Effective use of the predatory species, Phytoseiulus persimilis and Stethorus punctillum in controlling the two-spotted mite Tetranychus urticae in croton greenhouse

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Research Article

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Abstract

The croton, *Codiaeum variegatum* is a beautiful tropical plant. The two-spotted mite, *Tetranychus urticae* Koch, causes economic damage to the croton. The present study aimed to control *T. urticae* by releasing two predatory species, *Phytoseiulus persimilis* Athias Henriot and *Stethorus punctillum* Weise, depending on the suitable temperature and humidity of each predator. The experiments were carried out at commercial plastic greenhouses for two seasons. Once the mite *T. urticae* infestation was detected, the predators were released. The predator, *P. persimilis* was released three times, with a rate of 9/m^2. It increased when the temperature and humidity ranged from 25.5–29°C and 72.8–86.4% RH, respectively, while it disappeared when the temperature increased above 30°C. There was a negative correlation between *P. persimilis* and temperature, but a positive correlation with relative humidity. The predator, *S. punctillum*, was released twice, at a rate of 0.5/m^2. It increased when the temperature and humidity ranged from 31.8–35°C and 65–77% RH, respectively. There was a positive correlation between *S. punctillum* and temperature, but a negative correlation with relative humidity. In the two seasons, the population numbers of the mite *T. urticae* decreased gradually to nil/cm^2/croton leaf in the biological greenhouse. In the control greenhouse, the population of *T. urticae* increased to 59.47 ± 6.34 and 85.63 ± 10.45/cm^2/croton leaf in 2019 and 2020, respectively, at the end of the season. Integration of the two predators, *P. persimilis* and *S. punctillum* was able to achieve effective control of *T. urticae* when used according to the suitable temperature for each predator in croton greenhouses.

Introduction

Croton, *Codiaeum variegatum* (Euphorbiaceae family), is one of the beautiful indoor and outdoor plants that need agricultural care. Moreover, it is a small tree or shrub grown in the garden as an ornamental or pot plant. Croton leaves come in a variety of sizes, shapes, and colors. Croton leaf extracts have a wide range of medicinal properties, including purgative, sedative, anti-cancer and antiviral (Deshmukh and Borle 1975; Kupchan et al.1976).

It's also a rich source of alkaloids, and flavonoid secondary metabolites (Simona et al. 2008). Cuttings, grafting, seeds, and air layering are all used to grow croton. Each year, twenty plants can be grown from the shooting tips of one mother/stock plant (Nasib et al. 2008).

Codiaeum variegatum is severely harmed by temperatures below 50°F (10°C). Crotons have one of the highest transpiration levels of frequently cultivated leaf plants and frequent watering is required (Robert and Lance 2003). It grows well in humid environments (Nasib et al. 2008).

The two-spotted spider mite, *Tetranychus urticae* Koch, is a major pest of many greenhouse plants, ornamental nurseries, and field crops. *Tetranychus urticae* can cause webbing, fine stippling, leaf yellowing, leaf dropping, and even plant death (Helle and Sabelis 1985). Chemical control is difficult due to its resistance to many acaricides (Carbonaro et al. 1986). The infestation of *T. urticae* causes significant economic losses to many crops, including croton (Cashion et al. 1994; Robert and Lance 2003).

Several successful biological control programs have demonstrated that greenhouses are suitable for using programs of biological control agents because they are closed systems with barriers against natural enemy dispersal (Perdikis et al. 2008). Accordingly, several native parasitoids and predators in agro-ecosystems can be used to control crop pests in greenhouses (Nicoli and Burgio 1997).

The biological control of spider mites on crops and ornamentals using predatory mites has been the subject of numerous studies (Gerson and Weintraub 2007; Weintraub and Palevsky 2008). The predator *Phytoseiulus persimilis* Athias Henriot (Phytoseiidae) has been the most popular predator for biological control of spider mites for decades. It only eats spider mites, mainly those of the genus *Tetranychus* (McMurtry and Croft 1997). Environmental factors like humidity and temperature have an impact on *P. persimilis'* ability to perform and reproduce (Zhang 2003).

Stenseth (1979) found that in climate-controlled growth chambers with low humidity and high temperatures, this predator is ineffective at controlling spider mites. In regions with these typical climatic conditions, such as the inland arid regions of the Mediterranean basin, the predator *P. persimilis* is unable to control spider mite populations (Escudero and Ferragut 2005; Weintraub et al. 2006; Weintraub and Palevsky 2008). Additionally, *P. persimilis* is unable to persist and cannot survive in a plant environment with low densities of spider mites (Schausberger and Walzer 2001; Blümel and Walzer 2002).

The adults and larvae of *Stethorus* species (Coleoptera: Coccinellidae) are specialist predators of spider mites (Bailey and Caon 1986; Biddinger et al. 2009). *Stethorus* species were suggested as potential biological control agents for spider mites in crops (Hull et al. 1976, 1977).

The *Stethorus* species are small and exceptionally well adapted for living and searching for prey. Also, they are winged predators that can quickly reach spider mite hotspots, which is a benefit that can be used in biological control programs (Rott and Ponsonby 2000).

Stethorus punctillum Weise, activity increases significantly as temperatures increase to 30°C, and it has successfully developed to an adult stage between 14 and 34°C. Also, it was effective at relative humidity levels ranging from 30 to 90%, implying that RH had no effect (Rott and Ponsonby 2000). The predator, *S. punctillum* successfully developed to the adult stage between 14 and 34°C (Roy et al. 2002).

