Exp Appl Acarol (2016) 70:189–204
DOI 10.1007/s10493-016-0073-y

Evaluation of site-specific tactics using bifenazate and *Neoseiulus californicus* for management of *Tetranychus urticae* (Acari: Tetranychidae) in strawberries

Ruohan Liu[1] · Teresia W. Nyoike[2] · Oscar E. Liburd[3]

Received: 12 February 2016 / Accepted: 12 July 2016 / Published online: 8 August 2016
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Abstract Greenhouse and field experiments were conducted to evaluate the effectiveness of site-specific tactics for management of the twospotted spider mite, *Tetranychus urticae* Koch, a major pest of greenhouse and field-grown strawberries (*Fragaria x ananassa* Duchesne). Two site-specific (spot) treatments, the miticide bifenazate (Acramite®) and the predatory mite *Neoseiulus californicus* McGregor, were compared with whole-plot treatments of bifenazate or *N. californicus* to determine whether *T. urticae* could be effectively managed in field-grown strawberry using only site-specific tactics. Additionally, the cost of site-specific tactics was compared with whole-plot treatments to determine the economic value of using site-specific management tactics for *T. urticae* in strawberries. In the greenhouse, all treatments equivalently reduced the number of *T. urticae* below control. In the field during the 2011–2012 season, more *T. urticae* eggs and motiles were in the whole-plot treatments of both *N. californicus* and bifenazate in the mid-season and late season, respectively, compared with the spot treatments. With the exception of site-specific *N. californicus* during the 2011–2012 field season, there were no differences in marketable yields between plots with site-specific treatments and whole-plot management. An economic analysis demonstrated a significant cost savings (75.3 %) with site-specific treatments of *N. californicus* compared with whole-plot application of *N. californicus*. Similarly, a 24.7 % reduction in cost was achieved in using site-specific bifenazate compared with whole-plot application of bifenazate. The findings indicate that site-specific treatments with *N. californicus* and bifenazate are competitive alternatives to whole-field application for *T. urticae* management in strawberries.

Keywords Strawberry · *Tetranychus urticae* · Site-specific management · Predatory mite

✉ Oscar E. Liburd
oeliburd@ufl.edu

[1] Monsanto, 800 N. Lindbergh Blvd, St. Louis, MO 63141, USA

[2] BASF Corporation, Research Triangle Park, Durham, NC 27709, USA

[3] Entomology and Nematology Department, University of Florida, 1881 Natural Area Drive, Steinmetz Hall, Gainesville, FL 32611, USA

Introduction

The twospotted spider mite, *Tetranychus urticae* Koch (Prostigmata: Tetranychidae), is one of the most destructive pests that affect field-grown and greenhouse strawberry (*Fragaria* spp.) (Walsh et al. 2002; Cloyd et al. 2006). Twospotted spider mites damage strawberry plants by feeding on the chlorophyll in the mesophyll area of the leaf, reducing the plants' ability to photosynthesize, which subsequently inhibits vegetative growth, flower development and yield (Sances et al. 1981; Nyoike and Liburd 2013). At present, the most commonly used tactic for management of *T. urticae* in strawberry in Florida, USA, is to apply miticides prophylactically on a weekly or biweekly basis to the entire field (Liburd et al. 2007). The standard miticide that is used for the management of *T. urticae* in Florida is bifenazate (Acramite®) due to its effectiveness and relatively low price; however, bifenazate can only be used 2× in the growing season. Therefore, growers apply abamectin (Temprano®), spiromesifen (Oberon®), bifenthrin (Brigade®) and hexythiazox (Savey®) when bifenazate cannot be used. These methods of control can be effective, but costly; with a high risk of other problems including pesticide resistance, secondary pest outbreaks and negative effects on non-target organisms such as pollinators (Bernardi et al. 2013).

Twospotted spider mite distribution patterns are typically clumped and these clumping patterns in the field are referred to as 'hot spots' (Flint 2012; Nyoike 2012). If management tactics for *T. urticae* are directed towards these hot spots in strawberry fields, it may be possible to reduce mite populations to levels achieved with whole field application. This, in turn, could significantly reduce the cost of mite management and miticide residues on the fruit.

Site-specific pest management uses spatial information on the pests' distribution in the field to apply management tactics to a much smaller area than that of the whole field (Plant 2001; Nyoike 2012). Management tactics are applied to areas where they are needed and in this case where pests' densities are above the economic threshold level. Site-specific management decisions rely heavily on the results from sampling programs. These strategies can benefit the grower by substantially reducing the cost of inputs, including pesticides, consequently decreasing the selection pressure for resistant gene flow (Dunley and Croft 1992).

Neoseiulus californicus McGregor (Mesostigmata: Phytoseiidae) has been recommended as an additional tool for inclusion in strawberry IPM programs in Florida (Schausberger and Walzer 2001; Rhodes et al. 2006; Rhodes and Liburd 2006; Fraulo and Liburd 2007) due to its efficacy on *T. urticae* with minimal disturbance on the arthropod community structure (Fraulo et al. 2008). In the absence of spider mite prey, *N. californicus* can survive on other diets including pollen (Walzer et al. 2001), which makes the predatory mite well adapted to the annual raised-bed system used for growing strawberry in Florida.

Bifenazate is a reduced-risk miticide with excellent activity against motile stages of *T. urticae* (Liburd et al. 2007), but is restricted to two sprays (per growing season) to avoid possible resistance development. It is registered for many fruit crops, including strawberry, apple and a selected number of stone fruits. Bifenazate is a hydrazine compound from carboxylic acid ester and works as a GABA (gamma-aminobutryric acid) synergist on *T. urticae* by inducing a conformational change, which modifies the magnitude of the GABA response (Hiragaki et al. 2012).

Our hypothesis is that we can achieve comparable levels of control for *T. urticae* in strawberry using site-specific management tactics with bifenazate or *N. californicus* compared with whole-field application. Furthermore, we hypothesize that there will be no reduction in marketable yield as a result of site-specific management tactics in experimental plots. The primary objective of this study was to evaluate the potential of using site-specific management tactics of bifenazate and *N. californicus* for *T. urticae* control in strawberry production as an alternative to whole-field management tactics. Secondly, to investigate the economic feasibility of using site-specific treatment within the strawberry system.

Materials and methods

Colony

A *T. urticae* colony was reared on bean (*Phaseolus* spp.) plants in a predatory mite-free laboratory. The colony was kept under a photoperiod of 14:10 L: D cycle at room temperature ($\sim$21 °C) between 70 and 80 % RH. Potted bean plants used for the colony were watered 2–3$\times$ per week and replaced every 4–5 days to allow mites to have access to new leaves for feeding.

