



# Effect of sweet alyssum on efficacy and sustainability of *Neoseiulus californicus* (Acari: Phytoseiidae) for biological control of spider mites in multiple cultivars of organic strawberries

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## ABSTRACT

The twospotted spider mite, *Tetranychus urticae* Koch (Acari: Tetranychidae) is one of the major pests of strawberry in the United States, causing losses of photosynthetic efficiency of up to 30% in strawberries. The predatory mite, *Neoseiulus californicus* (McGregor) (Acari: Phytoseiidae) is one of the primary biological control agents used against *T. urticae* in strawberries in Florida. We hypothesized that intercropping sweet alyssum (*Lobularia maritima* (L.)) would provide pollen as an alternative food source for *N. californicus*, allowing populations to survive longer when their preferred prey, spider mites, is not available. To test this hypothesis, three treatments including sweet alyssum in combination with *N. californicus* mites, *N. californicus* mites (only), and a miticide regimen were evaluated in a field study. We found that *N. californicus* mites consistently controlled the spider mite population when populations were high compared to a miticide regimen (control). Furthermore, there was no evidence that the presence of sweet alyssum affected the efficacy or sustainability of *N. californicus*. The implications of these results are discussed for organic growers who favor banker planting.

## 1. Introduction

The twospotted spider mite, *Tetranychus urticae* Koch (Acari: Tetranychidae), is one of the major pests of strawberry in the United States. This mite has a host range of over 1100 plant species and can cause severe yield losses at high population levels (Migeon and Dorkeld 2020; Sances et al., 1979). Infestations usually start from infested host plants surrounding the field or from populations that hitchhike from greenhouse distributors on the strawberry plugs. Twospotted spider mite causes damage to strawberries by feeding on the underside of the leaves, destroying the mesophyll of the plant, and reducing photosynthetic efficiency (Nyoike and Liburd 2013; Sances et al., 1979, 1982).

Conventional strawberry production primarily uses various synthetic miticides to keep spider mite populations below the economic threshold (Liburd et al., 2007). Organic farmers have a limited range of tactics that can be employed for mite control compared to conventional growers. Some examples of the available organic-labeled compounds include Aramite (cinnamon and clove oil, ExcelAg, Miami, FL) and Cosavet DF (sulfur, Sulphur Mills Ltd., Mumbai, India). Horticultural oils like Aramite clog the spiracles of mites, which causes the mites to suffocate. Sulfur abrades the mites' exoskeletons, causing the mites to dry out.

Both groups of products require the mites to come into direct contact with them.

An alternative option to miticides is to use a biological control agent such as the predatory mite *Neoseiulus californicus* (McGregor) (Acari: Phytoseiidae). *Neoseiulus californicus* has characteristics of both a type II and type III predator. They prefer spider mites and can also feed on other small arthropods and pollen when spider mite populations are inadequate (McMurtry et al., 2013). Because *N. californicus* can feed on pollen, banker plants (plants that provide a food source to beneficial arthropods in an agricultural system) providing a suitable source of pollen could be used in a biological control program using this predatory mite.

Sweet alyssum, *Lobularia maritima* (L.) (Brassicales: Brassicaceae), is commonly used as an insectary plant in greenhouse systems and annual crops in milder climates, as their continuous production of flowers provides a supply of pollen and nectar to many beneficial species, most notably *Orius* species (Hemiptera: Anthocoridae) and syrphid flies, important predators of thrips, whiteflies, and aphids (Amorós-Jiménez et al., 2014; Pumariño and Alomar, 2012). Ragusa et al. (2009) found that sweet alyssum also has the potential to sustain the predatory mite *N. californicus* for a short period of time. While the *N. californicus* mites were not able to reproduce by feeding on sweet alyssum pollen alone,

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females were able to survive on the pollen for the full duration of the experiment (~10 days). The primary objective of the research detailed in this manuscript was to assess the effect of sweet alyssum on augmenting *N. californicus* performance in organic strawberry cultivars. This project studied the effect of sweet alyssum (*L. maritima* (L.)) as a banker plant in the strawberry production system and evaluated the efficacy of sweet alyssum in providing an alternative habitat for the predatory mite *N. californicus* and therefore increasing biological control.

## 2. Materials and methods

### 2.1. Study site

Two seasons of field experiments were conducted in 2019–2020 and 2020–2021 at the University of Florida's Plant Science Research and Education Unit (PSREU) (29°24'28.0"N 82°08'50.1"W), near Citra, Florida. In North-Florida, the strawberry season starts by planting strawberry plugs in mid-October, with regular harvesting beginning in mid to late December until early April. A 0.1-ha experimental area was designated for both years of the experiment.

### 2.2. Plant culture

Organic sweet alyssum (*Lobularia maritima*) seeds (Johnny's Selected Seeds, Winslow, ME) were planted in a greenhouse in 72-plug trays filled with organic soil (Miracle-Gro Lawn Products Inc., Marysville, OH) and grown for three weeks before transplanting into the field. Strawberry transplants from the cultivars 'Strawberry Festival', 'Sweet Sensation', 'Florida Brilliance', and 'Florida Beauty' were acquired from Production Lareault Inc., Lavaltrie, Quebec.