According to the previous reports, the optimal temperature of the two predatory species of mites, *P. persimilis* and *S. punctillum* differs. The optimal temperature of *P. persimilis* was 27^{o}C. It is sensitive to temperatures above 30°C and will stop feeding at about 35°C. On the other hand, the winged predator *S. punctillum* activity increased up to 30°C. Therefore, the current study aims to improve the control of *T. urticae* by releasing the two predatory species, *P. persimilis* and *S. punctillum*, according to the suitable temperature for each predator in the croton greenhouse.

Materials And Methods

Greenhouses

The experiments were carried out at Egypt Green Farm, Mansouria, Giza, Egypt, using two plastic greenhouses of 540 m2 (60 m × 9 m) planted with croton plants *C. variegatum* cv. Petra was used to releasing predators. A smaller greenhouse planted with about 100 croton plants was used as the experimental control. The daily minimum and maximum temperatures and humidity were recorded using sensors installed in the greenhouse.

A randomized complete block design with three replicates per treatment was used in each greenhouse. Each replicate included 200 plants (50 plants/row) and was conducted in a separate tunnel, leaving one tunnel as a buffer between replicates.

Predator's colonies

The predator, *P. persimilis* was obtained from the Plant Protection Research Institute, Agriculture Research Center, Giza, Egypt. The predator, *S. punctillum* was collected from an eggplant greenhouse infested with the two-spotted mite, *T. urticae*.

Experiment

The two predatory species, *P. persimilis* and *S. punctillum*, are released dependent on their optimal temperature and humidity to control *T. urticae* in the croton greenhouse. The rate of release of the predator *P. persimilis* was 9 individuals/m^2 from March to May (Fig. 1). The rate of release of the predator *S. punctillum* was 0.5/m^2 from June to August (Fig. 1). The timing of the predator's releases was determined according to the average temperature and humidity in the greenhouse and the weekly average number of mites/plant.

The application's effectiveness in the greenhouse was estimated by the weekly count of the targeted pest, *T. urticae*, for two years, from the 15th of February to the 15th of September in 2019 and 2020.

The population density of *T. urticae* was counted at 3 levels (top, middle, and lower) of the plant, using a one-square-cm lens (25 plants/replicate).

Statistical analysis

Data were coded and entered using the statistical package SPSS version 22. Data were statistically described in terms of mean and standard deviation for quantitative variables. Analysis of variance (ANOVA) was applied using the Holm-sidak method, P 0.05.

The analysis becomes available using SigmaPlot V12.5 and MiniTab V18.1 software. The correlation coefficient between parameters was done in all the surveyed experiments during the study period using MINITAB Release 14 computer program.

Results

Two predators, *P. persimilis* and *S. punctillum* were released to control the population of two-spotted mite *T. urticae* in the croton greenhouse. The release was based on the suitable temperature of the predators as well as the temperature and humidity conditions in the croton greenhouse.

Climatic conditions

Mean weekly temperatures ranged from 26.54 to 31.62 ^{o} C during the experiment period. Minimum and maximum weekly temperatures ranged between 25.2–27.5 ^{o} C and 29.45–34.7 ^{o} C, respectively. Minimum and maximum weekly relative humidity ranged between 58.6–54.10% and 88.20–96.30%, respectively, during the experiment period.

The population of the two-spotted mite T. urticae **and the two predators**

In 2019, the population of *T. urticae* started on March 7th with 1.2 ± 0.28 mites/cm^2/croton leaf in the predators' greenhouse (Fig. 2A). Once the mite infestation was detected, the predator, *P. persimilis* was released 3 times on March 7th, 14th and April 4th. After the three releases, the population of *P. persimilis* increased to a peak of 10.2 ± 0.52 predators/cm^2/croton leaf on April 18th. The increase of the predator, *P. persimilis* led to a decrease in the population of *T. urticae* to 2.3 ± 0.33 mites/cm^2/croton leaf on May 23rd (Fig. 3A). After that, the population of the predator, *P. persimilis* decreased gradually to reach zero on May 23rd which gave a chance for *T. urticae* to increase again.

The temperature and humidity values from March 7th to May 9th, 2019 ranged from 25-29.8°C and 65-86.4% RH, respectively, which was suitable for the predator's activity. The predator disappeared on May 23th when the temperature reached 34.65°C, which wasn't suitable for the predator's activity.

On June 27th, the population of *T. urticae* reached 15.4 ± 0.5 mites/cm^2/croton leaf in the predators' greenhouse, which was controlled by the release of the predator *S. punctillum* 2 times on June 27th and July 4th. After that, the population of *S. punctillum* increased to a peak of 7.2 ± 0.48 predators/cm^2/croton leaf on July 18th. After that, the population of *T. urticae* decreased gradually to reach nil/cm^2/croton leaf in the predators' greenhouse at the end of the season (Fig. 3A).

From July 4th to August 3rd, the temperature and humidity ranged from 33.7–35.8°C and 59.7–72.3% RH, respectively.

In the control greenhouse, the population of the *T. urticae* started with 4.2 ± 0.51 mites/cm^2/croton leaf on March 7th and increased through the season to reach 70.5 ± 6.34/cm^2/croton leaf at the end of the season (Fig. 2A).

The population of *T. urticae* decreased significantly ($P < 0.001$) in the predators' greenhouse and increased significantly ($P < 0.001$) in the control greenhouse.

There was a significant interaction between treatments (F = 5322.9, df = 1, p   0.001). There was also a significant interaction between inspection date and treatments (p   0.001).

