ORIGINAL RESEARCH PAPER



Seasonal adult occurrence of a coccidophagous ladybird, *Telsimia* nigra (Coleoptera: Coccinellidae), in citrus groves in central Japan

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Abstract

A coccidophagous ladybird, *Telsimia nigra* (Weise) (Coleoptera: Coccinellidae), may play a significant role in suppressing the abundance of scale insects belonging to Diaspididae (Hemiptera: Coccoidea) in citrus groves in Japan, and therefore might need to be protected from pesticide applications. Seasonal changes in the abundance of *T. nigra* adults were monitored using yellow sticky traps set inside citrus tree canopies and by beating citrus foliage in citrus groves with different pesticide application levels in Shizuoka City, central Japan. *Telsimia nigra* adults were caught on sticky traps in pesticide-free, organic, and reduced-pesticide groves, with far more adults in a pesticide-free grove, but were rarely recorded in conventionally managed groves. A larger number of *T. nigra* adults were captured on the traps from late March to early May, with a peak numbers in early to mid-April in the pesticide-free grove, and afterward adults were continually detected in much smaller numbers until November. Some adults were trapped mainly in April–May and October in organic and reduced-pesticide groves, with more adults collected by beating citrus foliage tended to be more abundant in April and November in the pesticide-free grove. Thus, *T. nigra* adults occurred most abundantly in April–May and in smaller numbers in October–November in citrus groves in Shizuoka City. This result suggests that the application of pesticides that are harmful to ladybirds should be avoided in April–May and autumn to conserve *T. nigra* populations in citrus groves in central Japan.

Keywords Beating method · Citrus · Seasonal occurrence · Sticky trap · Telsimia nigra

Introduction

Several species of scale insects belonging to Diaspididae (Hemiptera: Coccoidea), chiefly the San Jose scale *Comstockaspis perniciosa* (Comstock) and the California red scale *Aonidiella aurantii* (Maskell), cause severe damage to the growth and fruit production of citrus trees in Japan (Korenaga et al. 2001). A ladybird, *Telsimia nigra* (Weise) (Coleoptera: Coccinellidae), attacks various species of diaspidid scales in Japan (Kurosawa et al. 1985), such as the mulberry scale *Pseudaulacaspis pentagona* (Targioni Tozzetti) on mulberry (Yasuda 1981) and *Comstockaspis macroporana* (Takagi) on *Castanea crenata* Siebold et Zucc. (Urano 2015). Thus, *T. nigra* may play an important role in

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depressing the abundance of the diaspidid scales on citrus trees.

There have only been a few studies concerning the occurrence of the ladybird T. nigra in citrus groves in Japan (Nakao 1975; Nakao and Ohgushi 1976; Nohara 1963). Moreover, these studies investigated the abundance of T. nigra adults only during a relatively short period of time. More reliable information on T. nigra occurrence in citrus groves is necessary to propose how to effectively utilize the ladybird species as a biological control agent against the diaspidid scales on citrus trees. Particularly, the seasonal prevalence of the ladybird occurrence on citrus trees needs to be accurately grasped, because this information is essential for determining the months in which the ladybird is more abundant and therefore application of pesticides that are potentially harmful to the ladybird should be avoided to conserve its populations in citrus groves. In addition, T. nigra occurrence needs be monitored in citrus groves with various levels of pesticide application, as its abundance may differ among the citrus groves.

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It has been reported that setting yellow sticky traps inside canopies of citrus trees is a useful way to monitor the seasonal occurrence of adults of the three ladybird species, *Platynaspidius maculosus* (Weise), *Cryptolaemus montrouzieri* Mulsant, and *Serangium japonicum* Chapin (all, Coleoptera: Coccinellidae) in citrus groves (Kaneko 2013, 2017a, b, respectively). Compared with beating citrus foliage, monitoring using yellow sticky traps captured more ladybird adults and more clearly indicated peaks in the number of caught ladybird adults (Kaneko 2017a, b).

The present study conducted monitoring of the seasonal abundance of *T. nigra* adults by placing yellow sticky traps inside citrus tree canopies and by beating citrus foliage for 4 years in three experimental citrus groves, and with sticky traps for 2 years in nine commercial citrus groves in Shizuoka City, central Japan. These groves varied in pesticide application levels. Based on these results, I describe the relevance between *T. nigra* adult abundance and pesticide application levels across the citrus groves in Shizuoka City and document seasonal changes in the ladybird adult abundance in the groves. I then determine the months in which pesticides that are detrimental to ladybird adults should be prohibited to protect *T. nigra* populations in citrus groves.

