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Effect of *Prosopis juliflora* and *Ziziphus joazeiro* plant extracts on *Stethorus tridens* predatory behaviour on *Tetranychus bastosi*

Cinara Wanderléa Felix BEZERRA, Carlos Romero Ferreira de OLIVEIRA, Cláudia Helena Cysneiros MATOS, Thieres George Freire da SILVA, Josias Jordão Andrade ALVES, Kelem S. FONSECA
Universidade Federal Rural de Pernambuco, Unidade Acadêmica de Serra Talhada, Pernambuco, Brazil

Abstract

Stethorini are unique among Coccinellidae and specifically prey on mites, particularly on Tetranychidae. Furthermore, they are commonly used in biological control programs in several regions around the world. Here, the selectivity of LC_{50} and LC_{90} aqueous extracts of *Prosopis juliflora* L. and *Ziziphus joazeiro* Mart. estimated for the pest mite *Tetranychus bastosi* Tuttle, Baker et Sales (Acari Tetranychidae), was assessed for its effects on the predator *Stethorus tridens* Gordon (Coleoptera Coccinellidae). The effect of the extracts used as a preventive or as a curative treatment on the predation of *S. tridens* on *T. bastosi*, and the repellent effect on larvae and adults of the predator were evaluated. Phenolic compounds present in each extract were analysed. LC_{50} of the *P. juliflora* extract did not affect the predation of *S. tridens* on *T. bastosi.* Conversely, LC_{90} of this extract, as well as LC_{50} and LC_{90} of the *Z. joazeiro* extract significantly reduced prey consumption rate of *T. bastosi* by the predator. LC_{90} of both extracts repelled *S. tridens* larvae, whereas only that of the *P. juliflora* extract caused repellency in adult predators. Significant levels of gallic acid and caffeic acid were found in the *P. juliflora* extract, while in the *Z. joazeiro* extract, gallic acid was identified in significant quantities mainly at LC_{90}. The *P. juliflora* extract was selective to *S. tridens* when used at LC_{50}, which may enable the use of this extract together with *S. tridens* for integrated management of *T. bastosi.*

Key words: Stethorini, alternative control, physic nut, mite, integrated pest management.

Introduction

Coccinellidae (Coleoptera) comprise a group of predators that are important agents in the biological control of various agricultural pests (He *et al*., 2020). The tribe Stethorini is represented by predators of the genera *Stethorus* Weise and *Parasthethorus* Pang et Mao, which are specialized in the consumption of phytophagous mites, with a particular preference for Tetranychidae. Ladybugs of the genus *Stethorus* have a wide geographical distribution. They are commonly used in biological control programs for Tetranychidae because of their high predatory potential, as they can consume large numbers of mites at different life stages (Biddinger *et al*., 2009; Costa *et al*., 2020).

In Brazil, Costa *et al*. (2017) recorded the presence of the predator *Stethorus tridens* Gordon, associated with the phytophagous mite *Tetranychus bastosi* Tuttle, Baker et Sales (Acari Tetranychidae) in physic nut (*Jatropha curcas* L.) plantations in the semiarid region of Pernambuco. The authors evaluated the functional response of the predator and verified a daily consumption of approximately 50 *T. bastosi* individuals, thus demonstrating its potential for the control of this particular pest.

Currently, control of *T. bastosi* is achieved with synthetic acaricides that are not registered with the Ministry of Agriculture, Livestock and Supply (MAPA, 2003). Therefore, methods are urgently required that can be used as an alternative to such synthetic products. Furthermore, studies have shown that plant extracts may have a certain potential to combat *T. bastosi* (Xavier *et al*., 2015; Ferraz *et al*., 2017; Santos *et al*., 2019). However, there is a lack of research available focused on evaluating the selectivity of plant extracts to the natural enemies of *T. bastosi*, such as *S. tridens.*

Therefore, this study aimed to evaluate whether the extracts of *Prosopis juliflora* (Sw) DC. (Mesquite) and *Ziziphus joazeiro* Mart. (Juazeiro) (used at LC_{50} and LC_{90} for *T. bastosi*) are selective to *S. tridens*. The scope of this study was to determine whether the use of these extracts can be combined with that of coccinellids for an effective control of the mite pest.

Materials and methods

Rearing of *T. bastosi* and *S. tridens*

The initial populations *T. bastosi* and *S. tridens* were obtained from an experimental area located at the Academic Unit of Serra Talhada (UAST/UFRPE), Serra Talhada, Pernambuco at 07°59'31"S 38°17'54"W, at an elevation of 429 m a.s.l.