Neoseiulus californicus was obtained from Koppert Biological Systems (Romulus, MI, USA) and applied to the respective treatments within 48 h of their arrival dates. A pretest was done by placing 30 mites in a Petri dish and observing them under a 10$\times$ compound microscope for 2–3 min to determine the percentage of *N. californicus* that were active prior to the application of the treatment.

Greenhouse experiment

The study was conducted in the Small Fruit and Vegetable IPM (SFVIPM) greenhouse at the University of Florida in Gainesville, FL, USA. Bare-root transplants of the cultivar 'Sweet Charlie' were transplanted into 1–l plastic pots (Nurseries Supplies, Chambersburg, PA, USA) filled with potting mix (Jungle Growth, Statham, GA, USA). Strawberry plants were managed following the standard growing procedures recommended for North-central Florida including fertilizer and fungicide applications, with the exception of insecticides and miticides (Whitaker et al. 2014). Plants were used when five mature trifoliate leaves were present.

One hundred and fifty uniform sized strawberry plants (with five trifoliates) were chosen randomly from plants grown in the greenhouse. Five plants were used for each treatment, with four plants surrounding the fifth (middle plant). In site-specific treatments, only the middle plants were infested with adult *T. urticae*, so as to create an artificial 'hot spot'. Each potted plant within a replicate was spaced $\sim$15 cm from the adjacent plant. In addition, each treatment was separated by a vertical oriented plastic sheet with a 0.5 m buffer zone. Adult *T. urticae* spider mites were released in all pots onto strawberry plants $\sim$2 weeks before treatments were applied to allow mites to reproduce on the plants and get acclimated. Clean leaf discs (12 mm in diameter) were used to transfer mites of the same age from bean plants (colony) onto strawberry plants and approximately 10 mites per disc per plant were used.

Plastic bottles (250 ml) with 5-mm openings in the covers were used to release mites in the greenhouse. Bottles were shaken gently directly over strawberry plants and *N. californicus* fell directly onto strawberry leaves.

The experiment was arranged in a completely randomized design with five treatments and six replicates. Treatments included: (1) *N. californicus* motiles (all stages except eggs) were released at 10:1 ratio (*T. urticae*: *N. californicus*) based on sampling results on all five plants (whole plot release); (2) site-specific *N. californicus* at 10:1 ratio, with motiles released on only the middle plant (hot spot); (3) bifenazate, applied using a 13 L (3.5 g) backpack sprayer fitted with XR Teejet nozzle (11004 VK) (spraysmarter.com). Bifenazate was applied at the manufacturer's recommended rate of 1.12 kg/ha on all (5) plants (whole plot release); (4) site-specific bifenazate, used at the recommended rate and applied using a backpack sprayer (only on the middle plant); and (5) untreated plants (control).

Sampling

Six trifoliate leaves were randomly taken from the middle canopies of each treatment once per week for 6 weeks, placed into quarter Ziploc storage bags (Glad®, Oakland, CA, USA) and brought back to the SFVIPM laboratory for analysis. The trifoliates were visually inspected under a dissecting microscope (10×) (Leica MZ12.5, McBain Instruments, Chatsworth, CA, USA). The number of *T. urticae* motiles (mites in all developmental stages except eggs) and eggs as well as motiles *N. californicus* were assessed and recorded.

Data analysis

Data from the different treatments were square-root transformed for motiles and eggs and analyzed using repeated measures ANOVA with mean separation using Least Significant Differences (LSD; $\alpha = 0.05$) to show treatment differences at different time points (PROC MIXED, SAS Institute 2007).

Field experiment

Two field experiments were conducted at the Plant Science Research and Education Unit (PSREU) at the University of Florida, in Citra, FL (82.17°W, 29.41°N). Strawberry cultivar 'Festival' was planted on 20 October 2011, and 16 October 2012. Festival was used in the field because this is the standard strawberry variety grown throughout the state of Florida. Plot size was 7.6 × 7.6 m^2 with a 6.1 m buffer-zone (bare soil without vegetation) (Rhodes and Liburd 2006). Each plot included six beds with 12 rows of strawberry plants. Before planting, the field was treated with a granulated fertilizer (10–10–10) (N–P–K) at a rate of 560 kg/ha and the soil was treated with fumigant 50:50 (Methyl-bromide: Chloropicrin) at a rate of 448 kg/ha. Beds were covered with black polyethylene mulch and plants were managed according to standard Florida strawberry practices (Whitaker et al. 2014) except that no insecticides were applied to the research plots and the only miticide used was bifenazate in certain treatments as part of the experiment.

Strawberry (bare root transplants) was planted manually by hand. The overhead irrigation was set to run for the first 10 days between 09:00 and 12:00 and 14:00 to 17:00 h after transplanting. After establishment, strawberry plants were irrigated by the drip tape on a timer 3× a day for a half hour at the rate of 8.7 L per 100 m (0.65 gal/100 ft.). Fertilizer was applied through the drip irrigation every week starting from one week after

transplanting, with fertilizer 6–0–8 (N–P–K) at the rate of 8.5 kg/ha until harvesting. When harvesting started, the nitrogen was increased to 12.4 kg/ha. Fungicides were applied weekly throughout the season in a rotation of several different products (Abound® [azoxystrobin], T-Methyl G-Pro® [Thiophanate-methyl], Cabrio® [Pyraclostrobin], Pristine® [pyraclostrobin + boscalid], Abound® [Azoxystrobin], Switch® [Cyprodinil + Fludioxonil], Bumper® [propiconazole], and Flint® [Trifloxystrobin]. None of the fungicides used are known to have any significant effect on *T. urticae* and *N. californicus* populations. Weeds were controlled by hoeing between rows and using an s-tine around the border of the plot. Strawberries were harvested once per week beginning in January and increased to 2× per week in February to reduce the opportunity for damage by birds and other vertebrates.

Experimental design was a randomized complete block with five treatments and four replicates (totaling 20 plots). Treatments included: (1) releases of motiles *N. californicus* over the entire plot (whole plot); (2) releases of motiles *N. californicus* only to 'hot spots' based on sampling (site-specific); (3) application of bifenazate to the entire plot; (4) site-specific application of bifenazate to 'hot spots' based on sampling; and (5) untreated control.