Experimental plots were 7 × 7.6 m planted with 3 double row beds of strawberries spaced at 35 cm between plants along the row. Plots were spaced at 6 m apart to provide a buffer zone between them. Each bed in the plot contained one of the three cultivars: 'Sweet Sensation', 'Florida Brilliance', and 'Strawberry Festival'. For the 2020-21 growing season, the cultivar 'Strawberry Festival' was replaced with 'Florida Beauty' strawberries, as 'Strawberry Festival' was not available from the distributor that year. Soil type is sandy loam (75%–25% sand to loam, respectively) with low organic matter.

### 2.3. Experimental design

Experimental design was a randomized complete block, with four replicates of three different treatments. Treatments included: 1) release of *N. californicus* predatory mites in plots with three 0.6 m wide clusters of 10 sweet alyssum (*Lobularia maritima*) plugs planted evenly along each bed (1 cluster at each end and a third cluster in the middle of each bed) of the plot, 2) release of *N. californicus* predatory mites, but without the addition of sweet alyssum clusters, and 3) miticide application, detailed below, as a grower's standard control. Sweet alyssum clusters were planted in the miticide application treatment (treatment 3) plots in the 2019/2020 season but not in the 2020/2021 season. The sweet alyssum was planted in the miticide treatment plots in the 2019/2020 season for consistency. However, there were production issues in the greenhouse for the 2020/2021 season, so sweet alyssum was not planted in the miticide treatment plots. Sweet alyssum plants were transplanted into the field plots 2 weeks after the strawberry plants were planted.

### 2.4. Predatory mites

*Neoseiulus californicus* mites were purchased in 500-ml bottle shaker formulation (Koppert Biological Systems, Howell, MI) containing 25,000 *N. californicus* mites and vermiculite as the carrier. A sample (5-ml) from the bottle was placed under the dissecting microscope and the

predatory mites were observed to confirm that they were actively moving prior to release. *Neoseiulus californicus* was released in the field on the day of arrival by scattering the bran on top of the strawberry foliage. Each bed (~7.6-m<sup>2</sup>) was treated with approximately 15-ml of bran. Approximately 100 *N. californicus* motiles were released per square meter. The release rate used was based on the rate recommended for introductions (25–125 mites/m<sup>2</sup> in the case of Koppert Biological Systems).

### 2.5. Miticide regimen

The miticide regimen consisted of Aramite (cinnamon and clove oil, ExcelAg, Miami, FL) and Cosavet DF (sulfur, Sulfur Mills Ltd., Mumbai, India) in rotation applied at the recommended rates of 3.51 L/ha and 11.21 kg/ha, respectively. Applications occurred every 10 days and the products were rotated in order to prevent the development of resistance. Miticides were applied 6 times total (3 applications of each miticide). Miticide and predatory mite treatments began once the spider mite population on the strawberries reached an average of 20 motiles (mites of any life stage other than eggs) per trifoliolate leaf to use these treatments as a preventative rather than curative measure (Nyoike and Liburd 2013).

### 2.6. Sampling

The sampling of *T. urticae* and *N. californicus* was conducted between December and March for both field-seasons. Each week, four mature trifoliolate leaves were sampled from each cultivar in each plot. Leaves were cut from plants and placed into press and seal bags and transported to the laboratory under cool conditions. Leaves were examined for *T. urticae* and *N. californicus* under a dissecting microscope (Leica MZ 12 5, Leica Microsystems, Houston, TX). From these leaf samples, motiles, as defined above, and eggs of both mite species were counted and recorded.

### 2.7. Yield

Strawberries were harvested twice per week from all plants, separated by treatment/plot and cultivar. Harvesting was conducted from when the first sign of ripe berries appeared in the field. Ripe fruit was classified as when more than 85% of the fruit was red. Harvesting continued until the plants stopped producing marketable fruit in March/April of each year.

Strawberry yield was categorized into marketable and unmarketable yield. Marketable fruits consisted of berries weighing more than 10 g and without visible damage. Berries weighing less than 10 g, or showing signs of pest, weather, or disease damage were considered unmarketable and were discarded. The marketable fruits were weighed, and the total weight of marketable strawberries from each cultivar in each treatment plot was recorded.

### 2.8. Statistical analysis

Spider mite population data from each season was analyzed using a generalized linear model through the statistics program R 4.3.1. The model included the treatment, cultivar, and date collected as fixed effects. A quasipoisson distribution was used as a correction for overdispersion to account for the clustered spider mite populations. Statistical difference was determined using an ANOVA and post-hoc comparisons among treatments were assessed using the emmeans package and function. Results were considered significant when  $P \leq 0.05$ .

For analysis of the predatory mite populations, both motile and egg numbers were too low to analyze weekly data. Therefore *N. californicus* motiles and *N. californicus* eggs totaled over each season were analyzed using a generalized linear model with treatment and cultivar as fixed

effects in a two-way ANOVA. Tukey’s HSD was used for post-hoc comparisons. Results were considered significant when  $P \leq 0.05$ .