Once in the laboratory, *T. bastosi* individuals were kept in Gerbox® boxes (12 × 12 × 5 cm) containing 4-cm thick polyethylene foam inside. This foam was covered with filter paper on which a leaf of jack bean *Canavalia ensiformis* (L.) DC. (Fabaceae) was placed with its adaxial surface facing down. The foam was moistened with distilled water as required to maintain full leaf turgidity. Hydrophilic cotton moistened in distilled water was placed around the leaf to prevent mites from escaping. As leaves lost turgidity, they were replaced with new ones.

Rearing of the predator was performed as previously reported by Costa *et al*. (2017), using plastic containers (11.0 × 11.0 × 3.0 cm) containing a *T. bastosi*-infested leaf of jack bean (*C. ensiformis*) attached by the petiole to a glass container (5 mL) filled with water. The upper end of the plastic container was sealed with organza fabric to prevent insects from escaping. The leaf was changed every two days to maintain sufficient *T. bastosi*

available as food. The mites and predatory insects were kept in a BOD climatic incubator at 27 ± 2 °C and 70% ± 10% relative humidity, under a 12-h photoperiod.

Collection and preparation of plant extracts

Leaves of *P. juliflora* (Fabaceae) and *Z. joazeiro* (Rhamnaceae), were regularly collected in the morning from UAST/UFRPE. Plants were in the vegetative state at sampling. Leaves were packed in paper bags, labelled, and transported to the Arthropod Ecology Center/UAST/UFRPE (07°59'31"S 38°17'54"W, and an elevation of 429 m a.s.l.). Once in the laboratory, the plant material was washed with distilled water and disinfected with active chlorine (0.05%) for 20 minutes (Vieira *et al*., 2006). Subsequently, samples were subjected to drying at room temperature (25 °C) for five hours and packed in Kraft paper bags for oven-drying under forced air circulation at 50 °C for 48 hours. After this procedure, leaves were ground to a fine powder with a Willey knife mill prior to further preparation of the aqueous extract.

Lethal concentrations of *Z. joazeiro* and *P. juliflora* extracts estimated for *T. bastosi* were used (Nascimento *et al*., 2018; Santos, 2018), corresponding to LC_{50} and LC_{90}, i.e., the concentrations that suffice to kill 50% and 90% of the insect population, respectively: *S. juazeiro* extract, LC_{50} = 11.87% (m/v) and LC_{90} = 54.96% (m/v); *P. juliflora* extract, LC_{50} = 53.45% (m/v) and LC_{90} = 85.35% (m/v). For each extract, these concentrations were prepared from a stock solution using 100 g of leaf powder for 500 mL of distilled water. For LC_{50} and LC_{90} of the *P. juliflora* extract, 26.7 and 42.7 mL of the stock solution were used, respectively, and the fin al solution was brought to a volume of 100 mL with distilled water. For the LC_{50} and LC_{90} of *Z. joazeiro* extract, 6.0 and 27.5 mL of the stock solution were used, respectively, and brought to a final volume of 100 mL with distilled water.

Identification and quantification of phenolic compounds present in LC_{50} and LC_{90} of aqueous extracts of *P. juliflora* and *Z. joazeiro*

For each plant extract, 7.5 mL of each lethal concentration of the aqueous extracts were placed in a 15-mL falcon tube and centrifuged (Hettich-Universal 320 R Centrifuge) at 10,000 rpm for 21 minutes at 2 °C to separate the supernatant from the precipitate. The supernatant was then transferred to 1.5 mL vials and analysed by HPLC.

Gallic and caffeic acid were identified and quantified using a Thermo Scientific Ultimate 3000 HPLC controlled by Chromeleon Chromatography Management System software, with a C18 column (250 mm × 4.6 mm; 5 µm). The isocratic method was used, whereby the mobile phase consisted of acidified water, 2% acetic acid (phase A) and pure methanol (phase B), at a flow rate of 1 mL/min, wavelength (λ) of 270 nm, injection volume of 20 µL and run time of 10 minutes. The standards used in the standard curves were gallic and caffeic acid at the concentrations of 0.05, 0.10, 0.15, 0.20 and 0.25 mg/mL. The same conditions were followed. Thus, the samples were injected into the HPLC and, based on the comparison of the retention time of the gallic and caffeic acid standards, they were identified and quantified.

