

**Original article (Orijinal araştırma)**

**Contact toxicity of pine, laurel and juniper essential oils to spirodiclofen-resistant and -susceptible *Tetranychus urticae* Koch, 1836 (Acari: Tetranychidae) populations**

Çam, defne ve ardıç eterik yağlarının spirodiklofen'e dirençli ve hassas *Tetranychus urticae* Koch, 1836 (Acari: Tetranychidae) popülasyonlarına kontak toksisitesi

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**Abstract**

*Tetranychus urticae* Koch, 1836 (Acari: Tetranychidae) causes significant losses in agricultural production because of it is a polyphagous pest and develops resistance to pesticides in a short time. This study was conducted in 2017-2018 in Isparta, Turkey. Contact toxicities of pine, laurel and juniper essential oils to different developmental stages of a highly spirodiclofen-resistant and a spirodiclofen-susceptible population of *T. urticae* were investigated. The aim was to contribute to the development of alternative methods to control in resistant populations. The essential oil solutions were applied using a spray tower at 100 kPa to the leaf surface at 1.2-1.6 mg/cm<sup>2</sup>. The experiments were conducted with three replicates, with 15 individuals each treatment replicate. The highest mortality in adults was 100% for pine, juniper and laurel oil in the susceptible population, and 59.5% for pine oil, 57.5% for laurel oil and 51.2% for juniper oil in the spirodiclofen-resistant population. In addition, the highest mortality for nymphs was 81.6% for pine oil, 95.2% for laurel oil, 95.7% for juniper oil in the susceptible population, and 50.0% for pine oil, 56.3% for laurel oil and 58.0% for juniper oil in the spirodiclofen-resistant population. In toxicity tests on egg the highest mortality was lower than 55.0% in both populations. As a result, the three essential oils were showed to have a significant effect in the *T. urticae* population with high level resistance to spirodiclofen.

**Keywords:** essential oil, juniper, laurel, pine, spirodiclofen, *Tetranychus urticae*

**Öz**

*Tetranychus urticae* Koch, 1836 (Acari: Tetranychidae) polifag bir zararlı olması ve pestisitlere kısa sürede direnç geliştirmesinden dolayı tarımsal üretim alanlarında önemli kayıplara neden olmaktadır. Bu çalışmada 296 kat spirodiclofen dirençli ve hassas *T. urticae* popülasyonlarında çam, defne ve ardıç uçucu yağlarının zararlarının farklı dönemleri üzerindeki kontakt toksisiteleri araştırılmıştır. Denemeler 2017-2018 yılları arasında yürütülmüştür. Bu çalışmada dirençli popülasyonlarda alternatif mücadele yöntemlerinin geliştirilmesine katkı sağlanması amaçlanmıştır. Uçucu yağ çözeltileri ilaçlama kulesi yardımıyla 100 kPa basınçta yaprak yüzeyine 1.2-1.6 mg/cm<sup>2</sup> olacak şekilde püskürtülmüştür. Denemelerde her doz için 3 tekrerrür ve her tekrerrürde 15 birey kullanılmıştır. Uçucu yağların *T. urticae* popülasyonlarının her ikisinde de larva ve erginler üzerindeki en yüksek etkileri 20 ml/l konsantrasyonda ve 96. saat sonunda elde edilmiştir. Erginlerde en yüksek etki hassas popülasyon için, çam, ardıç ve defne yağında %100; spirodiclofen dirençli popülasyonda ise çam yağında %59.5, defne yağında %57.5, ardıç yağında %51.2 olarak belirlenmiştir. Larvalarda ise en yüksek etki hassas popülasyon için, çam yağında %81.6, defne yağında %95.2, ardıç yağında %95.7; spirodiclofen dirençli popülasyonda ise çam yağında %50.0, defne yağında %56.3, ardıç yağında %58.0 olarak belirlenmiştir. Toksikite testlerinde yumurtalar üzerindeki en yüksek ölüm oranları her üç uçucu yağ için de %55.0'ten düşük bulunmuştur. Sonuç olarak spirodiclofen'e yüksek seviyede dirençli bir *T. urticae* popülasyonunda bu üç uçucu yağın kayda değer bir etkisi olduğu bulunmuştur.

