Effect of pyroligneous extract of *Acacia mearnsii* on *Tetranychus urticae* (Koch, 1836) (Acari, Tetranychidae) and *Neoseiulus californicus* (McGregor, 1954) (Acari, Phytoseiidae)

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Resumo

Efeito do extrato pirolenhoso de Acacia mearnsii sobre Tetranychus urticae (Koch, 1836) (Acari: Tetranychidae) e Neoseiulus californicus (McGregor, 1954) (Acari: Phytoseiidae). Este trabalho teve como objetivo determinar o efeito do Extrato Pirolenhoso Decantado (EPD) de acácia-negra sobre Tetranychus urticae (Koch, 1836) (Acari: Tetranychidae) e seu predador, Neoseiulus californicus (McGregor, 1954) (Acari: Phytoseiidae). Seis diferentes concentrações foram testadas, nomeadas, EPD: água 1:600, 1:300, 1:150, 1:75, 1:38 e 1:19 e o tratamento controle com água destilada. Cinco fêmeas de T. urticae e N. californicus foram transferidas para arenas, sendo mantidas por um período de oito dias, totalizando cinquenta fêmeas. O EPD causou alta mortalidade para N. californicus, enquanto que para T. urticae a mortalidade foi significativa em altas concentrações (1:75, 1:38 e 1:19) e em apenas uma concentração intermediária (1:150). Para T. urticae, o EPD provocou baixa mortalidade em baixas concentrações. Para N. californicus, o EPD foi prejudicial em todas concentrações, demonstrando ser prejudicial a este ácaro predador e pouco eficiente no controle de T. urticae.

Palavras-chave: Acacia mearnsii; MIP; Seletividade

Abstract

This work aimed to determine the effect of decanted pyroligneous extract (DPE) of black *Acacia mearnsii* on the mites *Tetranychus urticae* (Koch, 1836) (Acari: Tetranychidae) and *Neoseiulus californicus* (McGregor, 1954) (Acari: Phytoseiidae). Six different concentrations were tested, namely, DPE: water 1:600, 1:300, 1:150, 1:75, 1:38 and 1:19 and a control treatment with distilled water. Five females of *T. urticae* and *N. californicus* were transferred to arenas, totaling 50 females per treatment, for a period of eight days. DPE caused high mortality in *N. californicus*, while in *T. urticae*, high mortality was observed only at high (1:75, 1:38 and 1:19)

and intermediate (1:150) concentrations. Notably, DPE was slightly harmful to *T. urticae* at lower concentrations and harmful to *N. californicus* at all concentrations, indicating that it would have little efficacy in the control of *T. urticae*.

Key words: Acacia mearnsii; IPM; Selectivity

Introduction

The smoke resulting from the burning of wood in charcoal production kilns is the main residue of this economic activity, which is released into the atmosphere, acting as a pollutant. Gas emission-reducing processes are usually not used of. However, there is a way to use the smoke and residue produced to prepare a decanted pyroligneous extract (DPE). Accordingly, the smoke is collected and transformed by condensation into a liquid, which has a variety of potential uses in agriculture, such as in eucalyptus (SILVA et al., 2006) and lettuce (MASCARENHAS et al., 2006) crops. In addition, when the extract is produced, the pollution caused by kilns is reduced. According to Miyasaka et al. (2001), crude DPE cannot be used on agricultural crops without undergoing a purification process, mainly to eliminate the tar, which is solubilized immediately after obtaining the product.

The control of pests and disease in plants, including mites, has been done with pesticide application, which can select resistant individuals and may contribute to increasing production costs, and in such case, the products cannot be sold as organics (ALVES et al., 2005). DPE applied in 300- to 600-fold dilutions is promising in the control of pests and diseases and may be used alone or combined with other plant extracts (MAEKAWA, 2002). From this point of view, the use of DPE could be an alternative in integrated pest management (IPM). Besides the effects reported for DPE, little scientific information is available to support its use and to elucidate its mechanisms of action, especially in relation to the protection of plants against pests and diseases (CAMPOS NETO et al., 1993).

Phytophagous mites are among the groups most studied due to their agricultural importance and because they are found on various plant species of economic importance, causing potential damage to them (MORAES; FLECHTMANN, 2008). *Tetranychus* *urticae* is considered one of the main pests that attack various crops in Brazil, including cotton, grapevines, strawberry and others. In general, the damage caused by this species is featured by flecking, discoloration (bronzing) and scorching of leaves. Injury can lead to loss of leaves and even plant death. For their control, large quantities of pesticides are used by farmers (MORAES; FLECHTMANN, 2008).