Assessment of the effect of plant extracts on predation of *T. bastosi* by *S. tridens*

The curative and preventive effects of the aqueous extracts of *Z. joazeiro* and *P. juliflora* on predatory activity of *S. tridens* on *T. bastosi* were evaluated. In the preventive tests, mites and predator were introduced to the test arenas after plant extract application. Whilst in the curative test both were introduced to the arenas prior to application of the extracts. The experimental arenas used in these were prepared as described in previous tests.

Preventive test

After the immersion of the physic nut leaf discs in the treatments (plant extracts or distilled water) and assembly of the experimental arenas, 50 specimens of *T. bastosi* were placed on the leaf discs. After 30 minutes of exposure of the mites to the plants extract treatments, one specimen of *S. tridens* was introduced per arena.

Curative test

After the experimental arenas were assembled, 50 specimens of *T. bastosi* and *S. tridens* were placed on each leaf disc and then 2 mL of plant extracts or distilled water were applied using a manual sprayer.

Twenty-four and 48 hours after treatment, the mites consumed by the predator were counted (Sarmah *et al*., 2009). At the 24-h count, additional mites were introduced to maintain a population density of 50 mites per treatment at the start of the second evaluation period.

The experiment was laid out in a completely randomized design with three treatments for each extract (extract of *P. juliflora*: LC_{50}, LC_{90} and control; extract of *Z. joazeiro*: LC_{50}, LC_{90} and control) and 10 replications. The experimental jars were kept in BOD incubators at 27 ± 2 °C, 70 ± 5% RH, and under a 12-h photoperiod. Data were submitted to the Shapiro-Wilk normality test ($P \leq 0.05$) and subsequently submitted to analysis of variance (ANOVA). Means were compared by Tukey's test at 5% level of probability.

Repellency test

Assessment of repellency of *P. juliflora* and *Z. joazeiro* extracts on *S. tridens* larvae

Physic nut leaf-discs (9 cm ⌀) were thoroughly washed with distilled water. One half of the disc was then immersed in one of the plant extracts at either LC_{50} or LC_{90}, and the other half was immersed in distilled water (control) for five seconds. After this procedure, the leaf discs were placed in Petri dishes, and the experimental jars were set up as in the tests described before. Ten larvae (L3 and L4) were then released on to the midrib of the leaf disc such that they could choose between the treated and the untreated side. The 3rd and 4th instars of *Stethorus* are significantly larger and more agile than the 1st and 2nd instars. The experimental jars were then covered with organza fabric. Assessment of repellency effects started 30 minutes after and then every 12 hours, for 48 hours, the number of larvae present on each half of the leaf discs were counted. All jars were kept in BOD incubators under controlled conditions (27 ± 2 °C, 70 ± 5% RH, and under a 12 h photoperiod). The experiment was laid as a completely randomised design with 10 replications.

Assessment of repellency of *S. tridens* adults by *P. juliflora* and *Z. joazeiro* extracts

Three plastic jars (100 mL) interconnected by a 1-cm diameter hose were used to assess to repellency of *S. tridens* adults by the *P. juliflora* and *Z. joazeiro* extracts. Leaves of physic nut infested with mites (approximately 200 mites) from the stock population were placed inside the jars. In one of the jars, 2 mL of one of either LC_{50} or LC_{90} of the extracts of *P. juliflora* or *Z. joazeiro* was sprayed, while water was spayed in the other jar (control). Then, 10 *S. tridens* adults were released in the central jar and the number of adults in each jar was counted after 48 hours.

Repellency index

The repellency index (RI) was calculated by the formula RI = 2G/(G + P), where: G = % of insects in the treatment and P = % of insects in the control. RI = 1 indicates similar repellency of plant extract and control (neutral treatment), RI > 1 indicates lower treatment repellency relative to control (attractive treatment), and RI < 1 corresponds to higher treatment repellency relative to control (repellent treatment). This index is an adaptation of the formula cited from Lin *et al*. (1990) for consumption index. Treatment means were compared by the t-test at 5% of probability.

The Safety Interval (SI) was calculated as the sum of the value of RI + the respective standard deviation, where values > 1 indicate lower repellency, values = 1 indicate neutral repellency, and values < 1 correspond to higher repellency compared to untreated jars (Kogan and Goeden, 1970).