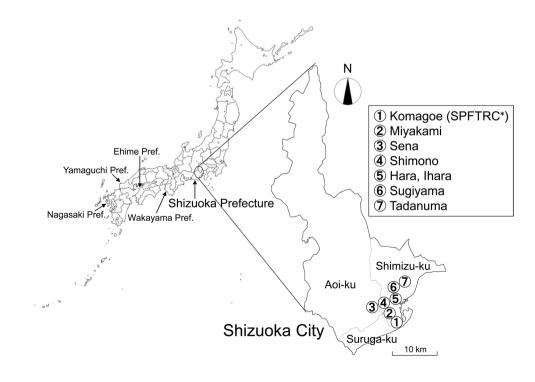
Materials and methods

Adult occurrence in experimental citrus groves

The abundance of *T. nigra* adults was monitored in three experimental groves (each more than 0.05 ha; Table 1)

Fig. 1 Map showing citrus groves where adults of the coccidophagous ladybird *Telsimia nigra* were monitored in Shizuoka City, central Japan. (Asterisk) Shizuoka Prefectural Fruit Tree Research Center cultivating Satsuma mandarin, *Citrus unshiu* (Swingle) Marcow., at the former Shizuoka Prefectural Fruit Tree Research Center (N 34°58′24″, E 138°29′32″) in Komagoe, Shizuoka City, central Japan (Fig. 1, no. 1). The three groves were labeled as follows: KO-1(f) where no pesticide had been applied for more than 20 years (hereafter referred to as pesticide-free grove); KO-2(r) [ca. 15 m away from KO-1(f)] where pesticide application was reduced ~ 50% less than that in conventionally managed groves, with insecticides being sprayed in early to mid-June (petroleum oil and Imidacloprid) and late August to early September (Spinosad) (hereafter referred to as reduced-pesticide grove); and KO-3(c) [ca. 100 m away from KO-1(f)] that was conventionally managed (hereafter referred to as conventional grove).

Monitoring using sticky traps was undertaken in the three groves continuously from January 2009 to December 2012. Five citrus trees, which were ~ 5 m apart from one another, were randomly chosen at the center of each grove at the end of December 2008, and these trees were continuously used for monitoring until the end of the survey. Inside each tree canopy, a single piece of double-sided, yellow sticky card trap [a yellow adhesive sheet ('IT-sheet', SANKEI CHEMI-CAL CO., LTD.) of 100 × 200 mm attached on each side of a yellow acrylic plate of 200 × 200 mm] was set in a vertical position 1.3–1.5 m above the ground using a wooden pole and metal fittings. These traps were replaced at an interval of 6-8 days (generally 7 days) from March through November or of 12-16 days (generally 14 days) from December through February, and the number of T. nigra adults captured on the collected traps was counted under a binocular



microscope (SZX12, OLYMPUS). For each of the dates at which the traps were replaced in each citrus grove, the mean number of captured *T. nigra* adults per trap per day was calculated by dividing the total number of adults on the traps collected at the date by the number of traps and the period (days) from the date at which the traps had been newly set until the date at which the traps were collected. Seasonal changes in the mean number of *T. nigra* adults per trap per day for 4 years in the three groves are shown in Fig. 2.

Sampling by beating citrus foliage was conducted in the three groves in 2009-2012 (from April through November of each year; Table 2). This was done twice a month: early (between the 6th and 8th, generally the 7th) and late (between the 22nd and 24th, generally the 23rd) in the month. Ten citrus trees were randomly selected at the center of each grove on each sampling date; the trees inside which sticky traps were set were not included in this survey. Then, two canopy portions (each involving ~ 100-200 leaves) on opposite sides (180° apart) were chosen on each tree. Citrus leaves and twigs of each portion were beaten 10 times using a wooden pole (1.5 cm in diameter, 1 m in length) and the insects that fell from the foliage were caught using an insect net (20 cm in diameter) held under each portion. Thus, a total of 200 times of beating were done for a single sampling in each grove. The number of T. nigra adults collected in the insect net was counted under the binocular microscope. Seasonal changes in the number of T. nigra adults collected through 200 times of foliage beating for four years in the three groves are described in Fig. 3.

