Analysis by DC-EPG of the resistance to *Bemisia tabaci* on an *Mi*-tomato line

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Abstract

The tomato *Mi* gene confers resistance to nematodes, *Meloidogyne* spp., and to the potato aphid, *Macrosiphum euphorbiae* (Thomas). Previous greenhouse choice assays with *Bemisia tabaci* (Gennadius) showed that tomato commercial varieties carrying this gene had significantly lower values of host suitability and whitefly reproduction than varieties lacking *Mi*. This indicated that *Mi*, or another gene in its region, could regulate partial resistance. In order to characterise this resistance, probing and feeding behaviour of *Bemisia tabaci* B-biotype was studied with DC Electrical Penetration Graph (EPG) technique on the near-isogenic tomato lines Moneymaker (without *Mi*) and Motelle (carrying *Mi*). Significant differences ($P < 0.05$) between tomato lines were found in EPG parameters related to epidermis and/or mesophyll tissues. On Motelle, a lower percentage of whiteflies achieved phloem phase and they made more probes before attaining first phloem phase, had a higher ratio (number of probes before first phloem phase)/(total number of probes), had a longer total duration of non-probing time, and a longer time before making the first intracellular puncture and before making the first phloem phase. In contrast, most of the parameters related to phloem phase were found not to differ significantly between these near-isogenic lines. The behavioural data strongly suggest that the partial resistance in the variety Motelle is due to factors in the epidermis and/or mesophyll that inhibit the whiteflies from reaching phloem sieve elements. However, once the stylets reach a sieve element, whitefly behaviour did not differ between the two varieties. Thus, phloem sap of the two varieties appears to be equally acceptable to the whiteflies. Further studies are necessary to provide a better understanding of these mechanisms of resistance to whiteflies in tomatoes.

Introduction

The sweetpotato whitefly, *Bemisia tabaci* (Gennadius) is a devastating pest of vegetables, ornamentals, and agronomic crops throughout the tropical and subtropical regions of the world. Losses caused by *B. tabaci* to tomato, *Lycopersicon esculentum* Mill., are the result of direct feeding damage, deposition of large quantities of honeydew, and transmission of several plant viruses (Moriones et al., 1993; Bedford et al., 1994; Markham et al., 1994). Because of environmental concerns on the use of pesticides in agriculture, alternate means of *B. tabaci* control are being sought. As an alternative approach to chemical control, host plant

resistance has been increasingly recognised as being one of the most desirable and economic control tactics in the management of whiteflies.

The *Mi* gene of tomato was reported to confer effective field resistance to three root-knot nematode species, *Meloidogyne incognita* Chitwood, *M. javanica* Chitwood and *M. arenaria* Chitwood (Gilber & McGuire, 1956; Braham & Winsted, 1957; Roberts & Thomason, 1986). It also was reported that *Mi* controlled resistance to reproduction of a race and one isolate of another race of *M. chitwoodi* Golden in the greenhouse (Brown et al., 1997). The *Mi* gene was cloned (Milligan et al., 1998) and described as dominant, or semi-dominant in tomato, and was

introduced into tomato from the wild species *Lycopersicon peruvianum* using embryo rescue of an interspecific cross of this wild species with *L. esculentum* (Smith, 1944). Tomato lines that carried the nematode-resistance gene, *Mi*, displayed resistance in the field to the potato aphid *Macrosiphum euphorbiae* (Thomas). In the greenhouse this insect had significantly higher mortality and lower fecundity on tomato lines carrying-*Mi* than on near isogenic lines lacking-*Mi* (Kaloshian et al., 1997). Finally, *Mi* was reported to confer resistance against the potato aphid (Rossi et al., 1998). Our recent greenhouse choice assays with *B. tabaci* demonstrated that tomato varieties carrying this gene showed significantly lower values of host suitability and whitefly reproduction than varieties lacking *Mi* (Nombela et al., 2000), indicating that *Mi* or another gene in its region, could regulate partial resistance to whiteflies.