**Anahtar sözcükler:** uçucu yağ, ardıç, defne, çam, spirodiklofen, *Tetranychus urticae*

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## Introduction

*Tetranychus urticae* Koch, 1836 (Acari: Tetranychidae) is a major pest that feeds on about 1000 plants from 250 families around the world (Migeon & Dorkeld, 2011). To control this pest, chemical methods including the insecticide/acaricide applications with varying modes of action and formulation are used (Pavliidi et al., 2017). However, due to its parthenogenetic reproduction and short life cycle, *T. urticae* can develop resistance to pesticides after a few applications. For this reason, the research on alternative control methods for *T. urticae* has gained importance in recent years. As an alternative to synthetic pesticides some compounds extracted from plants are being investigated. Essential oils are the most important herbal products that can be used for pest control due to their monoterpene and diterpene content. Essential oils and their components are effect insect physiology, behavior and biology (Singh & Upadhyay, 1993). Also, it has been shown that essential oils have insecticidal, acaricidal, ovicidal, antifeedant and repellent effects on many harmful species (Govindarajan et al., 2016; Reddy et al., 2016). In comparison to synthetic pesticides, essential oils are substances that have minimal effect on human and non-target organisms, on ecological balance and do not lead to resistance development in the pests (Isman et al., 2011).

Pine oil and its components are widely used in the cosmetics, perfumery and foods industry as raw material and aroma (Yang et al., 2010). Laurel oil, usually obtained by hydrodistillation and steam distillation, is used in the cosmetic industry and in the treatment of some diseases in medicine (Hafizoglu & Reunanen, 1993). Juniper oil has diuretic and antiseptic effects due to containing terpene hydrocarbons, such as sabinene, thujone,  $\alpha$ -pinene, myrcene and limonene (Pepeljnjak et al., 2005). There have been some studies on the effects of pine, laurel and juniper essential oils on insects. Shaaya et al. (1997) evaluated the fumigant effect of laurel essential oil against to the pests, *Oryzaephilus surinamensis* (L., 1758) (Coleoptera: Silvanidae), *Sitophilus oryzae* (L., 1763) (Coleoptera: Curculionidae), *Rhyzopertha dominica* (F., 1792) (Coleoptera: Bostrichidae). Karci & Isikber (2007) report that the fumigant effects of pine, laurel and juniper essential oils on *Tribolium confusum* Jacquelin du Val, 1863 (Coleoptera: Tenebrionidae) eggs is low.

The mechanism of the contact effect of the essential oil and their components in insects is on the octopamine system. Octopamine receptors have a role in the central nervous system, such as neurotransmitters, neurohormones and neuromodulators. When an essential oil or its components contact the body of the insect, it is recognized by the octopamine system, which is followed by hyperactivity, the heart rate accelerates and excessive stress occurs in legs and abdomen and then the insect rapidly falls to ground and dies. These symptoms are reported to be generated by octopamine receptors (Kostyukovsky et al., 2002).

Spirodiclofen is located within the spirocyclictetronicacaricide group (in Group 23 of the IRAC MoA list). Spirodiclofen, and is efficacious against pest mite species such as Tetranychidae. It reduces fertility and the number of eggs deposited by females (IRAC, 2019).

The aim of this study was to contribute to the development of alternative control methods for resistant *T. urticae* populations. The contact toxicity of different concentrations of pine, laurel and juniper essential oils on different developmental stages of spirodiclofen-resistant and spirodiclofen-susceptible populations of *T. urticae* were investigated.

## Material and Methods

### Populations and rearing

Spirodiclofen-resistant and spirodiclofen-susceptible populations of *T. urticae* were used in this study (conducted in 2017-2018 in Isparta, Turkey). The susceptible population of *T. urticae* was obtained

in 2001 from Rothamsted Experimental Station (UK) and long-term rearing has continued in a climate room. The spiroadiclofen-resistant population of *T. urticae* was collected from a pepper greenhouse in which intensive chemical control was applied in 2017. *Tetranychus urticae* populations were rearing on bean plants at  $26\pm 2^{\circ}\text{C}$ , 50-60% RH, and 16:8 h L:D photoperiod conditions in a controlled climate room.

### Acaricide and essential oils

A commercial acaricide, Envidor<sup>®</sup> SC 240 (Bayer Crop-Science), with spiroadiclofen as the active ingredient was used.

The names, scientific names and families of the pine, laurel and juniper oils used in the study are given in Table 1. Pine, laurel and juniper oils were provided by a commercial supplier, Botalife Company (Turkey). The essential oils had been obtained by hydro-Clevenger method and the green parts of the plants was used. The components of the essential oils were not determined.