Results

Phenolic compounds present at the lethal concentrations of the aqueous extracts of *P. juliflora* and *Z. joazeiro* were identified and quantified. In the LC_{50} of the *P. juliflora* extract, gallic acid (retention time 3.36 minutes) was detected at a concentration of 0.23 mg/mL (figure 1A). In turn, LC_{90} had a gallic acid concentration of 0.32 mg/mL, i.e., 1.36 times higher than that at LC_{50} (figure 1B). Meanwhile, the presence of caffeic acid was recorded at a concentration of 0.07 and 0.11 mg/mL for LC_{50} and LC_{90}, respectively, with the corresponding absorbance values occurring in 6.3 min on average (figure 1A and 1B). LC_{50} and LC_{90} showed gallic acid concentrations of 0.05 mg/mL and 0.25 mg/mL, respectively, for the *Z. joazeiro* extract with a retention time 3.33 minutes (figure 1C and 1D).

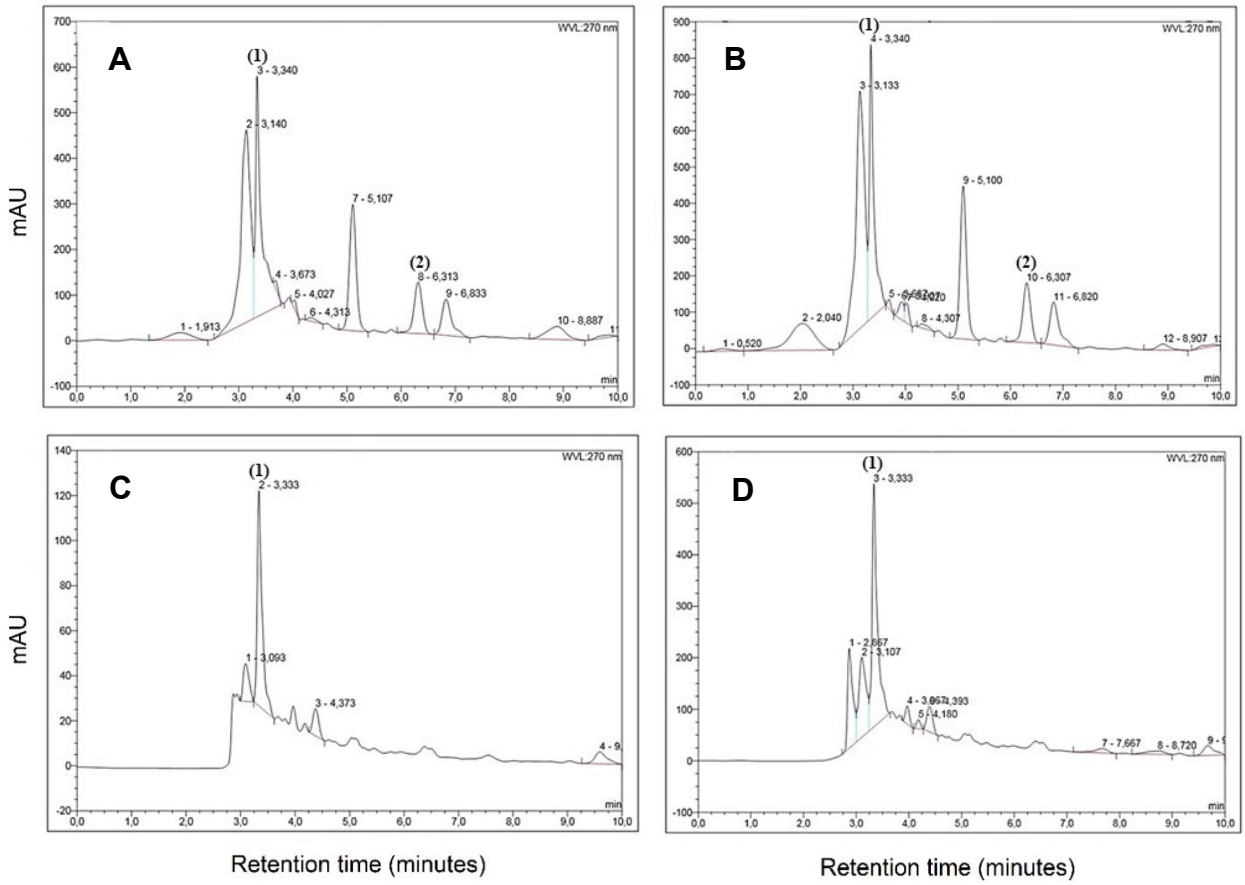

Figure1. HPLC chromatographic profile of fractions and compound retention of *P. juliflora* and *Z. joazeiro* extracts. **A.** Chromatograph of LC_{50} of *P. juliflora* extract; **B.** Chromatograph of LC_{90} of *P. juliflora* extract; **C.** Chromatograph of the LC_{50} of *Z. joazeiro* extract; **D.** Chromatograph of LC_{90} of *Z. joazeiro* extract. (1) Gallic acid, (2) Caffeic acid, at the corresponding retention times: f 3.34 and 6.30 minutes, respectively.