Adult occurrence in commercial citrus groves

Monitoring of T. nigra adults using sticky traps was carried out in 2011-2012 (from April through November of each year) in nine commercial C. unshiu groves (each more than 0.1 ha; Table 3). The groves were located in the southern part of Shizuoka City, mostly in Shimizu-ku, one of the major citrus-producing areas in Shizuoka Prefecture (Fig. 1, nos. 2–7). The groves included three groves in Sena-1, Shimono, and Sugiyama [hereafter referred to as SE-1(o), SH(o), and SU(o)] that were organically managed by spraying only lime sulfur or petroleum oil once or twice a year (in March and/or June) (hereafter referred to as organic grove). The groves also included one grove in Tadanuma-1 [hereafter referred to as TA-1(r)] where pesticide application was reduced by ~ 50% compared with that in conventionally managed groves, with insecticides being sprayed in early January (petroleum oil) and late June (Buprofezin) (referred to as reduced-pesticide grove). In addition, they included five groves in Miyakami, Sena-2, Hara, Ihara, and Tadanuma-2 [hereafter referred to as MI(c), SE-2(c), HA(c), IH(c), and TA-2(c)] that were conventionally managed (referred to as conventional grove).

Three citrus trees, which were ~ 5 m apart from one another, were randomly selected at the center of each grove at the end of March 2011, and these trees were continuously utilized for monitoring until late November 2012. A single piece of double-sided, yellow sticky card trap was set inside each tree canopy, as previously described. These traps were replaced at an interval of 12-16 days (generally 14 days), and the number of T. nigra adults captured on the collected traps was counted under the binocular microscope. For the four citrus groves [SE-1(o), SH(o), SU(0), and IH(c)] where one or more T. nigra adults were caught in 2011–2012, the mean number of T. nigra adults per trap per day at each trap replacement date was calculated in the same way as done for the experimental citrus groves. Seasonal changes in the mean number of T. nigra adults per trap per day for 2 years in the four groves are shown in Fig. 4.

Results

Adult abundance in citrus groves

Among the three experimental citrus groves in Komagoe, a large number of *T. nigra* adults were captured on sticky traps in the pesticide-free grove [KO-1(f)], with some adults in the reduced-pesticide grove [KO-2(r)] and a few adults in the conventional grove [KO-3(c)] (Table 1).

Beating citrus foliage in the three experimental groves in Komagoe showed a difference in the number of collected *T. nigra* adults among these groves (Table 2), which was similar to the difference in the number of adults caught on sticky traps among the groves (Table 1).

A small number of *T. nigra* adults were trapped in all of the three organic commercial citrus groves in Shizuoka City (Table 3). However, *T. nigra* adults were rarely

Table 1Mean (\pm SD) number of captured *Telsimia nigra* adults persticky trap in each experimental citrus grove in Komagoe, ShizuokaCity in 2009–2012

Year	No. of	Citrus grove						
	traps	KO-1(f) ^a	KO-2(r)	KO-3(c)				
2009	5	21.2 ± 9.1	1.6 ± 1.4	0.2 ± 0.4				
2010	5	43.8 ± 13.1	1.8 ± 1.6	0				
2011	5	30.4 ± 15.3	2.0 ± 0.9	0.2 ± 0.4				
2012	5	14.0 ± 10.1	1.0 ± 1.1	0				

Traps were replaced at an interval of ca. 7 days (March–November) or ca. 14 days (December–February) from January through December ^aLetters in parentheses mean: f pesticide-free, r reduced-pesticide, c conventional

2012

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Year No. of Citrus grove trees KO-1(f)^a KO-2(r) KO-3(c) 0 2009 10 71 0 2010 10 129 2 0 2011 181 7 10 1

 Table 2
 Total numbers of *Telsimia nigra* adults collected by beating citrus foliage in each experimental citrus grove in Komagoe, Shizuoka City in 2009–2012

Two-hundred times of beating were done early and late in each month (from April through November)

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^aLetters in parentheses mean: f pesticide-free, r reduced-pesticide, c conventional

captured on sticky traps in the reduced-pesticide and conventional commercial groves.

Seasonal adult occurrence in citrus groves

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A larger number of *T. nigra* adults were caught on sticky traps from late March to early May, with a peak of their numbers appearing in early to mid-April in the pesticide-free experimental grove [KO-1(f)] (Fig. 2). From June through November, *T. nigra* adults were almost constantly trapped in much smaller numbers in the grove. Some adults were recorded in April–May and October in the reduced-pesticide grove [KO-2(r)], with more adults in the former months. One adult was noted each in July 2009 and October 2011 in the conventional grove [KO-3(c)].

