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Acyl sugars and whitefly (*Bemisia tabaci*) resistance in segregating populations of tomato genotypes

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ABSTRACT. The wild tomato, *Solanum pennellii*, is an important source of resistance genes against tomato pests. This resistance is due to the presence of acyl sugars (AS), which are allelochemicals that have negative effects on arthropod pests. There are no commercially available tomato cultivars that exhibit significant levels of resistance to arthropod pests. Therefore, this study evaluated resistance to whitefly (*Bemisia tabaci*) in F_2 and F_2RC_1 tomato genotypes with high AS levels from a cross between *Solanum lycopersicum* 'Redenção' and the *S. pennellii* accession, LA-716. Plants were exposed to *B. tabaci* biotype B at the pre-flowering stage. In both generations, there were significant, negative correlations between AS content and oviposition preference and nymph development. Whitefly exhibited a lower preference for oviposition and produced fewer nymphs in genotypes with high AS levels and the wild parent *S. pennellii* than in the low AS-level genotypes and Redenção cultivar, demonstrating that the breeding program was effective in transferring resistance to the F_2 and F_2RC_1 generations. RVTA-2010-pl#31 and RVTA-2010-pl#94 in the F_2 population

are promising genotypes that produced materials with high AS levels in the F_2RC_1 generation (RVTA-2010-31-pl#177 and RVTA-2010-94-pl#381).

Key words: *Solanum pennellii*; Plant breeding; Resistance to pests; Allelochemical; *Solanum lycopersicum*

INTRODUCTION

According to the World Production Estimate of Tomatoes for Processing (WPTC, 2015), over 42 million tons of tomatoes were processed in 2015, an increase of 6.8% over the previous year. The largest global producers are the US (30.7%), China (15.4%), Italy (12.3%), Spain (6.5%), and Turkey (5.9%). Brazil currently ranks eighth in the production of tomatoes for processing, with about 1.4 million tons.

Tomatoes play significant economic and social roles in Brazil (ABCSEM, 2010), and account for 16% of the country plant-related gross domestic product. Because tomato cultivation requires a large number of workers, it is estimated that about 300,000 jobs in agriculture depend on it (Geraldini et al., 2011).

However, tomatoes are susceptible to a variety of arthropod pests, which increase production costs directly by damaging the product and indirectly by requiring the use of chemicals for their management. One of the main pests that damages tomato crops and reduces yield is the whitefly (*Bemisia tabaci* biotype B) (Srinivasan et al., 2012). Direct damage occurs when whitefly suck the sap from phloem in plant tissues, which causes a series of physiological disorders such as the irregular ripening of fruit and withered leaves. Direct damage is associated with the appearance of sooty mold (*Capnodium* spp), a fungus that causes the browning of leaves and fruit. The appearance of this fungus is caused by sugar secreted from epidermal tissues after *B. tabaci* biotype B feeding, which interferes with plant photosynthesis and promotes the establishment of viral infections, causing indirect damage (Firdaus et al., 2012).

To prevent pests, pesticides have been applied indiscriminately. However, they encourage the selection of individuals resistant to the main active ingredients, cause environmental pollution, and are health risks to humans (Forget et al., 1993; Isman, 2006). Therefore, alternative methods of controlling *B. tabaci* biotype B are required, in order to reduce the amount of chemicals used in the management of arthropod pests (Sayeda et al., 2009). The use of tomato genotypes that are resistant to arthropod pests has shown promising results (Toscano et al., 2002; Fancelli et al., 2003; Maluf et al., 2010; Dias et al., 2013; Lucini et al., 2015). Varietal resistance is a viable alternative within an integrated pest management strategy, because the resistance levels exhibited by commercial cultivars, particularly those that are cultivated for industrial processing, are inadequate.