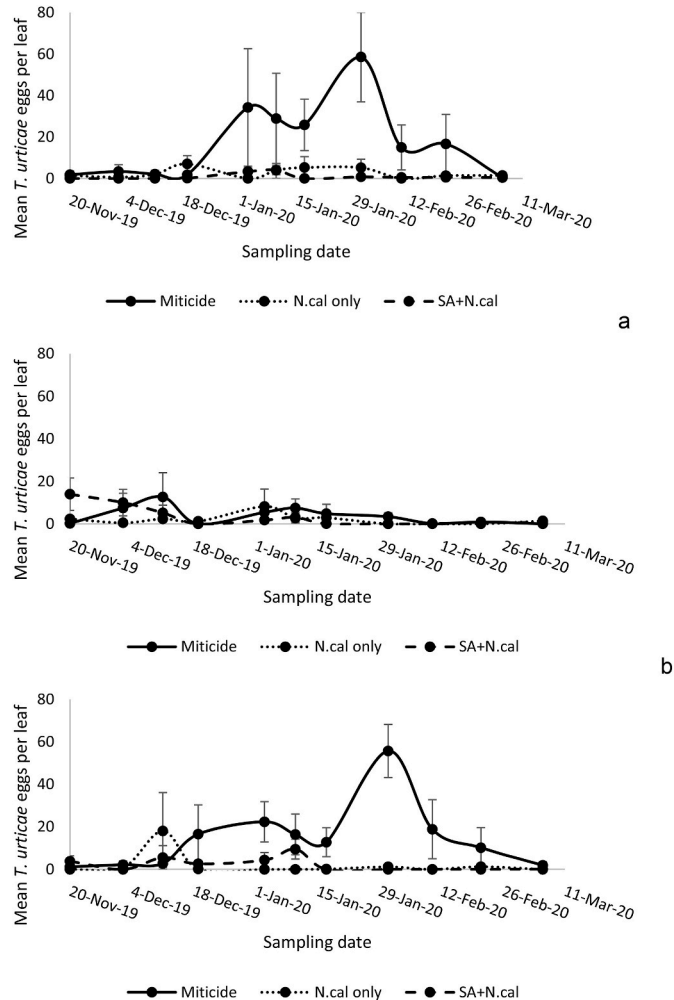
For yield from each season, a linear model was run, with the same fixed effects as the spider mite models, but with the block as a random effect. Statistical difference was determined using an ANOVA and post-hoc comparisons among treatments were assessed using the emmeans package and function. Results were considered significant when  $P \leq 0.05$ .

### 3. Results

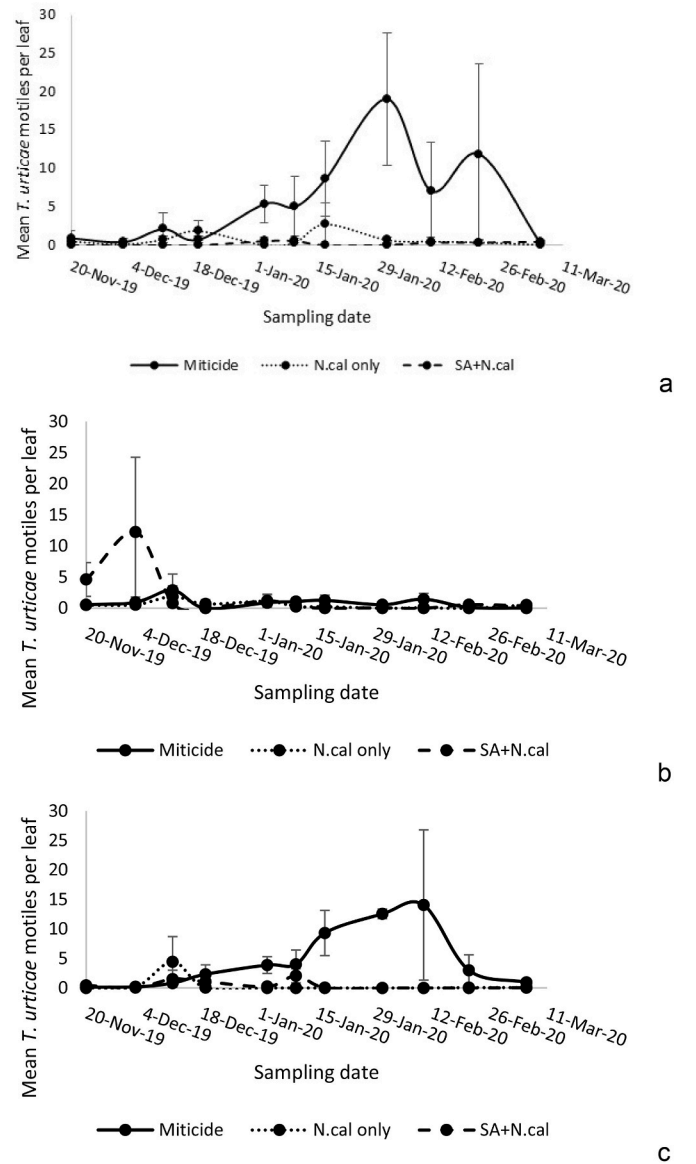
#### 3.1. 2019–2020 season

##### 3.1.1. Spider mites

Spider mite eggs and motiles were observed in all treatments from the beginning of data recording in mid-November (Figs. 1 and 2). Spider mite populations peaked approximately around the beginning of February in the ‘Florida Brilliance’ and ‘Sweet Sensation’ cultivars. The mite population in the ‘Strawberry Festival’ cultivar stayed consistently low throughout the entire season. The interaction between date and treatment was significant for both eggs ( $\chi^2 = 8.99$ ,  $df = 1$ ,  $P = 0.003$ ) and motiles ( $\chi^2 = 16.68$ ,  $df = 1$ ,  $P < 0.0001$ ). The interaction between



**Fig. 1.** Mean *T. urticae* egg numbers per leaf from each treatment in the a) ‘Brilliance’, b) ‘Festival’, and c) ‘Sensation’ cultivars from the 2019–2020 field season. Error bars represent SEMs. (Miticide = miticide regimen treatment, N. cal only = *N. californicus* only, and SA+N.cal = *N. californicus* and sweet alyssum).