Table 1. Plant essential oils tested

| Essential oils |                            |              |
|----------------|----------------------------|--------------|
| Name           | Scientific name            | Family       |
| Pine oil       | <i>Pinus sylvestris</i>    | Pinaceae     |
| Laurel oil     | <i>Laurus nobilis</i>      | Lauraceae    |
| Juniper oil    | <i>Juniperus oxycedrus</i> | Cupressaceae |

### Acaricide bioassay for spiroadiclofen

LC<sub>50</sub> values of susceptible and resistant *T. urticae* populations were determined in order to determine to spiroadiclofen resistance ratio. The spiroadiclofen dose that gives 90% mortality was chosen as the first dose. Seven doses were prepared by two-fold serial dilution from the first dose. All the bioassay tests were conducted as a control plus seven spiroadiclofen doses, all with three replicates. To provide sufficient humidity, 3-cm diameter bean leaf discs were placed into 9 cm diameter Petri dishes which had a saturated cotton in the base. Twenty-five adult female individuals were transferred onto the leaf discs. Two mL of the acaricide doses were applied by spray tower at a pressure of 100 kPa. Only pure water was applied to the control. Mortality assessment was made after 24 h.

### Contact toxicity tests for essential oils

In contact toxicity tests, the Miresmailli et al. (2006) method was used for adults and nymphs, and Badawy et al. (2010) method for eggs. The same stage, eggs, nymphs or adults, were used. In the experiments, 15 adult female individuals were transferred to a prepared Petri dish as described above. Concentrations of 1, 5, 10 and 20 mL/l of pine, laurel and juniper essential oils were used. The essential oils were dissolved in 0.3% Tween 20 solution prepared with purified water. For the control, only the Tween solution was used. All the experiments were conducted with three replicates each containing 15 individuals. The essential oil concentrations were sprayed onto the leaf surface at 1.2-1.6 mg/cm<sup>2</sup> at 100 kPa using a spray tower (Mansour et al., 1986). Mortality assessments for nymphs and adults were made after 24, 48 and 96 h. For eggs, the assessments were made when all eggs had hatched in the control.

### Statistical analysis

LC<sub>50</sub> values were calculated in the POLO computer package program (LeOra Software, 1994). The spiroadiclofen resistance ratio was determined by the ratio of the LC<sub>50</sub> value of the resistant and susceptible populations. The mortality percentages from the contact toxicity experiments were calculated using the Abbott formula (Abbott, 1925). The data were arcsin transformed. The contact effects of

essential oils on the nymph and adult stages of the pest were analyzed using three-way ANOVA. In order to determine the toxic effect of the essential oils on the eggs, two-way ANOVA was used for essential oil and concentration data. Tukey multiple comparison test was used to compare the differences between the means.

## Results

### Resistance ratio in *Tetranychus urticae* greenhouse population

LC<sub>50</sub> values determined against spiroadiclofen for the field population and for the susceptible population are given in Table 2. A high level (296-fold) of spiroadiclofen-resistance was determined for the *T. urticae* population collected from pepper greenhouse.

Table 2. LC<sub>50</sub> values determined against spiroadiclofen in resistant and susceptible populations of *Tetranychus urticae*

| Population  | n*  | Slope±SE  | LC <sub>50</sub> (mg a. i l <sup>-1</sup> ) | R** |
|-------------|-----|-----------|---|-----|
| Resistant   | 605 | 2.35±0.37 | 210<br>(143-324)                            | 296 |
| Susceptible | 607 | 2.72±0.46 | 0.71<br>(0.30-0.99)                         | -   |

\*: number of individuals used in the experiment;

\*\* : resistance ratio.

### Contact toxicity of the essential oils on *Tetranychus urticae* populations

#### Contact toxicity on adult

The contact toxicity on the adults in the susceptible and resistant populations of essential oils are given in Table 3. The effects on adults for all concentrations of essential oils were found to be higher in the susceptible population than the resistant population. Highest mortality of adults with the essential oils was at 20 mL/l after 96 h. In the susceptible population, 100% mortality was observed at 20 mL/l after 96 h for all three essential oils. In the resistant population, the mortality was 59.5% for pine oil, 57.5% for laurel oil and 51.2% for juniper oil.