Wild tomato species, such as *Solanum pennellii*, are used as a source of resistance against pests and diseases. Resistance to pests obtained from *S. pennellii* has been linked to the presence of allelochemicals called acyl sugars (ASs). These allelochemicals have deleterious effects during certain stages of pest development, and can even prevent oviposition and feeding (Muigai et al., 2002; Resende et al., 2002a; Silva et al., 2009; Lucini et al., 2015). According to Resende et al. (2009), genotypes in breeding programs should be selected for high AS levels for pest resistance. By backcrossing, advanced lineages can be used for the formation of a new hybrid.

Therefore, the objective of this study was to evaluate the resistance of different tomato genotypes (with contrasting AS levels) and their parents (*Solanum lycopersicum* 'Redenção' and *S. pennellii*) to whitefly (*B. tabaci* biotype B) nymphs.

MATERIAL AND METHODS

Experimental location

The study was conducted at the Plant Physiology/Horticulture Laboratory, Department of Agronomy, Universidade Estadual do Centro-Oeste, Brazil at 1100 m above mean sea level and 25°23'01"S and 51°29'46"W.

Obtaining the F_2 and F_2RC_1 segregating populations

The segregating F_2 population was obtained from an interspecific cross between Redenção cultivar (a *S. lycopersicum* lineage with low AS levels and processing characteristics) and the wild *S. pennellii* accession LA-716 (a genotype with high AS levels and a source of resistance). For this, AS levels in 400 plants of the F_2 population, 40 *S. pennellii* plants (medium to high AS content), and 40 *S. lycopersicum* plants (Redenção cultivar, medium to low AS content) were measured using three samples of fully expanded leaflets from the upper third of the plants, following the methodology proposed by Resende et al. (2002a).

After the AS quantification of the F_2 population, the genotypes RVTA-2010-pl#31, RVTA-2010-pl#44, RVTA-2010-pl#83, RVTA-2010-pl#75, and RVTA-2010-pl#94 were selected for high AS levels, and the genotypes RVTA-2010-pl#33, RVTA-2010-pl#36, RVTA-2010-pl#39, and RVTA-2010-pl#73 for low AS levels.

The RVTA-2010-pl#31, RVTA-2010-pl#83, and RVTA-2010-pl#94 genotypes had high AS levels in the F_2 generation and originated from an interspecific cross between Redenção and the *S. pennellii* accession LA-716. Baier et al. (2015) previously selected these cultivars for resistance to spider mites (*Tetranychus urticae*), and by Dias et al. (2013) for resistance to the tomato pinworm (*Tuta absoluta*); these cultivars were cloned and backcrossed with the recurrent parent, Redenção. The cloning was performed using axillary buds taken from each genotype, which were transplanted into 60-cell polystyrene trays and identified. The clones of each genotype were then transplanted into 7-dm^3 pots. Cloning was conducted simultaneously with the sowing of Redenção. The backcrosses were realized when the selected genotypes (RVTA-2010-pl#31, RVTA-2010-pl#83, and RVTA-2010-pl#94) were at the flowering stage. The fruit from the backcross of each genotype was collected separately, and F_1RC_1 seeds were removed and sowed. The plants were inbred to obtain the F_2RC_1 generation, in which the AS content was again measured.

Quantification of AS content and selection of F_2RC_1 tomato genotypes

A total of 420 genotypes of the F_2RC_1 generation, 40 *S. pennellii* plants (medium to high AS content), and 40 *S. lycopersicum* plants (Redenção, medium to low AS content) were grown in a greenhouse. Parental *S. lycopersicum* and *S. pennellii* were used as standards in the AS quantification, as they contained low and high levels, respectively. To determine AS levels, six leaf discs of each plant were removed with a 0.9525-cm diameter punch and placed in test tubes. Subsequently, the ASs were extracted by the addition of dichloromethane and a colorimetric reaction using the Somogyi-Nelson test. The samples were analyzed in a UV visible Spectrophotometer model Cary 60 UV-Vis (Agilent Technologies, USA) and the absorbance was measured at a wavelength of 540 nm (Nelson, 1944). The AS concentrations in the leaflets were directly proportional to the absorbance values; the higher the absorbance values the higher the AS

levels. From the absorbance values, the AS content (mmol/cm) calculated.