**Fig. 2.** Mean *T. urticae* motile numbers per leaf from each treatment in the a) ‘Brilliance’, b) ‘Festival’, and c) ‘Sensation’ cultivars from the 2019–2020 field season. Error bars represent SEMs. (Miticide = miticide regimen treatment, N. cal only = *N. californicus* only, and SA+N.cal = *N. californicus* and sweet alyssum).

date and cultivar, however, was not significant for both eggs ( $\chi^2 = 2.79$ ,  $df = 1$ ,  $P = 0.09$ ) and motiles ( $\chi^2 = 2.03$ ,  $df = 1$ ,  $P = 0.15$ ). This was also true for the interaction between week, treatment, and cultivar for both eggs ( $\chi^2 = 0.63$ ,  $df = 1$ ,  $P = 0.43$ ) and motiles ( $\chi^2 = 2.11$ ,  $df = 1$ ,  $P = 0.15$ ).

Overall, there were significantly higher numbers of spider mite eggs (mean  $\pm$  SEM) in the miticide treatment ( $11.9 \pm 3.0$ ) compared with the *N. californicus* alone ( $2.2 \pm 0.7$ ) and *N. californicus* + sweet alyssum ( $2.2 \pm 0.6$ ) treatments ( $\chi^2 = 55.32$ ,  $df = 1$ ,  $P < 0.0001$ ). There were also significantly higher numbers of spider mite eggs (mean  $\pm$  SEM) in the ‘Brilliance’ ( $6.8 \pm 3.0$ ) and ‘Sensation’ cultivars ( $6.3 \pm 2.2$ ) compared with ‘Festival’ ( $3.0 \pm 0.7$ ) treatments ( $\chi^2 = 8.17$ ,  $df = 1$ ,  $P = 0.004$ ). The treatment\*cultivar interaction was also significant ( $\chi^2 = 6.87$ ,  $df = 1$ ,  $P = 0.009$ ). Looking at treatment differences within cultivars, spider mite eggs were significantly higher in the miticide-treated plots compared to both predatory mite treatments in all cultivars except for ‘Strawberry Festival’, where it was not significantly different from the plot treated

with sweet alyssum and *N. californicus* (Table 1). Comparing the two plots treated with *N. californicus* (with and without sweet alyssum), there was no difference in the amount of spider mite eggs in any cultivar (Table 1).

Like eggs, overall, there were significantly higher numbers of spider mite motiles (mean ± SEM) in the miticide treatment (3.7 ± 1.1) compared with the *N. californicus* alone (0.6 ± 0.1) and *N. californicus* + sweet alyssum (0.9 ± 0.4) treatments ( $\chi^2 = 42.77$ , df = 1,  $P < 0.0001$ ). There were also significantly higher numbers of spider mite eggs (mean ± SEM) in the ‘Brilliance’ (2.2 ± 1.0) and ‘Sensation’ cultivars (1.9 ± 0.8) compared with ‘Festival’ (1.1 ± 0.4) treatments ( $\chi^2 = 5.84$ , df = 1,  $P = 0.004$ ). The treatment\*cultivar interaction was also significant ( $\chi^2 = 12.94$ , df = 1,  $P = 0.0003$ ). Looking at treatment differences within cultivars, spider mite motiles were significantly higher in the miticide-treated plots compared to both predatory mite treatments in all cultivars except for ‘Strawberry Festival’, where it was not significantly different from the other treatments (Table 2). Comparing the two plots treated with *N. californicus*, there was no significant difference in spider mite motiles in any cultivar in the sweet alyssum plots compared to those without sweet alyssum (Table 2).

### 3.1.2. Predatory mites

For predatory mite eggs, there were higher numbers of eggs in the miticide treatment compared with the *N. californicus* + sweet alyssum treatment ( $F = 3.90$ ; df = 2, 27;  $P = 0.033$ ). There was an average ± SEM of 0.8 ± 0.4, 0.6 ± 0.2, and 0.4 ± 0.3 eggs in the miticide, *N. californicus* only, and *N. californicus* + sweet alyssum treatments respectively. Higher numbers of eggs were recorded from ‘Sensation’ compared with both ‘Brilliance’ and ‘Festival’ ( $F = 4.89$ ; df = 2, 27;  $P = 0.015$ ). There was an average ± SEM of 0.4 ± 0.2, 0.3 ± 0.1, and 1.1 ± 0.3 eggs in the ‘Brilliance’, ‘Festival’, and ‘Sensation’ cultivars respectively. There was no interaction between treatment and cultivar ( $F = 1.20$ ; df = 4, 27;  $P = 0.33$ ).

