



Antibiosis of strawberry genotypes to the spotted spider mite

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ABSTRACT: The spotted spider mite, *Tetranychus urticae* (Koch, 1836), is one of the most important pests of strawberry production systems worldwide. Because plant resistance is an important integrated management strategy, the present study investigated the effects of 12 strawberry genotypes on the biological characteristics of this pest under laboratory conditions (25 ± 2 °C and RH $60 \pm 10\%$; 12:12-hour photophase). The experiment was carried using 12 treatments (genotypes), divided into 20 replications each, in a completely randomized design. The response variables were the biological parameters of the spider mite and its survival rate. The development and survival of *T. urticae* were influenced by different strawberry genotypes. The Camarosa cultivar together with genotypes Selection 05 and 2017-04-03 negatively affected the development and survival of *T. urticae*. The Selection 02 genotype had greatest susceptibility to the spider mite, which allowed fast development and high survival rate. The reproductive parameters of *T. urticae* were affected differently as a function of the strawberry genotype, with the Camarosa cultivar and the genotypes Selection 05 and 2017-04-03 being unfavorable to development, suggesting a possible resistance based on antibiosis.

Key words: *Tetranychus urticae* (Koch, 1836), biology, plant resistance, integrated pest management.

Antibiose de genótipos de morangueiro ao ácaro-rajado

RESUMO: O ácaro-rajado, *Tetranychus urticae* (Koch, 1836), é uma das pragas mais importantes dos sistemas de produção de morangos em todo o mundo. Considerando a resistência de plantas como importante estratégia de manejo integrado, o presente estudo investigou os efeitos de 12 genótipos nas características biológicas desta praga, em condições de laboratório (25 ± 2 °C e UR $60 \pm 10\%$; fotofase de 12 horas). O experimento foi realizado em condições de Laboratório sendo utilizado 12 tratamentos (genótipos), divididos em 20 repetições cada, num delineamento inteiramente casualizado. As variáveis resposta foram os parâmetros biológicos do ácaro-rajado e taxa de sobrevivência. O desenvolvimento e a sobrevivência de *T. urticae* foram influenciados pelos diferentes genótipos de morangueiro. A cultivar Camarosa junto ao genótipo Seleção 05 e 2017-04-03 afetaram negativamente o desenvolvimento e a sobrevivência de *T. urticae*. O genótipo Seleção 02 demonstrou maior suscetibilidade ao ácaro-rajado, o qual apresentou rápido desenvolvimento e elevada taxa de sobrevivência. Conclui-se que os parâmetros reprodutivos de *T. urticae* são afetados em função do genótipo de morangueiro, sendo 'Camarosa' e os genótipos Seleção 05 e 2017-04-03 desfavoráveis ao desenvolvimento, sugerindo uma possível resistência do tipo antibiose.

Palavras-chave: *Tetranychus urticae* (Koch, 1836), biologia, resistência de plantas, manejo integrado de pragas.

INTRODUCTION

The first strawberry hybrids, *Fragaria* × *ananassa* (Duchesne), were developed in the eighteenth century in European gardens. In turn, the first cultivars (Keens Imperial and Keens Seedling) were launched in 1814 and 1823, respectively, and are the precursors of current materials. The fruit is popular throughout the world and is cultivated on all continents except Antarctica. This situation is due to the high adaptability developed by breeders over the years, making strawberry cultivation possible in a wide range of climates, from cold to temperate and subtropical (SIMPSON, 2018).

Various pests can attack the plants during cultivation, including aphids, thrips, caterpillars

and mites (BERNARDI et al., 2015; BOTTON et al., 2016). These arthropods limit the crop yield, requiring the adoption of strategies to reduce losses and assure fruit quality, without the presence of toxic residues (BENATTO et al., 2021).