Based on the absorbance values, plants from the F_2RC_1 population were selected for their AS levels. The genotypes selected for high AS levels were RVTA-2010-31-pl#177, RVTA-2010-83-pl#357, RVTA-2010-31-pl#310, RVTA-2010-31-pl#319, RVTA-2010-83-pl#346, RVTA-2010-31-pl#347, RVTA-2010-94-pl#378, and RVTA-2010-94-pl#381, while those selected for low AS levels were RVTA-2010-94-pl#50, RVTA-2010-94-pl#95, RVTA-2010-94-pl#258, and RVTA-2010-94-pl#272.

Whitefly resistance test in F_2 tomato genotypes

Tomato genotypes of the F_2 generation (Baier et al., 2015) with different AS levels. These genotypes, together with the F_1 hybrid (*S. lycopersicum* 'Redenção' x *S. pennellii* LA-716), were cloned using their axillary rooting buds in 60-cell plastic trays that were filled with a commercial substrate and grown in a greenhouse. For the parents, seeds were sown in a 128-cell polystyrene tray with commercial substrate.

The transplantation of selected F_2 genotypes, together with the F_1 hybrid and the parents (*S. lycopersicum* 'Redenção' and *S. pennellii* LA-716), was conducted in 7-dm^3 polyethylene pots filled with soil and substrate at a 1:1 ratio. The genotypes were kept in a greenhouse until the phenological stage of pre-flowering, before being subjected to whitefly (*B. tabaci* biotype B) infestation. The whitefly had been reared on sweet potato (*Ipomoea batatas*) clones in a greenhouse.

Randomized block design with four replications and 12 treatments, and each plot contained one genotype. The plants were infested with whitefly for 48 h, before being removed and kept in another compartment of the greenhouse.

The genotypes were evaluated for oviposition rates and the number of nymphs present on the leaflets. At each assessment, three leaflets from the top third, the middle third, and the bottom third of each plant were removed and taken to the laboratory. Evaluations were performed at 7 and 20 days after infestation. The first evaluation consisted of marking leaves in each third of the plant, in order to use the same leaves in the second evaluation. Eggs and nymphs were counted over a 2-cm^2 area of the leaf using a binocular stereoscopic microscope.

Whitefly resistance test in F_2RC_1 tomato genotypes

The bioassay was conducted in a greenhouse, a randomized block design was used in 14 treatments, and four replications, and each pot considered a block. The treatments were eight F_2RC_1 tomato genotypes that had been selected for high AS levels, four F_2RC_1 genotypes that had been selected for low AS levels, and the parents, the *S. pennellii* accession LA-716 and the commercial cultivar Redenção (*S. lycopersicum*).

A creation of whitefly (*B. tabaci* biotype B) was made in a wooden cage (1.0 x 1.0 x 1.20 m) with an anti-aphid screen, and the whitefly population was maintained on Santa Clara tomatoes.

The F_2RC_1 genotypes used for the resistance test were first cloned using their axillary buds in a 45-cell polypropylene tray, and the parents were sown in 128-cell polystyrene trays with commercial substrate. When the clones had well-developed root systems and the parents had three to five true leaves, they were transplanted into 7-dm^3 polypropylene vessels filled with a 1:1 ratio of commercial substrate and corrected by soil testing.

Forty-five days after transplantation, the number of eggs and nymphs on three leaves from the upper third, middle third, and bottom third of plants that had no pest or disease damage were counted.

The infestation was conducted by the removal of all of the pots covered with an anti-

aphid screen, and placed in the rows of the blocks where the F_2RC_1 genotypes with contrasting AS levels and parents were. The number of eggs and nymphs were counted at 2, 20, and 25 days after infestation. For this, a marked leaflet from each third of the plant was removed and taken to the Plant Physiology/Horticulture Laboratory. Counting was performed on the abaxial surface of the leaflet over a 2-cm^2 area with a stereomicroscope. The number of eggs and nymphs present at each evaluation was recorded as the average in each of three leaflets that belonged to each genotype within the block.