In 2020, the infestation of *T. urticae* started on February 20th, with 4.63 ± 2.03 and 3.67 ± 1.22 / cm^2/croton leaf in predators and control greenhouses, respectively (Fig. 2B). The population of the mite *T. urticae* increased to 20.23 ± 3.3 /cm^2/croton leaf on March 5th, in the predators' greenhouse. The predator, *P. persimilis* was then released three times on March 5th, 12th and 19th. After the three releases, the population of *P. persimilis* increased to a peak of 12.5 ± 0.4 predators/cm^2/croton leaf on March 26th. The increase of the predator, *P. persimilis* led to a decrease in the population of *T. urticae* to 3.7 ± 0.3 mites/cm^2/croton leaf on May 7th (Fig. 3B). At the same time, the population in the control greenhouse increased to reach 50.27 ± 3.24/cm^2/croton leaf (Fig. 2B).

The predator *P. persimilis* appeared and its population density increased when the temperature and humidity ranged from 25.5–29°C and 72.8–86.4% RH, respectively, from February 27th to April 23rd, while it disappeared when the temperature increased above 30°C and the humidity decreased.

About two months after the last release of the predator *P. persimilis*, the mite *T. urticae* population appeared with 10.2 ± 2.56 /cm^2/croton leaf in the predators' greenhouse, which was controlled by the release of the predator *S. punctillum* twice on May 28th and July 4th. After that, the population of *S. punctillum* increased to a peak of 9.9 ± 0.5 predators/cm^2/croton leaf on June 25th (Fig. 3B). After that, the population of *T. urticae* decreased gradually to reach nil/cm^2/croton leaf in the predators' greenhouse at the end of the season (Fig. 3B) and the population increased in the control greenhouse to 85.63 ± 10.45/cm^2/croton leaf (Fig. 2B).

From May 21st to July 23th the temperature and humidity ranged from 31.8–35°C and 65–77% RH, respectively. This range of temperature and humidity was suitable for the development of the predator, *S. punctillum*.

The population of *T. urticae* decreased significantly (P < 0.001) in the predators' greenhouse and increased significantly (P < 0.001) in the control greenhouse.

There was a significant interaction between treatments (F = 2813.6, df = 1, p   0.001). There was also a significant interaction between inspection date and treatments (p   0.001).

The correlation coefficient of the mite T. urticae **population with abiotic factors**:

The mite *T. urticae* incidence had a significant positive correlation (r = 0.707) with the weather parameter temperature on croton during 2019, whereas the mite population had a negative correlation (r = -0.638) with relative humidity (Fig. 4A & 5A). The same pattern of mite incidence on the croton was also recorded in 2020. The mite population had a significant positive correlation (r = 0.784) with temperature, whereas it had a negative correlation (r = -0.669) with relative humidity (Fig. 4B & 5B).

The correlation coefficient of the predators' population with abiotic factors:

A negative correlation was found between the predator, *P. persimilis* and temperature (r =-0.435 and – 0.527) in 2019 and 2020, respectively (Fig. 6A&B), while the same predator had a positive correlation with relative humidity (r = 0.378 and 0.332) in 2019 and 2020, respectively (Fig. 7A&B).

A positive correlation was found between the predator, *S. punctillum* and temperature (r = 0.498 and 0.522) in 2019 and 2020, respectively (Fig. 6A&B), while the same predator had a negative correlation with relative humidity (r = -0.405 and – 0.345) in 2019 and 2020, respectively (Fig. 7A&B).

Discussion

The present study showed that the release of the two predators, *P. persimilis* and *S. punctillum*, depending on the suitable temperature and humidity of each predator, was successful in controlling the population of the mite *T. urticae* in croton greenhouses.

The results of this study showed that the mite, *T. urticae* population occurred throughout the year, and there was a positive correlation with temperature and a negative correlation with relative humidity.

These findings agree with Sarkar and Somchowdhary (1989), Rajakumar et al. (2005), Yadavbabu & Manjunatha (2007) Mahato et al. (2008) and Meena et al. (2013) who reported that the mite, *T. urticae*, the incidence was recorded on orchids all year round, indicative of overlapping generations; however, two distinct population peaks were recorded during April and May. There was a significant positive correlation

with mite incidence and maximum and minimum temperatures, daylight had a non-significant positive correlation and rainfall and relative humidity had a non-significant negative correlation with mite incidence.

In this study, once the mite infestation was detected, the predators *P. persimilis* and *S. punctillum* were released. Cashion et al. (1994) used *P. persimlis* to control two-spotted spider mites on Croton. The use of the predator has succeeded in reducing the number of acaricide applications by 87 to 92% in Croton. Parvin and Haque 2008, reported that the early release of the 3 predators; *Scolothrips sexmaculatus* Pergande, *P. persimilis* and *S. punctillum* has been shown to keep the mite population lower than their late release. All three predators may be used as bio-control agents against the two-spotted spider mite.

In the present study, the population density of the predator *P. persimilis* increased when the temperature and humidity ranged from 25.5–29°C and 72.8–86.4% RH, respectively, while it disappeared when the temperature increased above 30°C and the humidity decreased. There was a negative correlation between the predator, *P. persimilis* and temperature, but a positive correlation with relative humidity.

According to Pruszynski (1976), *P. persimilis* stopped feeding at about 35°C (95°F), and it was more sensitive to temperatures over 30°C (86°F) than the prey. The predator, *P. persimilis* development rates were optimum at 27°C (at 60 to 85% RH) when compared to temperatures ranging from 15 to 27°C, and spider mite control was most successful at this temperature.

Rojas et al. (2013) reported that the predator *P. persimilis* could develop and reproduce at all tested temperatures from 21 to 31^{o}C. However, temperature influenced the development time, survival, and fecundity of *P. persimilis*. The authors concluded that *P. persimilis* survived much better at temperatures below 29^{o}C.

