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Biology of *Bemisia tabaci* (Genn.) B biotype (Hemiptera, Aleyrodidae) on tomato genotypes

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ABSTRACT: Brazil is one of the main tomato (*Solanum lycopersicum*) producers worldwide. Nevertheless, considerable part of the production is lost due to *Bemisia tabaci* (Genn.) B biotype attacks. Resistant plants can be an important method for controlling this pest in an integrated pest management. Tests for evaluating some biological aspects of *B. tabaci* were carried out on 18 tomato genotypes, in controlled laboratory greenhouse conditions. Thirty-day-old plants placed in plastic cages were infested with 20 whitefly pairs each, for 24 h. The development of at least 30 eggs in three leaflets per plant (repetition) was observed until adult emergence. The development period of insects grown in LA1335, PI365928 and LA722 genotypes took three days longer when compared to the ones grown in PI134418 (20.3 days). The highest mortality rate of whitefly nymphs occurred in PI365928, LA1335 and LA722 genotypes (63.8, 54.5 and 53.3%, respectively), and the smallest ones in IAC294 and IAC68F-22-2 genotypes (4.9 e 6.2%, respectively). LA1335, PI365928 and LA722 genotypes presented moderate feeding nonpreference and/or antibiosis-based resistance to *B. tabaci* B biotype. Key words: insecta, whitefly, antibiosis, antixenosis

Biologia de *Bemisia tabaci* (Genn.) Biótipo B (Hemiptera, Aleyrodidae) em genótipos de tomateiro

RESUMO: O Brasil é um dos maiores produtores mundiais de tomate (*Solanum lycopersicum*), porém grande parte da produção é perdida devido ao ataque de *Bemisia tabaci* (Genn.) biótipo B. Entre as táticas de controle dessa praga num manejo integrado de pragas, pode-se relacionar a resistência de plantas. Ensaios para avaliar alguns aspectos biológicos de *B. tabaci* foram realizados com 18 genótipos de tomateiro, em condições controladas de laboratório (casa de vegetação). Plantas com 30 dias de idade foram colocadas em gaiolas plásticas e infestadas com 20 casais de moscas-brancas cada, durante 24h. Acompanhou-se então o desenvolvimento de pelo menos 30 ovos em três folíolos por planta (repetição) até a emergência dos insetos. Os insetos criados nos genótipos LA1335, PI365928 e LA722 apresentaram prolongamento de três dias no período de desenvolvimento, em relação aos criados em PI134418 (20,3 dias). As maiores taxas de mortalidade das ninfas de mosca-branca ocorreram nos genótipos PI365928, LA1335 e LA722 (63,8, 54,5 e 53,3%, respectivamente) e as menores, nos genótipos IAC294 e IAC68F-22-2 (4,9 e 6,2%, respectivamente). Os genótipos LA1335, PI365928 e LA722 apresentara do tipo não-preferência para alimentação e/ou antibiose a *B. tabaci* biótipo B. Palavras-chave: Insecta, mosca-branca, antibiose, antixenose

Introduction

Brazil produced around 3.35 million tons of tomato (Solanum lycopersicum) (= Lycopersicon esculentum Mill) (Peralta et al., 2006) in 2007, being considered at that time, one of the major producers of this product worldwide (Agrianual, 2009). Nearly 28% of the production is used in the industry, and 72% in natura consumption. Nowadays, the whitefly Bemisia tabaci (Genn.) B biotype is one of the main pests both for industrial processing and for fresh tomato. In addition to direct damages caused by extracting large quantities of phloem sap, it also transmits plant viruses (Villas Bôas, 2005). This is the main limiting factor for tomato crop in many producing regions (Markham et al., 1994; Amari et al., 2008; García-Cano et al., 2008), interfering in the tomato production chain, which holds great economical and social importance in Brazil (EMBRAPA/CNPH, 2006). Losses due to virus transmission range from 40 to 70%, if the plants are infected within 5-6 weeks after germination (Villas Bôas, 2005).