Statistical analysis

In order to test the normality and homogeneity of the data, Lilliefors (Sprent and Smeeton, 2007) and Bartlett (Steel et al., 1997) tests were used, respectively. Analysis of variance was then conducted, and the means were compared by a Scott-Knott test using the statistical program, SISVAR (Ferreira, 2008). Associations between AS levels and the resistance genotypes were estimated using Pearson correlations, and the significance of the correlations was calculated using the Student *t*-test in the ASSISTAT program (Silva and Azevedo, 2009). Pest resistance levels were compared between the parents (*S. lycopersicum* and *S. pennellii*) and the genotypes with different AS levels in both generations using SISVAR.

RESULTS

Whitefly resistance in F_2 tomato genotypes

After seven days of exposure to *B. tabaci* biotype B, the genotypes with high AS levels had significantly lower oviposition rates than the genotypes selected for low AS levels and the parent, Redenção (Table 1). They did not significantly differ from those on the wild parent, *S. pennellii*. After 20 days of infestation, whitefly exhibited the lowest preference for oviposition on the RVTA-2010-pl#94 genotype that had high AS levels, and all of the genotypes selected for high AS levels had significantly lower oviposition rates than the genotypes selected for low AS levels and Redenção.

After seven days of exposure, RVTA-2010-pl#94, RVTA-2010-pl#75, RVTA-2010-pl#83, RVTA-2010-pl#44, and RVTA-2010-pl#31, which had high AS levels, had 75, 77, 71, 75, and 68%, respectively, fewer eggs than Redenção. After 20 days of infestation, RVTA-2010-pl#94 had 78% fewer eggs than the commercial material.

Seven days after exposure, the average number of nymphs was not significantly different between genotypes with high AS levels and the LA-716 accession (Table 1). The genotypes with high AS levels were less attractive to the whitefly for oviposition than Redenção and those selected for low AS levels (RVTA-2010-pl#39 and RVTA-2010-pl#73). Twenty days after infestation, the average number of nymphs on the genotypes with high AS levels did not differ from that on LA-716. However, these genotypes were superior to the materials selected for low AS levels and Redenção, demonstrating their effectiveness in reducing the number of whitefly nymphs on the surface of the leaflets.

Seven days after infestation, RVTA-2010-pl#94, RVTA-2010-pl#75, RVTA-2010-pl#83, RVTA-2010-pl#44, and RVTA-2010-pl#31, which had high AS levels, had 74, 76, 73, 70, and 69%, respectively, fewer nymphs than Redenção; after 20 days of exposure, they had 82, 79, 71, 79, and 75%, respectively, fewer nymphs than Redenção.

The F_1 hybrid (Redenção x LA-716) had an AS level of 62. 83 mmol/cm, which was similar to that of Redenção (58.34 mmol/cm) but differed to that of LA-716 (214.54 mmol/cm). After 20

days, the F_1 hybrid had a similar mean number of eggs as Redenção, but after seven days, it was similar to LA-716. These results may be due to the presence of other unknown components that also promoted resistance, but were not selected during the breeding process.

Table 1. Acyl sugar content and mean number of whitefly (*Bemisia tabaci*) eggs and nymphs in 2-cm² sections of F_2 *Solanum lycopersicum* and *Solanum pennellii* leaflets.