The abiotic conditions for optimal performance of *P. persimilis* are temperatures between 15°C and 27°C, and relative humidity of 60–90%. low temperatures (15^{o}C) to avoid the economic damage of *T. urticae*. High temperatures (27 ^{o}C) and low relative humidity (40 ~ or below) reduce the vitality of *P. persimilis* and favour development of *T. urticae* (Stenseth 1979). While it is an excellent predator under these climatic regimes, its efficacy under hot and dry conditions is often insufficient (Force 1967; Nihoul 1992; Skirvin and Fenlon 2003; Escudero and Ferragut 2005; Weintraub and Palevsky 2008).

In the present study, the population density of the predator *S. punctillum* increased when the temperature and humidity ranged from 31.8–35°C and 65–77% RH, respectively. There was a positive correlation between the predator, *S. punctillum* and temperature, but a negative correlation with relative humidity.

Rott and Ponsonby (2000) found that the activity of *S. punctillum* increased significantly at higher temperature levels but had no effect on relative humidity.

Studies investigating the effect of temperature on coccinellid species have shown that fecundity and oviposition have increased with increasing temperatures up to 30°C (Ponsonby and Copland 1998) and an increase in larval developmental rate was found to be at 28°C (Ponsonby and Copland 1996; Lamana and Miller 1998). *Stethorus punctillum* developed successfully to an adult stage between 14 and 34°C, but survival was low at both ends of the temperature range. The optimal temperature for immature survival ranged from 16 to 32°C. Eggs did not hatch at 12°C or 36°C (Roy et al. 2002).

This indicates that temperature level variations can influence predation, reproduction, and development of coccinellids and that the thermal requirements of each coccinellid species have to be considered in the biological control programs (Obrycki and Kring 1998).

Conclusion, the present study provided an effective method for controlling the two-spotted mite *T. urticae* in croton greenhouses by using the two predators, *P. persimilis and* S. *punctillum*, which have different optimum temperature and relative humidity values. The timely use of the two predators based on their temperature and humidity-dependent activities has proved to be effective in controlling the mite *T. urticae* during the two seasons 2019–2020.

Declarations

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Competing interests: There is neither conflict of interest.

Authors' contributions: I designed, created, wrote, and approved the manuscript.

Data Availability: The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Ethics approval: Not applicable – the study was conducted on insect species that are abundant in the ecosystem and does not require ethical approval.

Consent to participate: Not applicable.

Consent for publication: The manuscript has not been published in completely or in part elsewhere.

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Figures

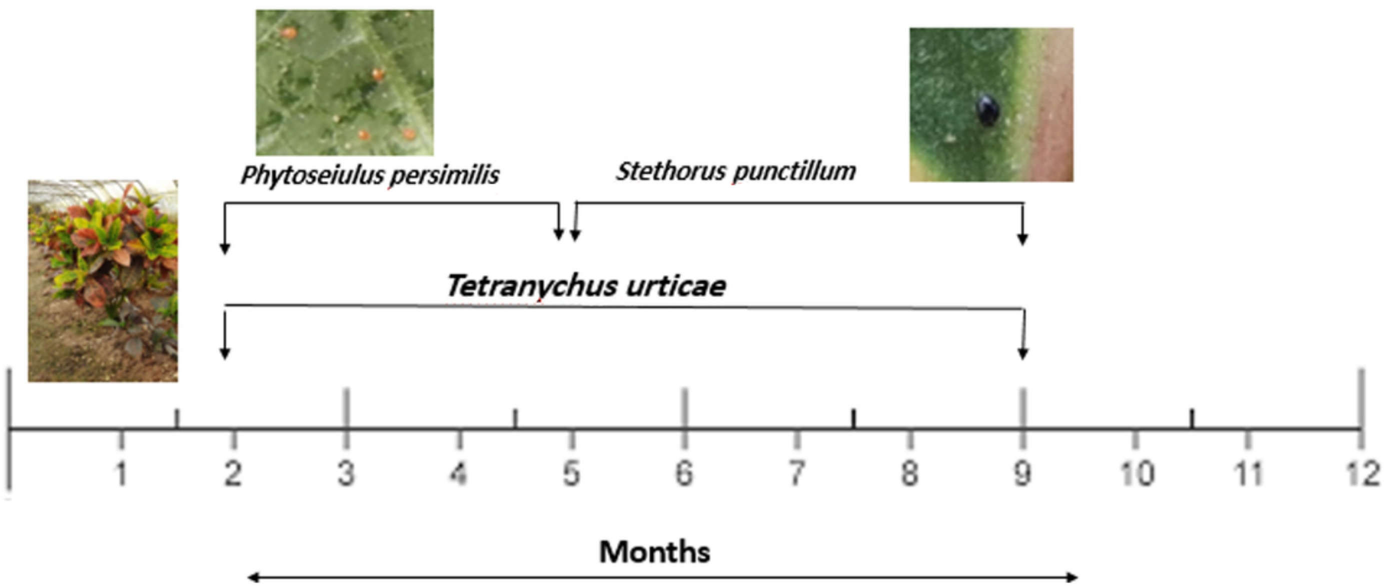
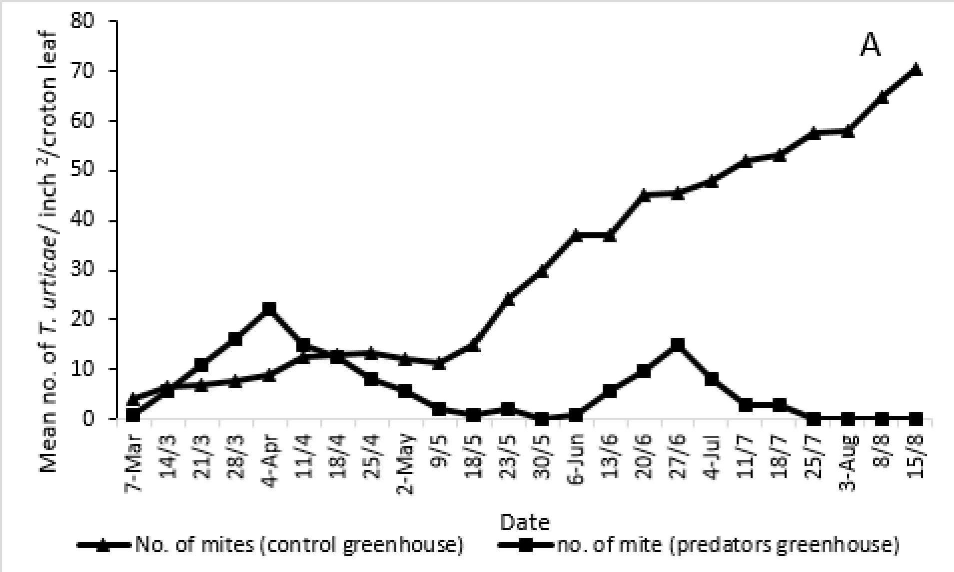


