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Integration of the predatory mite *Neoseiulus californicus* with petroleum spray oil treatments for control of *Tetranychus marianae* on eggplant

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Abstract

The red spider mite, *Tetranychus marianae* McGregor (Acari: Tetranychidae), a pest of several vegetable crops and perennials, is widespread in the Pacific Islands, like the Mariana Islands, where it was first reported. Recently, *T. marianae* has become a serious pest, particularly affecting eggplant. Efforts were therefore made to develop an integrated approach to control of *T. marianae*. First, indoor experiments on eggplant, *Solanum melongena* L. (Solanaceae), determined the optimal combination of petroleum spray oil (PSO; Volck® oil spray) with release of *Neoseiulus californicus* (McGregor) (Acari: Phytoseiidae) for the management of *T. marianae*. Additionally, studies evaluated the viability of the predatory mites on Guam. The combination of *N. californicus* with PSOs produced significant control of *T. marianae* and did not affect the survival of *N. californicus*. Although dicofol and carbaryl reduced populations of *T. marianae*, they caused high mortality of *N. californicus*. Integrating PSO with *N. californicus* yielded better control of *T. marianae* than did *N. californicus* alone. The release of *N. californicus* at 200 individuals per plant reduced populations of *T. marianae* more than did other release rates. In addition, *N. californicus* was able to survive and become established after being released on Guam.

Introduction

In the Mariana Islands farmers have been encouraged to grow many vegetables, including eggplant (*Solanum melongena* L., Solanaceae), to increase self-sufficiency by reducing the importation of vegetables into the region and thereby reducing the probability of introduction of invasive species. Eggplant has been widely grown on commercial farms and in house backyards (Schlub and Yudin 2002). The spider mite *Tetranychus marianae* McGregor (Acari: Tetranychidae) severely damages eggplant in most parts of the Marianas (Reddy, Kikuchi, and Remolona 2011), but no other key pests require control. Little research has been carried out on control of spider mites in the Pacific Islands. The majority of growers in this region use carbaryl and dicofol insecticides for the control of spider mites, applying as many as 15–20 sprays (average of 15 sprays) each cropping period. These pesticides are not only expensive but also pose ecological and toxicological hazards. An ecologically sound and cost-effective integrated control programme is therefore needed.

An integrated control approach including the use of biopesticides and biological control can be a viable alternative (Reddy 2001). The combination of a predatory mite, *Phytoseiulus persimilis* Athias-Henriot (Acari: Phytoseiidae), with petroleum spray oil (PSOs or horticultural mineral oils) has proven effective in controlling mites on vegetables and ornamental crops (Nicetic, Watson, Beattie, Meats, and Zheng 2001). However, Esguerra and Del Rosario (2007) reported that *P. persimilis* failed to establish itself after release in the Republic of Palau. They pointed out that *P. persimilis* could not survive the hot, humid and wet conditions of tropical islands, but a closely related species, *Neoseiulus longispinosus* (Evans) (Acari: Phytoseiidae), readily established in the field (Kongchuensin, Charanasri, and Takafuji 2005). Moreover, many pesticides are reported to be harmless to adult females, eggs and immature life stages of *N. longispinosus* (Kongchuensin and Takafuji 2006). Another species closely related to *N. longispinosus* is *Neoseiulus californicus* (McGregor) (Acari: Phytoseiidae). Originating in California, this predatory mite is tolerant of many pesticides and feeds on both the spider mite and the broad mite (Fraulo, McSorley, and Liburd 2008).

Because of their low toxicity to natural enemies of pests and safety for the environment, PSOs are compatible with integrated pest management (Beattie and Smith 1996). They are effective against a wide range of horticultural pests, including spider mites (Nicetic et al., 2001). PSOs are less disruptive to predatory mites than some commonly used synthetic miticides, and they are generally much safer to use from environmental and human-health perspectives (Davidson, Dibble, Flint, Marer, and Guye 1991; Johnson 1994). Because of their short-term residual activity, PSOs do not severely affect populations of beneficial arthropods, although predators and parasitoids may be killed on contact when sprayed directly (Davidson et al. 1991).

We undertook the present study to evaluate use of PSOs and *N. californicus* for the management of *T. marianae* in the Marianas and other tropical regions where eggplant is grown, specifically (1) to determine the interaction between *N. californicus* and PSOs and (2) to evaluate the viability of the predatory mites in their new environment to make sure they can multiply and survive.