As in the greenhouse study, bifenazate was applied using a 13 L (3.5 g) backpack sprayer fitted with XR Teejet nozzle (11004 VK) (spraysmarter.com). Two applications were made (per spot/plot) during the entire season according to the manufacturer's recommendation at a rate of 1.12 kg/ha. *Neoseiulus californicus* was applied at a ratio 10:1 (*T. urticae*: *N. californicus*). For site-specific treatments, bifenazate and *N. californicus* were only applied when naturally-occurring *T. urticae* population exceeded the threshold (30 or more motiles per trifoliate) based on sampling results (Nyoike and Liburd 2013).

Similar to the greenhouse study, plastic bottles (250 ml) with 5 mm openings in the covers were used to release mites in the field. Bottles were shaken gently directly over strawberry plants and *N. californicus* fell directly onto strawberry leaves. During the 2011–2012 field season, *N. californicus* was released twice (treatment dates: 13 Jan. 2012 and 08 Feb. 2012) for whole-plot treatment, and bifenazate was sprayed 2× (treatment dates: 13 Jan. 2012 and 23 Feb. 2012) for the whole-plot treatment. Hot spots were chosen if four or more plants within a 0.092 m^2 were infested with *T. urticae* motiles or eggs and had exceeded the threshold of 30 or more motiles per trifoliate (Nyoike and Liburd 2013). The number of hot spots and plants sampled varied within replicate plots but an average of six plants per hot spot was sampled. Four hundred and thirty-two hot spots were treated with site-specific *N. californicus* treatments (treatment dates: 13 Jan. 2012, 29 Jan. 2012 and 21 Feb. 2012), and 416 hot spots were sprayed in site-specific bifenazate treatments (treatment dates: 13 Jan. 2012, 29 Jan. 2012 and 21 Feb. 2012).

In the 2012–2013 field season, *N. californicus* (whole-plot treatment) were only applied once (28 Jan. 2013) for the entire field season because the population of *T. urticae* in this treatment remained low and did not warrant a second application. The single application of *N. californicus* reduced the cost of this treatment during the second year. Bifenazate was applied twice for the whole-plot treatment (treatment dates: 28 Jan. 2013 and 08 Feb. 2013) according to the manufacturer's recommendation and at a rate of 1.12 kg/ha. Similar to 2011–2012, hot spots were chosen if 4 or more plants within a 0.092 m^2 were infested with *T. urticae* motiles or eggs and had exceeded the threshold of 30 or more motiles per trifoliate. Bifenazate was applied 2× per spot/plot. The location of hot spots within site-specific treatment varied and 568 hotspots were treated for bifenazate in site-specific treatment (treatment dates: 28 Jan. 2013, 08 Feb. 2013 and 24 Feb. 2013), and 536 were

treated with *N. californicus* for site-specific treatments (treatment dates: 28 Jan. 2013, 11 Feb. 2013 and 24 Feb. 2013).

Sampling

Each week, 10 trifoliates were taken randomly from each plot and brought back to the SFVIPM laboratory. Trifoliates were taken from the middle canopies of the plants (Croft and Coop 1998; Sances et al. 1981). The *T. urticae* and *N. californicus* motiles and eggs were counted under a dissecting microscope (10×) and recorded.

Yield

Fruits were harvested 2× per week. During the 2010–2011 field season, strawberries were harvested from 25 Dec. 2010 to 15 March, 2011. During the 2012–2013 field season, harvesting was conducted from 10 Jan. 2013 to 15 March 2013. Fruits from the two inner beds (four rows) were weighed, and marketable fruits were selected according to grading standards for strawberries in Florida (a single berry weighs 10 g or more without physical injury) and weighed separately (Nyoike and Liburd 2013).

Economic analysis

The quantity of pesticides applied and number of *N. californicus* released in individual plots and hotspots were calculated and recorded. Economic analysis was based on the assumption of 300 plants per plot. We measured the time it took to treat individual plots with bifenazate and *N. californicus*. We then calculated the cost of treating individual plots with bifenazate at the manufacturers recommended rate ($156 per ha), and *N. californicus* at the recommended curative rate ($1877 per ha) (Koppert Biological Systems, Howell, MI, USA). We used the average cost for unskilled farm labor in North-central Florida of $10 per hour to calculate the cost of applying bifenazate and releasing *N. californicus*.

Data analysis

The entire field season was divided into four categories based on when sampling/monitoring began, including: (1) pretreatment, (2) early season, (3) middle season, and (4) late season (Fraulo and Liburd 2007). The data for the number of *T. urticae* motiles and eggs were square-root transformed to meet the assumptions of normality and analyzed using the PROC MIXED procedure for repeated measures to evaluate the interaction of different treatments with time (PROC MIXED, SAS Institute 2007). LSD test was performed to separate means among treatments ($\alpha = 0.05$). Data for unmarketable and marketable yields of different treatments were square-root transformed to meet the assumptions of normality and analyzed using ANOVA followed by LSD test with mean separation. Calculations for the economic analysis were done using Excel 2010 (Microsoft, Redmond, WA, USA) and data were subjected to ANOVA followed by LSD to separate treatment means (SAS Institute 2007).

Results

Greenhouse experiment

The control had significantly higher *T. urticae* eggs ($F_{4,174} = 25.24$; $P < 0.0001$) and motiles ($F_{4,174} = 42.09$; $P < 0.0001$) compared with all other treatments (Fig. 1a, b). Each week (except week 4) *T. urticae* egg numbers were significantly higher in the control compared with all other treatments (Table 1). Similarly, each week significantly higher motile numbers were recorded in the control compared with all other treatments (Table 1). There were no differences among all the other treatments. Alternatively, both whole plot and site-specific treatments of *N. californicus* remained significantly ($P < 0.0001$) below the control and was not different from any bifenazate treatments.

Field experiments

2011–2012 field season

Prior to the application of treatments, there was no difference in *T. urticae* egg and motile numbers among the plots. Also, early in the season *T. urticae* egg and motile populations were low and no statistical difference was recorded among the treatments; for eggs ($F_{4,236} = 1.87$; $P = 0.94$) and motiles ($F_{4,236} = 1.94$; $P = 0.92$) (Fig. 2a, b). The *T. urticae* population reached its highest point during the mid-season (2 Feb 2012–27 Feb. 2012) and significant differences were observed among treatments. Whole-plot treatments of *N. californicus*, site-specific *N. californicus*, and bifenazate were not significantly