Figure 1

A timeline representing the release of predatory species *Phytoseiulus persimilis* and *Stethorus punctillum* to control the two-spotted mite *Tetranychus urticae* in a croton greenhouse



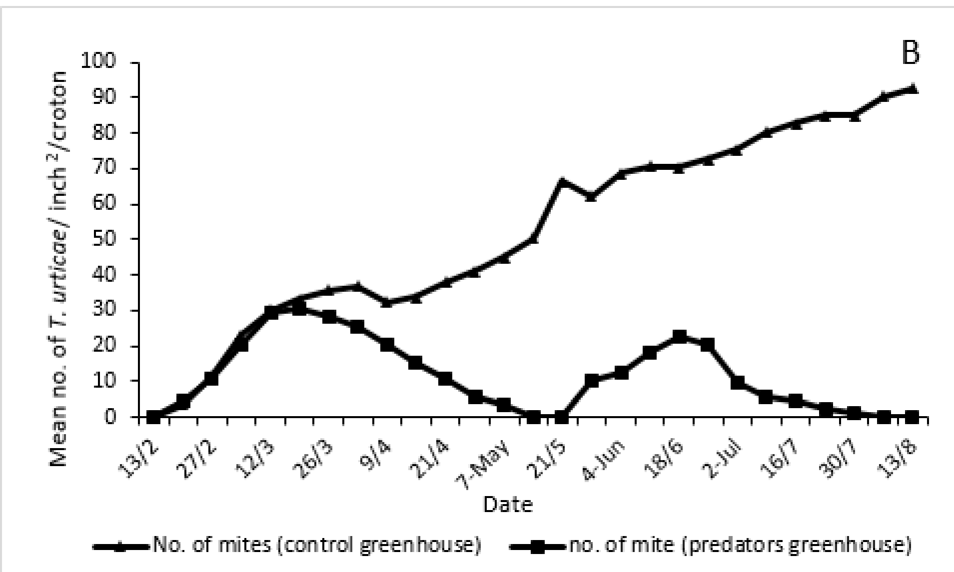


Figure 2

The comparison between the population of *T. urticae* in predators' greenhouse and control greenhouse, A) 2019 and B) 2020