Phytoseiid mites are predators of spider mites and other small mites and insects on plants. Several members of this family are of great importance in the biological control of spider mites and thrips in greenhouse crop production (ZHANG, 2003). *Neoseiulus californicus*, a phytoseiid mite commonly found in native plants and some crops in the state of Rio Grande do Sul (FERLA et al., 2007; MARCHETTI; FERLA, 2011), is a selective predator of tetranychid mites (McMURTRY; CROFT, 1997), and it is used commercially as a biological control agent in Brazil and other countries (WATANABE et al., 1994; FERLA et al., 2007; MCMURTRY et al., 2013).

Due to the importance of the mites in agroecosystems, the results obtained in this study may support control strategies that take into account the presence of natural enemies in agricultural environments or that aim to preserve natural enemies using alternative products to control mite pests. The aim of this study was to evaluate in the laboratory the action of DPE on *T. urticae* and its selectivity, with regard to its predator, *N. californicus*.

Material and Methods

The study was conducted in the Acarology Laboratory of UNIVATES University Center, Lajeado, state of Rio Grande do Sul, between July and October of 2010. The colonies were started with *T. urticae* specimens from strawberry plants (*Fragaria* sp.), from Lajeado County, Rio Grande do Sul State and *N. californicus* from a grapevine of the Merlot variety, from Bento Gonçalves County, state of Rio Grande do Sul. Both species were collected in 2010 February and kept in the laboratory on bean leaves (*Phaseoulus vulgaris* L.).

Neoseiulus californicus colonies were kept in rearing units on a sponge moistened with distilled water in plastic trays with bean leaves infested with *T. urticae*, covered with cardboard. The units were kept in a climatic chamber at 25 ± 1 °C, $80 \pm 5\%$ RH and 12-h photoperiod. *Tetranychus urticae* rearing was carried out in similar trays on bean leaves.

DPE of acacia charcoal (*Acacia mearnsii* De Wild.) obtained from acacia wood burning was stored in a polyethylene drum, covered and allowed to stand for about 100 days to obtain the aqueous phase of the product. It was tested at six concentrations, diluted in water: 1:600; 1:300, 1:150, 1:75, 1:38 and 1:19. Distilled water was used as the control.

In the experiment, we used fertilized females of *N. californicus* and *T. urticae*, respectively seven and fifteen days old. *Tetranychus urticae* was kept in arenas of 2.0 cm in diameter cut into bean leaves, while *N. californicus* fed on *T. urticae* throughout the experiment.

Arenas were cut and immersed in the DPE solutions and control for approximately eight seconds, withdrawn and dried at room temperature on a paper towel for about one hour. The dried treated arenas were placed on a moistened sponge in a Petri dish of 6.5 cm in diameter. Five females, apparently healthy, were removed from the rearing colonies and transferred to an arena using a fine paint brush. The experimental design was completely randomized with ten replicates with five mites per replicate.

After the females were transferred to treated circles, the evaluation was daily, where we counted the number of dead females and the eggs laid during a period of eight days. The eggs laid were transferred from the treated arenas to discs also treated to evaluate viability.

The effect of DPE was evaluated as female mortality in the treatment, where mortality was corrected by the control for *N. californicus* and *T. urticae*. The effect on reproduction was determined only with *N. californicus* (OVERMEER; VANZON, 1982; HASSAN et al., 1985; OVERMEER, 1988; BAKER et al., 1992b). The formula used to calculate total effect was $E = 100\% - (100\% - M_c) \times E_r$, in which E = total effect, $M_c =$ corrected mortality (ABBOTT, 1925) and $E_r =$ reproduction effect. The reproduction effect was calculated as oviposition of females in each treatment divided by mean control oviposition ($E_r = R_{treatment}/R_{control}$). The mean oviposition of females (R) was obtained using the formula: R = total viable eggs/number of females alive at the end of the test.

The total effect obtained for each product was classified on a scale of 1 to 4, according to the criteria established by IOBC/WPRS (BAKER et al., 1992a; HASSAN et al., 1994): class 1: E < 30% (innocuous, harmless); class 2: 30% < E < 79% (slightly harmful); class 3: 80% < E < 99% (moderately harmful) and class 4: E > 99% (harmful).