#### Contact toxicity on nymph

The contact toxicity on the nymph in the susceptible and resistant populations of essential oils are given in Table 4. The contact toxicity of essential oils on the nymphs of *T. urticae* were found to be similar to the adults. For both populations, when the concentrations of essential oils increased, the mortality on nymphs increased. However, the mortality of the resistant nymphs was found to be lower than the susceptible nymphs. According to the concentrations of 20 mL/l of essential oils after 96 h counting results the mortality was 92.6%, 95.2% and 98.4% for pine oil, laurel oil and juniper oil, respectively, in the susceptible population, and 50.0%, 56.3%, and 58.0% for pine oil, laurel oil and juniper oil, respectively, in the resistant population.

Table 3. Contact toxicity of essential oils on adults of susceptible and resistant populations of *Tetranychus urticae*

| Time (hour) | Concentration (ml/l) | Mortality (%) |     |             |    |             |    |
|-------------|----------------------|---------------|-----|-------------|----|-------------|----|
|             |                      | Pine          |     | Laurel      |    | Juniper     |    |
| Susceptible |                      |               |     |             |    |             |    |
| 24          | 1                    | 24.49±0.45    | aD* | 4.45±0.25   | cE | 10.64±0.33  | bF |
|             | 5                    | 34.69±0.25    | bC  | 8.89±0.85   | cD | 48.94±2.15  | aD |
|             | 10                   | 48.98±0.65    | bC  | 8.89±0.55   | cD | 68.09±1.45  | aC |
|             | 20                   | 77.55±0.48    | bB  | 46.67±0.24  | cB | 100.00±0.65 | aA |
| 48          | 1                    | 97.73±1.15    | aA  | 11.36±1.65  | bD | 20.00±0.65  | bE |
|             | 5                    | 97.73±0.23    | aA  | 15.91±1.45  | cD | 66.67±0.95  | bC |
|             | 10                   | 100.00±0.65   | aA  | 27.27±0.89  | cC | 82.22±0.75  | bB |
|             | 20                   | 100.00±0.45   | aA  | 90.91±0.48  | aA | 100.00±0.23 | aA |
| 96          | 1                    | 100.00±0.25   | aA  | 30.95±0.88  | bC | 37.78±0.35  | bD |
|             | 5                    | 100.00±0.55   | aA  | 30.95±0.25  | bC | 95.56±1.75  | aA |
|             | 10                   | 100.00±1.25   | aA  | 90.48±0.65  | aA | 100.00±0.25 | aA |
|             | 20                   | 100.00±0.63   | aA  | 100.00±0.75 | aA | 100.00±0.45 | aA |
| Resistant   |                      |               |     |             |    |             |    |
| 24          | 1                    | 0.00±0.00     | bE  | 0.00±0.00   | bE | 2.13±0.35   | aF |
|             | 5                    | 0.00±0.00     | cE  | 2.33±0.45   | bD | 6.38±0.55   | aE |
|             | 10                   | 2.17±0.25     | bD  | 2.33±0.65   | bD | 8.51±0.75   | aD |
|             | 20                   | 4.35±0.55     | bD  | 13.95±1.25  | aC | 10.64±1.45  | aD |
| 48          | 1                    | 7.50±0.75     | aD  | 2.33±1.35   | cD | 4.65±1.75   | bE |
|             | 5                    | 25.00±0.56    | aC  | 4.35±0.75   | cD | 10.25±0.65  | bD |
|             | 10                   | 30.00±0.68    | aC  | 9.30±0.25   | cC | 16.28±0.33  | bC |
|             | 20                   | 42.50±0.25    | aB  | 34.38±2.15  | bB | 16.28±0.70  | cC |
| 96          | 1                    | 29.73±1.45    | aC  | 7.50±1.10   | bC | 20.93±0.65  | aC |
|             | 5                    | 40.54±0.70    | aB  | 42.50±0.75  | aB | 23.46±1.15  | bC |
|             | 10                   | 45.95±0.65    | bB  | 60.00±0.44  | aA | 39.53±0.45  | bB |
|             | 20                   | 59.46±0.60    | aA  | 57.50±0.78  | aA | 51.16±0.95  | aA |

\*: Different lowercase letters in the same line and different uppercase letters in the same column indicate that the means are significantly different according to essential oils and application doses, respectively ( $p < 0.05$ ).