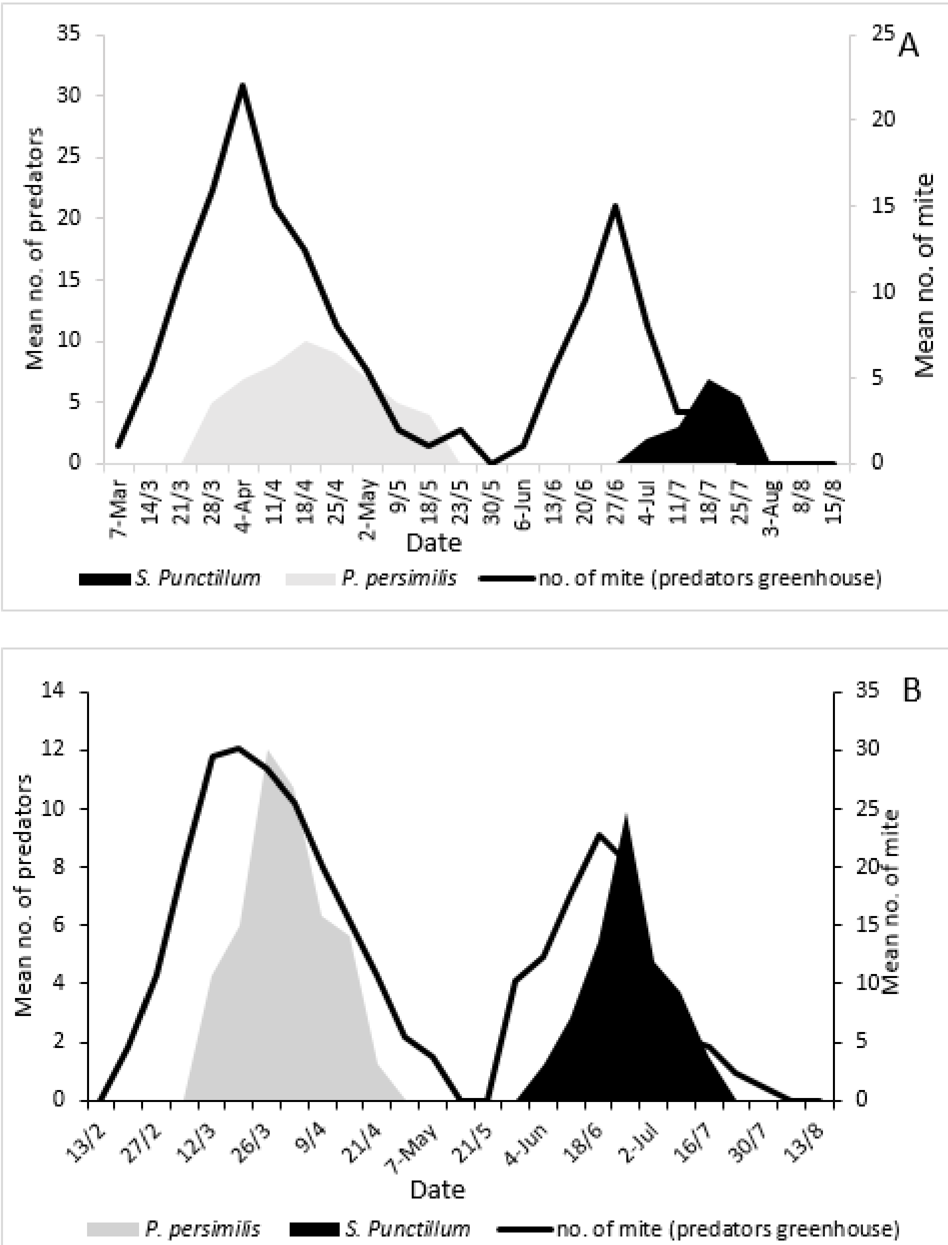


Figure 3

The combination between the population of *T. urticae* and the population of the two predators, *P. persimilis* and *S. punctillum* in predators' greenhouse, A) 2019 and B) 2020

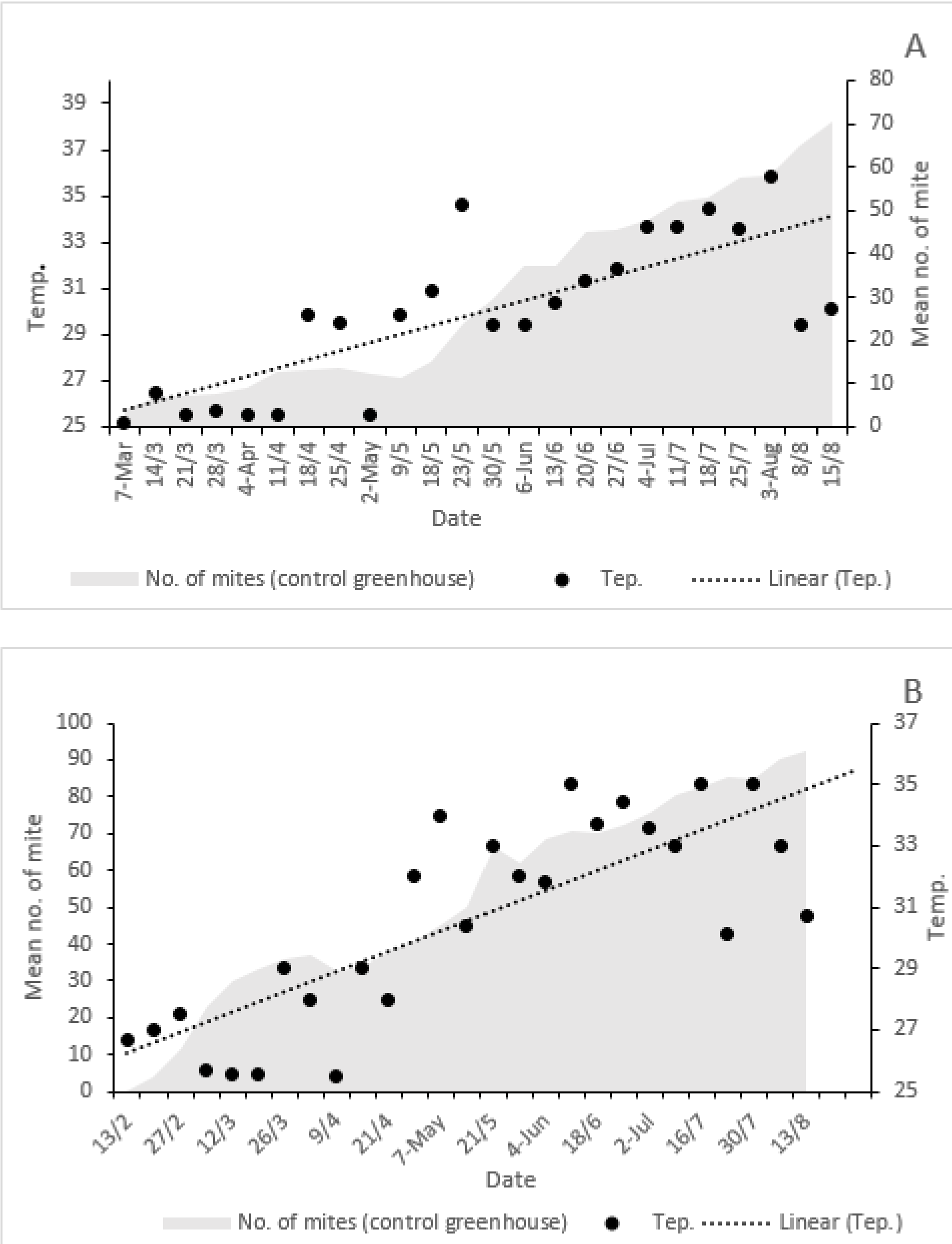
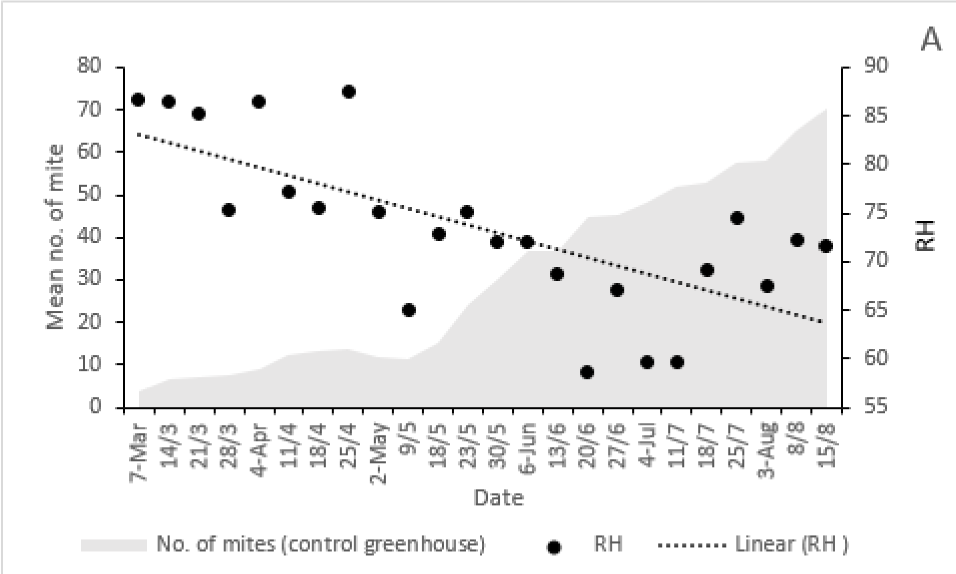