The results for corrected mortality were subjected to ANOVA and the means compared with the Tukey test at 5% probability with the Bioestat 5.0 statistics program.

Results and Discussion

DPE caused high mortality in *N. californicus* (100%) at all concentrations studied, while in *T. urticae*, only 1:150 and higher concentrations caused high mortality (Table 1). At concentrations of 1:600 and 1:300, *T. urticae* mortality was lower, i.e., 70 and 54%, respectively. The treatment with low concentration, beyond to show reduced effect on *T. urticae* mortality. These treatments proved to be sublethal to this phytophagous mite, and their physiologic effect was increased fecundity. According to this work, DPE could eliminate a natural enemy of *T. urticae*. In the presence of abundant food and absence of natural enemies, there would be an increase in populations of pest species. Thus, these results suggest limiting the use of DPE, but further studies are needed, mainly in the field.

All treatments were shown to be harmful to *N. californicus*. The results obtained demonstrated that DPE at the concentrations used was not selective for *N. californicus*, thus opening the possibility to test this product at lower concentrations. However, we also found that these lower concentrations were innocuous to *T. urticae*, invalidating their use in IPM.

Comparing the two species evaluated, DPE showed, in general, a greater harmful effect on the predator species than phytophagous species. Against *T. urticae*, 1:600, 1:300 and 1:75 DPE was slightly harmful, while the other concentrations were considerably harmful, and thus, DPE toxicity increased with concentration, except for 1:75. Against *N. californicus*, DPE was harmful at all concentrations.

By the fifth day, for *T. urticae*, the highest concentrations caused complete mortality, and at 1:150, just by the seventh day, while the other concentrations did not cause complete mortality. In *T. urticae* oviposition, the greatest effect was observed at the highest concentration, where the number of eggs laid/day by females was lower than that at other concentrations. During the eight-day observation period, there were small oscillations in the number of eggs at every concentration, with a gradual decrease, except at the 1:75, where there was an increase. At the highest concentration, 1:19, a daily average of 5.5 eggs was observed, compared to 11 eggs per day in the control; in other words, in presence of DPE there was a fifty percent reduction in oviposition.

All predator mites died at the end of the study. During the observation period, the oviposition rate was affected by DPE, where it was lowest at the highest concentration. Regarding the mortality of *N. californicus*, we found that although all predators died at the end of the test, there was variation in the time of complete mortality, i.e., DPE action was more rapid at the higher concentrations. At the lowest concentration (1:600), complete mortality was reached by the eighth day.

The effect of DPE on *N. californicus* oviposition was substantial, where there was complete mortality on the second day at the highest concentration (1:19), with no oviposition observed. Egg number increased with decreasing concentration. Immediately before reaching complete mortality, there was an increase in egg number at the lower concentrations. This increase in oviposition may have been due to the females being in an adverse environment and as a response to enhance fecundity. This phenomenon may be explained by the sublethal action of DPE on biological parameters of this predatory mite, resulting in physiological changes that can alter fertility, reducing it (O'BRIEN et al., 1985)

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or enhancing it (ATALLAH; NEWSON, 1966; ABLES et al., 1977).

Comparing the effects of DPE on *T. urticae* and *N. californicus* oviposition, there was a greater effect on *N. californicus*, because all concentrations showed a lower number of eggs/day. Ferla and Moraes (2006) studied the effects of different insecticides and acaricides on predator mites *Euseius concordis* (Chant, 1959) and *N. anonymus* (Chant & Baker, 1965) to evaluate selectivity, and also found that the majority of products were not selective and caused higher toxicity with increasing concentrations.

Alves et al. (2007) observed that DPE caused significant mortality in *Brevipalpus phoenicis* (Geijskes, 1939) (Acari: Tenuipalpidae) only at concentrations equal to or greater than 1:150. Likewise, in the present study, DPE caused high mortality in *T. urticae* just at the same high concentrations, higher than that recommended by producers. Therefore, the results obtained with DPE confirm previous studies. DPE has been demonstrated to be harmful to *N. californicus* and only slightly harmful to *T. urticae* at low concentrations, and therefore, it should not be used in IMP of *T. urticae*, since it is also harmful at higher concentrations.

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