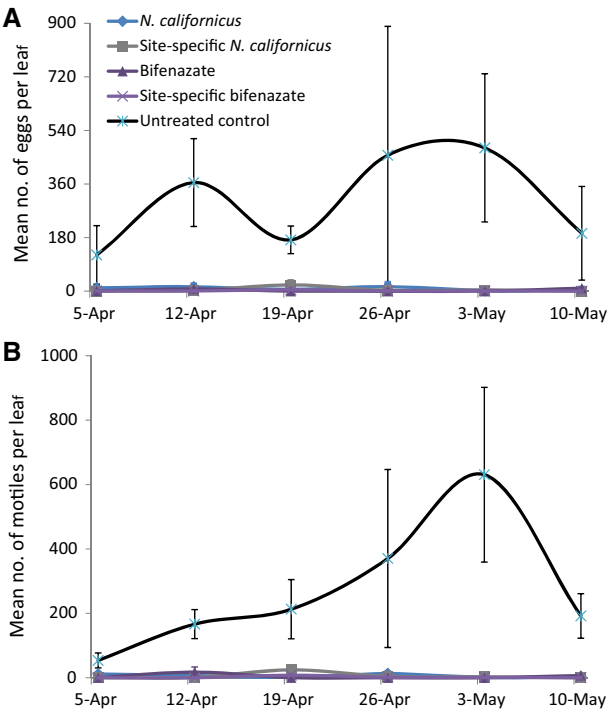

Fig. 1 Mean (±SEM) numbers of *Tetranychus urticae* motiles and eggs in greenhouse trials. **a** Weekly observations of *T. urticae* eggs per leaf for each treatment. **b** Weekly observations of *T. urticae* motiles per leaf for each treatment

Table 1 Weekly levels of significance for *Tetranychus urticae* when treatments were compared with the control in a greenhouse study

Week	*df*	*F*	*P*
1			
Egg	4,29	2.65	0.048
Motile	4,29	4.40	0.0053
2			
Egg	4,174	8.70	<0.0001
Motile	4,174	12.15	<0.0001
3			
Egg	4,348	7.90	<0.0001
Motile	4,348	13.10	0.0002
4			
Egg	4,522	1.70	0.17
Motile	4,522	4.72	0.0038
5			
Egg	4,696	8.45	<0.0001
Motile	4,696	14.80	<0.0001
6			
Egg	4,870	3.16	0.025
Motile	4,870	15.45	<0.0001

Data were square-root transformed to meet normality assumptions for motiles and eggs and analyzed using repeated measures ANOVA

different. However, site-specific bifenazate and the control had a significantly lower population of *T. urticae* eggs than whole-plot treatment of *N. californicus* ($F_{4,792} = 9.36$; $P < 0.0001$) (Fig. 2a). For motiles, site-specific *N. californicus* and the control had significantly fewer *T. urticae* than the whole-plot treatment of *N. californicus* ($F_{4,792} = 10.2$; $P < 0.0001$) (Fig. 2b). None of the other treatments were significantly different from each other. During the late season the number of *T. urticae* in plots treated with bifenazate increased significantly above all other treatments for eggs ($F_{4,592} = 13.29$; $P < 0.0001$) and motiles ($F_{4,592} = 11.62$; $P < 0.0001$), and there were no differences among the other treatments (Fig. 2a, b).

2012–2013 field season

There were no differences among plots prior to treatment; however, during early season site-specific *N. californicus* had significantly more *T. urticae* eggs and motiles than whole-plot treatments of *N. californicus*, bifenazate, and site-specific bifenazate; for eggs ($F_{4,356} = 2.96$; $P = 0.04$), and motiles ($F_{4,356} = 3.21$; $P = 0.023$) (Fig. 3a, b). The whole-plot treatment of *N. californicus* and site-specific bifenazate had similar amounts of *T. urticae* motiles and were significantly lower than site-specific *N. californicus* and the control ($F_{4,356} = 3.28$; $P = 0.0001$) (Fig. 3b). There was no difference between the site-specific treatment of *N. californicus* and the control for eggs and motiles (Fig. 3a, b).

During the middle season (28 Jan. 2013–23 Feb. 2013), all treatments had significantly fewer eggs ($F_{4,792} = 4.06$; $P = 0.0002$) and motiles ($F_{4,792} = 4.08$; $P = 0.0002$) than the control. Whole-plot treatments of *N. californicus* were significantly lower than site-specific *N. californicus* and site-specific bifenazate for eggs ($F_{4,536} = 4.13$; $P < 0.0001$) (Fig. 3a). There were no differences between site-specific *N. californicus*, whole plot application of bifenazate and site-specific bifenazate (Fig. 3a). For motiles, whole-plot treatments of *N.*

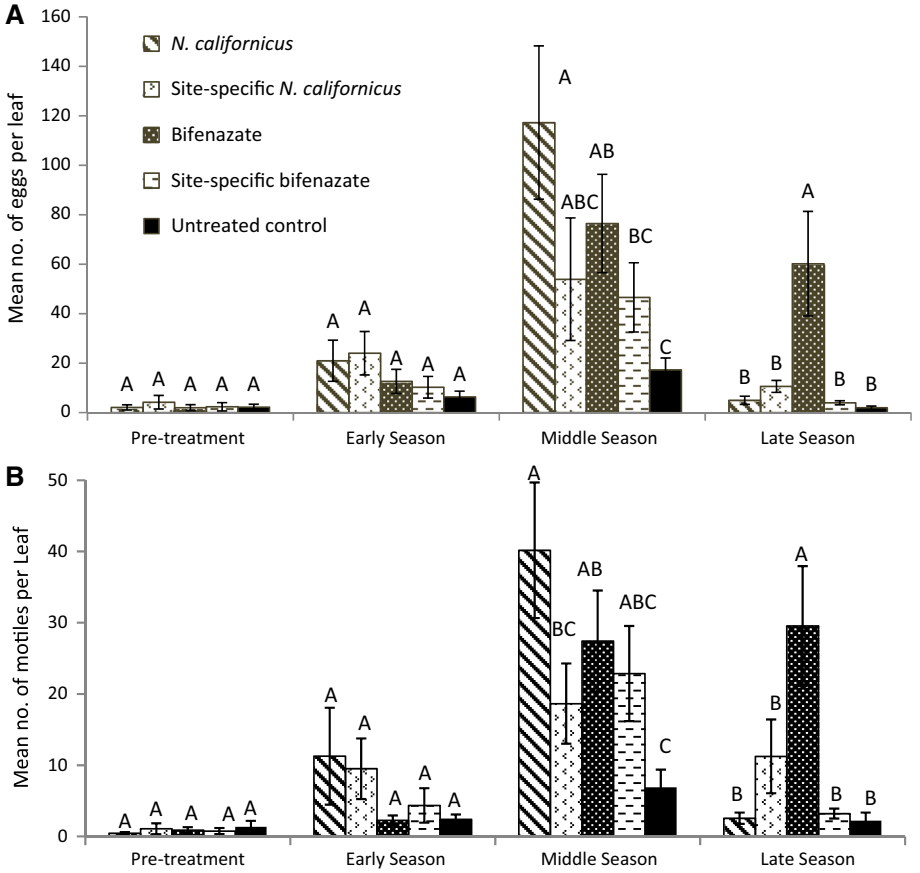

Fig. 2 Mean (±SEM) numbers of *Tetranychus urticae* **a** eggs and **b** motiles per leaf for four periods (pre-treatment, early-season, mid-season and late-season) of the 2011–2012 field season. Treatment means within a period capped by the *same letters* are not significantly different

californicus were significantly lower than site-specific *N. californicus* and the whole-plot treatment of bifenazate ($F_{4,536} = 4.11$; $P < 0.0001$) (Fig. 3b). There was no significant difference between site-specific *N. californicus* and whole-plot treatment of bifenazate (Fig. 3b). These trends continued during the late season where *T. urticae* motile population was low in all treatments except the control.