Materials and methods

Eggplant seedlings

Eggplant seeds of the Pingtung Long variety (Known-You Seed Co., Kaohsiung, Taiwan) were sown in trays (40 × 30 cm) and raised in a nursery shade house (30–32°C, 60–80% RH, 14:10 h light:dark photoperiod). The seedlings were watered by means of microsprays and were grown for 40 days.

Study site descriptions

Experiments were performed in the shade house on the premises of the Western Pacific Quarantine Biocontrol Laboratory at the University of Guam. The shade house was 100 m² in area; the walls and roof were constructed of shade cloth. The first experiment was conducted during the dry season (January–May 2010) and then repeated during the wet season (July–November 2010). The second experiment was conducted during the dry season (February–June 2011) and then repeated during the

wet season (August–December 2011). Before each experiment, eggplant seedlings raised in the nursery were transplanted to individual pots (20 cm diameter × 30 cm deep) filled with Miracle Grow potting soil. The potting soil was disease- and weed-free and also helped hold moisture while providing nutrients. All suggested agronomic practices were followed (Schlub and Yudin 2002).

Chemicals

The PSO used in all the experiments was Volck oil spray® (petroleum oil 97%, other ingredients 3%; The Ortho Group, Marysville, OH) and was applied at 20 ml/liter of water. Carbaryl (Sevin) was applied at 1.5 ml/liter and dicofol (Dicofol 3) at 2 ml/liter.

Mite cultures

All mite cultures were held at $28 \pm 2^\circ\text{C}$, under a 14:10 h light:dark cycle, and at 72–86% relative humidity.

Phytophagous mite culture

Laboratory cultures of *T. marianae* were originally collected from eggplant crops in growers' fields in Yigo and Dededo (Guam, USA). Cultures had been maintained on potted eggplant plants in cages in the shade house for two years and supplemented yearly with additional field-collected mites. These mite cultures have been confirmed and identified as *T. marianae* (Reddy et al. 2011).

Predatory mite culture

Laboratory cultures of *N. californicus* were originally obtained from Biocontrol Network, Brentwood, TN. Cultures had been maintained for six months with monthly additions from Biocontrol Network. These predatory mite cultures were fed mixed life stages of *T. marianae* three times per week on eggplant before the study period.

Experiment 1: interaction between the predatory mite (N. californicus) and PSO

The first experiment investigated two factors: (1) any adverse effect of PSO on *N. californicus* and (2) whether the efficacy of a combined PSO–*N. californicus* control strategy against *T. marianae* was greater when *N. californicus* was applied alone or in combination with dicofol or carbaryl.

The layout of the experiment was a randomised block design of four treatments with 10 replicates each. The plants were grown in pots under natural light and temperature conditions. For each treatment, 10 pots were used as replicates. The pots were tagged and covered with cages that prevented mite migration, and four treatments were applied: (1) *N. californicus* alone, (2) *N. californicus* and PSO, (3) *N. californicus* and dicofol and (4) *N. californicus* and carbaryl.

Three eggplant leaves, each bearing 200 nymphs and adults of *T. marianae*, were placed in each potted plant 15 days after transplanting (DAT); 50 nymphs and adults

of *N. californicus* were added in a similar way 30 DAT; and PSO was applied 45 and 55 DAT with a two-nozzle lawn and garden sprayer.

Experiment 2: survival rate and feeding potential of the predatory mite (N. californicus)

The second experiment evaluated the viability of the introduced predatory mite in its new environment (Guam) to ensure that it could survive and become established.

The layout of the experiment was a randomised block design of five treatments with 10 replicates each. The plants were again grown in pots under natural light and temperature conditions. The pots were tagged and covered with cages that prevented mite migration. Three eggplant leaves, each bearing about 500 nymphs and adults of *T. marianae*, were placed in each potted plant 15 DAT; 0 (control), 50, 100, 150 or 200 nymphs and adults of *N. californicus* were added to each plant 30 DAT. For each number of added predatory mites, 10 pots were used as replicates. Individuals of *T. marianae* and *N. californicus* were counted on plants at 30-day intervals during the entire crop duration. Eggplants were harvested when the fruit surface was glossy and tender and before the seeds within the fruit became brown.