For predatory mite motiles, there were no differences among treatments ( $F = 0.73$ ; df = 2, 27;  $P = 0.49$ ). There was an average ± SEM of 0.8 ± 0.2, 0.6 ± 0.2, and 0.5 ± 0.2 motiles in the miticide, *N. californicus* only, and *N. californicus* + sweet alyssum treatments respectively. Higher numbers of motiles were recorded from ‘Sensation’ compared with both ‘Brilliance’ and ‘Festival’ ( $F = 4.06$ ; df = 2, 27;  $P = 0.029$ ). There was an average ± SEM of 0.5 ± 0.2, 0.4 ± 0.1, and 1.0 ± 0.2 motiles in the ‘Brilliance’, ‘Festival’, and ‘Sensation’ cultivars respectively. There was no interaction between treatment and cultivar ( $F = 0.17$ ; df = 4, 27;  $P = 0.95$ ).

### 3.1.3. Yield

Yield during the 2019–2020 field season peaked in early February, but there was also a small peak in production in early December. There was no significant difference between the yields from any treatment in

**Table 1**

Estimated marginal means and standard error (SE) of *T. urticae* eggs collected during the 2019–2020 field-season across cultivars and treatments. A (\*) indicates a significant difference between the two treatments at  $P \leq 0.05$ . Sweet alyssum = SA, *N. californicus* = N.cal.

Comparison	Estimate	SE	df	Z-ratio	P-value
<b>Brilliance</b>					
Miticide vs. N.cal only *	14.571	3.167	Inf	4.601	<0.001
Miticide vs. (SA+N.cal) *	16.275	3.027	Inf	5.377	<0.001
N.cal only vs. (SA+N.cal)	1.704	1.319	Inf	1.292	0.4
<b>Festival</b>					
Miticide vs. N.cal only	2.022	1.699	Inf	1.19	0.46
Miticide vs. (SA+N.cal)	0.506	1.913	Inf	0.264	0.96
N.cal only vs. (SA+N.cal)	-1.517	1.621	Inf	-0.935	0.62
<b>Sensation</b>					
Miticide vs. N.cal only *	12.662	2.908	Inf	4.355	<0.001
Miticide vs. (SA+N.cal) *	12.281	2.941	Inf	4.176	<0.001
N.cal only vs. (SA+N.cal)	-0.381	1.484	Inf	-0.257	0.96

**Table 2**

Estimated marginal means and standard error of *T. urticae* motiles collected during the 2019–2020 field season across cultivars and treatments. A (\*) indicates a significant difference between the two treatments at  $P \leq 0.05$ . Sweet alyssum = SA, *N. californicus* = N.cal.

Comparison	estimate	SE	df	Z-ratio	P-value
<b>Brilliance</b>					
Miticide vs. N.cal only *	4.856	1.164	Inf	4.173	<0.001
Miticide vs. (SA+N.cal) *	5.392	1.113	Inf	4.842	<0.001
N.cal only vs. (SA+N.cal)	0.536	0.441	Inf	1.215	0.44
<b>Festival</b>					
Miticide vs. N.cal only	0.361	0.545	Inf	0.662	0.79
Miticide vs. (SA+N.cal)	-0.965	0.763	Inf	-1.266	0.41
N.cal only vs. (SA+N.cal)	-1.327	0.71	Inf	-1.868	0.15
<b>Sensation</b>					
Miticide vs. N.cal only *	4.195	1.043	Inf	4.022	<0.001
Miticide vs. (SA+N.cal) *	4.139	1.049	Inf	3.946	<0.001
N.cal only vs. (SA+N.cal)	-0.056	0.445	Inf	-0.127	>0.99

any cultivar during this field season.

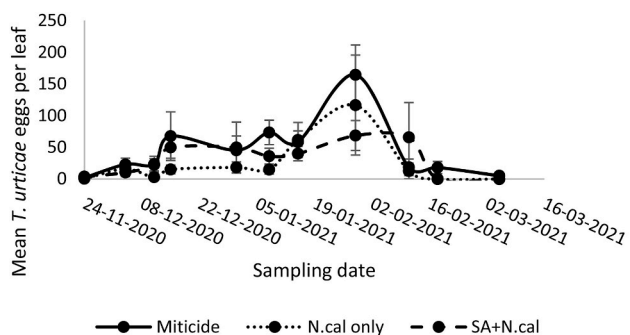
### 3.2. 2020–2021 season

#### 3.2.1. Spider mites

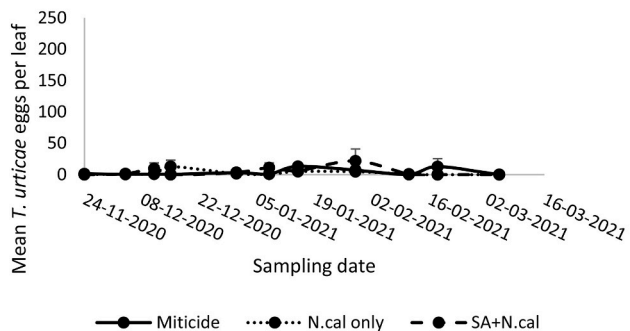
Spider mite eggs and motiles were observed in all treatments from the beginning of data recording in mid-November (Figs. 3 and 4). Based on the observations in the miticide-treated plots, spider mite populations peaked approximately around the beginning of February in the ‘Florida Beauty’ cultivar. The mite population in the ‘Florida Brilliance’ and ‘Sweet Sensation’ cultivars stayed consistently low throughout the entire season, with a small peak recorded at the end of January. The interaction between date and treatment was not significant for both eggs ( $\chi^2 = 0.0004$ , df = 1,  $P = 0.95$ ) and motiles ( $\chi^2 = 0.001$ , df = 1,  $P = 0.97$ ). The interaction between date and cultivar, was also not significant for both eggs ( $\chi^2 = 0.04$ , df = 1,  $P = 0.83$ ) and motiles ( $\chi^2 = 0.16$ , df = 1,  $P = 0.69$ ). This was also true for the interaction between week, treatment, and cultivar for both eggs ( $\chi^2 = 0.005$ , df = 1,  $P = 0.95$ ) and motiles ( $\chi^2 = 0.25$ , df = 1,  $P = 0.62$ ).