During the late season the number of *T. urticae* eggs and motiles remained low but the control had significantly higher numbers than all other treatments (eggs: $F_{4,592} = 20.53$; $P < 0.0001$, motiles: $F_{4,592} = 26.01$; $P < 0.0001$) (Fig. 3a, b). Site-specific *N. californicus* had significantly higher numbers of *T. urticae* eggs than whole-plot treatments of *N. californicus* and bifenazate ($F_{4,473} = 84.31$; $P < 0.0001$) (Fig. 3a). There were no differences in egg numbers between whole-plot treatments of *N. californicus* and bifenazate, and site-specific bifenazate. Similarly for motiles, whole-plot treatments of *N. californicus* and bifenazate were not significantly different to site-specific bifenazate (Fig. 3b). However, site-specific *N. californicus* had significantly more motiles than whole-plot treatments of *N. californicus*.

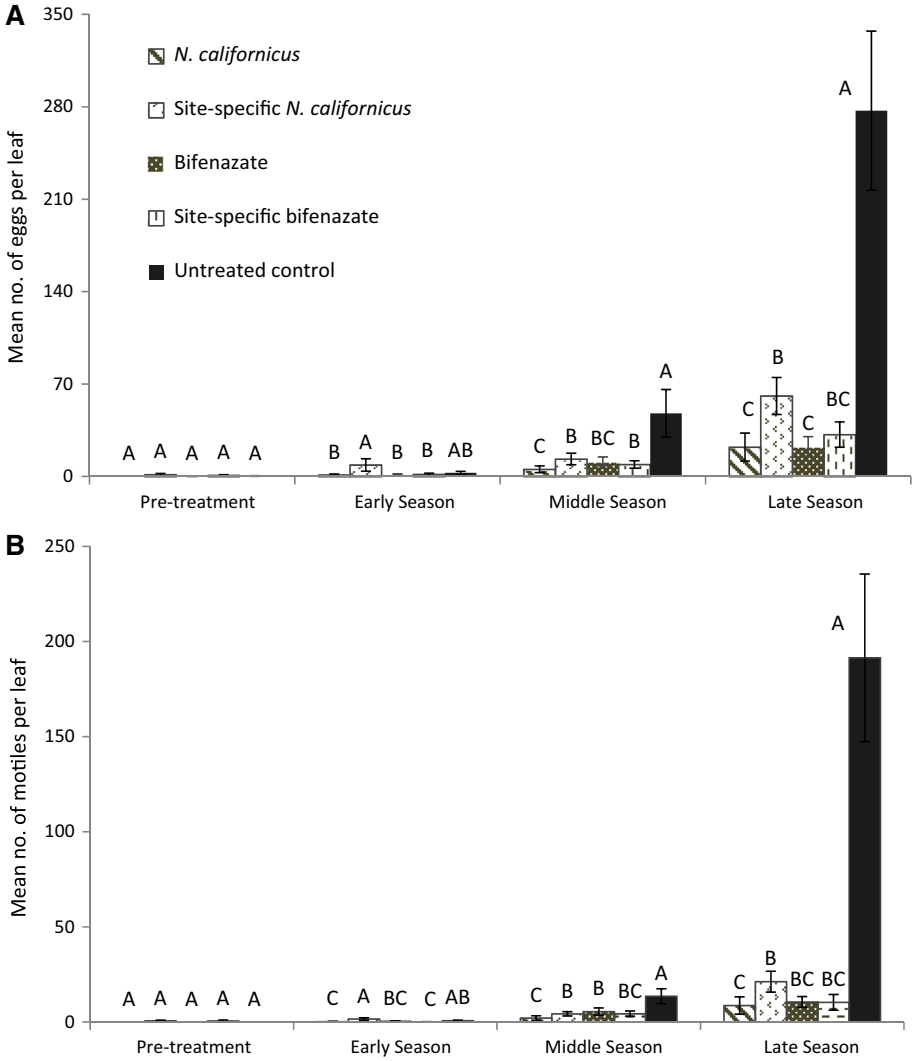

Fig. 3 Mean (±SEM) numbers of *Tetranychus urticae* **a** eggs and **b** motiles per leaflet for four periods (pre-treatment, early-season, mid-season and late-season) of the 2012–2013 field season. Treatment means within a period capped by the *same letters* are not significantly different

Yield

In the 2011–2012 field season, the average marketable yield for *N. californicus*, bifenazate (whole plot application), and site-specific bifenazate treatments were significantly higher than the site-specific *N. californicus* and control treatments ($F_{4,592} = 4.15$; $P = 0.0002$) (Table 2). The control treatment had significantly more unmarketable yield compared with all other treatments ($F_{4,592} = 2.85$; $P = 0.006$) (Table 2).

During 2012–2013, all of the treatments had significantly higher yields than control plots ($F_{4,352} = 3.87$; $P = 0.03$) (Table 2). There was no significant difference in

Table 2 Mean (±SE) total marketable and unmarketable strawberry yield (kg) from each treatment plot during the 2011–2012 and 2012–2013 field seasons

Tactic	2011–2012		2012–2013	
	Marketable	Unmarketable	Marketable	Unmarketable
Neoseiulus californicus	1326.4 ± 95.0 a	93.6 ± 11.4 b	1974.6 ± 114.5 a	71.3 ± 7.8 b
Site-specific *N. californicus*	1082.0 ± 108.5 b	91.3 ± 10.4 b	1792.0 ± 101.2 a	66.4 ± 11.9 b
Bifenazate	1419.6 ± 108.5 a	131.7 ± 21.6 b	1883.0 ± 114.9 a	77.5 ± 9.2 b
Site-specific bifenazate	1310.5 ± 81.8 a	87.4 ± 10.2 b	1720.6 ± 107.0 a	80.2 ± 9.7 b
Control	1042.8 ± 80.1 b	271.9 ± 36.3 a	1552.3 ± 104.0 b	125.4 ± 14.8 a

Means within a column followed by the same letter are not significantly different (LSD test; $P < 0.05$)

marketable yield between whole-plot treatments and site-specific treatments. Unmarketable yields in control plots were significantly higher than all other treatments ($F_{4,352} = 5.23$; $P < 0.0001$) (Table 2).