To determine *T. marianae* and *N. californicus* population levels, we randomly selected three leaves on each plant, one each from the top, middle and bottom of the plant. On the underside of each of these leaves, we counted motile *N. californicus* and *T. marianae* with the aid of a magnifying lens.

Statistical analysis

All statistical analyses were carried out with SAS Version 9.3 (SAS Institute 2009). The number of *N. californicus* and *T. marianae* on 30 leaves in each treatment was square-root transformed before each set of data was subjected to ANOVA. A 5% level of significance was used for all the analyses. Treatment means were compared to all means (different releases and sampling dates together) with a Tukey's test.

Results

Experiment 1: interaction between the predatory mite (N. californicus) and PSO

The results of the first experiment indicated that the combination of *N. californicus* and PSO spray was the most effective ($F_{3,18} = 92.4$, $P < 0.05$, Figure 1) in reducing the *T. marianae* population in both trials, followed in order by *N. californicus* alone, *N. californicus* and dicofol and *N. californicus* and carbaryl. Treatment with the combination of *N. californicus* and PSO caused 100% mortality of *T. marianae* after the second spray, in both dry-season and wet-seasons trials. In contrast, PSO alone did not significantly affect *N. californicus* populations ($F_{3,22} = 78.6$, $P > 0.05$, Figure 2), even after the second spray in both of the trials, but treatment with either dicofol or carbaryl significantly reduced populations of *N. californicus* in both trials.

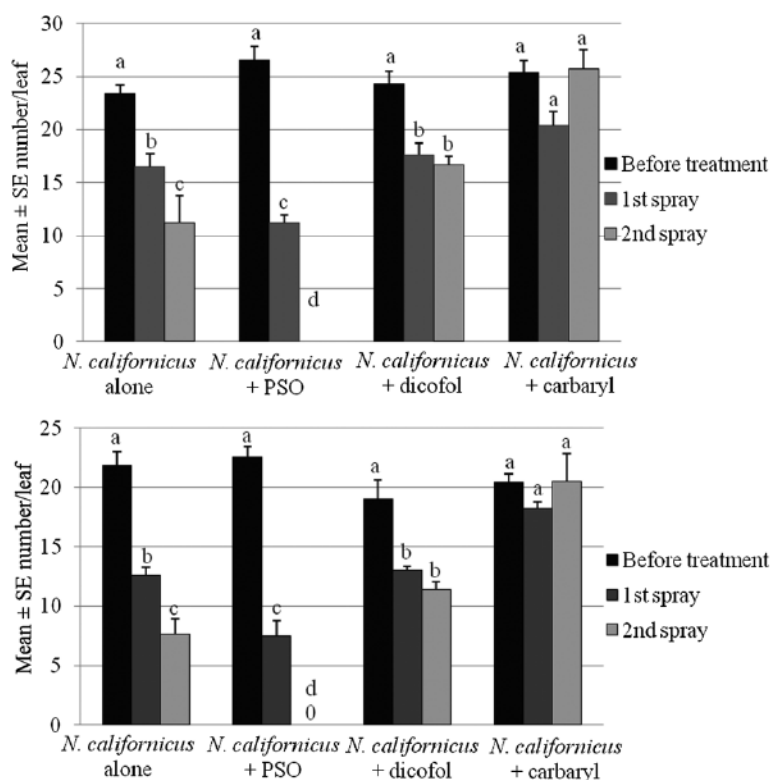


Figure 1. Effect of petroleum spray oil (PSO) and dicofol and carbaryl sprays on numbers of the phytophagous mite *Tetranychus marianae* on eggplant, in the presence of the predatory mite *Neoseiulus californicus* during the dry (upper panel) and wet (lower panel) seasons. Bars represent numbers of *T. marianae* before spray treatment (black), after the first spray treatment (45 days after transplantation, DAT; mid grey), and after the second spray treatment (55 days DAT, light grey). Means marked by the same letters do not differ significantly ($P > 0.05$, One-way ANOVA, Tukey's test). Each treatment value represents the mean ($\pm$ SE) of 10 replications.