Foliage beating indicated that *T. nigra* adults tended to be more abundant on citrus trees in April and November in the pesticide-free experimental grove [KO-1(f)] (Fig. 3). Adults of *T. nigra* were almost constantly captured in other months in the grove, with slightly fewer adults in midsummer (from late July to late August). A few adults were collected between early April and early June and between early September and early November in the reduced-pesticide grove [KO-2(r)]. Only one adult was found in late September 2011 in the conventional grove [KO-3(c)]. More *T. nigra* adults tended to be caught on sticky traps in late April to early May in the three organic and one conventional, commercial citrus groves in Shizuoka City (Fig. 4). A few adults were also detected in late September to early October.

Discussion

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The present study found adults of the coccidophagous ladybird *T. nigra* in seven out of the 12 citrus groves in Shizuoka City, central Japan (Tables 1, 2, 3). Nakao (1975) noted the presence of *T. nigra* adults in citrus groves in Wakayama Prefecture and Ehime Prefecture, both western Japan, and in Nagasaki Prefecture, southern Japan (see Fig. 1). Nohara (1963) captured *T. nigra* adults in citrus groves in Yamaguchi Prefecture, located between Wakayama Prefecture and Nagasaki Prefecture (see Fig. 1). Thus, these three studies suggest that *T. nigra* is widely distributed across citrus groves in central to western and southern Japan.

The present survey in Shizuoka City recorded a number of *T. nigra* adults in the pesticide-free, organic, and reduced-pesticide citrus groves, whereas it rarely collected the adults in the conventional groves (Tables 1, 2, 3). Nakao and Ohgushi (1976) caught some *T. nigra* adults in two pesticide-free citrus groves, but no adults in 15 conventional groves in Nagasaki Prefecture. These two studies suggest that *T. nigra* seldom occurs in conventional citrus groves in Japan. Further studies are necessary to clarify whether the absence of *T. nigra* in conventional citrus groves is caused by direct detrimental impacts of pesticide applications on the ladybird individuals on citrus trees or results from the absence of its prey diaspidid scales on the trees due to the pesticide applications.

Some *T. nigra* adults were captured in the organic and reduced-pesticide citrus groves, but the adult numbers in these groves were much smaller than those in the pesticide-free grove (Tables 1, 2, 3). In the organic and reduced-pesticide groves, no insecticide was applied in April–May when *T. nigra* adults were abundant in the pesticide-free grove (Figs. 2, 3). On the other hand, insecticides (petroleum oil,

 Table 3
 Mean (±SD) number of captured Telsimia nigra adults per sticky trap in each commercial citrus grove in Shizuoka City in 2011–2012

Year	No. of traps	Citrus grove								
		Organic			Reduced-pesticide	Conventional				
		SE-1(o) ^a	SH(o)	SU(o)	TA-1(r)	MI(c)	SE-2(c)	HA(c)	IH(c)	TA-2(c)
2011	3	0	1.0 ± 0	5.7 ± 2.1	0	0	0	0	0.3 ± 0.5	0
2012	3	1.3 ± 1.2	0.7 ± 0.5	1.0 ± 0.8	0	0	0	0	0	0

Traps were replaced at an interval of ca. 14 days from April through November

^aLetters in parentheses mean: o organic, r reduced-pesticide, c conventional

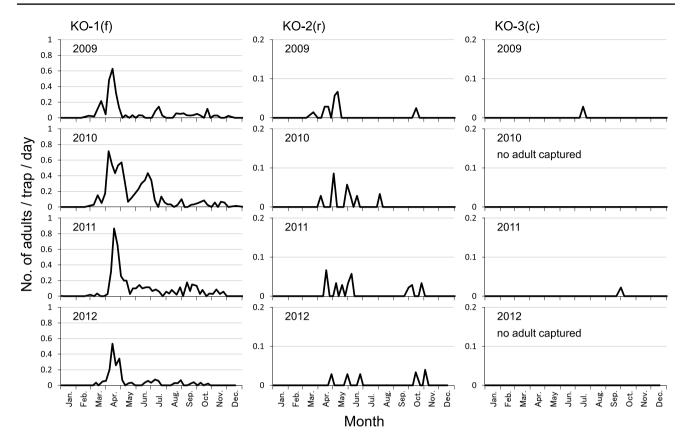


Fig. 2 Seasonal changes in the mean number of captured *Telsimia nigra* adults per sticky trap per day in a pesticide-free experimental citrus grove [KO-1(f)], a reduced-pesticide grove [KO-2(r)], and a conventional grove [KO-3(c)] in Komagoe, Shizuoka City in 2009–2012. The mean numbers were calculated as described in the main