For the 2020–2021 field season, overall, there were no differences in spider mite eggs among treatments ( $\chi^2 = 1.41$ , df = 1,  $P = 0.24$ ). There were a mean ± SEM of 18.8 ± 6.6, 10.4 ± 4.0, and 14.7 ± 5.1 spider mite eggs in the miticide, *N. californicus* alone, and *N. californicus* + sweet alyssum treatments respectively. Higher numbers of eggs were recorded from the ‘Beauty’ (33.4 ± 6.2) cultivar compared with the ‘Brilliance’ (3.9 ± 1.0) and ‘Sensation’ (6.8 ± 1.4) cultivars ( $\chi^2 = 81.99$ , df = 1,  $P < 0.0001$ ). The treatment\*cultivar interaction was not significant ( $\chi^2 = 1.33$ , df = 1,  $P = 0.25$ ). Looking at treatment differences within cultivars, the only cultivar where a significant difference in spider mite eggs occurred was in the ‘Florida Beauty’ cultivar, where the treatment with only *N. californicus* mites had a significant reduction in spider mite egg numbers compared with the miticide control treatment (Table 3). Between the two *N. californicus* treatments (with and without sweet alyssum), there was no significant difference in the control of spider mite eggs in any cultivar (Table 3).

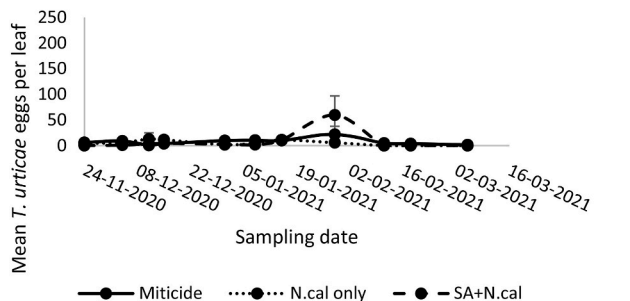
Like the egg data, overall, there were no differences in spider mite motiles among treatments ( $\chi^2 = 1.43$ , df = 1,  $P = 0.23$ ). There were a mean ± SEM of 10.4 ± 3.8, 5.6 ± 2.3, and 7.9 ± 2.8 spider mite eggs in the miticide, *N. californicus* alone, and *N. californicus* + sweet alyssum treatments respectively. Higher numbers of eggs were recorded from the ‘Beauty’ (18.3 ± 3.6) cultivar compared with the ‘Brilliance’ (1.7 ± 0.5) and ‘Sensation’ (3.9 ± 1.1) cultivars ( $\chi^2 = 70.74$ , df = 1,  $P < 0.0001$ ). The treatment\*cultivar interaction was also significant ( $\chi^2 = 3.72$ , df = 1,  $P = 0.05$ ). Looking at treatment differences within cultivars, the only cultivar where a significant difference in spider mite motiles occurred was in the ‘Florida Beauty’ cultivar, where both *N. californicus* treatments had a significant reduction in spider mite motile numbers compared with the miticide control treatment (Table 4). Between the



a



b



c

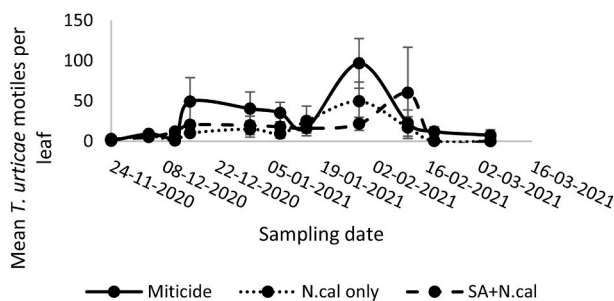
**Fig. 3.** Mean *T. urticae* egg numbers per leaf from each treatment in the a) ‘Beauty’, b) ‘Brilliance’, and c) ‘Sensation’ cultivars from the 2020–2021 field season. Error bars represent SEMs. (Miticide = miticide regimen treatment, N. cal only = *N. californicus* only, and SA+N.cal = *N. californicus* and sweet alyssum).

two *N. californicus* treatments (with and without sweet alyssum), there was no significant difference in the control of spider mite motiles in any cultivar (Table 4).