Economic analysis

The total cost of treating site-specific *N. californicus* during the 2011–2012 field season was ~$45.02. During the 2012–2013 field season the total cost of treating site-specific *N. californicus* increased to $53.77 (Table 3). Alternatively, the total cost of treating the whole plot with *N. californicus* during the 2011–2012 field season was $87.46; the price dropped significantly during the 2012–2013 field season to $43.78 because only one application of *N. californicus* was made (Table 3). Overall the cost of treating whole plots with *N. californicus* was significantly higher than site-specific *N. californicus* ($F_{3,11} = 3.67$; $P < 0.001$) (Fig. 4). There was no significant difference in the cost of

Table 3 Costs of different treatments in strawberry during the 2011–2012 and 2012–2013 field seasons in Citra, Florida

Tactic	Season	Actual area treated (ha)	Cost per ha (US$)	Cost of treatment (US$)	Total labor used (h)	Total labor costs (US$)	Total cost (US$)	Avg cost (US$)	% reduction
Site-specific *Neoseiulus californicus*	2011–2012	0.008	1877.40	15.02	3.00	30.00	45.02		
	2012–2013	0.01	1877.40	18.77	3.50	35.00	53.77	49.40	
N. californicus	2011–2012	0.046	1877.40	86.36	0.11	1.10	87.46		
	2012–2013	0.023	1877.40	43.18	0.06	0.60	43.78	65.62	75.28
Bifenazate	2011–2012	0.046	156.00	7.18	0.06	0.60	7.78		
	2012–2013	0.046	156.00	7.18	0.06	0.60	7.78	7.78	
Site-specific bifenazate	2011–2012	0.0039	156.00	0.61	0.1	1.00	1.61		
	2012–2013	0.0053	156.00	0.83	0.14	1.40	2.23	1.92	24.68

The average cost was derived from calculating the mean total cost from the 2011–2012 and the 2012–2013 field season; % reduction was calculated by comparing the cost of site-specific *N. californicus* versus whole-plot treatment with *N. californicus*, and site-specific bifenazate versus whole-plot treatment with bifenazate

treating the whole plot with bifenazate compared with site-specific application of bifenazate ($F_{3,11} = 1.2$; $P = 0.09$) (Fig. 4). The cost of treating the whole plot with bifenazate during the 2011–2012 and 2012–2013 field seasons was \$7.78 (Table 3). The cost of applying site-specific bifenazate in the 2011–2012 field season was \$1.61. However, the cost increased to \$2.23 during the 2012–2013 field season as more areas were treated (Table 3). The mean reduction in the total cost of using the site-specific *N. californicus* versus whole plot application of *N. californicus* is 75.3 % (Table 3). The mean reduction in using site-specific bifenazate versus whole plot application of bifenazate is 24.7 % (Table 3).

Discussion

Our results from greenhouse and field studies supported the hypothesis that similar levels of *T. urticae* control can be achieved irrespective of using site-specific management tactics of bifenazate or *N. californicus* and whole-field application of bifenazate or *N. californicus*. One of the reasons why site-specific management was as effective as whole field application is related to the ecology of *T. urticae* by forming hot spots in strawberry fields (Nyoike 2012). These hot spots allow for targeted application of treatments in concentrated areas where *T. urticae* population exceeds the threshold (Nyoike and Liburd 2013).

The results were less apparent for the site-specific *N. californicus* treatment especially during the 2012–2013 season when a higher population of *T. urticae* was recorded in site-specific plots compared with whole field application of *N. californicus*. Twospotted spider mite hot spots were widely distributed throughout the *N. californicus* site-specific treated plots during the 2012–2013 season (536 hot spots treated in the 2012–2013 season vs. 432 during the 2011–2012 season). Many of these hot spots were never treated because they did not reach the minimum threshold of 30 or more mites to warrant an application of *N. californicus*. These untreated hot spots may have caused an explosion in the *T. urticae* population resulting in higher numbers in site-specific plots. Fraulo and Liburd (2007) found that introducing *N. californicus* early in the season before *T. urticae* reaches the economic threshold level will prevent yield reduction, and essentially averting economic damage. Therefore, reducing the threshold so that more hot spots could be treated may remedy the slight difference between the site-specific *N. californicus* treatment and whole-plot treatment. Regardless, the findings from our site-specific bifenazate plots clearly demonstrate that there were no reductions in marketable yield. Previous research by

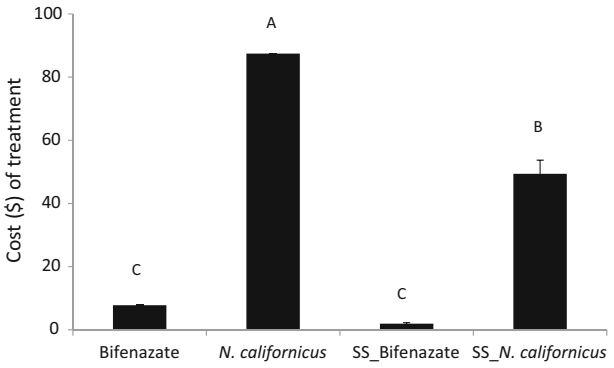

Fig. 4 Mean (±SEM) cost (USD) of using whole-plot treatments of bifenazate and *Neoseiulus californicus* versus site-specific treatments bifenazate and *N. californicus*

Nyoike and Liburd (2013) and Fraulo and Liburd (2007) indicated that *T. urticae* population that develops later in the season has less impact on marketable yields compared with those observed during early and mid-season.

Greenhouse experiment

With the exception of the control, there was no significant difference among any of the treatments evaluated (site-specific or whole plot), demonstrating that the same levels of control can be achieved using site-specific tactics or whole-plot treatments. Although *N. californicus* in site-specific treatments were only released in the center (hot spots) where *T. urticae* releases were initially done our sampling data indicated that they were able to move to adjacent plants where the *T. urticae* population had begun to colonize and reduce *T. urticae* densities below the control. Similarly, for the site-specific bifenazate treatment, only the middle plant 'hot spot' was sprayed where the initial release was done. The treatment of bifenazate to the center plant was sufficient to prevent the build-up of *T. urticae* populations; therefore, mite populations in this treatment remained below the control for the six weeks. Fraulo et al. (2008), Liburd et al. (2007), Rhodes et al. (2006), Rhodes and Liburd (2006), and Schausberger and Walzer (2001) have previously demonstrated the effectiveness of *N. californicus* and bifenazate for *T. urticae* management in strawberries. Bernardi et al. (2013) demonstrated the effectiveness of using predatory mites in combination with azadirachtin to manage *T. urticae* in strawberries in Brazil.