Experiment 2: survival rate and feeding potential of the predatory mite (*N. californicus*)

The results of the second experiment demonstrated that *N. californicus* was able to survive and become established after being released on Guam. The addition of *N. californicus* at 200 individuals per plant reduced the population of *T. marianae* ($F_{4,34} = 101.7$, $P < 0.05$, Figure 3) more effectively than addition of 150, 100 or 50 individuals. On the control plants (on which no predators were released), high numbers of *T. marianae* were recorded in both trials. In addition, when 200 *N. californicus* individuals were released per plant, significant numbers ($F_{4,22} = 87.2$, $P < 0.05$, Figure 4) remained 90 DAT in both trials, showing that the species successfully established itself. Finally, addition of 200 *N. californicus* individuals significantly ($F_{4,44} = 122.3$, $P < 0.05$, Figure 5) increased the marketable eggplant yield from 430 (in the control plots) to 3640 g per plant in the dry season and from 680 to 3240 g in the wet season.

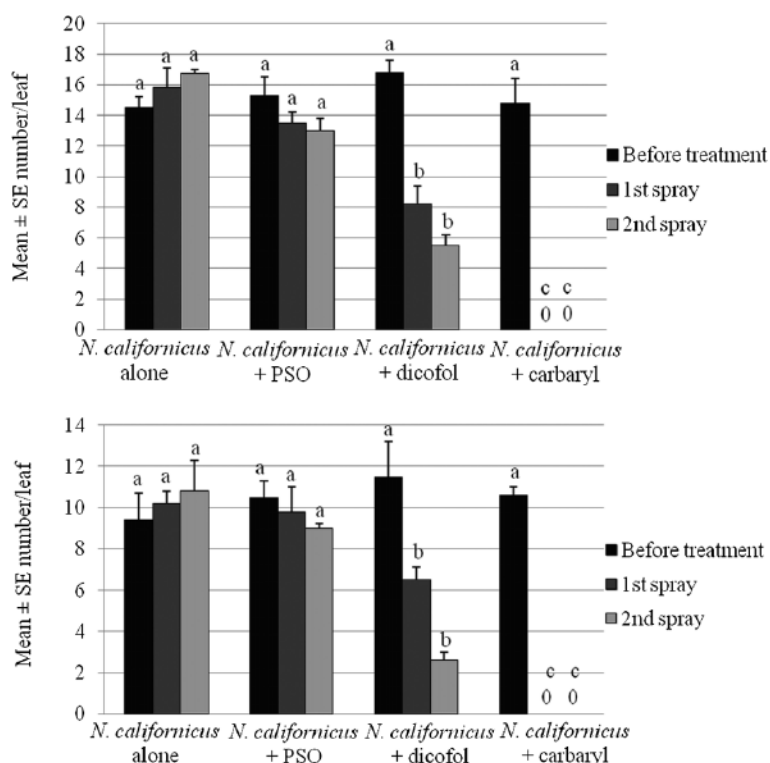


Figure 2. Effects of the treatments shown in Figure 1 on numbers of *N. californicus* on eggplant.

Discussion

Predatory mites have been used as an alternative to miticides on a variety of crops (Beard 1999; Hoddle, Aponte, Kerguelen, and Heraty 1999). *Neoseiulus californicus* prefers tetranychid mites as food, but because it is a generalist, it will also consume other mite species, small insects such as thrips, and even pollen when the primary prey is unavailable (Castagnoli and Simoni 2003). In addition, *N. californicus* does well in areas with at least 60% humidity and has reportedly thrived in warmer climate conditions (Monetti and Croft 1997). Gotoh, Suwa, and Kitashima (2004) reported that this predatory mite prefers higher temperatures. The authors reported that more than 97.3% of *N. californicus* eggs hatched, and more than 81.6% of newly hatched larvae attained maturity at temperatures between 15 and 35°C. As these conditions are very similar to those of Guam, *N. californicus* would be an ideal biocontrol agent for release on spider and broad mites in eggplant fields on Guam and nearby islands.

Our results indicated that PSO was not detrimental to *N. californicus*. A number of other studies have explicitly observed the effects of spray oils on predatory mites. For example, Nicetic et al. (2001) reported that 0.5% PSO applied fortnightly to roses gave excellent protection from *Tetranychus urticae* Koch (Acarina: Tetranychidae) but did not affect predatory mites, *P. persimilis* (Acarina: Phytoseiidae). Moreover, Morse, Bellows, Gatson, and Iwata (1987) found that the predatory mite *Euseius stipulatus* Athias-Henriot (Acarina: Phytoseiidae) was not significantly affected by