Mites feed on the leaf cell content. The resulting mesophyll perforation causes the leaflets to take on a bronze color and fall prematurely, reducing fruit production and even killing the plant if high infestation is not controlled (BERNARDI et al., 2015; BOTTON et al., 2016). Tetranychidae is considered one of the most important families of polyphagous and phytophagous mites, characterized by the formation of webs on the plants. *Tetranychus urticae* (Koch, 1836), or the spotted spider mite, is

the leading strawberry pest in Brazil (MORAES & FLECHTMANN, 2008).

There are various strategies to manage *T. urticae*. The main one involves the application of chemical pesticides (ATTIA et al., 2013; SHEN et al., 2021). However, the use of biological products (VIDRIH et al., 2021) such as essential oils (FAROUK et al., 2021) is gaining traction as a sustainable and ecologically friendly method, because the excessive application of chemical acaricides leads to environmental pollution and resistance. Indeed, control methods based only on the use of synthetic acaricides often are unable to maintain the number of mites below levels that cause economic damage (ATTIA et al., 2013), so it is necessary to use complementary control measures, such as the cultivation of resistant genotypes (varietal control) (BROWN et al., 2018; WANG et al., 2018).

Varietal control consists of the cultivation of genotypes that have a negative influence on the characteristics of arthropod pests (biology, ecology and/or morphology), reducing their population to levels below the threshold of economic damage (PAINTER, 1951). Among the plant resistance mechanisms, more specifically constitutive resistance, i.e., resistance that is present in plants irrespective of external factors, is antibiosis (SMITH & CLEMENT, 2012). Antibiosis influences the biological parameters of the pest, such as by altering its biological cycle (BALDIN et al., 2019).

In light of these factors, and since plant resistance is an important management tactic, the objective of this study was to evaluate the presence of resistance via antibiosis to *T. urticae* of various strawberry genotypes (international and national, of the Crop Improvement Program of the Embrapa Temperate Climate research unit).

MATERIALS AND METHODS

The experiment was conducted under laboratory conditions at the Eliseu Maciel School of Agronomy, Federal University of Pelotas (UFPEL), located in the municipality of Pelotas, Rio Grande do Sul, Brazil, with 9 genotypes developed by the Strawberry Improvement Program of the Embrapa Temperate Climate research unit, namely: Selection 25; Selection 09; Selection 22; Selection 05; Selection 06; Selection 03; Selection 12; 2017-04-03; Selection 02; and three imported cultivars: 'Camarosa', 'Fronteras' and 'Camino Real'. All plants were kept under the same cultivation conditions. The experimental design was completely randomized,

with 12 treatments (genotypes) and a total of 20 repetitions for each, according to the method adapted from KARLEC et al. (2017).

The mites used were collected from strawberry plants of the Merced cultivar, in commercial fields in the municipality of Pelotas, Rio Grande dos Sul, Brazil (31°39'40" S; 52°25'51" W). Mite specimens were taken to the laboratory to compose the stock population, maintained on common bean plants (*Phaseolus vulgaris* L.), cultivated in pots in a greenhouse.

Leaf samples were collected from the middle part of the strawberry plants of each genotype grown in the field. In the laboratory, we analyzed them visually to assure the absence of contaminants (such as mite eggs and adults). Arenas were formed with leaf disks with diameter of 2.2 cm of each genotype, arranged individually in Petri dishes (10 cm diameter) on cotton moistened with distilled water, with the abaxial face upward. Each dish composed a repetition. A previously fertilized female mite was released using a no. 2 brush on each leaf disk, where she remained for 24 hours for oviposition. After this period, one egg was individualized per leaf disk (removing the female and other eggs with a brush) for subsequent observation of the development stages of the mite.

The dishes were placed in a biological oxygen demand (BOD) incubator at a temperature of 25 ± 1 °C, RH = $70 \pm 10\%$ and 12:12h photophase, for daily evaluation of the following parameters: duration (days) of the immature stages (egg, larva, protonymph and deutonymph), and adult stage, pre-oviposition and oviposition periods, fertility (total number of eggs/female), survival (%) of each stage during the life cycle, and sex ratio.