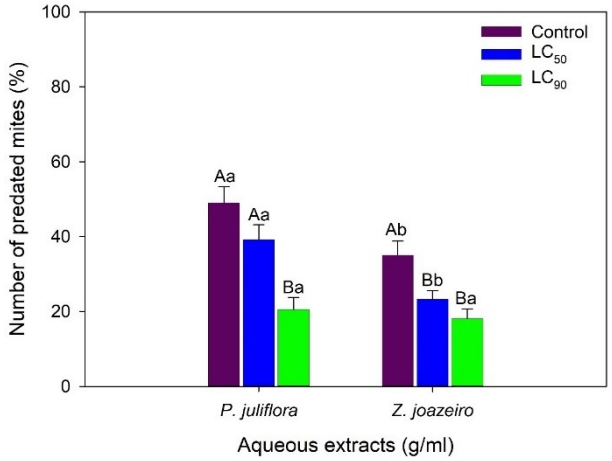

Figure 2. Curative application test. Proportion/number of *T. bastosi* mites predated by *S. tridens* under different concentrations of *P. juliflora* and *Z. joazeiro* extracts as a function of time. Capital letters indicate comparison of means within extracts and lowercase letters indicate comparison of means between extracts. Means were separated by Tukey's test at 5% level of probability.

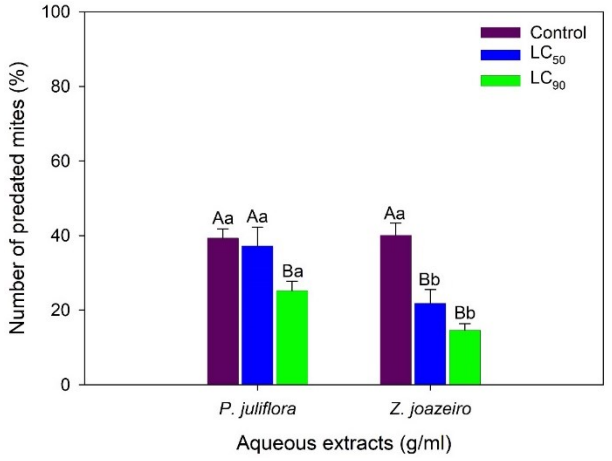

Figure 3. Preventive application test. Proportion/number of *T. bastosi* mites predated by *S. tridens* under different concentrations of *P. juliflora* and *Z. joazeiro* extract as a function of time. Capital letters indicate comparison of means within extracts and lowercase letters indicate comparison of means between extracts. Means were separated by Tukey's test at 5% level of probability.

Assessment of the curative effect revealed that *P. juliflora* and *Z. joazeiro* extracts had an effect on the predation of *T. bastosi* by *S. tridens* ($F_{1,58}$ = 28.8; P ≤ 0.001). When using LC_{90} of the *P. juliflora* extract, there was a significant reduction in the daily consumption of mites by *S. tridens*, but at the LC_{50} concentration consumption did not differ significantly from that observed for the control (figure 2). There was no significant difference in predation at LC_{50} or LC_{90} of the *Z. joazeiro* extract. However, both concentrations led to a level of prey consumption significantly below that observed in the control treatment (figure 2).

As in the curative test, an effect of *P. juliflora* and *Z. joazeiro* extracts was observed on the predation of *S. tridens* on *T. bastosi* ($F_{1,58}$ = 28,8; P ≤ 0.001) in the preventive test. The predation of *S. tridens* on *T. bastosi* (approximately 40%) was significantly lower (by nearly 25%) upon treatment with the *P. juliflora* extract at LC_{90}, than with LC_{50} or in the control, which did not differ significantly (figure 3). The effects of LC_{50} and LC_{90} did not significant differ for *Z. joazeiro*, but they both significantly suppressed predation of *S. tridens* on *T. bastosi* relative to the control treatment (figure 3).

When assessing the effect of plant extracts on their attractiveness to larvae and adults of *S. tridens*, we found no significant difference between treatments and LC_{50} of either extract or the control, neither for larvae (figure 4) nor for adults (figure 5). These concentrations were classified as neutral with regard to SI (tables 1 and 2). Conversely, LC_{90} of the *P. juliflora* extract repelled both larvae (figure 4) and adult insects (figure 5), wherein this concentration was classified as repellent with regard to SI (tables 1 and 2). In turn, LC_{90} of the *Z. joazeiro* extract repelled only the adults of *S. tridens* (figure 5, table 2) but did not influence the response of the larvae (figure 4).