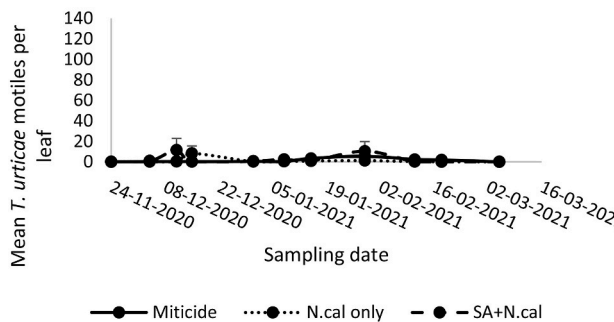
### 3.2.2. Predatory mites

For predatory mite eggs, there were no differences among treatments ( $F = 1.22$ ;  $df = 2, 27$ ;  $P = 0.31$ ). There was an average  $\pm$  SEM of  $0.8 \pm 0.4$ ,  $0.8 \pm 0.3$ , and  $1.4 \pm 0.5$  eggs in the miticide, *N. californicus* only, and *N. californicus* + sweet alyssum treatments respectively. Higher numbers of eggs were recorded from ‘Beauty’ compared with ‘Brilliance’ ( $F = 3.93$ ;  $df = 2, 27$ ;  $P = 0.032$ ). There was an average  $\pm$  SEM of  $1.9 \pm 0.6$ ,  $0.4 \pm 0.2$ , and  $0.7 \pm 0.5$  eggs in the ‘Brilliance’, ‘Festival’, and ‘Sensation’ cultivars respectively. There was no interaction between treatment and cultivar ( $F = 0.47$ ;  $df = 4, 27$ ;  $P = 0.76$ ).

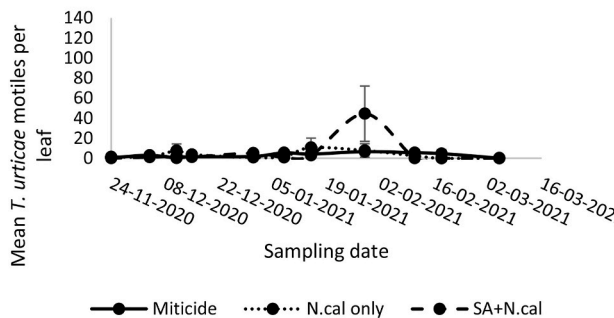
For predatory mite motiles, there were also no differences among treatments ( $F = 2.65$ ;  $df = 2, 27$ ;  $P = 0.09$ ). There was an average of  $1.1 \pm 0.5$ ,  $2.3 \pm 0.8$ , and  $2.8 \pm 1.2$  motiles in the miticide, *N. californicus* only, and *N. californicus* + sweet alyssum treatments respectively. Higher numbers of motiles were recorded from ‘Beauty’ compared with



a



b



c

**Fig. 4.** Mean *T. urticae* egg numbers per leaf from each treatment in the a) ‘Beauty’, b) ‘Brilliance’, and c) ‘Sensation’ cultivars from the 2020–2021 field season. Error bars represent SEMs. (Miticide = miticide regimen treatment, N. cal only = *N. californicus* only, and SA+N.cal = *N. californicus* and sweet alyssum).

**Table 3**

Estimated marginal means and standard error (SE) of *T. urticae* eggs collected during the 2020–2021 field season. A (\*) indicates a significant difference between the two treatments at  $P \leq 0.05$ . Sweet alyssum = SA, *N. californicus* = N. cal.

Comparison	estimate	SE	df	Z-ratio	P-value
Beauty					
Miticide vs. N.cal only *	21.796	7.725	Inf	2.821	0.013
Miticide vs. (SA+N.cal)	13.819	8.161	Inf	1.693	0.21
N.cal only vs. (SA+N.cal)	-7.977	6.902	Inf	-1.156	0.48
Brilliance					
Miticide vs. N.cal only	1.093	2.348	Inf	0.465	0.89
Miticide vs. (SA+N.cal)	-1.279	2.752	Inf	-0.465	0.89
N.cal only vs. (SA+N.cal)	-2.372	2.574	Inf	-0.922	0.63
Sensation					
Miticide vs. N.cal only	2.055	3.324	Inf	0.618	0.81
Miticide vs. (SA+N.cal)	-0.334	3.623	Inf	-0.092	>0.99
N.cal only vs. (SA+N.cal)	-2.389	3.367	Inf	-0.709	0.76

**Table 4**

Estimated marginal means and standard error SE of *T. urticae* motiles collected during the 2020–2021 field season. A (\*) indicates a significant difference between the two treatments at  $P \leq 0.05$ . Sweet alyssum = SA, *N. californicus* = N. cal.

Comparison	estimate	SE	df	Z-ratio	P-value
<b>Beauty</b>					
Miticide vs. N.cal only *	14.405	4.833	Inf	2.981	0.008
Miticide vs. (SA+N.cal) *	10.761	5.053	Inf	2.13	0.084
N.cal only vs. (SA+N.cal)	-3.645	4.102	Inf	-0.888	0.65
<b>Brilliance</b>					
Miticide vs. N.cal only	0.213	1.214	Inf	0.176	0.98
Miticide vs. (SA+N.cal)	-1.161	1.517	Inf	-0.765	0.72
N.cal only vs. (SA+N.cal)	-1.374	1.474	Inf	-0.932	0.62
<b>Sensation</b>					
Miticide vs. N.cal only	-0.376	1.974	Inf	-0.19	0.98
Miticide vs. (SA+N.cal)	-2.164	2.23	Inf	-0.97	0.6
N.cal only vs. (SA+N.cal)	-1.789	2.281	Inf	-0.784	0.71

both 'Brilliance' and 'Sensation' ( $F = 17.58$ ;  $df = 2, 27$ ;  $P < 0.0001$ ). There was an average of  $4.9 \pm 1.1$ ,  $0.4 \pm 0.1$ , and  $0.9 \pm 0.3$  motiles in the 'Brilliance', 'Festival', and 'Sensation' cultivars respectively. There was no interaction between treatment and cultivar ( $F = 0.90$ ;  $df = 4, 27$ ;  $P = 0.48$ ).

