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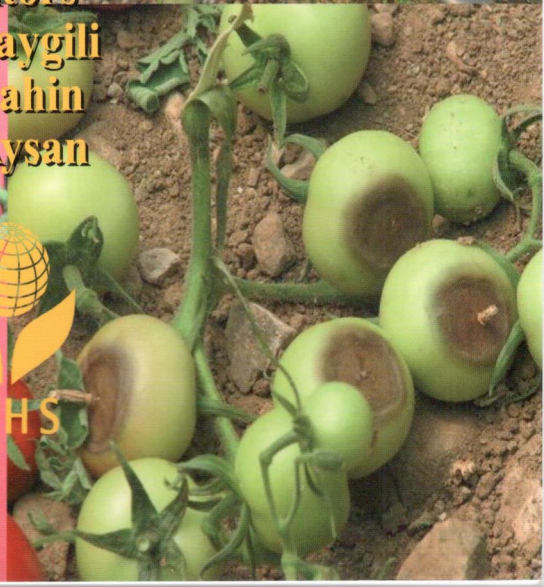
*Phytophthora infestans*



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# Research on Tomato Resistance to the Virus-Transmitter Whitefly *Bemisia tabaci* Undertaken during the Last Years in Madrid (Spain)

M. Muñoz and G. Nombela<sup>§</sup>

Department of Plant Protection

Instituto de Ciencias Agrarias - Centro de Ciencias Medioambientales

Consejo Superior de Investigaciones Científicas (CSIC)

Serrano 115 Dpdo. Madrid-28006

Spain

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## Abstract

Whitefly *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae) is an important pest of horticultural crops worldwide, including tomato plants, whose damage may be directly through phloem feeding or indirectly by the transmission of plant viruses such as Tomato Yellow Leaf Curl Viruses. Although control of *B. tabaci* is based mainly on the application of insecticides, most of these chemicals are environmentally toxic and whiteflies appear to be resistant to many of them. Among the most outstanding alternative tools to insecticides, the use of plant resistance, either innate or induced, is outlined as one of the strategies of efficient and environmentally respectful control.

In different studies carried out by our group in Madrid during the last years (with collaboration of other researchers from Spain and USA), we obtained that the tomato gene *Mi-1.2* is responsible for innate resistance to both B- and Q- biotypes of *B. tabaci*, and the *Rme1* locus is also required for this resistance. Moreover, BTH or a previous infestation by the potato aphid *Macrosiphum euphorbiae* induces resistance to *Bemisia tabaci* in susceptible tomato plants.

## INTRODUCTION

Tomato *Lycopersicon esculentum* L. (Solanaceae) cropping is widely distributed, being one of the economically most important in Mediterranean areas. One of the limiting factors affecting the success of this crop is the existence of insect pests. Among them, whitefly *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae) is one of the most outstanding problems. Damage caused by this insect to commercial tomato may be directly through phloem feeding or indirectly by the transmission of plant viruses such as Tomato Yellow Leaf Curl Viruses (TYLCV's). The complex of geminiviruses called TYLCV, transmitted in a persistent and circulative manner by *B. tabaci*, are some of the most devastating viruses of cultivated tomato, causing economic losses of up to 100% in many tropical and subtropical regions. Severe outbreaks of these viruses have occurred recently in Southern Europe and USA and appear linked with the progressive increase of *B. tabaci* populations worldwide. So, efficient management of the virus diseases requires also the management of their vector insects. Often, control of *B. tabaci* is mostly based on the application of insecticides, but whiteflies appear to be resistant to many of the chemicals employed and most of these products are environmentally toxic, being recommended the employment of any measure different from the conventional chemical treatments. Among these measures, the use of plant resistance is outlined as one of the strategies for an efficient and environmentally respectful pest control. There are two kinds of plant resistance: 1) natural, innate or congenital, and 2) induced or acquired. Regarding the first case, there are plant species or even plant varieties with a high resistance level selected along an evolution process. Some tomato varieties carry one of the best-characterized resistance genes so far: the *Mi-1* gene, which is responsible for the resistance to certain species of root-knot nematodes, aphids and whiteflies. However, induced resistance is acquired in plants after being attacked, in such a way that in many

<sup>§</sup> gnombela @ ccma.csic.es

occasions if a plant survives an attack, it defends itself much better from a later attack of the same organism, and even from other biological agents. Acquired or induced resistance can be expressed locally, at the site of primary inoculation (LAR), but also systemically, in tissues far away from the initial treatment (SAR).

Consequently with the importance of the efficiency in the use of resistant varieties, the interest in the study of host plant resistance, based on detailed insect-plant interaction studies, has recently increased. In the present work we summarize the main results on the topic of tomato resistance (innate and induced) to *B. tabaci*, obtained by our research group in Madrid during the last years, with collaboration of other researchers from Spain and USA.

## MATERIALS AND METHODS

Adult whiteflies (*B. tabaci*) were used in these studies, from two populations of the B- and Q-biotype, respectively, which had been reared for more than 50 generations in our laboratory on tomato cv. Marmande. In addition, one aphid clone (*Macrosiphum euphorbiae*), established from a single *Virginoparous aptera* female and reared on tomato plants cv. Marmande, was used for the last study.