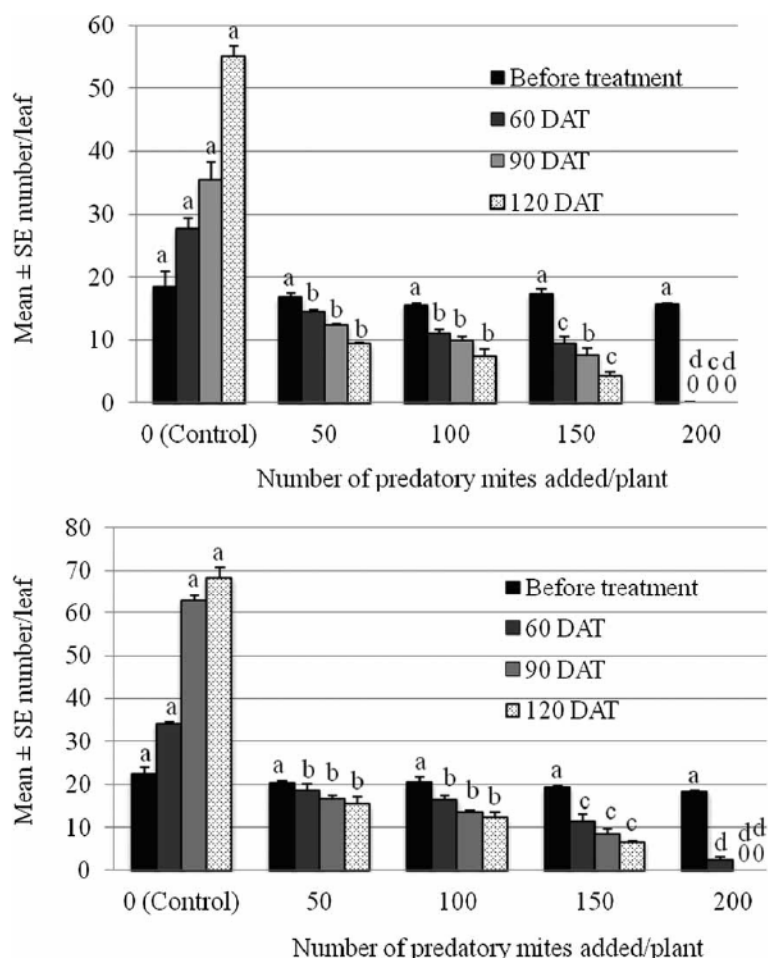


Figure 3. Effects of the addition of different numbers of the predatory mite *N. californicus* on numbers of the phytophagous mite *T. marianae* on eggplant in the dry (upper panel) and wet (lower panel) seasons. DAT, days after transplantation. Means marked by the same letters do not differ significantly ($P > 0.05$, one-way ANOVA, Tukey's test). Each treatment value represents the mean ($\pm$ SE) of 10 replications.

oil spray. Davidson et al. (1991) reported that *Galendromus* (*Typhlodromus*) *occidentalis* Nesbitt was not affected by 5% oil spray applied to almond trees, but Stanyard, Foster, and Gibb (1998) found that oil reduced the number of predatory mites, *Neoseiulus* (*Amblyseius*) *fallacis* (Garman) (Acarina: Phytoseiidae), on apple trees after several consecutive sprays, although it did not eradicate the population.

Our results revealed that, although synthetic chemicals such as carbaryl and dicofol slightly reduced numbers of *T. marianae*, they also caused mortality of *N. californicus*. Modern synthetic chemicals are still being used in parts of the world to control mite incidence on various crops. Although the label on carbaryl does list mites, the product is well known to induce mite outbreaks. Many farmers in Guam are compelled to use repeated applications because of the ineffectiveness of these chemicals and the eventual mite population buildup. In addition, carbaryl is known