The data were analyzed regarding normal distribution of the residuals and homoscedasticity of variance by the Shapiro-Wilk and Bartlett tests, respectively. When the assumptions of analysis of variance were not satisfied, the nonparametric Kruskal-Wallis test with separation of means of the ranks and the Dunn test were applied, in all cases with 5% significance. The viability (%) was compared by the Fisher test. All the statistical tests were carried out with the R software, version 3.6.3 (R DEVELOPMENT CORE TEAM, 2020).

RESULTS AND DISCUSSION

According to our results, the strawberry genotypes tested influenced the development time of the spotted spider mite only in the larval period

($X^2 = 30.174$, $df = 11$, $P = 0.0015$). The duration of the other stages (egg, deutonymph, protonymph and adult) were not significantly different ($P \geq 0.05$) (Table 1).

The genotype 2017-04-03 (3.50 days) and the cultivar Camarosa (3.46 days) ($P \leq 0.05$) stood out for having the longest larval period, while the genotype Selection 02 (1.83 days) had the shortest duration. A previous study indicated that two strawberry cultivars (Marak and Sequioa) could influence the duration of the nymph stages of *T. urticae* (REZAIE et al., 2013). A shorter development time means faster population growth of the mite (ADANGO et al., 2006), making the plant more susceptible to damages.

MONTEIRO et al. (2014) and KARLEC et al. (2017) also observed significant differences in the larval period of the Camarosa cultivar in comparison with different genotypes, inferring that the longer larval duration is possibly associated with a high adaptive cost. In this case, the plant does not supply sufficient nutrients for normal development of the mites, indicating the presence of resistance by antibiosis (KARLEC et al., 2017).

In the egg-adult ($X^2 = 26.2925$, $df = 11$, $P = 0.01$) and pre-oviposition ($X^2 = 22.313$, $df = 11$, $P = 0.02206$) periods, there were significant differences between the treatments (Table 2). The egg-adult was longest in the Selection 05 genotype (13.62 days), significantly different in relation to the Selection 02 genotype (10.27 days) ($P \leq 0.05$), which had the shortest period. This suggested that Selection 02 is more susceptible to the spotted spider mite, because it reaches the adult stage more rapidly. According to SEDARATIAN et al. (2009), a plant on which an

arthropod pest develops faster is more susceptible, justifying the importance of the egg-adult period. Among the other genotypes, there were no significant pairwise differences.

For the pre-oviposition period ($X^2 = 22.313$, $df = 11$, $P = 0.02206$), the cultivar Camarosa (3.88 days) and genotypes Selection 05 (3.40 days) and Selection 06 (3.33 days) had the longest durations. In turn, Selection 09 (1.14 days) had the shortest duration, differing significantly ($P \leq 0.05$). A short pre-oviposition period is beneficial to the spotted spider mite by resulting in faster and more efficient reproduction (SEDARATIAN et al., 2009).

In the larval stage, the highest survival rates were observed for the mites on the genotypes Selection 03 (100%), Selection 12, Selection 02, Camino Real and Fronteras (94.74%), with significant differences in relation to the lowest survival rates, of Camarosa (68.42%) ($P = 0.02905$) and Selection 05 (57.89%) ($P = 0.03602$) (Table 3). Similar results were reported for Camarosa by KARLEC et al. (2017), who concluded that the larval stage has the highest sensitivity within the mite's life cycle.

For the egg-adult period, Camino Real (90%) had the greatest survival, differing significantly from Camarosa ($P = 0.03987$) and Selection 05 ($P = 0.01857$) (50% and 40%, respectively), which had the lowest rate (Table 3).

An explanation for the results is that plants in general, including strawberry, contain defensive substances in the leaves, such as alkaloids, phenols, terpenoids and quinones, which alter the development of arthropods, even killing them, a phenomenon

Table 1 - Duration of the immature and adult stages of *tranychus urticae* in days (\pm standard error), submitted to different strawberry genotypes under controlled laboratory conditions ($25^\circ \pm 3^\circ\text{C}$; RH = $70\% \pm 10\%$; 12:12 hour photoperiod).