Genotype	Acyl sugar content (mmol/cm)	No. of eggs		No. of nymphs	
		7 days	20 days	7 days	20 days
S. pennellii LA-716	214.54	1.00[a]	1.00[a]	1.00[a]	1.00[a]
RVTA-2010-pl#94 (high)	179.89	1.91[a]	2.36[b]	1.10[a]	1.73[a]
RVTA-2010-pl#75 (high)	196.81	1.89[a]	3.61[c]	1.00[a]	2.05[a]
RVTA-2010-pl#83 (high)	221.53	2.32[a]	3.68[c]	1.13[a]	2.85[a]
RVTA-2010-pl#44 (high)	221.59	1.97[a]	4.08[c]	1.25[a]	2.06[a]
RVTA-2010-pl#31 (high)	196.33	2.55[a]	3.37[c]	1.33[a]	2.40[a]
RVTA-2010-pl#36 (low)	41.63	4.78[b]	8.84[e]	1.50[a]	5.12[b]
RVTA-2010-pl#33 (low)	40.92	6.45[c]	8.54[e]	1.89[a]	5.46[d]
RVTA-2010-pl#39 (low)	42.16	6.66[c]	8.64[e]	2.87[b]	4.73[b]
RVTA-2010-pl#73 (low)	41.75	7.56[d]	8.65[e]	3.60[b]	5.29[b]
F_1 (Redenção x LA-716)	62.83	4.66[b]	6.71[d]	1.41[a]	4.51[b]
Redenção	58.34	7.87[d]	10.97[f]	4.24[b]	9.77[c]
CV %		53.14	44.83	42.59	44.97
Linear correlations		-0.91**	-0.91**	-0.68*	-0.79*
Parameter		Estimates			
C1- Genotypes (high) *vs* Genotypes (low)		-4.23**	-5.25**	-1.30	-2.93**
C2- LA-716 *vs* Genotypes (high)		-1.13	-2.42**	-0.16	-1.22
C3- LA-716 *vs* Genotypes (low)		-5.36**	-7.67**	-1.47	-4.15**
C4- Redenção *vs* Genotypes (high)		5.74**	7.54**	3.23**	7.55**
C5- Redenção *vs* Genotypes (low)		1.51	2.30**	1.93	4.62**

Means followed by the same letter did not differ from each other according to the Scott-Knott test ($P < 0.05$). CV = coefficient of variation. **,*Significant at $P \leq 0.01$ and $P \leq 0.05$, respectively, by the Student *t*-test.

The linear correlations that compared AS levels with average oviposition rates and number of nymphs revealed the important role of ASs in decreasing the average number of ovipositions and nymphs (Table 1). The correlations were significant and negative for the average number of eggs ($r = -0.91$; $P < 0.001$) and for the average number of nymphs at 7 and 20 days ($r = -0.68$ and -0.79, respectively; $P < 0.05$). From the C1 contrast (Table 1), we can infer that the genotypes with high AS levels were more effective in decreasing preference for oviposition at 7 and 20 days, and reduced the number of viable nymphs after 20 days, than the genotypes with low AS levels.

The C2 contrast showed that, on average, the genotypes selected for high AS levels did not differ from the *S. pennellii* accession LA-716 in the average number of eggs at seven days and the average number of nymphs after 7 and 20 days. However, the C3 contrast revealed significant differences between the genotypes selected for low AS levels and LA-716 in the number of eggs (7 and 20 days) and nymphs (20 days) after infestation (Table 1).

The comparison between the cultivar Redenção and genotypes with high AS levels (C4 contrast) showed an increase in the average number of eggs and nymphs in all evaluations. Therefore, crossing *S. pennellii* with Redenção enabled the F_2 population to obtain genotypes with a satisfactory level of resistance to *B. tabaci* biotype B, because there were significant differences at all evaluations in the numbers of eggs and nymphs, with the lowest in the genotypes with high AS levels. Finally, the C5 contrast showed that the genotypes with low AS levels had similar mean numbers of eggs and nymphs after seven days of infestation as Redenção (Table 1).

Whitefly resistance test in F_2RC_1 tomato genotypes

Whitefly oviposition preference was significantly influenced at all evaluations by the levels of AS in the F_2RC_1 genotypes, which were obtained by the crossing of the wild accession LA-716 (*S. pennellii*) with the commercial line, Redenção (*S. lycopersicum*) (Table 2). The whitefly least-preferred the genotypes with high AS levels for oviposition, and most-preferred the genotypes with low AS levels and Redenção.