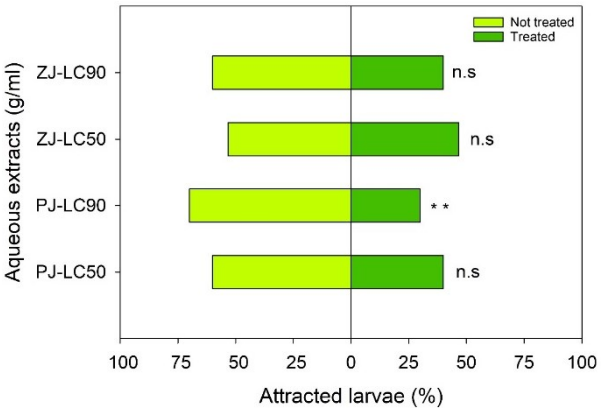

Figure 4. Percentage of *S. tridens* larvae (L3 and L4) attracted when exposed to different concentrations of *P. juliflora* and *Z. joazeiro* extracts. *Significant differences between treated and not treated leaves (t-test, P < 0.05). n.s. = not significant (t-test, P < 0.05).

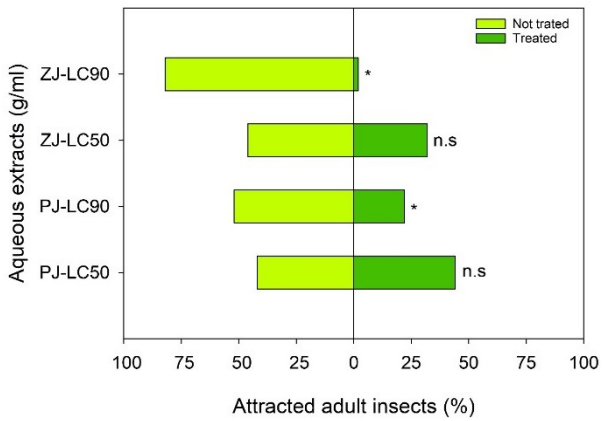

Figure 5. Percentage of adult *S. tridens* insects attracted when exposed to different concentrations of *P. juliflora* and *Z. joazeiro* extracts. *Significant differences between treated and not treated leaves (t-test, P < 0.05). n.s. = not significant (t-test, P < 0.05).

Table 1. Repellency index (%) lethal concentrations of aqueous extracts *P. juliflora* and *Z. joazeiro* in the larvae of *S. tridens.*

Time (hours)	Extracts	Concentration (g/ml)	Not treated μ ± SD	Treated μ ± SD	RI μ ± SD	SI	(t)
0.5	*P. juliflora*	LC_{50}	8.0 ± 0.0	2.0 ± 0.0	0.50 ± 0.00	R	*
		LC_{90}	7.6 ± 1.5	2.3 ± 1.5	0.47 ± 0.35	R	*
	Z. joazeiro	LC_{50}	6.0 ± 2.0	4.0 ± 2.0	1.33 ± 0.40	N	n.s.
		LC_{90}	9.3 ± 0.6	6.7 ± 5.8	0.13 ± 0.10	R	*
12	*P. juliflora*	LC_{50}	8.6 ± 1.5	1.3 ± 1.5	0.31 ± 0.031	R	*
		LC_{90}	6.3 ± 0.6	3.6 ± 0.6	0.73 ± 0.12	R	*
	Z. joazeiro	LC_{50}	5.6 ± 1.5	4.3 ± 1.5	1.53 ± 0.12	N	*
		LC_{90}	7.6 ± 0.6	2.3 ± 0.6	0.47 ± 0.35	R	*
24	*P. juliflora*	LC_{50}	7.3 ± 1.5	2.6 ± 1.5	0.73 ± 0.31	N	*
		LC_{90}	7.3 ± 0.5	4.6 ± 0.6	0.78 ± 0.12	R	n.s.
	Z. joazeiro	LC_{50}	5.0 ± 1.0	5.0 ± 1.0	2.0 ± 0.20	N	n.s.
		LC_{90}	7.3 ± 0.6	2.6 ± 0.6	0.53 ± 0.12	R	*
36	*P. juliflora*	LC_{50}	7.6 ± 0.6	2.3 ± 0.6	0.61 ± 0.12	R	*
		LC_{90}	7.6 ± 0.6	3.6 ± 0.6	0.65 ± 0.12	R	n.s
	Z. joazeiro	LC_{50}	8.0 ± 0.0	2.0 ± 0.0	0.5 ± 0.0	R	*
		LC_{90}	7.6 ± 0.6	2.3 ± 0.6	0.47 ± 0.12	R	*
48	*P. juliflora*	LC_{50}	6.0 ± 1.0	4.0 ± 1.0	1.33 ± 0.20	N	n.s.
		LC_{90}	7.0 ± 0.0	4.3 ± 0.0	0.76 ± 0.00	R	n.s
	Z. joazeiro	LC_{50}	5.3 ± 0.6	4.7 ± 0.6	1.75 ± 0.12	N	n.s.
		LC_{90}	6.0 ± 0.0	4.0 ± 0.0	0.8 ± 0.00	R	*