Treatment	Period (Days)				
	Egg ^{ns}	Larva [*]	Protonymph ^{ns}	Deutonymph ^{ns}	Adult ^{ns}
Selection 09	4.00 \pm 0.21	3.06 \pm 0.41 ab	2.67 \pm 0.36	2.73 \pm 0.26	11.18 \pm 2.48
Selection 05	4.11 \pm 0.22	3.27 \pm 0.32 ab	2.70 \pm 0.28	3.25 \pm 0.68	14.00 \pm 3.34
Selection 03	3.56 \pm 0.12	2.28 \pm 0.23 ab	2.41 \pm 0.25	2.69 \pm 0.28	16.44 \pm 2.43
Selection 02	4.11 \pm 0.10	1.83 \pm 0.24 b	2.00 \pm 0.18	2.53 \pm 0.19	15.87 \pm 2.15
Selection 06	3.53 \pm 0.14	2.82 \pm 0.35 ab	2.60 \pm 0.18	2.38 \pm 0.20	16.08 \pm 2.17
Selection 12	3.84 \pm 0.17	2.00 \pm 0.22 ab	2.24 \pm 0.21	3.00 \pm 0.28	17.38 \pm 1.92
Selection 22	3.85 \pm 0.08	2.71 \pm 0.37 ab	2.07 \pm 0.24	2.46 \pm 0.21	13.15 \pm 2.18
Selection 25	3.95 \pm 0.22	2.53 \pm 0.24 ab	2.63 \pm 0.53	2.64 \pm 0.26	11.07 \pm 2.09
2017-04-03	3.63 \pm 0.13	3.50 \pm 0.41 a	2.43 \pm 0.22	2.44 \pm 0.18	14.63 \pm 2.08
Camarosa	4.11 \pm 0.26	3.46 \pm 0.35 a	2.25 \pm 0.29	3.50 \pm 0.51	16.80 \pm 2.38
Camino Real	4.26 \pm 0.18	2.44 \pm 0.26 ab	2.67 \pm 0.31	2.39 \pm 0.21	10.78 \pm 1.89
Fronteras	4.05 \pm 0.17	2.28 \pm 0.19 ab	2.53 \pm 0.28	2.79 \pm 0.25	14.57 \pm 1.68

*Different lowercase letters in the column indicate significant differences by the Dunn test ($P \leq 0.05$).

^{ns}Not significant.

Table 2 - Sex ratio and duration of the egg-adult, pre-oviposition and oviposition periods (\pm standard error), along with the total average number of eggs paid by the female mite on leaf disks of each genotype.

Treatment	Sex Ratio ^{ns}	-----Period (days)-----			Total number of eggs/female ^{ns}
		Egg-Adult*	Pre-oviposition*	Oviposition ^{ns}	
Selection 09	0.64	12.00 \pm 0.60 ab	1.14 \pm 0.43 b	5.29 \pm 1.98	12.14 \pm 4.86
Selection 05	0.63	13.62 \pm 1.00 a	3.40 \pm 1.19 a	10.00 \pm 3.09	25.60 \pm 6.62
Selection 03	0.63	10.94 \pm 0.49 ab	3.00 \pm 0.71 ab	13.90 \pm 2.98	36.70 \pm 8.80
Selection 02	0.73	10.27 \pm 0.24 b	2.54 \pm 0.30 ab	12.73 \pm 1.44	38.09 \pm 5.01
Selection 06	0.69	11.00 \pm 0.24 ab	3.33 \pm 0.74 a	13.22 \pm 2.48	26.89 \pm 7.83
Selection 12	0.69	10.81 \pm 0.48 ab	2.36 \pm 0.15 ab	16.00 \pm 1.72	31.18 \pm 4.05
Selection 22	0.62	10.69 \pm 0.32 ab	2.63 \pm 0.47 ab	9.75 \pm 2.20	30.75 \pm 6.93
Selection 25	0.64	10.93 \pm 0.41 ab	2.44 \pm 0.57 ab	7.78 \pm 2.73	18.67 \pm 6.77
2017-04-03	0.81	11.94 \pm 0.47 ab	2.62 \pm 0.57 ab	12.69 \pm 2.22	24.69 \pm 3.97
Camarosa	0.80	13.30 \pm 1.10 ab	3.88 \pm 0.65 a	14.63 \pm 2.23	40.25 \pm 9.20
Camino real	0.78	11.72 \pm 0.51 ab	1.79 \pm 0.27 ab	7.14 \pm 1.64	21.57 \pm 4.76
Fronteras	0.64	11.36 \pm 0.47 ab	2.44 \pm 0.59 ab	10.78 \pm 1.95	29.22 \pm 6.84