### 3.2.3. Yield

Yield in the 2020–2021 season slowly increased over the month of February and peaked in mid-March. There was no significant difference between the yields of any treatment in any cultivar during this field season.

## 4. Discussion

This study had three criteria for assessing the effect of sweet alyssum on augmenting *N. californicus* treatments for control of *T. urticae*: 1) determining whether *N. californicus* populations with the addition of sweet alyssum in strawberries caused a greater decrease in the spider mite population compared with the miticide regimen used (control), 2) whether sweet alyssum planting allowed the populations of the predatory mite *N. californicus* to survive longer in strawberry fields, and 3) whether the effect of the two previous criteria affected yield in strawberries. For the first criterion, determining if sweet alyssum led to a decrease in spider mites, the *N. californicus* mites consistently controlled the spider mites during the 2019–2020 season compared to the miticide control. However, no evidence was found that the sweet alyssum influenced the efficacy of the predatory mite treatments. This was not surprising, as both miticides used had a contact mode of action and spider mites tend to congregate on the underside of the leaves, so the spider mites may not have come into contact with the miticide even though attempts were made to target mites on the underside of leaves. Alternatively, the predatory mites can move freely and find *T. urticae* feeding on the underside of strawberry leaves. The predatory mites kept the *T. urticae* population in the 'Beauty' cultivar low compared with the miticide treatment in the 2020–2021 season. The spider mite population was consistently low throughout the 2020–2021 season on the 'Brilliance' and 'Sensation' cultivars. This was possibly due to the colder weather during the 2020–2021 season compared to 2019–2020 which could have limited the reproduction of the spider mites on those cultivars (Nyoike and Liburd 2013; Hoque et al., 2008). It should be noted that *T. urticae* numbers remained below the economic threshold of 30 motiles per leaf in all treatments and cultivars except in the miticide treatment in the 'Beauty' cultivar in 2020–2021.

For the second criterion, determining whether sweet alyssum increased the longevity of *N. californicus* populations, no differences were observed between *N. californicus* treatments with and without sweet alyssum. There are several possibilities for why these results were observed. First, low numbers of predatory mites were observed on leaf

samples. Secondly, *N. californicus* mites only begin to feed on plant pollen once prey populations drop to below numbers able to sustain *N. californicus* survival (estimated below 10 mites/trifoliolate) (Khana-mani et al., 2017). Spider mite populations remained present and observable for most of the season in all treatments and cultivars, and indeed, predatory mites were observed on the strawberry leaves over the entire duration of the experiment. It is possible that addition of the sweet alyssum was not necessary, as the less voracious feeding habits of *N. californicus* compared to other predatory mites like *P. persimilis* allowed the spider mites to remain at numbers able to sustain *N. californicus* populations (Gilstrap and Friese 1985). Thirdly, *N. californicus* mites were found in the miticide treatment plots, which indicates that they may have moved to find more spider mite prey. Both the horticultural oil and sulfur-based miticides are contact based and have a short residual time, which may explain the minimal effects of the miticide treatments on the *N. californicus* mites moving into the miticide treated plots. Lastly, there is recent evidence by Yuan et al. (2021) that *N. californicus* is capable of feeding on strawberry pollen and is even capable of reproducing on it. If *N. californicus* feeds on strawberry pollen in the field in times of starvation, then that renders the practice of banker or companion planting for augmentation purposes in strawberries less important, as *N. californicus* mites already have an alternative food source.

The third and final criterion was to determine the effect of sweet alyssum and *N. californicus* on yield. Overall, there was no difference in the marketable yield from the strawberry plants in any treatment during both seasons. There are several reasons this could have occurred. First, there were multiple pests present in the field besides spider mites that were present in large enough numbers and left uncontrolled due to the nature of the experiment, such as strawberry seed bugs *Neopamera bilobata* (Say) (Hemiptera: Lygaeidae), which caused direct damage to strawberries and left them unmarketable (Talton et al., 2020). This damage could have masked some of the effects of the spider mite damage on the yield. There was also a prevalence of fungal infection (*Botrytis cinerea*) on many berries, which prevented some of the berries from being counted as marketable. One of the miticides used in the regimen was elemental sulfur, which, in addition to being used as a miticide, is also used in many agricultural systems as a fungicide (Cooper and Williams 2004). Applying sulfur could have protected some berries in the miticide-treated plots against fungal infection and obscured yield differences caused by the differing spider mite populations.

Environmental stress in the colder 2020–2021 season may have played a part in reducing strawberry yield during that season. We observed higher overall marketable strawberry yields during the 2019–2020 season compared to the 2020–2021 season. The lower temperatures in 2020–2021 could also have limited the yield loss from the spider mites as their population would also have been severely limited by the cold (Nyoike and Liburd 2013).

Overall, the results obtained from this study indicate that there is no evidence that the usage of sweet alyssum has the potential to increase the sustainability for *N. californicus* to manage *T. urticae* populations in strawberries. The fact that *N. californicus* is able to feed on strawberry pollen warrants further study, as it is possible that using banker plants or companion planting is not needed for *N. californicus* in strawberries if they can sustain themselves from strawberry pollen rather than from pollen from an insectary plant.

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### Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence

the work reported in this paper.

### Data availability

Data will be made available on request.

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