Detailed behavioural studies can play an important role in determining mechanisms of plant resistance to whiteflies. As phloem sap feeders, whiteflies penetrate their mouthparts into the leaf tissue, covering the distance from the epidermis to the phloem vessel intercellularly, and finally feed on substances in the sieve elements (Pollard, 1955). In this way whiteflies can locate possible plant resistance factors at the successive levels of the tissues encountered from the surface to the phloem sieve element. Thus, the study of stylet penetration behaviour can provide clues for determining what kind of resistance mechanisms may be involved. No technique has been more useful in revealing detailed information about stylet penetration behaviour of homopteran insects than the electrical penetration graph (EPG) technique, which was first developed for aphids by McLean & Kinsey (1964) using an AC circuit and later modified by Tjallingii (1978) using a DC circuit. However, only recently this technique has been used to study the feeding behaviour of adult whiteflies *Trialeurodes vaporariorum* (Westwood) and *Bemisia tabaci* (Gennadius) on a DC system (Janssen et al., 1989; Lei et al., 1996, 1998, 1999; Jiang et al., 1999, 2000; Johnson & Walker, 1999) and for *Bemisia argentifolii* Bellows & Perring and *Parabemisia myricae* (Kuwana) with an AC system (Walker & Perring, 1994).

Little is known about the mechanisms of plant resistance to whiteflies (Berlinger, 1986). Of special relevance to this, the mechanisms of plant resistance to aphids have been extensively studied by using the EPG technique (Montllor & Tjallingii, 1989; Kimmins, 1989; Cole et al., 1993; Cole, 1994; Wilkinson

& Douglas, 1998; Sauge et al., 1998). By analysing 'non-sequential' (van Helden & Tjallingii, 1993) and 'sequential' parameters obtained from EPG experiments or combined with other analytical methods (Caillaud et al., 1995), EPG data can be used as an indicator for tissue location of plant resistance factors (Tjallingii, 1995). For example, EPG data from a study on lettuce suggest that resistance to the aphid *Nasonovia ribisnigri* (Mosley) is in the phloem of the resistant variety (van Helden & Tjallingii, 1993). Similarly, behavioural analysis of *Aphis gossypii* Glover feeding on melon showed that expression of the Vat resistance gene produces alterations of chemical compounds in the phloem of a resistant melon line (Chen et al., 1996). In contrast, EPG analysis of resistance in tomato and sweet pepper against greenhouse whiteflies suggests that resistance is based on factors present in the surface/epidermis and/or in the mesophyll layer of pepper plants (Lei et al., 1999).

The aim of this study was to characterise the mechanisms of the tomato resistance to *B. tabaci* previously observed in a number of *Mi*-carrying commercial varieties. Therefore, the probing and feeding behaviour of *B. tabaci* B-biotype was studied on the two near isogenic tomato lines Moneymaker (without *Mi*) and Motelle (carrying *Mi*) by using DC electrical penetration graphs. Consequently, the possible resistance factors at the successive levels of tissues in tomato plants were located. Moreover, EPG data were compared with the greenhouse choice assays in this paper.

Materials and methods

Whitefly colony and tomato plants. A Spanish population of *B. tabaci*, previously identified by Guirao et al. (1997) as B-biotype, was reared on *Lycopersicon esculentum* Mill. tomato var. Río Fuego at 27:23 °C (day:night) and at a photoperiod of L16:D8 h. Plants of the tomato lines Motelle (carrying *Mi* gene) and Moneymaker (without *Mi* gene) were grown in pots in an insect-proof chamber with artificial light at a photoperiod of L16:D8 at 26±1 °C and used at 3–4 true leaf stage as test plants. Moneymaker and Motelle are near-isogenic lines and differ only in the presence of a 650 kb introgressed region from *L. peruvianum* containing the *Mi* gene, in chromosome 6 of Motelle. Individual 2–3-day-old female whiteflies were kept at 4 °C for 5–10 s and then transferred to a stereo-microscope (Nikon SMZ-2T, Nikon Crop. Tokyo, Japan) and immobilised using a small vac-

uum pump (Vacuum pump A 3S, Tokyo Rikakikai Co. LTD, Japan). Subsequently, a 10 μ m gold wire was attached to the dorsum of whiteflies using a small silver paint droplet (solvent: isopropyl alcohol and methyl ethyl ethylketone, Pelco® Coloidal Silver, Ted Pella, Inc., Reeding, CA). After wire attachment, whiteflies were maintained inside a glass Petri dish for about 1-h acclimation and then connected to the amplifier for the test.