RI: Repellency index (R=2G/(G+P), G = number of treated insects, P = number of not treated insects (control); SD: standard deviation; SI: security interval (IR+SD). Values ≥ 1 do not indicate repellency and ≤ values indicate repellency. *Means in the rows differ significantly by t-test (t) at 5% probability. n.s.= not significant.

Table 2. Repellency index (RI) lethal concentrations of the aqueous extracts of *P. juliflora* and *Z. joazeiro* in adults of *S. tridens.*

Extracts	Concentration (g/mL)	Not Treated μ ± SD	Treated μ ± SD	RI μ ± SD	SI	(t)
P. juliflora	LC_{50}	4.2 ± 3.4	4.4 ± 2.6	0.821 ± 0.71	N	n.s.
	LC_{90}	5.2± 1.9	2.2 ± 1.8	0.048 ± 0.06	R	*
Z. joazeiro	LC_{50}	4.6 ± 3.0	3.2 ± 3.8	1.023 ± 0.67	N	n.s.
	LC_{90}	8.2 ± 1.3	0.2 ± 0.4	0.595 ± 0.51	N	*

RI: Repellency index (R=2G/(G+P), G = number of treated insects, P = number of not treated insects (control); SD: standard deviation; SI: security interval (IR+SD). Values ≥ 1 do not indicate repellency and ≤ values indicate repellency. *Means in the rows differ significantly by t-test (t) at 5% probability. n.s.= not significant.

Discussion

The high levels of gallic acid found in the lethal concentrations (LC_{50} and LC_{90}) of *P. juliflora* and *Z. joazeiro* extracts may be associated with antioxidant, anti-parasitic, and antibacterial effects. These effects are common, as observed by Brito *et al*. (2015) but studies demonstrating the insecticidal activity of gallic acid are still scarce. Caffeic acid found in *P. juliflora* extract acts as an active intestinal protease inhibitor (Joshi *et al*., 2014), and *Helicoverpa armigera* Hubner larvae fed with caffeic acid showed changes in gene expression and protease activities (Joshi *et al*., 2014). However, this effect of caffeic acid on insects has been little studied (Joshi *et al*., 2014; Singh *et al*., 2021), and no studies have shown the effect of caffeic acid on Coccinellidae, specifically on members of the genus *Stethorus*. Similarly, Santos *et al*. (2013) attributed an increase in time for development of the fall armyworm (*Spodoptera frugiperda* Smith) to the effect of gallic acid present in manioc leaves (*Manihot esculenta* Crantz), which, additionally, reduced the fertility of the insect. Furthermore, increased mortality of leafcutter ants (*Atta sexdens* L.) was also reported in response to the action of gallic acid (Santos *et al*., 2013). Consistently, Palido *et al*. (2017) showed that some phenolic compounds strongly inhibit tyrosinase, which is a key enzyme involved in the process of insect metamorphosis. The authors proposed that some caffeic acid derivatives may be directly linked to this inhibitory effect.

Based on the above, research has been directed at evaluating the potential of plant extracts for agricultural pest control and on how this control method can be incorporated into Integrated Pest Management (IPM) programs. Concerning pest mites, work with plant extracts has aimed to demonstrate their efficacy as natural acaricides (Siqueira *et al*., 2014; Xavier *et al*., 2015; Ferraz *et al*.,

2017). However, few studies have evaluated their effects on the natural enemies associated with these mites. Thus, Sarmah *et al*. (2009), Sarmah (2016), and Handique *et al*. (2017) observed a reduction in the feeding of predators of the genus *Stethorus* when they were exposed to different concentrations of plant extracts, with a reduction in mite consumption during a 48-h exposure.