Table 4. Contact toxicity of essential oils on nymphs of susceptible and resistant populations of *Tetranychus urticae*

| Time (hour) | Concentration (ml/l) | Mortality (%) |     |            |    |            |    |
|-------------|----------------------|---------------|-----|------------|----|------------|----|
|             |                      | Pine          |     | Laurel     |    | Juniper    |    |
| Susceptible |                      |               |     |            |    |            |    |
| 24          | 1                    | 13.95±1.25    | bE* | 22.92±0.75 | aE | 20.41±0.45 | aE |
|             | 5                    | 53.49±0.25    | bC  | 64.58±0.46 | aC | 67.35±0.75 | aC |
|             | 10                   | 55.81±0.55    | bC  | 70.83±0.85 | aB | 69.39±0.15 | aC |
|             | 20                   | 62.79±0.85    | bB  | 79.17±0.43 | aB | 79.59±0.65 | aB |
| 48          | 1                    | 18.60±0.88    | cD  | 29.79±0.54 | bE | 36.73±0.48 | aD |
|             | 5                    | 67.44±0.68    | aB  | 70.21±0.64 | aB | 71.43±0.35 | aB |
|             | 10                   | 74.42±0.15    | bB  | 76.60±0.25 | bB | 83.67±0.13 | aB |
|             | 20                   | 76.74±0.78    | bB  | 82.98±1.45 | aA | 87.76±0.78 | aB |
| 96          | 1                    | 23.68±0.65    | cD  | 47.62±1.74 | aD | 36.96±0.36 | bD |
|             | 5                    | 68.42±0.35    | bB  | 83.33±1.58 | aA | 78.26±0.98 | aB |
|             | 10                   | 81.58±0.25    | bA  | 85.71±0.35 | bA | 91.30±0.25 | aA |
|             | 20                   | 92.58±1.35    | aA  | 95.24±0.75 | aA | 98.41±0.35 | aA |
| Resistant   |                      |               |     |            |    |            |    |
| 24          | 1                    | 6.00±0.25     | bE  | 10.00±0.25 | aD | 9.80±0.350 | aE |
|             | 5                    | 14.00±0.36    | bD  | 24.00±0.45 | aC | 11.76±0.45 | bE |
|             | 10                   | 32.00±0.78    | aB  | 36.00±0.85 | aB | 27.45±0.75 | bC |
|             | 20                   | 40.00±0.55    | bB  | 52.00±0.65 | aA | 39.22±1.45 | bB |
| 48          | 1                    | 8.00±1.15     | bE  | 14.00±0.23 | aD | 10.00±1.75 | bE |
|             | 5                    | 18.00±1.85    | bD  | 28.00±1.15 | aC | 14.00±0.25 | bE |
|             | 10                   | 36.00±0.55    | aB  | 40.00±0.85 | aB | 30.00±0.36 | bC |
|             | 20                   | 48.00±0.36    | bA  | 56.00±0.45 | aA | 44.00±0.46 | bB |
| 96          | 1                    | 12.00±0.74    | aD  | 14.58±0.78 | aD | 14.00±0.75 | aE |
|             | 5                    | 26.00±0.89    | aC  | 29.17±0.89 | aC | 20.00±0.15 | bD |
|             | 10                   | 38.00±1.85    | bB  | 45.83±0.25 | aB | 34.00±0.25 | bC |
|             | 20                   | 50.00±0.48    | bA  | 56.25±0.15 | aA | 58.00±0.45 | aA |

\*: Different lowercase letters in the same line and different uppercase letters in the same column indicate that the means are significantly different according to essential oils and application doses, respectively ( $p < 0.05$ ).

### Contact toxicity on egg

The contact toxicity on egg stage in the susceptible and resistant populations of essential oils are given in Table 5. The contact toxicity on egg stage of pine, laurel and juniper essential oils in susceptible population were found to be higher than the resistant population. At 20 mL/l, the mortality was 48.9, 48.9 and 55.3% for juniper oil, pine oil and laurel oil, respectively, in the eggs of susceptible population, and 14.9, 23.3 and 41.1% for juniper oil, pine oil and laurel oil, respectively, in the eggs of resistant population.

