This suggests that Phyto traps are not suitable for measuring the population dynamics of *N. womersleyi* on apple trees in summer; however, it is possible that *N. womersleyi* used Phyto traps as overwintering sites, because the increasing number of adult female *N. womersleyi* collected in the traps was synchronized with increases in the number of diapausing female *T. urticae* in October. In addition, more adult female *N. womersleyi* were collected in Phyto traps than from rough bark (Table 2 and Fig. 2). Kawashima and Amano (2006) reported that *T. vulgaris* can overwinter in Phyto traps (Koike et al., 2000) on Japanese pear until early spring; therefore, installation of artificial microstructures such as Phyto traps in apple orchards may maintain a higher population density of predacious mites through the winter and thus may enable the *N. womersleyi* population to better control the size of the *T. urticae* population. Detailed investigations are necessary to determine whether the presence of artificial microstructures can in fact increase the population density of *N. womersleyi* in apple orchards in early spring.

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