Bonsignore and Vacante (2012) evaluated the influence of nine biopesticides on adults and larvae of *Frankliniella occidentalis* (Pergande) and its predator *Orius laevigatus* (Fieber). Negative effects on predator dynamics were evident only with the use of some products. The treatments were not effective for the control of *F. occidentalis* and the botanical insecticides rotenone and neem reduced the number of *O. laevigatus* adults. In turn, Sayed *et al*. (2020) evaluated the effect of five botanical extracts (*Psiadia penninervia*, *Salvia officinalis*, *Ochradenus baccatus*, *Pulicaria crispa* and *Euryops arabicus*) on the bean aphid, *Aphis craccivora* Koch and the 2nd larval instar of the green lacewing, *Chrysoperla carnea* (Stephens) under laboratory conditions. The *O. baccatus* extract had a strong effect on aphids and was more selective for the predator.

In this study, LC_{50} of the aqueous extract of *P. juliflora* was selective to the predator *S. tridens*, as it did not interfere significantly with its predatory activity on *T. bastosi*, compared to the control treatment. This result suggests that the plant extract may be effectively combined with the predator *S. tridens*, as the latter was not harmed by the plant extract at the LC_{50} for the mite. In contrast, LC_{90} significantly reduced the number of mites consumed by the predator and therefore, its use cannot be recommended for the control of this pest in association with *S. tridens*.

Neither of the *Z. joazeiro* extract concentrations tested were selective to *S. tridens*, and they significantly reduced its predatory potential. A possible explanation for this finding is that there was a reduction in the palatability of *T. bastosi* to *S. tridens*. According to laboratory observations, mites exposed to LC_{90} of *Z. joazeiro* and *P. juliflora* extract were rejected immediately. Furthermore, the predator also rejected the mites that had were exposed to LC_{50} of the extracts. On the other hand, mites which were not exposed to the extracts were consumed immediately by the predator.

It is important to point out that the reduction in the predatory action of *S. tridens* on *T. bastosi* caused by the *Z. joazeiro* extract is due to the presence of chemical compounds that act on this predator. Caffeic acid, for example, may have acted directly in the intestine of the predator by inhibiting the enzymes involved in its detoxification. This effect may be due to the sequential binding of multiple molecules of caffeic acid that directly induce conformational changes in proteases, thus causing a significant decrease in their activities and, ultimately intensifying the insecticidal effect of the compound (Joshi *et al*., 2014).

Although inhibition of intestinal enzymes is a positive action for pest control, it is important to determine if these effects also occur on predators, because some products may have low selectivity. In this sense, the combined use of extracts with high levels of caffeic acid and biological control agents in IPM programs would be unfeasible.

As for larval and adult predator repellency caused by LC_{90} of the *P. juliflora* extract and the adult repellency caused by LC_{90} of the *Z. joazeiro* extract observed in this study, it is important to identify the substances in the composition of the extract that caused this effect (Bonsignore and Vacante, 2012; Siqueira *et al*., 2014). For example, Kumral *et al*. (2013) observed similar effects of sublethal doses of an ethanolic extract of *Datura stramonium* leaves (Solanaceae) on the mite *Panonychus ulmi* (Koch) (Acari Tetranychidae) and its predator *Stethorus gilvifrons* (Mulsant) (Coleoptera Coccinellidae). They found that low concentrations of the extract (13.72 mg/L) caused repellency of *S. gilvifrons* adult, an effect that the authors attributed directly to the chemical composition of the plant.

Research aimed at assessing the selectivity of insecticides/acaricides of botanical origin as biological control agents is of great relevance, as botanical extracts vary in their potential suitability for use in IPM programs (Poderoso *et al*., 2016). Although some products are selective, others may harm biological control agents, rendering their combined use impossible. As our knowledge and understanding of these phenomena expand, we will be able to determine the situations in which the combination of different methods may allow the design and implementation of IPM programs.

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Authors' addresses: Cláudia Helena Cysneiros MATOS (corresponding author: claudia.matos@ufrpe.br), Cinara Wanderléa Felix BEZERRA, Carlos Romero Ferreira de OLIVEIRA, Thieres George Freire da SILVA, Josias Jordão Andrade ALVES, Kelem S. FONSECA, Universidade Federal Rural de Pernambuco, Unidade Acadêmica de Serra Talhada, 38293205, Serra Talhada, Pernambuco, Brazil.

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