EPG recording. The feeding behaviour of *B. tabaci* was electrically monitored using a DC-system with a 10⁹ Ohm input resistance in a Faraday cage (Tjallingii, 1990). A Giga-4 DC-amplifier was connected to a microcomputer using an A/D card (sample rate of the A/D card = 100 Hz) (ACM-12-8, Strawberry Tree, Sunnyvale, CA), as well as a strip-chart recorder (Konik, KNK-801-201, Japan). The output signals were stored using the A=cm² acquisition software version 1.0 (Febvay, 1994). A wired individual whitefly was connected to the amplifier and placed on the abaxial surface of the fully expanded second leaf of a tomato test plant (leaf turned over and fixed by Parafilm so abaxial surface faced up). The EPG signal was monitored for 6 h per whitefly. A total of 25 whiteflies on Motelle and 26 on Moneymaker plants were recorded, each whitefly individual was recorded on a single tomato plant.

The analysis of EPG waveforms was carried out using the Macstylet software version 2.0 (Febvay et al., 1996). The following waveforms, partially correlated with behavioural events in greenhouse whiteflies (Janssen et al., 1989) and in *B. tabaci* (Jiang et al., 1999), were characterised: waveform C, inter-cellular stylet pathway; pd, short intracellular puncture during pathway; waveform G, xylem ingestion; and waveform F, mechanical probing difficulties. Also, waveform E(pd)₁, which was correlated with salivation into phloem sieve elements (Jiang et al., 2000), and E(pd)₂, being regarded as ingestion from phloem that is comparable to E2 of aphids (Prado & Tjallingii, 1994) were characterised. EPG parameters were carefully extracted based on previous work on aphids (Tjallingii, 1995; Caillaud et al., 1995), choosing only those parameters that can provide information on locating plant resistance factors in different tissues. Table 1 shows possible plant tissues involved as well as all parameters that were classified into two catalogues: non-phloem phase (including pathway phase and xylem phase) and phloem phase.

Statistical analysis. Comparison of mean EPG parameters from two isogenic tomato lines was performed according to the Mann-Whitney U test using Statview software (Abacus Concepts, 1987). In the case of percentages, the chi-square test was used at 0.05 and 0.001 levels.

Results

Parameters related to non-phloem phase (including pathway and xylem ingestion phases) (Table 2). A number of EPG parameters related to pathway phase were found to differ significantly between Money-maker (without *Mi*) and Motelle (carrying *Mi*). On Motelle, a lower percentage of whiteflies achieved phloem phase ($P < 0.01$) and they made more probes before attaining the first phloem phase ($P < 0.05$), had a higher ratio (number of probes before the 1st phloem phase)/(total number of probes) ($P < 0.01$), had a longer total duration of non-probing time ($P < 0.05$), and a longer time before making the 1st intracellular puncture ($P < 0.05$) and the 1st phloem phase ($P < 0.01$). However, parameters more directly related with surface factors (parameters 1, 2 and 3) as well as those related to waveforms G (xylem ingestion) and F (mechanical difficulty) did not differ significantly between the two tomato lines (Table 2).

Parameters related to phloem phase (Table 3). Except for the time that whiteflies took from start of the 6h experiment to the 1st sustained phloem ingestion ($P < 0.01$) and the percentage of whiteflies showing sustained ingestion ($P < 0.05$), most of the parameters related to phloem phase were found not to differ significantly between these two near-isogenic tomato lines. Moreover, the total time that whiteflies spent ingesting phloem sap was almost the same in Motelle as that in Money-maker. Similarly, the numbers of E(pd)₁ and E(pd)₂ were almost the same in both tomato line (parameters 20, 23, Table 3.).

Discussion and conclusions

Comparison of EPG profile with greenhouse preference study. Our previous preference study with the B-biotype of *B. tabaci* showed that significantly fewer Motelle plants than Money-maker plants were infested by females in a greenhouse choice test. Also, the daily

Table 1. EPG parameters and possible relations to resistance factors in specific plant tissue