Two days after exposure to whitefly, RVTA-2010-31-pl#177, RVTA-2010-83-pl#357, RVTA-2010-31-pl#310, RVTA-2010-31-pl#319, RVTA-2010-83-pl#346, RVTA-2010-31-pl#347, RVTA-2010-94-pl#378, and RVTA-2010-94-pl#381, which had high AS levels, had 89, 80, 98, 76, 70, 85, 85, and 87%, respectively, fewer eggs than Redenção; similar results were obtained at 20 days (93, 81, 93, 95, 88, 86, 98, and 95%, respectively, fewer eggs than Redenção) and 25 days (95, 95, 91, 67, 49, 77, 91, and 91%, respectively, fewer eggs than Redenção). After two days of whitefly exposure, there were no significant differences in the number of nymphs present (Table 2), probably because there was not enough time for the eggs to develop into nymphs.

The F_2RC_1 genotypes with high AS levels were effective in reducing the number of nymphs on the leaflets, with results similar to those obtained in the wild parent in all evaluations performed, and significantly differed from the genotypes selected for low AS levels and Redenção (Table 2). At 20 days, no nymphs were found on RVTA-2010-31-pl#177, RVTA-2010-83-pl#357, and RVTA-2010-94-pl#381, which had high AS levels. The other genotypes with high AS levels (RVTA-2010-31-pl#310, RVTA-2010-31-pl#319, RVTA-2010-83-pl#346, RVTA-2010-31-pl#347, and RVTA-2010-94-pl#378) had 92, 97, 71, 83, and 97%, respectively, fewer nymphs than Redenção. After 25 days of exposure, there were 95, 95, 91, 67, 49, 77, 91, and 91% fewer nymphs in genotypes with high AS levels (RVTA-2010-31-pl#177, RVTA-2010-83-pl#357, RVTA-2010-31-pl#310, RVTA-2010-31-pl#319, RVTA-2010-83-pl#346, RVTA-2010-31-pl#347, RVTA-2010-94-pl#378, and RVTA-2010-94-pl#381, respectively) than Redenção.

Table 2. Acyl sugar content and mean number of whitefly (*Bemisia tabaci*) eggs and nymphs in 2-cm² sections of F_2RC_1 *Solanum lycopersicum* and *Solanum pennellii* leaflets.

Genotype	Acyl sugar content (mmol/cm)	No. of eggs			No. of nymphs	
		2 days	20 days	25 days	20 days	25 days
S. pennellii LA-716	243.00	0.00^a	0.08^a	0.00^a	0.00^a	0.00^a
RVTA-2010-31-pl#177 (high)	170.10	0.50^a	0.25^a	0.08^a	0.00^a	0.16^a
RVTA-2010-83-pl#357 (high)	188.91	0.91^a	0.67^a	0.33^a	0.00^a	0.16^a
RVTA-2010-31-pl#310 (high)	163.04	0.08^a	0.25^a	0.17^a	0.25^a	0.25^a
RVTA-2010-31-pl#319 (high)	256.32	1.08^a	0.17^a	0.67^a	0.08^a	1.00^a
RVTA-2010-83-pl#346 (high)	189.69	1.33^a	0.42^a	1.17^a	0.83^a	1.49^a
RVTA-2010-31-pl#347 (high)	181.47	0.66^a	0.50^a	0.83^a	0.50^a	0.66^a
RVTA-2010-94-pl#378 (high)	174.02	0.67^a	0.08^a	0.41^a	0.08^a	0.25^a
RVTA-2010-94-pl#381 (high)	190.48	0.58^a	0.16^a	0.08^a	0.00^a	0.25^a
RVTA-2010-94-pl#50 (low)	47.03	3.00^b	4.25^b	3.33^b	4.25^c	5.41^c
RVTA-2010-94-pl#95 (low)	56.83	1.80^b	4.42^b	3.33^b	1.99^b	2.58^b
RVTA-2010-94-pl#272 (low)	60.75	5.25^b	2.25^b	2.67^b	1.91^b	2.99^b
Redenção	56.05	4.50^b	3.50^b	3.17^b	3.00^b	2.92^b
CV %		120.70	139.82	87.58	118.94	95.78
Linear correlations		-0.78**	-0.90**	-0.89**	-0.87**	-0.82**
Parameter		Estimate				
C1- Genotypes (high) *vs* Genotypes (low)		-2.60	-3.33**	-2.71**	-2.50**	-3.13**
C2- LA-716 *vs* Genotypes (high)		-0.73	-0.23	-0.40	-0.22	-0.50
C3- LA-716 *vs* Genotypes (low)		-3.33	-3.57	-3.18**	-2.72	-3.82**
C4- Redenção *vs* Genotypes (high)		3.77	3.18	2.70	2.78	2.38
C5- Redenção *vs* Genotypes (low)		1.17	-0.14	-0.01	0.27	-0.75