Figure 4

The relation between *T. urticae* population and temperature A) 2019 and B) 2020



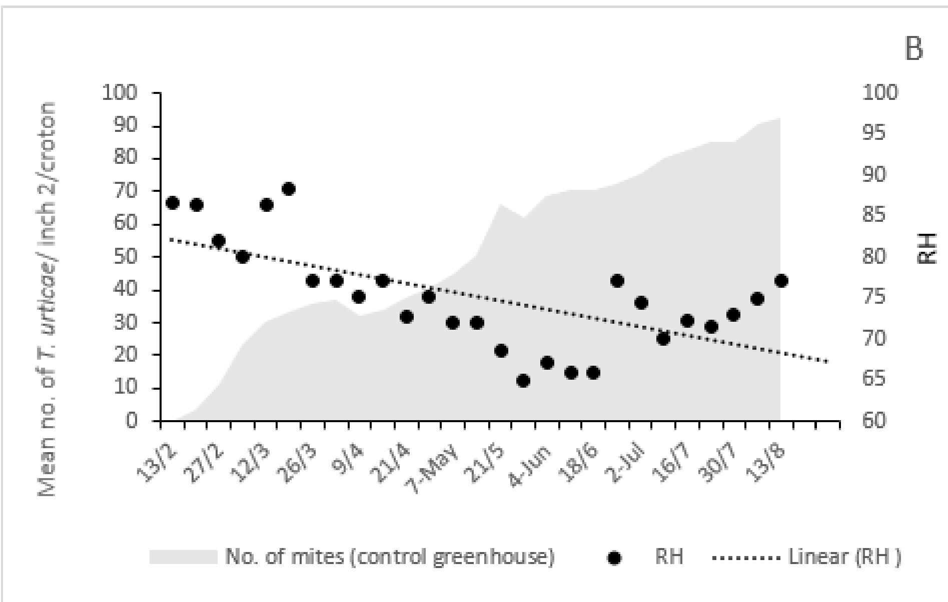


Figure 5

The relation between *T. urticae* population and relative humidity A) 2019 and B) 2020