EPG parameters	Tissue(s)/factors involved
Non-phloem parameters	
1. Time to the first probe	surface factors
2. Duration of 1st probe	surface factors + epidermis/mesophyll
3. Total number of probes	surface factors + all plant tissues
4. Number of probes made before attaining the first phloem phase (1st E(pd))	epidermis, mesophyll and other parenchyma
5. Ratio of probes before 1st E(pd)/total number of probes	epidermis, mesophyll and other parenchyma
6. Total duration of non-probing time	all plant tissues except phloem
7. Total duration of F	all plant tissues except phloem
8. Total duration of G	xylem factors
9. Time whitefly took to make its first intracellular puncture (1st pd) from the start of the 6h experiment (min)	all plant tissues except phloem
10. Time whitefly took to the 1st E(pd) from the start	all plant tissues except phloem
11. Time whitefly took to 1st E(pd) within probe	all plant tissues except phloem
12. % of whitefly reaching the phloem phase	epidermis, mesophyll and other parenchyma
Phloem parameters	
13. Duration between time to the 1st E(pd) and time to the first sustained ingestion (1st SI) (E(pd) ₂ > 15 min)	vascular tissue and sieve elements (initial factors)
14. Time whitefly took to 1st SI from start	all plant tissues
15. % of time whitefly spent in E(pd) ₂ after the 1st phloem phase	phloem factors
16. Duration of E(pd) ₁ preceding the first E(pd) ₂ > 15 min	phloem factors
17. Duration of E(pd) ₂ after the 1st SI	phloem factors
18. Total duration of E(pd) ₁	phloem factors
19. Mean duration of E(pd) ₁	phloem factors
20. No. of E(pd) ₁	phloem factors
21. Total duration of E(pd) ₂	phloem factors
22. Mean duration of E(pd) ₂	phloem factors
23. No. of E(pd) ₂	phloem factors
24. % of whiteflies showing sustained ingestion (E(pd) ₂ > 15 min).	all plant tissues

Table 2. Comparison of EPG parameters of B-biotype of *B. tabaci* on two isogenic tomato lines during 6-h recordings

EPG parameters	Moneymaker Mean±SE (n)	Motelle Mean±SE (n)
1. Time to the first probe (1st probe) (s)	39.09 ± 14.94 (25)	61.68 ± 25.76 (26) Ns
2. Duration of 1st probe (s)	89.48 ± 24.96 (25)	99.78 ± 31.11 (26) Ns
3. Total number of probes	44.36 ± 5.05 (25)	57.23 ± 8.38 (26) Ns
4. Number of probes made before attaining the first phloem phase (1st E(pd))	27.08 ± 3.65 (25)	53.00 ± 8.87 (26)*
5. Ratio of probe before 1st E(pd)/total number of probes	0.65 ± 0.06 (25)	0.86 ± 0.05 (26)**
6. Total duration of non-probe time (min)	65.81 ± 7.95 (25)	100.39 ± 11.50 (26)*
7. Total duration of F (min)	21.12 ± 4.00 (12)	32.54 ± 6.65 (19) Ns
8. Total duration of G (min)	20.31 ± 3.25 (9)	42.88 ± 15.62 (14) Ns
9. Time whitefly took to make its first intracellular puncture (1st pd) from the start of the 6h experiment (min)	94.31 ± 12.44 (24)	154.38 ± 20.32 (25)*
10. Time whitefly took to the 1st E(pd) from the start (min)	163.25 ± 20.01 (25)	272.16 ± 22.82 (26)**
11. Time whitefly took to 1st E(pd) within probe (min)	21.19 ± 2.28 (22)	38.97 ± 7.15 (13) Ns
12. % of whitefly reaching the phloem phase	88.00	50.00 **

Significant differences were compared at 0.05 and 0.01 level according to Mann–Whitney U test (Stateview, Abacus Concepts, 1987). In the case of Percentage or ratio, a chi-square test was used at 0.05 and 0.01 levels. Ns, no significant differences ($P > 0.05$), * $P < 0.05$ and ** $P < 0.01$.

Table 3. Comparison of EPG parameters of B-biotype of *B. tabaci* on two isogenic tomato lines during 6-h recordings