Field experiments

Consistent with the greenhouse results, both field seasons demonstrated the potential use of the site-specific management approach using *N. californicus* and bifenazate for control of *T. urticae* in strawberry. Except for the late season bifenazate (whole-plot treatment) where the mite population increased rapidly during the 2011–2012 field season, there were only few differences in *T. urticae* numbers between whole-plot treatments and site-specific treatments. *Neoseiulus californicus* was released 2× on 12 Jan. and 8 Feb. during the 2011–2012 field season. It appears that after the first release of *N. californicus*, adults did not become established because only a few were found just one week prior to the second release. The reason why *N. californicus* did not become established is unclear but could be related to environmental conditions in early January. Northern Florida does experience cool nights during mid-January that are not conducive for mite development (Nyoike and Liburd 2013). Regardless, since *N. californicus* did not become established during the first release the population of *T. urticae* (motiles and eggs) in the *N. californicus* whole plot application peaked and was significantly higher than the control during the mid-season. The reason why *T. urticae* population was higher in the whole plot compared with the control during mid-season is not certain but it is possible that after the first release of *N. californicus* these mite predators fed initially on the natural predators for *T. urticae* before they succumb to the mid-January temperatures. Predatory mite populations are known to feed on natural enemies of key pests resulting in a higher population of the pest compared to untreated blocks (Arévalo et al. 2009). After the second release, *N. californicus* became established and they effectively reduced the population of *T. urticae* in these plots by the late season.

Site-specific management approaches have been used previously to successfully map crop yields and reduce the application of pesticides and fertilizers in agriculture (Senay

et al. 2000; Bongiovanni and Lowenberg-DeBoer 2004). However, its use in insect pest management is a relatively new practice that is designed to treat only the areas where the pest is present in economically damaging numbers (as opposed to the entire field). The big advantage is that it can significantly reduce the cost of production including labor demands, pesticide residue on fruits, and selection pressure for resistance development among the mite population (Dunley and Croft 1992).

Bifenazate was applied twice during the field season (the maximum applications allowed) in the whole-plot treatment. The considerable increase in numbers of *T. urticae* in the late season of 2011–2012 suggests that bifenazate may have lost its effectiveness after a few weeks in the field. Heavy rainfall on 27 February (FAWN 2013), 3 days after the bifenazate application, may have reduced the residual time of the bifenazate. Alternatively, bifenazate is not systemic, so a lack of adequate coverage may have caused the treatment failure. In contrast, the site-specific bifenazate treatment was just as effective as the other treatments in controlling *T. urticae* populations during the 2011–2012 field season. Site-specific bifenazate application occurred 3× during the growing season. The treated areas were flagged so that they were not treated more than 2× per season. This may have provided better control. Also, it is much easier to ensure adequate coverage when treating a smaller area.

During the 2011–2012 field season plants in the control had numerically lower numbers of *T. urticae* compared with other treatments. The reason for the lower (numerical) mite numbers in the control is unclear. However, strawberry plants in one of the control replicate plots were smaller and did not look as healthy as the plants in other replicates. Further investigations of the strawberry roots and soil revealed that the nematode *Meloidogyne hapla*, which destroys the root hairs and interferes with nutrient uptake (Nyoike et al. 2012), was present in that replicate plot. This diagnosis was confirmed in the Nematode assay laboratory at the University of Florida. The nematode infested plants were stunted and may not have been able to support a high population of *T. urticae* due to smaller size and poor nutrient quality.

Throughout the 2012–2013 season, all treatments irrespective of site-specific or whole plot suppressed *T. urticae* numbers (motiles and eggs) below the control. Both site-specific and whole-plot treatments suppressed *T. urticae* numbers throughout the season. Two applications of bifenazate per plot/hot spot were sufficient to regulate the populations of *T. urticae* throughout the season.

Neoseiulus californicus adults became established for both whole plot and site-specific treatments when releases were made on 4 Jan. 2013, requiring no additional releases later in the season. During the late season, the population of *T. urticae* in the site-specific *N. californicus* treatment started to increase but a second release was not made because it was too close to the end of the season.

The population of *T. urticae* in treatment plots during the 2011–2012 field season was numerically higher than the 2012–2013 field season except on control plots during the late season. Environmental factors, especially temperature may have contributed to the differences in population between the field seasons. Nyoike and Liburd (2013) reported that the higher numbers of degree days experienced during warm temperatures supported a higher population of *T. urticae* in field-grown strawberries. Earlier, White and Liburd (2004) and Hart et al. (2002) reported higher reproduction rates for *T. urticae* with increased temperatures and reduced reproduction rates during cooler temperatures. During the 2011–2012 field season, the daily averages exceeded 22 °C (73 °F) in February (FAWN 2013) and the temperature remained high for the rest of the field season. In the 2012–2013 field season, the temperature fluctuated throughout the entire season. The

temperature was very stable during the early season with only 2 days below 0 °C, but remained cool during the middle and late season with 6 days below 0 °C. The colder temperatures throughout the middle and late 2012–2013 field season may have helped to suppress the *T. urticae* population.

Yield

With exception of the site-specific *N. californicus* treatment during the 2011–2012 season, marketable yields in site-specific plots were not different from whole-plot treatments, which further justifies the potential use of site-specific management tactics in strawberry fields. The control had the highest amount of unmarketable yields. The berries in these plots were smaller than average (10 g) and had the highest amount of physical injury. The control plots had numerically the lowest marketable yield but this was not different from site-specific treated plots of *N. californicus* during 2011–2012 field season.

Economic analysis

The results of the economic analysis demonstrated that the application of site-specific management tactics for bifenazate and *N. californicus* can reduce the cost of *T. urticae* management by as much as ~ 75 % for site-specific *N. californicus* treatments without any decrease in marketable yields. However, this will need further research to verify this theory since there was a reduction in yield during the first year of the study but not in the second year. Our data analysis indicated that this reduction and cost savings achieved from using site-specific management tactics was significant. Most strawberry growers would consider using site-specific pest management tactics as an alternative to whole farm pest management approaches if there are economic benefits in terms of yield or reduced costs from spaying less miticides on small areas versus whole plots, less demand on labor for tractor operators, and the environmental cost associated from spraying fewer pesticides in the environment.