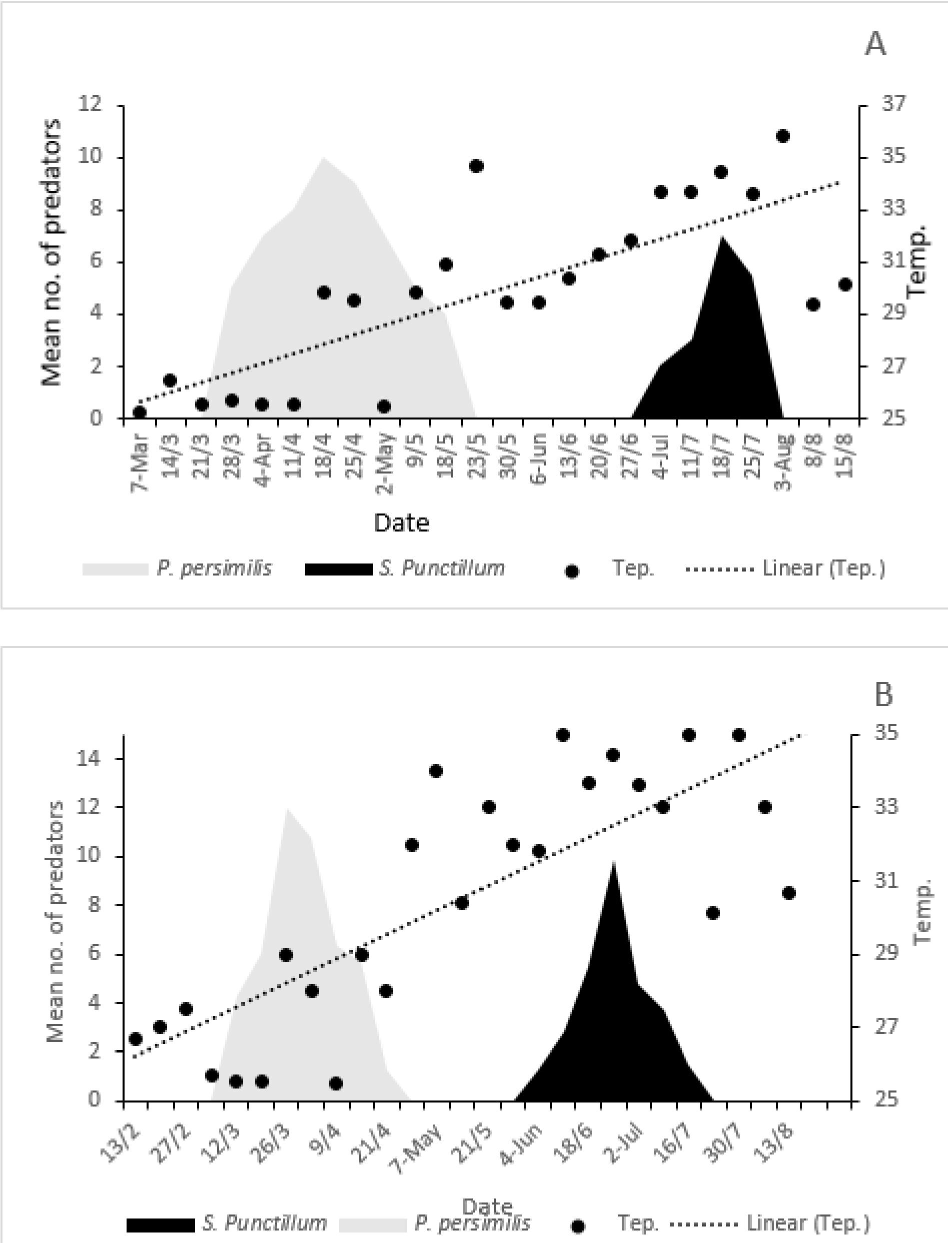
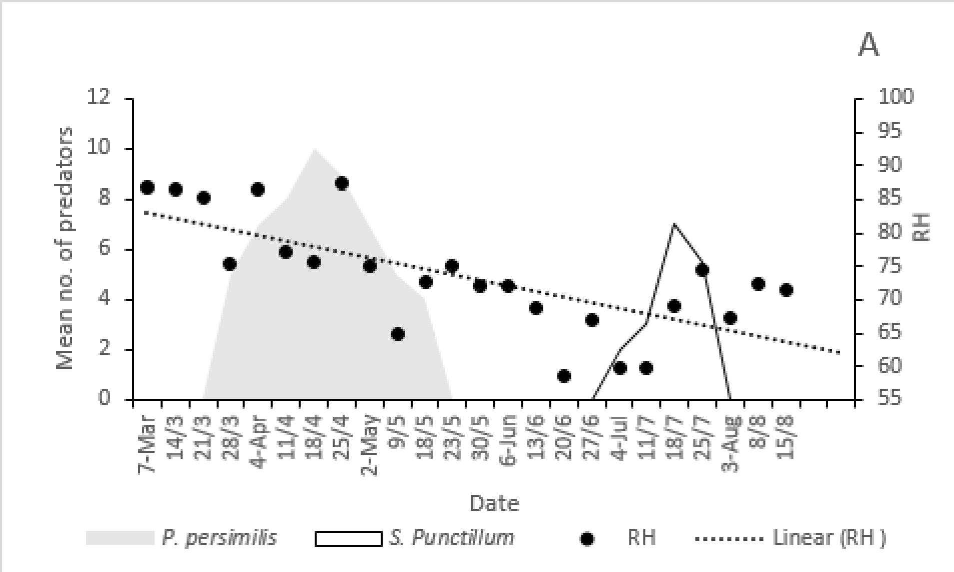


Figure 6

The relation between two predators, *P. persimilis* and *S. punctillum* population and temperature A) 2019 and B) 2020



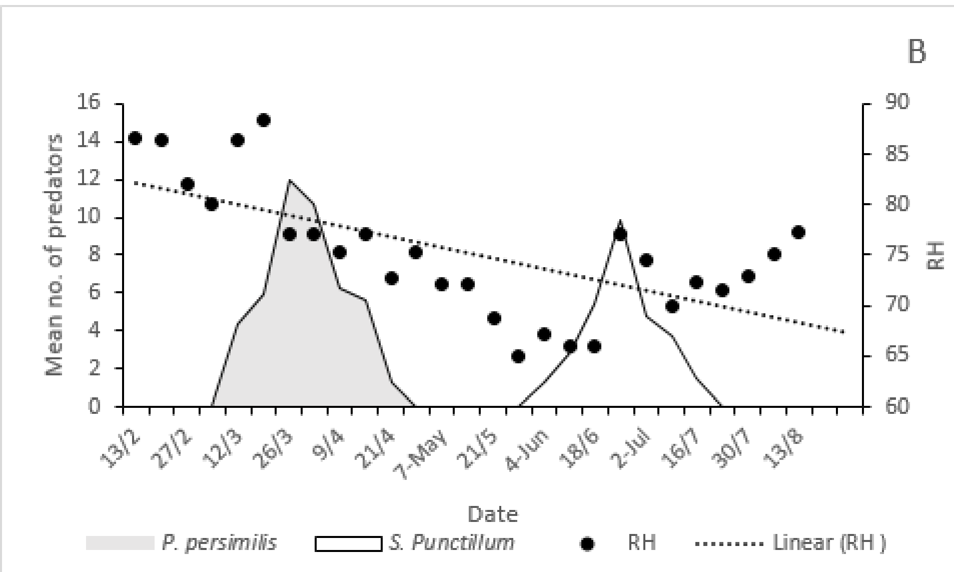


Figure 7

The relation between two predators, *P. persimilis* and *S. punctillum* population and relative humidity A) 2019 and B) 2020

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