EPG parameters	Moneymaker Mean $\pm$ SE (n)	Motelle Mean $\pm$ SE (n)
13. Duration between time to the 1st E(pd) and time to the first sustained ingestion (1st SI) (E(pd) ₂ >15 min) (min)	35.99 $\pm$ 17.79 (19)	11.34 $\pm$ 9.72 (11) Ns
14. Time whitefly took to 1st SI from the start of the 6h experiment (min)	188.62 $\pm$ 21.12 (25)	277.79 $\pm$ 21.45 (26)**
15. % of time whitefly spent in E(pd) ₂ after the 1st phloem phase	51.96 $\pm$ 28.31 (21)	62.4 $\pm$ 35.35 (13) Ns
16. Duration of E(pd) ₁ before E(pd) ₂ in first or in any E(pd) ₂ > 15 min (min)	3.17 $\pm$ 0.72 (22)	1.63 $\pm$ 0.43 (13) Ns
17. Duration of E(pd) ₂ after the 1st SI (min)	29.35 $\pm$ 9.18 (22)	13.39 $\pm$ 7.73 (13) Ns
18. Total duration of E(pd) ₁ (min)	5.85 $\pm$ 1.09 (22)	3.05 $\pm$ 0.87 (13) Ns
19. Mean duration of E(pd) ₁ (min)	2.73 $\pm$ 0.48 (22)	1.67 $\pm$ 0.44 (13) Ns
20. No. of E(pd) ₁	2.09 $\pm$ 0.24 (22)	2.08 $\pm$ 0.50 (13) Ns
21. Total duration of E(pd) ₂ (min)	106.44 $\pm$ 17.55 (21)	105.10 $\pm$ 24.00 (13) Ns
22. Mean duration of E(pd) ₂ (min)	64.88 $\pm$ 14.63 (21)	84.41 $\pm$ 25.14 (13) Ns
23. No. of E(pd) ₂	1.90 $\pm$ 0.25 (21)	1.69 $\pm$ 0.33 (13) Ns
24. % of whiteflies showing sustained ingestion (E(pd)>15 min)	76.00 (19/25)	42.30 (11/26)*

Significant differences were compared at 0.05 and 0.01 levels according to Mann–Whitney U test (Stateview, Abacus Concepts, 1987). In the case of percentage, a chi-square test was used at 0.05 level. Ns, no significant differences ($P > 0.05$), * $P < 0.05$ and ** $P < 0.01$.

number of females recorded on Motelle was significant less than on Moneymaker (Nombela et al., 2000). On the same two isogenic tomato lines, the EPGs now revealed that a significantly lower percentage of female whiteflies reached phloem on Motelle (50%) compared to Moneymaker (88%) within 6 h (Table 2). Also, a lower number of whiteflies in Motelle (11 out of 26) were able to show sustained ingestion from phloem than on Moneymaker (19 out of 25). This indicates that Motelle plants could drastically reduce the infestation rate and decrease the number of settling adult whiteflies, in agreement with the preference study, and further supports the fact that tested tomato lines carrying the *Mi* gene, display resistance to the B-biotype of *B. tabaci*.

Possible location of resistant factors at tissue levels.

The probing and feeding behaviour performance of whiteflies on the two isogenic tomato cultivars provide invaluable clues for understanding the mechanisms of plant resistance at tissue levels. As mentioned earlier, the whole process of whitefly probing starts with labial contact with the plant surface, followed by insertion of its stylets through successive tissue layers between the epidermis and phloem and, eventually targets on the sieve element of phloem. Therefore, plant tissue factors can play various roles in the initiation, maintenance, and cessation of each subsequent event in the probing process, especially the first phloem phase contact by whiteflies (Lei et al., 1999).

At the level of the surface/epidermis and/or mesophyll layers. On Motelle, whiteflies showed more probes before attaining the first phloem phase (parameter 4) and time whitefly took to the first phloem phase from the start (parameter 10) when compared with Moneymaker. The point that all these activities occurred before the first phloem contact, strongly suggests that the resistance factors are present on the surface/epidermis and/or mesophyll tissue layers in Motelle plants. Additionally, parameter 6 (total duration of non-probing time) also supports this point, i.e., whiteflies spent more time on non-probing activities (walking or resting), on Motelle than on Moneymaker, clearly indicating, again, a negative effect of the first encountered tissues to whitefly feeding on Motelle plants. Besides, detailed comparison of the percentage of short probes (less than 3 min) before the first phloem phase on Motelle (95%), compared to that on Moneymaker (85%) (not shown in the Tables), suggests that the plant epidermis might play an important role in Motelle resistance to whiteflies. Our close-up video observations, combined with stylet cutting, proved that whiteflies do not penetrate deeper than the epidermis in such a short time (Y. X. Jiang, unpublished). However, parameters more directly related with surface factors (parameters 1, 2 and 3) did not differ significantly between the two tomato lines (Table 2), which might indicate that these surface factors do not play a major role in the resistance, although the surface and epidermis factors cannot be separated completely based on EPG recordings. In addition, the role of resistance factors in mesophyll and non-vascular

tissues cannot be ignored as indicated by parameter 10: the time whiteflies took to reach the first phloem phase within a probe in Motelle (38.97 m) was much longer than that in Moneymaker (21.19 m).