Means followed by the same letter did not differ from each other according to the Scott-Knott test ($P < 0.05$). CV = coefficient of variation. **Significant at $P \leq 0.01$ by the Student *t*-test.

Significant negative correlations ($P < 0.01$) were found between the number of eggs at 2, 20, and 25 days of exposure (-0.78, -0.90, and -0.89, respectively) and nymphs at 20 and 25 days of exposure (-0.87 and -0.82, respectively) and AS levels. Therefore, the higher the AS levels, the lower the oviposition rates and number of nymphs (Table 2).

The C1 contrast demonstrated that the responses of genotypes with high AS levels were inversely proportional to those with low AS levels, suggesting that high AS levels were effective in reducing the number of eggs and nymphs after 20 and 25 days of infestation (Table 2). The C2 contrast showed that there was no statistical difference between LA-716 and the genotypes with high AS levels. The C3 contrast compared the results of the wild parent with the low-AS genotypes, and a significant difference was found in the numbers of eggs and nymphs after 25 days of exposure (Table 2). Finally, the C5 contrast showed that there was no statistical difference between the low-AS genotypes and Redenção (Table 2).

DISCUSSION

The tomato genotypes selected for high AS levels had similar values to those observed in the *S. pennellii* accession LA-716 (a parent with high AS levels), while those selected for low AS levels had values similar to the Redenção cultivar (*S. lycopersicum*) that had naturally low AS levels (Table 1). The F_1 hybrid between *S. lycopersicum* and *S. pennellii* had AS levels that were similar to those of Redenção. This was probably due to the type of inheritance involved in the genetic control of this trait, because a monogenic genetic inheritance is presumed where a major-effect gene, recessive, and low additive-effect genes are involved (Resende et al., 2002b).

The average number of whitefly eggs on the tomato leaflets was significantly lower in genotypes that had high AS levels in the F_2 generation (Table 1), as well as in the F_2RC_1 generation (Table 2). This demonstrates the important role that ASs play in whitefly control, by reducing the preference for oviposition. These sugars, which are glucose esters and sucrose that is exuded by glandular trichomes (type IV), are present in leaflets of wild *S. pennellii* and *Solanum pimpinellifolium*, and have a negative effect on the development of arthropod pests (Slocombe et al., 2008; Schilmiller et al., 2009; Rodíguez-López et al., 2011). They promote a type of resistance known as antixenosis, or no preference, in which the plant is not attractive to pests for oviposition, shelter, or food (Lucini et al., 2015).