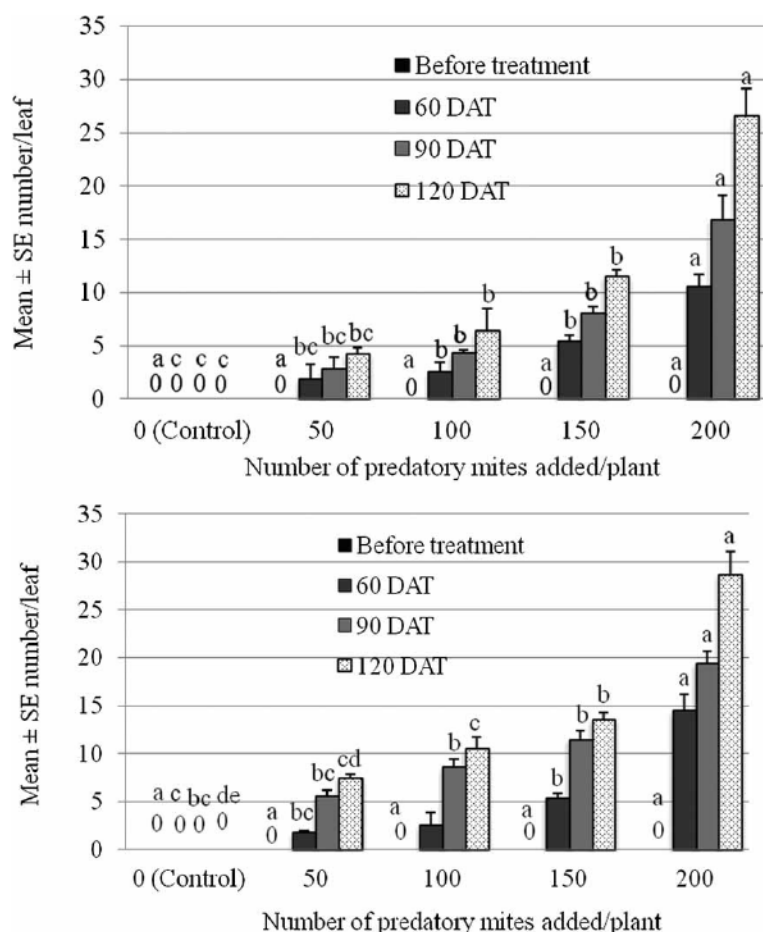


Figure 4. Numbers of the predatory mite *N. californicus* on eggplant at various intervals after release of different numbers of mites. Means marked by the same letters do not differ significantly ($P > 0.05$, one-way ANOVA, Tukey's test). Each treatment value represents the mean ($\pm$ SE) of 10 replications.

to cause mite populations to flare and become carbaryl resistant (Mullens, Velten, Hinkle, Kuney, and Szijj 2004; Shaw and Wallis 2008). Likewise, dicofol is widely known to cause the development of resistance in spider mites after only one or two applications and to destroy predatory mite populations (Dutcher 2007).

We have demonstrated that combining PSO treatment with *N. californicus* resulted in better control of *T. marianae* than using *N. californicus* alone. Our results agree with those of Nicetic et al. (2001), who reported that the combination of PSO with *P. persimilis* gave better control of *T. urticae* than did either treatment alone. An additional advantage is that the PSO simultaneously controls any accidental pests that may arrive during the crop-growing period. Therefore, the effects of integration of PSO with *N. californicus* are highly beneficial and recommended for the control of *T. marianae* on eggplant. Even though our experiments were conducted indoors, the results are expected to prove useful for managing this important mite pest in larger eggplant plantations.

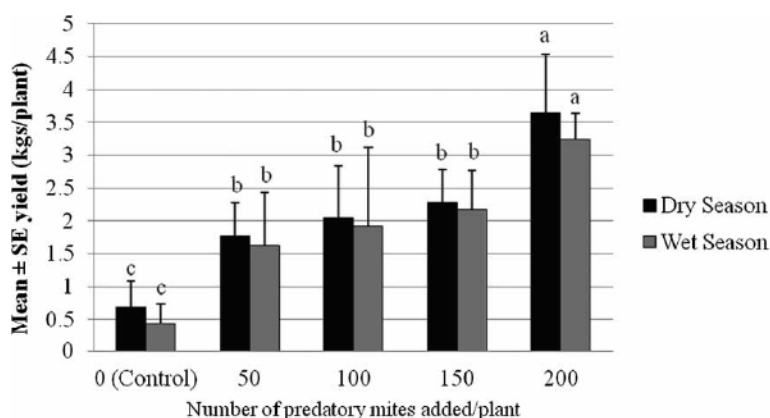


Figure 5. Effects of different numbers of *N. californicus* on the marketable yield of eggplant grown in the dry and wet seasons under indoor conditions. Means marked by the same letters do not differ significantly ($P > 0.05$, one-way ANOVA, Tukey's test). Each treatment value represents the mean ($\pm$ SE) of 10 replications.

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