*Different lowercase letters in the column indicate significant differences by the Dunn test ($P \leq 0.05$).

^{ns}Not significant.

directly related to antibiosis (RODRIGUEZ & RODRIGUEZ, 1981; HANLEY et al., 2007).

According to SEDARATIAN et al. (2009), an important defense mechanism of plants to *T. urticae* is the absence or low levels of the primary nutrients necessary for correct development of the mite's biological cycle. This was the case of the genotypes Florida Festival and IAC Campinas (KARLEC et al., 2017). Besides this, other factors can be involved, such as the presence of trichomes (glandular or non-glandular) on the leaves, directly affecting the mite's survival due to the release of substances associated with oxidation and polymerization of phenolic compounds, limiting the locomotion, or acting directly on the arthropod as a toxic effect (FIGUEIREDO et al., 2013).

Knowledge of the characteristics of the different strawberry genotypes is essential to formulate effective integrated pest management systems. We were able to identify different genotypes with lower or greater resistance to the spotted spider mite.

The introduction of the varietal control technique would allow reduced need for pesticides, helping to preserve the environment, besides not affecting the other pest control forms. This demonstrates the importance of ascertaining plants' resistance in phytosanitary crop management.

CONCLUSION

The cultivar Camarosa together with the genotypes Selection 05 and 2017-04-03 had the best

Table 3 - Survival (%) of the different stages of the spotted spider mite in the laboratory when submitted to different strawberry genotypes.

Treatment	-----Survival (%)-----						
	Egg	Larva*	Protonymph	Deutonymph	Egg-adult*		
Selection 09	95.00	84.21	ab	75.00	91.67	55.00	ab
Selection 05	95.00	57.89	b	90.91	80.00	40.00	B
Selection 03	90.00	100.00	a	94.44	94.12	80.00	ab
Selection 02	95.00	94.74	a	100.00	83.33	75.00	ab
Selection 06	95.00	89.47	ab	88.24	86.67	65.00	ab
Selection 12	95.00	94.74	a	94.44	94.12	80.00	ab
Selection 22	100.00	85.00	ab	88.24	86.67	65.00	ab
Selection 25	95.00	89.47	ab	94.12	87.50	70.00	ab
2017-04-03	95.00	84.21	ab	100.00	100.00	80.00	ab
Camarosa	95.00	68.42	b	92.31	83.33	50.00	b
Camino Real	95.00	94.74	a	100.00	100.00	90.00	a
Fronteras	95.00	94.74	a	83.33	93.33	70.00	ab

*Significant by the Fisher test Fisher ($P \leq 0.05$).

resistance by antibiosis to *T. urticae*, by negatively affecting the mite's biological parameters. Therefore, we can recommend the use of these genotypes to achieve resistance to the spotted spider mite as part of integrated pest management. We observed that the genotype Selection 02 was most susceptible to *T. urticae*, so it is not recommended for commercial planting.

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DECLARATION OF CONFLICT OF INTEREST

The authors declare no conflicts of interest. Sponsors did not have any role in the design of the study; collection, analysis and interpretation of the data; writing of the manuscript; and decision to publish the results.

AUTHORS' CONTRIBUTIONS

All the authors contributed equally to the work, independently of the topics addressed. All the authors critically reviewed and approved the final version of the manuscript.

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