The low *B. tabaci* biotype B oviposition rate on genotypes with high AS levels reduces the amount of chemical pesticides required, thereby reducing production costs. Lucini et al. (2015), working with F_2 genotypes of the same cross that were selected for high AS levels and infested with spider mites, demonstrated that the high resistance levels of these tomato genotypes are associated with the density of type IV trichomes and the AS concentration. Genetic studies have demonstrated high correlations between whitefly resistance and the presence of type IV trichomes (Fancelli and Vendramim, 2002; Muigai et al., 2003; Oriani and Vendramim, 2010; Rodríguez-López et al., 2011).

Maciel et al. (2009), Resende et al. (2009), and Silva et al. (2009) reported that tomato genotypes selected for high AS levels had low rates of *B. tabaci* biotype B oviposition, and the results of the present study support this observation.

Apart from RVTA-2010-pl#33 and RVTA-2010-pl#36, which were genotypes of the F_2 generation and had low AS levels (Table 2), the largest number of nymphs was found on genotypes that had low AS levels (Tables 1 and 2). Freitas et al. (2000) and Fancelli et al. (2005) also demonstrated the important role that ASs, which type IV trichomes produce, play in whitefly resistance. The genotypes that were selected for high AS levels in the F_2 and F_2RC_1 generations exhibited similar resistances to whitefly as *S. pennellii* (Tables 1 and 2), except for the average number

of eggs after seven days in the F_2 genotypes (Table 2). This is important, because it shows that the high AS levels present in the leaflets were the main resistance factor in the wild accession, and that the breeding program was effective in producing generations without decreasing resistance levels.

Our results confirm those obtained by Resende et al. (2009), who found that ASs are the main factor in tomato resistance to *B. tabaci* biotype B. The authors state that genotypes selected for high AS levels should be used in breeding programs, in order to obtain advanced lineages through backcrossing that can be used for further crosses to obtain hybrids (Resende et al., 2009). The significant, negative correlations we found between AS levels and whitefly oviposition rates and number of nymphs confirm the importance of ASs in reducing attacks by this pest species. Dias et al. (2013) found significant, negative correlations between AS content and the number of *T. absoluta* eggs and larvae on tomato leaflets. However, Resende et al. (2009) found no significant correlation between AS content and whitefly oviposition rates, but did find a significant negative correlation between AS levels and the number of nymphs. Baier et al. (2015) reported significant negative correlations between AS levels and the number of spider mites on tomato plants after 60 min of exposure to the mites.

The C1 contrast demonstrated the importance of ASs in pest resistance, confirming that they are an important form of resistance to whitefly (Maluf et al., 2010). Silva et al. (2009) investigated whitefly resistance in tomato hybrids that were obtained from lineages with high levels of zingiberene and ASs. The authors found that hybrids heterozygous for zingiberene or ASs had the same whitefly resistance as double-heterozygous hybrids, showing that these allelochemicals act similarly in terms of whitefly resistance in different genotypes, and do not exhibit any synergistic effects (Silva et al., 2009).

The present study demonstrated that the direct selection for genotypes with high AS levels is effective in selecting plants with a satisfactory level of whitefly resistance. This is favorable for breeding programs, because the selection of resistant genotypes dispenses with the need to test resistance to pests, thereby accelerating the development of materials. Our results are similar to those obtained by Resende et al. (2002b), who reported that tomato plants with high AS levels exhibited moderately high values of heritability with a simple inheritance pattern, which increases the effectiveness of selecting genotypes in segregating populations. Direct selection for high AS levels may be more efficient in obtaining tomato genotypes with high pest resistance than direct resistance (Gonçalves et al., 2007).

Therefore, the backcrosses were effective in maintaining the resistance genes. In addition to the direct selection for high AS levels, it is important to use resistance bioassays. In the F_2 generation, the genotypes RVTA-2010-pl#31 and RVTA-2010-pl#94 were selected with high AS levels and were resistant to whitefly. Genotypes of the F_2RC_1 generation with high AS levels (RVTA-2010-31-pl#177 and RVTA-2010-94-pl#381) could be used to obtain lineages that are resistant to whitefly.

Conflicts of interest

The authors declare no conflict of interest

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