text. Traps were replaced at an interval of ca. 7 days (March–November) or ca. 14 days (December–February) from January through December. Note that the scale of *y*-axes of the graphs differs between KO-1(f) and others

lime sulfur, or Buprofezin) had been sprayed in January, March, or June in the organic and reduced-pesticide groves to control scale insects including diaspidid scales, and therefore the abundance of diaspidid scales in these groves would have been much lower than that in the pesticide-free grove. The lower abundance of diaspidid scales may have led to the smaller number of *T. nigra* adults in the organic and reducedpesticide groves, compared with in the pesticide-free grove, despite of no insecticide application in April–May. Thus, to assess the influence of pesticide application schedules on *T. nigra* populations in citrus groves, we need to take into consideration both the direct impacts of the pesticide applications on the ladybird individuals and their indirect effects on the ladybirds through the changes in diaspidid scale abundance caused by them.

This study indicated that *T. nigra* adults were most abundant in April in the pesticide-free experimental citrus grove in Shizuoka City (Fig. 2). Similarly, more *T. nigra* adults were captured in late April to early May in organic commercial citrus groves in the city (Fig. 4). It is unclear why *T. nigra* adults occur more abundantly from early April to early

May in citrus groves. Kaneko et al. (2006) demonstrated that the seasonal occurrence of adults of a coccidophagous ladybird species roughly synchronized with that of crawlers (first-instar larvae) of its prey scale insect species. Specifically, the number of adults of the ladybird Pseudoscymnus hareja (Weise) (Coleoptera: Coccinellidae) caught on sticky traps inside tea tree canopies reached a peak an average of 7 days after the peak day in the number of crawlers of its prey scale insect *P. pentagona* trapped on the sticky sheets. This would be because larger numbers of P. hareja adults visited the tea trees mainly to deposit young larvae that primarily consume young larvae of the scale insect (Kaneko et al. 2006). Telsimia nigra attacks various species of scale insects belonging to Diaspididae (Kurosawa et al. 1985). The San Jose scale C. perniciosa is one of the predominant diaspidid scales on citrus in Japan (Korenaga et al. 2001) and was found in the pesticide-free citrus grove (S. Kaneko, personal observation). Therefore, T. nigra may have exploited the scale C. perniciosa as its main prey in the citrus grove. The scale overwinters mostly at the first-instar larval stage in Wakayama Prefecture, settling on citrus trees and forming

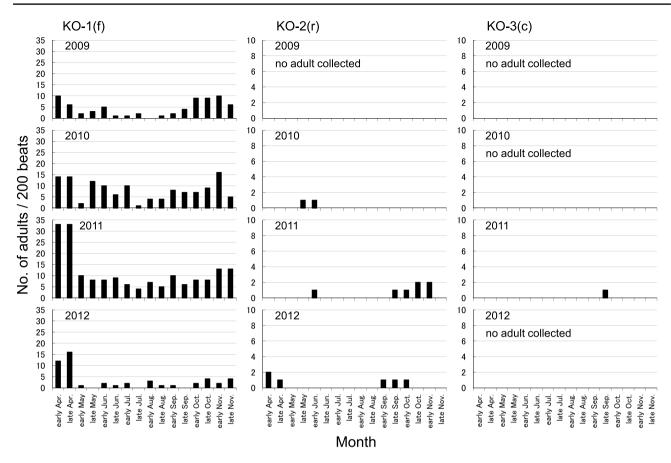


Fig. 3 Seasonal changes in the number of *Telsimia nigra* adults collected through 200 times of beating of citrus foliage in a pesticide-free experimental citrus grove [KO-1(f)], a reduced-pesticide grove [KO-2(r)], and a conventional grove [KO-3(c)] in Komagoe, Shi-

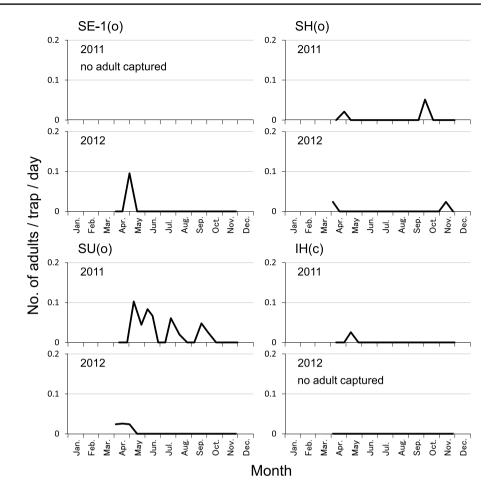
zuoka City in 2009–2012. Foliage beating was done early and late in each month (from April through November). Note that the scale of *y*-axes of the graphs differs between KO-1(f) and others