At the level of phloem tissue. In an EPG study, Gabrys et al. (1997) reported that less preferred plant parts (*Sinapis alba*) had deterrent effects at epidermal/mesophyll levels to *Brevicoryne brassicae*. Apart from that, repetitive failures to start ingestion, and reduced phloem sap ingestion were often typical features associated with resistance as reported in EPG studies performed on aphids/plant combinations (Campbell et al., 1982; van Helden & Tjallingii, 1993; Cole, 1994; Caillaud et al., 1995; Sauge et al., 1998; Kaloshian et al., 2000). However, the feeding and probing profile of whiteflies from our data on the two isogenic tomato lines revealed a drastically different picture than these aphids, i.e., most of the EPG parameters related to phloem phase did not differ significantly. Specifically, whiteflies spent a similar amount of time ingesting from phloem (duration of E(pd)₂, parameter 21, Table 3) in Motelle and Moneymaker, which strongly indicates that the acceptability of phloem sap is rather similar. Also, no significant differences were found in the duration between time to the first phloem phase and time to the first sustained ingestion (parameter 13), percentage of the time whiteflies spent on E(pd)₂ after the 1st phloem phase (parameter 15) and duration of E(pd)₂ after the first sustained ingestion (parameter 17) between Motelle and Moneymaker, again indicating that there are no differences in factors in phloem sap between the two isogenic lines that affected whitefly feeding behaviour on the short term, or long term. Evidence from qualitative analysis of honeydew of potato aphids feeding on tomatoes showed that some aphids ingest fluids from resistant tomato plants (with *Mi* gene) although significant reductions in honeydew production occurred in resistant plants (Kaloshian et al., 1997). A possible inference is that the aphids also encountered difficulties going through successive tissue layers (surface, epidermis and mesophyll) and had a low success rate reaching the phloem in the resistant variety, and thus, the delayed phloem ingestion caused less honeydew on *Mi* plants. However, whiteflies spent similar phloem sap ingestion times in resistant plants as in susceptible plants, once their stylets reached the phloem.

Interestingly, two exceptional parameters associated with phloem phase were found to differ sub-

stantially on the two isogenic tomato lines: time that whiteflies took from start to the first sustained phloem ingestion (parameter 14) and percentage of whiteflies showing sustained ingestion (parameter 24). A closer look at these two parameters revealed that all non-phloem phase factors rather than only phloem factors are actually involved. Indeed, the whitefly's stylets should go through the epidermis and mesophyll tissues before it eventually reaches the phloem and the whitefly shows sustained ingestion. Based on these data, although the role of initial resistance factors in phloem cannot be completely excluded, they probably do not play a major role in Motelle resistance to whiteflies.

Possible mechanisms of this tomato resistance. Resistance to nematodes *Meloidogyne* spp., conferred by *Mi* gene to tomato plants, is characterised by a localised region of necrotic plant cells (hypersensitive response), around the head of the invading nematode (Dropkin, 1969; Ho et al., 1992). The mechanisms of *Mi* resistance to potato aphid, *M. euphorbiae*, which drastically reduce longevity and fecundity, and alter feeding behaviour, are still unknown (Rossi et al., 1998). Our previous assays (Nombela et al., 2000) suggested the existence of a partial resistance against *Bemisia* in *Mi*-carrying tomato varieties when compared to varieties lacking *Mi*, although it is not demonstrated so far whether such a resistance is regulated by *Mi* or by another gene closely linked to it. The present EPG data confirm this resistance and demonstrate that resistance factors in Motelle are detected by *B. tabaci* early during the stylet penetration pathway, indicating that these factors might be present in the plant epidermis and/or mesophyll layers. Although aphids, whiteflies, and nematodes belong to different taxa, they do have some common feeding features, i.e. they penetrate their hosts intercellularly and eventually their stylets terminate at the phloem. Thus, it is likely that these organisms share the same recognition mechanisms in tomato, probably regulated by gene-for-gene interactions. A hypothesis for these common resistance mechanisms is that the recognition and initiation of a signal transduction cascade leading to resistance is triggered when aphids, whiteflies, or nematodes start penetrating into the tomato cells located on surface/epidermis and/or mesophyll tissues. This hypothesis needs to be proved by more experiments in the future. Therefore, more detailed studies including histochemical and molecular methods are necessary to provide a better understanding of these mechanisms of resistance in commercial tomato varieties.

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