Tomato varieties bearing the *Mi-1* gene (Motelle, Ronita and VFN8), other varieties lacking this gene (Moneymaker, Roma and Río Fuego), a transgenic line proceeding from Moneymaker and transformed with a 14.7-kb tomato genomic fragment carrying the gene *Mi-1.2*, as well as a defective mutant (*rme1*) in the background of the wild-type parent Motelle, were compared in the studies on the *Mi-1*-mediated resistance. For the studies on induced resistance, only susceptible tomato plants (cv. Marmande) were used.

Free-choice and no-choice assays were carried out under greenhouse and controlled conditions, respectively. Numerical data obtained from each assay were  $\log_{10}(x+1)$  transformed and analyzed by a one-way ANOVA. Means from different genotypes or treatments were compared by the Student's *t* or the Tukey HSD tests.

## RESULTS AND CONCLUSIONS

### The Root-Knot Nematode Resistance Gene *Mi-1.2* of Tomato is Responsible for Resistance against the Whitefly *Bemisia tabaci*

In free-choice assays, a shorter number of *Bemisia tabaci* (Q-biotype) adults were observed on tomato transgenic plants with the *Mi* gene than were on susceptible (cv. Moneymaker) plants (Fig. 1). Mean values of infested plants were significantly lower on these transgenic plants than on susceptible plants (Fig. 1). The mean number of pupae per plant or per leaf was higher on Moneymaker (Fig. 1). At the end of no-choice assays, significantly lower number of individuals of the next generation (L3+L4+adults or empty pupal cases) was recorded in the lacking-*Mi* transgenic plants (Fig. 2). Similar results were obtained for the B-biotype.

From these and other previous results comparing bearing- and lacking-*Mi-1* tomato varieties (Nombela et al., 2000, 2001) we concluded that the tomato gene *Mi-1.2* (the only gene of resistance to insects cloned up to date) is responsible for resistance to both B- and Q- biotypes of *B. tabaci* (Nombela et al., 2003).

### *Rme1* is Necessary for *Mi-1*-Mediated Resistance and Acts Early in the Resistance Pathway

Significantly shorter percentages of adults were daily observed on the *rme1* mutant plants ( $-Mi$ ) than on susceptible Moneymaker ( $-Mi$ ) or resistant Motelle ( $+Mi$ ) (Fig. 3). Differences between numbers of adults on *rme1* mutant and Motelle plants were also significant (Fig. 3). The mean number of eggs per plant on Moneymaker and plants carrying the *Rme1* gene were significantly higher than were on Motelle plants (Fig. 3).

In conclusion, the *Rme1* locus is also required for *Mi-1.2*-mediated resistance to *Bemisia tabaci* (Martinez De Iladurya et al., 2004)

## **Benzothiadiazole Induces Local Resistance to *Bemisia tabaci* (Hemiptera: Aleyrodidae) in Tomato Plants**

Benzo [1,2,3] thiadiazole-7-carbothioic acid-S-methyl ester (BTH) or acibenzolar-S-methyl (ASM) is the active ingredient of the Syngenta plant activator Bion<sup>®</sup>. Adult Q-biotype *B. tabaci* significantly preferred control plants (cv. Marmande) than those plants sprayed with Bion<sup>®</sup> at 0.2 g/l (Fig. 4). Consequently, a decrease in the number of empty pupal cases was also observed 23 days later in plants treated with Bion<sup>®</sup> at 0.2 g/l. In no-choice assays, the total numbers of immature individuals (eggs+L1+L2) on Bion<sup>®</sup>-treated leaves after 16 days was significantly lower than those on water-treated leaves from control plants (Fig. 4). This difference was mostly due to the number of L1 larvae. The acquired resistance was induced locally (LAR) because of the differences between Bion<sup>®</sup>-treated and non-treated leaves in the same plants, meanwhile no statistically significant differences were observed in no-treated leaves between Bion<sup>®</sup>-treated and control plants (Fig. 4).

So, BTH induces resistance to *Bemisia tabaci* in susceptible tomato plants (Nombela et al., 2005)

## **Resistance to *Bemisia tabaci* is Induced in Tomato Plants after Aphid Infestation**

*B. tabaci* (B biotype) numbers were significantly reduced when aphid attack (*Macrosiphum euphorbiae*) occurred between 1 and 18 hours prior to *B. tabaci* infestation, meanwhile reduction was not detectable when 4 days passed between aphid and whitefly infestations (Fig. 5). The resistance was both local (LAR) and systemically (SAR) induced because no differences were detected between aphid-infested leaves and the uninfested contiguous leaves in aphid-infested plants (Fig. 6).

From these results, the potato aphid *M. euphorbiae* induces plant resistance to *B. tabaci* in cv. Marmande tomato plants (Nombela et al., 2004).