The improvement of germplasm aiming to the development of resistant genotypes to *Bemisia* spp. may be an important tool in integrated pest management of whitefly (McAuslane et al., 1996), thus preventing unnecessary use of insecticides, which are still widely used in pest control. Nowadays, the cloned *Mi-1* gene is one of the most important tomato resistant genes as it confers resistant to B and Q biotypes of *B. tabaci* and also to nematodes (*Meloidogyne* spp.) and potato aphid [*Macrosiphum euphorbiae* (Thomas)] (De Ilarduya et al., 2004; Jiang et al., 2001; Muñiz and Nombela, 2001; Nombela et al., 2000, 2001, 2003). Several authors have observed that the tomato genotypes, LA716 (*S. pennellii* = *L. pennellii*), PI134417 and PI134418 (*S. habrochaites* f. glabratum = *L. hirsutum* f. glabratum) showed certain resistance to *B. tabaci* B biotype (Heinz and Zalom, 1995; Fancelli and Vendramim, 2002; Toscano et al., 2002; Muigai et al., 2003; Fancelli et al., 2003, 2005; Baldin et al., 2005). This resistance was associated to the presence of glandular trichomes that release exudates, type IV in *S. pennellii* (Nombela et al., 2000), and types IV and VI in *S. habrochaites* f. glabratum (Williams et al., 1980; Channarayappa et al., 1992). Low levels of feeding nonpreference and/or antibiosis were observed in PI127826, PI134417, PI134418 and LA444-1, which lengthened the cycle of the insect when compared with PI126931 (Baldin et al., 2005).

Therefore, this research aimed to evaluate some biological aspects of *B. tabaci* B biotype on 18 tomato genotypes.

Material and Methods

Eighteen tomato genotypes were evaluated: Santa Clara, Fanny (S. lycopersicum); VFNA (S. lycopersicum cerasiforme); LA716 (S. pennellii = L. pennellii); LA1963 (S. chilense = L. chilense); LA371, LA444-1, LA462 (S. peruvianum = L. peruvianum); IAC237, LA722, LA1335, NAV1062, PI126931, PI365928 (S. pimpinellifolium = L. pimpinellifolium); PI134417, PI134418 (S. habrochaites f. glabratum = L. hirsutum f. glabratum); IAC294 (S. habrochaites); IAC68F-22-2 (S. peruvianum × S. lycopersicum). The experiments were carried out in laboratory under the following conditions: $23 \pm 2^{\circ}$ C temperature, $70 \pm 10\%$ relative humidity and L13:D11h (Light:Dark) photoperiod.

The seeds of tomato genotypes were germinated in plastic trays containing a substrate composed of vermiculite, perlite, pinus bark and peat. Fifteen days after the sowing, the seedlings were transplanted into 0.5 L plastic pots containing Plantmax Hortaliças[®] substrate (one seedling per pot), irrigated with nutritive solution [1M KNO₃ = 5 mL L⁻¹; 1M KH₂PO₄ = 1 mL L⁻¹; 1M MgSO₄ = 2 mL L⁻¹; FeEDTA = 1 mL L⁻¹; micronutrients (H₃BO₃, MnCl₂×4H₂O, ZnCl₂, CuCl₂ and H₂MoO₄×H₂O) = 1 mL L⁻¹; 1M Ca(NO₃)₂ = 5 mL L⁻¹] (Sarruge, 1975) and maintained in greenhouse.

For whitefly rearing, a colony was initiated from a population previously characterized as *B. tabaci* (B biotype). The insects were reared on soybean [*Glycine max* (L.) Merrill] and painted spurge (*Euphorbia heterophylla* L.) plants kept in a greenhouse with anti-aphid screens.

For plant infestation, a transparent plastic cage (16 cm height and 13 cm diameter) was used. The cage had a plastic lid with a 6 cm diameter hole covered with an anti-aphid screen to facilitate the ventilation of the cage. Experimental insects were introduced into the cage through a hole on the side of the cage. The cages were placed in plastic pots containing 1-month-old tomato seedlings and held in place with the aid of a masking tape 4 cm width. Each plant was infested by 20 whitefly

pairs during 24 h, except LA716, PI134417 and PI134418 genotypes, for which 40 pairs were used during 72 h, in order to reach enough eggs to perform the experiment. Then, the adults were removed and the number of eggs found in the abaxial surface of three leaflets per plant was registered, containing at least 30 eggs in each.

The development of the immatures was observed until adult emergence, and the following parameters were evaluated: number of eggs, ecloded nymphs and emerged adults, in addition to the eggs viability and nymphs mortality. During the experiment, plants were kept in chambers growth of multi-tiered shelving without sidewall and with individual lighting control and irrigated with nutritional solution when needed.

The trials were set up in a randomized block design with six repetitions for each tomato genotype evaluated. The data obtained were first analyzed through the test of homocedasticity by Bartlett, which were transformed accordingly, and then the data were submitted to analysis of variance by the F-test and the means were compared by Tukey test ($p \le 0.05$).

Results and Discussion

The viabilities of the eggs were not different (Table 1). In tomato, high percentages (approximately 99%) of ecloded whitefly nymphs have also been verified by Hendi et al. (1985) in controlled conditions of temperature and humidity ($30 \pm 2^{\circ}$ C and $60 \pm 5^{\circ}$ %). In mung beans (*Phaseolus radiatus* L.), Verma et al. (1990) observed that this parameter ranged from 84% at 23°C to 92% at 27°C and Butler Jr. et al. (1983) reported that the lowest percentages of ecloded nymphs of *B. tabaci* were from 68% at 26.7°C to 75% at 32.2°C, in cotton plants (*Gossypium hirsutum* L.). Also, Wagner (1995) suggested that there was a decreasing tolerance of eggs when high temperatures were reached at the hottest times of the year.