Table 5. Contact toxicity of essential oils on egg stage of susceptible and resistant populations of *Tetranychus urticae*

| Concentration (ml/l) | Mortality (%) |     |            |    |            |    |
|----------------------|---------------|-----|------------|----|------------|----|
|                      | Pine          |     | Laurel     |    | Juniper    |    |
| Susceptible          |               |     |            |    |            |    |
| 1                    | 19.15±0.35    | aC* | 0.00±0.25  | bD | 21.28±0.75 | aD |
| 5                    | 23.46±0.78    | aB  | 11.11±0.85 | bC | 29.79±0.15 | aC |
| 10                   | 42.55±1.12    | aA  | 37.78±0.43 | bB | 46.81±0.45 | aB |
| 20                   | 48.94±1.63    | bA  | 48.89±0.45 | bA | 55.32±0.33 | aA |
| Resistant            |               |     |            |    |            |    |
| 1                    | 4.26±1.45     | bC  | 2.33±0.75  | cC | 14.89±1.25 | aC |
| 5                    | 10.64±0.45    | cB  | 16.28±0.45 | bB | 28.30±1.85 | aB |
| 10                   | 10.64±0.36    | cB  | 19.95±0.15 | bB | 31.06±0.25 | aB |
| 20                   | 14.89±0.85    | cA  | 23.25±1.15 | bA | 41.06±0.55 | aA |

\*: Different lowercase letters in the same line and different uppercase letters in the same column indicate that the means are significantly different according to essential oils and application doses, respectively ( $p < 0.05$ ).

### Discussion

Plant essential oil-containing pesticides or components can affect some pathogenic fungi that cause pre- and post-harvest diseases, agricultural pests, pests of stored products and urban pests (Koul et al., 2008). Essential oils consist of a mixture of hydrocarbons, terpenes, aldehydes, ketones, alcohols and phenol-like compounds. Each of these components, or their mixtures, can cause toxicity, repellent, behavioral, antifeeding and reproductive effects on some arthropod species (Lawless, 2002). Essential oils and their components are particularly suitable for use in IPM applications, as they cause many effects on pest species but are not deleterious to non-target organisms.

Studies showing that essential oils obtained from different plants are effective on phytophagous mite species and can be used as an alternative to synthetic acaricides. Lee et al. (1997) reported some of the monoterpenoids were lethal to the *T. urticae* at high concentrations; specially carvomenthenol and terpinen-4-ol. Rasikari et al. (2005) investigated contact toxicity of extracts from 67 species from six subfamilies of Australian Lamiaceae to *T. urticae* and determined some of the extracts had acaricidal effects. However, the number of studies on the effects of essential oils on acaricide-resistant mite populations has been limited. Han et al. (2010) reported essential oils from citronella Java, clover leaf, lemon eucalyptus, pennyroyal, peppermint and thyme showed acaricidal activity in the abamectin-resistant *T. urticae* population. In a study on eucalyptus oil components, menthol,  $\beta$ -citronellol and citral components were found to be effective in the chlorfenapyr-resistant CRT-53 population, geranyl acetate, citronellal and  $\alpha$ -terpinene components in the fenpropathrin-resistant FRT-53 population, and citronellyl acetate, citral, eugenol and geraniol components in the pyridaben-resistant PRT-53 population (Han et al., 2011).

Rauch and Nauen (2003) and Van Pottelberge et al. (2009) determined the resistance against spirodiclofen developed as a result of selection pressure in *T. urticae*. A highly spirodiclofen-resistant (296-fold) *T. urticae* population used this study. The findings showed that the acaricidal activity of pine, laurel and juniper oils in the resistant population was consistent with the literature. Therefore, it is considered that the spraying method leads to the toxic effect in susceptible and resistant populations of *T. urticae*, and that this method is easily applicable, especially in greenhouse production areas.

In order for plant-based products, such as essential oil, to be used for insecticide resistance management in *T. urticae*, the resistance mechanisms and the mode of action of insecticide resistance should be known. The resistance mechanisms against acaricides in *T. urticae* are usually the formation of target-site insensitivity, such as decreased sodium channel sensitivity caused by pyrethroids, or decreased acetylcholinesterase sensitivity caused by organic phosphates (Knowles, 1997). In the literature it has been reported that the effects of essential oils are on the octopaminergic system and GABA receptors (Kostyukovsky et al., 2002; Priestley et al., 2003). However, more studies on the effects of essential oils to these target regions are needed, and the possibility of resistance development must be considered.

In conclusion, pine, laurel and juniper oils had a greater effect on the susceptible population than on the spirodiclofen-resistant population of *T. urticae*, but the effect on the resistant population was significant. Provided no side effects to natural enemies and phytotoxicity are found, essential oils could prove useful for control of spirodiclofen-resistant populations of *T. urticae*, especially in greenhouses.

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