Overall, our studies confirmed that site-specific management tactics are a potential alternative for *T. urticae* management in strawberries in north-central Florida. A successful program will require continuous field monitoring for *T. urticae* to determine where treatment applications are needed.

Acknowledgments We would like to thank the Small Fruit and Vegetable IPM lab at the University of Florida assistance with field sampling. We would also like to thank Drs. Janine Razze, Elena Rhodes and Robert McSorley for providing earlier reviews of the manuscript. We thank the crew at the University of Florida, Plant Science Research and Education Unit in Citra for their assistance with maintenance of research plots. The project was funded by the Specialty Crop State Block Grant Program; contract # 00080893.

References

Arévalo A, Fraulo A, Liburd OE (2009) Management of flower thrips in blueberries in Florida. Fla Entomol 92:14–17

Bernardi D, Bottom M, Silva de Cunha U, Bernardi O, Malausa T, Garcia MS, Nava DE (2013) Effects of azadirachtin on *Tetranychus urticae* (Acari: Tetranychidae) and its compatibility with predatory mites (Acari: Phytoseiidae) on strawberry. Pest Manag Sci 69:75–80

Bongiovanni R, Lowenberg-DeBoer J (2004) Precision agriculture and sustainability. Precis Agric 5:359–387

Cloyd RA, Galle CL, Keith SR (2006) Compatibility of three miticides with the predatory mites *Neoseiulus californicus* McGregor and *Phytoseiulus persimilis* Athias-Henriot (Acari: Phytoseiidae). HortScience 41:707–710

Croft BA, Coop LB (1998) Heat units, release rate, prey density, and plant age effects on dispersal by *Neoseiulus fallacis* (Acari: Phytoseiidae) after inoculation into strawberry. J Econ Entomol 91:94–100

Dunley JE, Croft BA (1992) Dispersal and gene flow of pesticide resistance traits in phytoseiid and tetranychid mites. Exp Appl Acarol 14:313–325

FAWN (2013) Florida automated weather network report generator. University of Florida, http://fawn.ifas.ufl.edu/data/reports/, Gainesville, Florida

Flint ML (2012) IPM in practice: principles and methods of integrated pest management, 2nd edn. University of California, Oakland

Fraulo AB, Liburd OE (2007) Biological control of twospotted spider mite, *Tetranychus urticae* Koch, with predatory mite, *Neoseiulus californicus*, in strawberries. Exp Appl Acarol 43:109–119

Fraulo AB, McSorley R, Liburd OE (2008) Effect of the biological control agent *Neoseiulus californicus* (Acari: Physeiidae) on arthropod community structure in north Florida strawberry fields. Fla Entomol 91:436–445

Hart AJ, Bale JS, Tullett AG, Worland MR, Walters KFA (2002) Effects of temperature on the establishment potential of the predatory mite Amblyseius californicus Mcgregor (Acari: Phytoseiidae) in the UK. J Insect Physiol 48:593–599

Hiragaki S, Kobayashi T, Ochiai N, Toshima K, Dekeyser MA, Matsuda K, Takeda M (2012) A novel action of highly specific acaricide; bifenazate as a synergist for a GABA-gated chloride channel of *Tetranychus urticae* [Acari: Tetranychidae]. Neurotoxicology 33:307–313

Liburd OE, White JC, Rhodes EM, Browdy AA (2007) The residual and direct effects of reduced-risk and conventional miticides on twospotted spider mites, Tetranychus urticae (Acari: Tetranychidae) and predatory mites (Acari: Phytoseiidae). Fla Entomol 90:249–257

Nyoike TW (2012) Incorporating economic thresholds and geospatial information technology (git) into pest management for twospotted spider mites in strawberries. A Ph.D. dissertation submitted to the graduate school in partial fulfillment of the requirements for the Doctor of Philosophy degree. August 2012, University of Florida, Gainesville, Florida

Nyoike TW, Liburd OE (2013) Effect of twospotted spider mite, *Tetranychus urticae* Koch (Acari: Tetranychidae) on marketable yields of field-grown strawberries in north-central Florida. J Econ Entomol 106:1757–1766

Nyoike TW, Mekete T, McSorley R, Weibezahl-Kakarigi E, Liburd OE (2012) Confirmation of *Meloidogyne hapla* on strawberry in Florida using molecular and morphological techniques. Nematropica 42:253–259

Plant RE (2001) Site-specific management: the application of information technology to crop production. Comput Electron Agric 30:9–29

Rhodes EM, Liburd OE (2006) Evaluation of *N. californicus* and Acramite® for control of twospotted spider mites in strawberries in north-central Florida. J Econ Entomol 99:1291–1298

Rhodes EM, Liburd OE, Kelts C, Rondon SI, Francis RR (2006) Comparison of single and combination treatments of *Phytoseiulus persimilis*, *Neoseiulus californicus,* and Acramite (bifenazate) for control of twospotted spider mites in strawberries. Exp Appl Acarol 39:213–225

Sances FV, Wyman JA, Ting IP, van Steenwyk RA, Oatman ER (1981) Spider mite interactions with photosynthesis, transpiration and productivity of strawberry. Environ Entomol 10:442–448

SAS Institute (2007) PROC user's manual, version, 3rd edn. SAS Institute, Cary

Schausberger P, Walzer A (2001) Combined versus single species release of predaceous mites: predator–predator interactions and pest suppression. Biol Control 20:269–278

Senay GB, Ward AD, Lyon JG, Fausey NR, Nokes SE (2000) Manipulation of high spatial resolution aircraft remote sensing data for use in site-specific farming. T ASAE 41:489–495

Walsh DB, Zalom FG, Shaw DV, Larson KD (2002) Yield reduction caused by twospotted spider mite feeding in an advanced-cycle strawberry breeding population. J Am Soc Hort Sci 127:230–237

Walzer A, Blumel S, Schausberger P (2001) Population dynamics of interacting predatory mites, *Phytoseiulus persimilis* and *Neoseiulus californicus*, held on detached bean leaves. Exp Appl Acarol 25:731–743

Whitaker VM, Boyd NS, Peres NA, Smith HA (2014) (revised) Strawberry production. Publication # HS736. EDIS-IFAS. University of Florida, Gainesville. FL 32611

White JC, Liburd OE (2004) Effects of soil moisture and temperature on reproduction and development of twospotted spider mite (Acari: Tetranychidae) in strawberries. J Econ Entomol 98:154–158