The total development period was longer for insects reared on LA1335 (23.5 days), PI365928 and LA722 (23.4 days) genotypes, when compared with values recorded on PI134418, LA462, IAC68F-22-2, PI134417, LA1963, NAV1062, IAC294, LA371, LA444-1 and LA716, which ranged from 20.3 to 21.1 days. However, the genotype that presented the longest whitefly development period (LA1335) did not differ from PI126931, IAC237, 'Fanny' and 'Santa Clara'. The immature mortality rate was higher for insects raised on PI365928 (63.8%), LA1335 (54.5%) and LA722 (53.3%) and differed of the ones found on IAC294 (4.9%) and IAC68F-22-2 (6.2%), which presented the lowest nymphs mortality rates (Table 1).

Nombela et al. (2000) and Heinz and Zalom (1995) reported no oviposition on LA716 genotype, and others authors have excluded LA716 from the feeding nonpreference and/or antibiosis test, once the number of eggs required for the study of the whitefly biology was not reached, requisite for the performance of the experiment (Fancelli and Vendramim, 2002; Baldin et al., 2005). Such problem has also been noted in this test for LA716, PI134417 and PI134418 genotypes. Consequently,

Genotype -	Eggs		Nymphs		
	n	Viability	n	Mortality ¹	Development period
		%		%	days
LA1335	399	93.7 ± 2.5	389	54.5 ± 8.4 a	23.5 ± 0.5 a
PI365928	407	90.0 ± 6.3	360	63.8 ± 15.4 a	23.4 ± 0.6 ab
LA722	327	86.5 ± 7.3	293	53.3 ± 6.6 ab	23.4 ± 0.3 ab
PI126931	288	88.5 ± 4.9	252	38.6 ± 14.2 abc	22.7 ± 0.5 abc
IAC237	268	89.4 ± 4.5	239	$29.7 \pm 3.2 \text{ abc}$	22.3 ± 0.3 abcd
Fanny	431	97.4 ± 1.7	422	25.0 ± 7.4 abc	$22.2 \pm 0.4 \text{ abcd}$
Santa Clara	431	95.5 ± 1.4	419	30.7 ± 10.31 abc	22.2 ± 0.3 abcd
VFNA	329	97.2 ± 2.8	324	27.1 ± 10.6 abc	21.5 ± 0.6 bcd
LA716	642	94.1 ± 3.0	575	31.0 ± 11.1 abc	$21.1 \pm 0.8 \text{ cd}$
LA444-1	476	95.1 ± 1.2	445	32.3 ± 11.1 abc	$21.1 \pm 0.3 \text{ cd}$
LA371	361	97.3 ± 2.0	345	13.7 ± 5.3 abc	21.0 ± 0.5 cd
IAC294	428	98.8 ± 0.9	425	4.9 ± 2.3 c	$21.0 \pm 0.3 \text{ cd}$
NAV1062	340	89.7 ± 4.8	323	20.2 ± 13.5 abc	20.9 ± 0.9 cd
LA1963	541	98.4 ± 1.0	532	$32.0 \pm 7.8 \text{ abc}$	20.9 ± 0.5 cd
PI134417	221	96.2 ± 1.5	210	15.6 ± 5.6 abc	$20.9 \pm 0.3 \text{ cd}$
IAC68F-22-2	532	99.1 ± 0.6	527	6.2 ± 2.6 bc	$20.9 \pm 0.2 \text{ cd}$
LA462	432	97.4 ± 1.5	427	19.0 ± 6.3 abc	20.4 ± 0.3 d
PI134418	367	92.5 ± 3.3	341	$15.7 \pm 5.3 \text{ abc}$	20.3 ± 0.3 d
F		1 43 ^{NS}		2 71*	7 36*

Tab

Means followed by the same letters within columns are not different (Tukey's test, $p \le 0.05$). ¹Original data; transformed in arcsin $(x/100)^{1/2}$ for analysis.

we decided to leave the females confined for a longer period of time, three days on LA716 and two days on PI134417 and PI134418. Also we duplicated the number of infesting insects (40 pairs), in order to provide enough number of eggs to make possible the inclusion of these genotypes in this experiment. However, the LA716 genotype (S. pennellii) showed susceptibility in the feeding nonpreference and/or antibiosis test, once B. tabaci B biotype presented a short development (21.1 days) and a lower nymphs mortality rate (31%) (Table 1). Such fact has also been verified for PI134417 and PI134418 genotypes (S. habrochaites f. glabratum), on which the whiteflies presented short development periods (20.9 and 20.3 days, respectively) and low nymphs mortality (15.6 and 15.7%, respectively) (Table 1).