a wax cover (Matsuura and Hatta 1972). A first-instar larva becomes a second-instar larva in late March to early April, and then a male pupa in mid- to late April or a female adult in late April to early May, which produces crawlers from late May to mid-June in the prefecture (Matsuura and Hatta 1972). Thus, in April when T. nigra adults were noted in the largest numbers in the pesticide-free citrus grove, the scale population would consist of second-instar larvae, male pupae, and female adults, and it would be one month or more before the scale crawlers occur. It is therefore unlikely that the seasonal occurrence of T. nigra adults is synchronized with that of the scale crawlers. To elucidate the reason why T. nigra adults were most abundant in April in citrus groves, we would need to determine the main prey species of the coccidophagous ladybird, the main developmental stages of the prey scale insects utilized by each of the ladybird larvae and adults, and the seasonal occurrence of each developmental stage of the ladybird and the scale insects on citrus trees.

Sampling by beating citrus foliage showed that *T. nigra* adults tended to occur more abundantly on citrus trees not

only in April but also in November in the pesticide-free grove (Fig. 3). This result differed from that of monitoring using sticky traps in the same grove (Fig. 2). It is considered that sticky traps set inside citrus tree canopies capture insects actively flying or walking inside the canopies, including those blown off by strong winds, whereas beating citrus foliage collects insects staying or foraging on/beneath citrus leaves and twigs. Thus, *T. nigra* adults caught through foliage beating in November might be those that are overwintering on citrus trees. The overwintering of *T. nigra* adults on citrus trees needs to be verified by performing foliage beating in winter, from December through February, in the citrus grove.

The present study documented that *T. nigra* adults were most abundant from early April to early May, followed by in October–November in citrus groves with no or low levels of pesticide application in Shizuoka City (Figs. 2, 3, 4). In conventional citrus groves in Shizuoka Prefecture, insecticides are sprayed mostly from May through September to guard against the yellow tea thrips, *Scirtothrips dorsalis* Hood Fig. 4 Seasonal changes in the mean number of captured Telsimia nigra adults per sticky trap per day in three organic commercial citrus groves in Sena [SE-1(o)], Shimono [SH(o)], and Sugiyama [SU(o)], and one conventional grove in Ihara [IH(c)], Shizuoka City in 2011-2012. Only the commercial groves where one or more T. nigra adults were trapped in the 2 years are shown. The mean numbers were calculated as described in the main text. Traps were replaced at an interval of ca. 14 days from April through November



(Thysanoptera: Thripidae), scale insects, and other insect pests (Masui et al. 2018). These facts suggest that the application of insecticides that are harmful to coccidophagous ladybirds should be avoided particularly in May to protect T. nigra populations in citrus groves. In fact, some T. nigra adults were trapped in April-May in the reduced-pesticide citrus grove [KO-2(r)] where no insecticide was sprayed in the 2 months (Fig. 2). This result suggests that no insecticide application in May could contribute to the protection of T. nigra in citrus groves. Organophosphates or neonicotinoids are often applied in May in citrus groves in Shizuoka Prefecture (Masui et al. 2018) mainly to control adults of beetle pests, including the smaller green flower chafer, Oxycetonia jucunda (Falderman) (Coleoptera: Scarabaeidae) and a sap beetle, Epuraea (Epuraea) domina Reitter (Coleoptera: Nitidulidae), both of which visit citrus flowers and injure very young fruits. However, these types of insecticides impose highly lethal effects on adults of the coccidophagous ladybirds, Chilocorus kuwanae Silvestri (Coleoptera: Coccinellidae) and P. hareja (Ozawa and Uchiyama 2016, 2019, respectively). Therefore, in May we should either refrain from using any insecticides or replace these detrimental insecticides with those that effectively reduce the beetle pest abundance on citrus trees but have no or low negative impacts on coccidophagous ladybirds, such as an insecticide containing Cyantraniliprole that induces no or low mortality of *C. kuwanae* and *P. hareja* adults (Ozawa and Uchiyama 2019), in order to conserve *T. nigra* populations in citrus groves. Protective effects of the application of insecticides such as Cyantraniliprole on *T. nigra* populations in citrus groves and its indirect influence on the abundance of diaspidid scales in the groves through the changes in the ladybird abundance need to be evaluated.

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