## **ACKNOWLEDGEMENTS**

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**Figures**

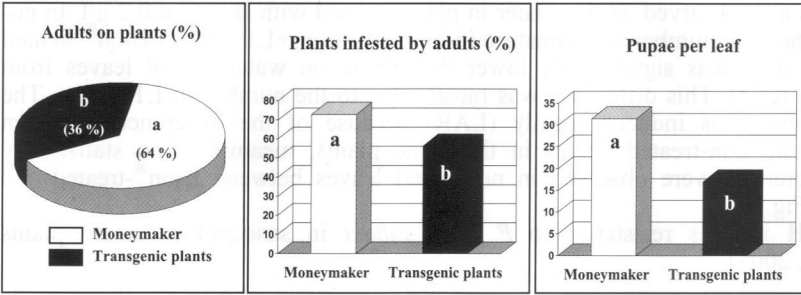


Fig. 1. Percentages of *B. tabaci* adults (Q biotype) on Moneymaker (- *Mi-1.2*) and transgenic (+ *Mi-1.2*) tomato plants, percentages of plants infested by adults during the free-choice assay, and mean numbers of pupae per leaf at the end of the free-choice assay.

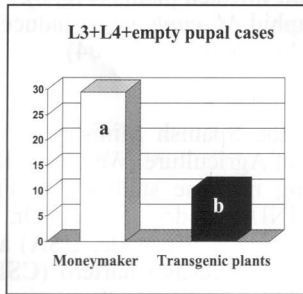


Fig. 2. Mean numbers of individuals of the next generation (L3+L4+adults or empty pupal cases) of *B. tabaci* (Q biotype) on Moneymaker (- *Mi-1.2*) and transgenic (+ *Mi-1.2*) tomato plants at the end of the no-choice assay.

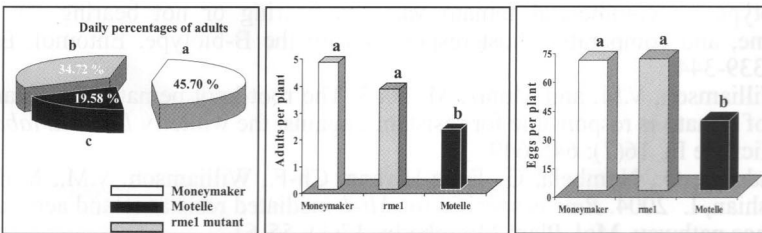


Fig. 3. Percentages of *B. tabaci* adults (B biotype) and mean numbers of adults per plant on Moneymaker (- *Mi-1.2*), Motelle (+ *Mi-1.2*) and rme1 mutant (+ *Mi-1.2*) plants during the free-choice assay, and mean numbers of eggs per plant in the no-choice assay.

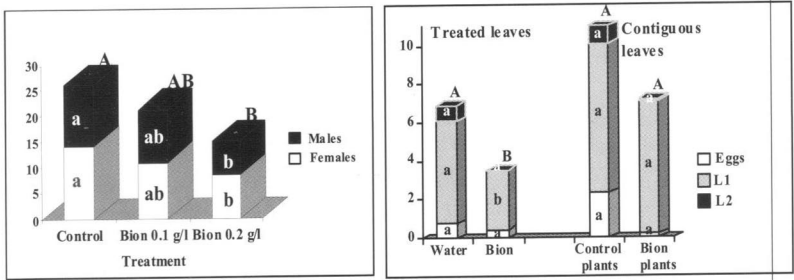


Fig. 4. Mean numbers of adults on Bion-treated (2 different doses) and water-treated (control) plants during a free-choice assay, and mean numbers of eggs, L1 and L2 on the treated and un-treated contiguous leaves of control and Bion-treated plants after 16 days of another no-choice assay.

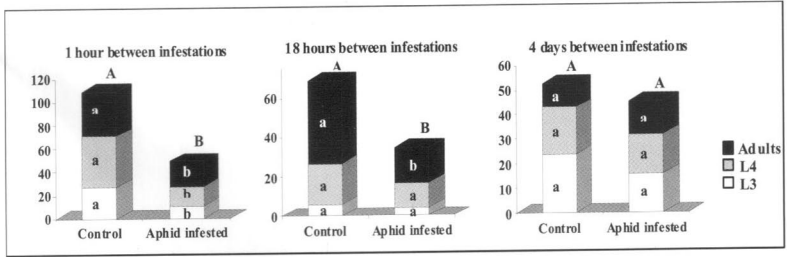


Fig. 5. Mean numbers of L3, L4 and new adults of *B. tabaci* (B biotype) on plants previously infested by aphids and un-infested control plants, at different time intervals after aphid infestation.

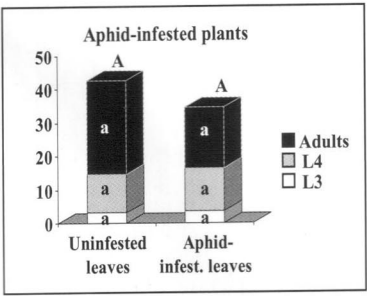


Fig. 6. Mean numbers of L3, L4 and new adults of *B. tabaci* (B biotype) on aphid-infested leaves and un-infested contiguous leaves of the aphid-infested plants.