8.55

F CV%

Since LA716, PI134417 and PI134418 presented high oviposition nonpreference resistance in relation to B. tabaci B biotype (Heinz and Zalom, 1995; Fancelli and Vendramim, 2002; Toscano et al., 2002; Muigai et al., 2003; Fancelli et al., 2003, 2005; Baldin et al., 2005), these genotypes would not have to spend extra energy in a second type of resistance: the feeding nonpreference and/ or antibiosis. Many times, the whitefly is inhibited to oviposite on LA716 genotype, and this antixenosis for oviposition is associated to the presence of glandular trichomes type IV (Williams et al., 1980; Heinz and Zalom, 1995; Nombela et al., 2000), in addition to its exudates entrap adults (Toscano et al., 2001; Freitas et al., 2002; Muigai et al., 2002; Fancelli et al., 2003, 2008).

4.33

54.59

Baldin et al. (2005) suggest that PI134417, PI134418 and LA444-1 genotypes presented feeding nonpreference and/or antibiosis, even though in low levels, since they prolong the development period of the whitefly (28.1 to 27.7 days) when compared to the most susceptible genotype, PI126931 (27.2 days). Nevertheless, the development periods of whiteflies obtained by Baldin et al. (2005) were considerably higher to the ones observed in this research - PI134418 (20.3 days), PI134417 (20.9 days), LA444-1 (21.1 days), and PI126931 (22.7 days) - all of them classified as susceptible. The experimental conditions in both experiments were similar (temperature from 25 \pm 2°C, relative humidity of 70 \pm 10% and photophase of 13h), which are in within the optimum range for its development, 20-30°C (Wang and Tsai, 1996).

Many authors have reported the influence of temperature on the development of B. tabaci. In cotton plants, the life cycle of the whitefly ranged from 17 days (30°C) to approximately 70 days (15°C) (Butler Jr. et al., 1983; Wagner, 1995); in eggplants (Solanum melongena L.), it ranged from 14 days (30°C) to 105 days (15°C) (Wang and Tsai, 1996). Temperatures over 35°C and extreme air relative humidity were also not favorable to the whitefly development (Avidov, 1957; Gerling et al., 1986; Horowitz, 1986; Wagner, 1995; Wang and Tsai, 1996), giving emphasis to low humidity.

The necessary time for whitefly to complete its development, also depends on its host. Coudriet et al. (1985) observed that the development occurred in a period which was 30% lower in lettuce (Lactuca sativa L.), cucumber (Cucumis sativus L.), eggplant and pumpkin (Cucurbita maxima Dene.) than in broccoli (Brassica oleracea L.) or carrot (Daucus carota L.). The development period of B. argentifolii (= B. tabaci B biotype) ranged from 17.3 days on eggplants to 20.9 days on bean (Phaseolus vulgaris L.) (Tsai and Wang, 1996), and was 23 days in poinsettia (Euphorbia pulcherrima Willd. ex Klotzsch) and 25 days in cotton (Bethke et al., 1991). In tomato plants, the development period was 20 days (Hendi et al., 1985; Islan and Shunxiang, 2007). These data were very similar to the ones observed in this experiment; however Islan and Shunxiang (2007) worked in the same conditions as this assay, while for Hendi et al. (1985), the temperature ranged between $30 \pm 2^{\circ}C$ and the relative humidity was $65 \pm 5\%$.

Resistant genotypes manifest its adverse effects especially related to insect biology, promoting an increase in the development period; mortality of immatures; mortality before reaching adulthood; size and weight reduction of the individuals; reduction of fecundity; alteration of sexual proportion; and decrease in the longevity of the insect (Painter, 1951; Beck, 1965; Lara, 1991). The development period of insects grown in LA1335, PI365928 and LA722 genotypes took three days longer when compared to the ones grown in PI134418 (20.3 days), on which the development period was shorter. The biggest mortality rates of whitefly nymphs also occurred in PI365928, LA1335 and LA722 genotypes (63.8, 54.5 and 53.3%, respectively) and the smallest in IAC294 and IAC68F-22-2 genotypes (4.9 and 6.2%, respectively).

It is difficult to determine what kind of resistance mechanism (feeding nonpreference and/or antibioses) is involved on LA1335, PI365928 and LA722 genotypes, since no technique such as electrical penetration graph (EPG) was available which could reveal detailed information about the insect feeding. Nevertheless, it can be inferred that LA1335, PI365928 and LA722 genotypes presented moderate feeding nonpreference and/or antibiosis resistance to *B. tabaci* B biotype.

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