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Source: Florida Entomologist, 100(2) : 481-484

Published By: Florida Entomological Society

URL: https://doi.org/10.1653/024.100.0213

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Bemisia tabaci MED (Q biotype) (Hemiptera: Aleyrodidae) in Florida is on the move to residential landscapes and may impact open-field agriculture

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Q biotype whiteflies, more properly known as *Bemisia tabaci* Mediterranean (MED) and classified as *B. tabaci* (Gennadius) (Hemiptera: Aleyrodiae) (Dennehy et al. 2005; McKenzie et al. 2009, 2012), have been in the United States for approximately a dozen years. First found on poinsettia during fall 2004 in a retail outlet in Arizona (Dennehy et al. 2005), *B. tabaci* MED has since been identified from greenhouse-grown ornamental horticulture plants in 26 states (McKenzie et al. 2012), including Florida (McKenzie et al. 2009). Indistinguishable morphologically from silverleaf whitefly (*B. tabaci* Middle Eastern Asia Minor 1 [MEAM1]), *B. tabaci* MED is extremely problematic to agricultural production because it has a high propensity to develop resistance to insect growth regulators (Horowitz et al. 2003) and neonicotinoid insecticides (Elbert & Nauen 2000; Horowitz et al. 2004). Both of these insecticide classes play crucial roles in controlling whiteflies in many different cropping systems including cotton (Ellsworth & Martinez-Carrillo 2001), vegetables (Palumbo et al. 2001), and ornamentals (McKenzie et al. 2014). Associated with the appearance of *B. tabaci* MED in the United States were reports of increasing problems in controlling whitefly infestations, primarily from ornamental growers. Population studies indicate *B. tabaci* MED was introduced into the United States on at least 3 separate occasions, and both eastern and western *B. tabaci* MED are found throughout the continental United States including Florida (Dickey et al. 2013). Previously, *B. tabaci* MED had been detected in North America only on ornamental plants and herbs in greenhouses and nurseries. This, however, changed in the spring of 2016, and the first detections of *B. tabaci* MED in residential landscapes and open field agricultural production are reported here.

Adults or immature stages of whiteflies collected were immediately placed in 95% ethanol for molecular analysis. If available, at least 12 whiteflies from each sample were used for species determination following the protocol developed by Shatters et al. (2009). DNA was extracted from individual whiteflies by placing a single whitefly in a 1.5 mL microcentrifuge tube, adding 50 µL DNA lysis buffer, and grinding with a pestle. The pestle was rinsed with an additional 50 µL DNA lysis buffer that was collected in the same tube. Tubes were placed in a metal boiling rack and boiled at 95 °C for 5 min and then placed in crushed ice for 5 min. Tubes were then centrifuged at 8,000 *g* for 30 s, and the supernatant (crude DNA lysate) was transferred to a new tube and stored at −80 °C for future processing.

We used species (biotype) specific polymerase chain reaction (PCR) primers that had been designed by Shatters et al. (2009) to recognize unique *mtCOI* gene regions within the MEAM1 (B biotype), NW biotype, and MED (Q biotype) species; these primers produce different-sized products depending on the source of the isolated template DNA and do not require DNA sequencing. The chosen *mtCOI* primer pairs amplified fragments of 303, 405, and 478 bp from DNA of the MED, NEW WORLD, and MEAM1 species, respectively. The 30 µL final volume PCRs were run using a PTC-0200 DNA Engine® Peltier thermal cycler (MJ Research, Inc., Waltham, Massachusetts) under the conditions described by Shatters et al. (2009).

PCR amplifications for the *mtCOI* gene were also performed using the Btab-Uni primer set described by Shatters et al. (2009) for whiteflies determined to be *B. tabaci* NW biotype in any environment or *B. tabaci* MED in unique environments (residential landscapes and open field agriculture) by the species specific primer cocktail. An *mtCOI* sequence analysis was performed first by PCR amplifying an approximately 700 to 800 bp *mtCOI* DNA fragment and then sequencing the PCR amplified DNA. The 30 µL PCRs were run using a T100™ Thermal Cycler (BIO-RAD Laboratories, Inc., Hercules, California) under the conditions described by Shatters et al. (2009). Prior to sequencing, the amplified products were cleaned using Montage® PCR cleanup filters (Millipore, Billerica, Massachusetts). Fifty ng of total whitefly genomic DNA were used in BigDye® (Applied Biosystems, Foster City, California) sequencing reactions. All sequencing was performed bidirectionally with the amplification primers and BigDye®Terminator v3.1 Cycle Sequencing Kits (Applied Biosystems, Foster City, California). Sequence reactions were analyzed on an Applied Biosystems® 3730XL DNA Analyzer (Applied Biosystems, Foster City, California) and were then compared and edited using Sequencher software (Gene Codes, Ann Arbor, Michigan). Biotype determination was based on direct sequence comparisons using the web based National Center for Biotechnology Information BLAST sequence comparison application (http://blast.ncbi.nlm.nih.gov/Blast.cgi), and sequences were deposited in GenBank.

Since the previous *B. tabaci* distribution surveys were done in Florida statewide (McKenzie et al. 2009) and in North America (McKenzie et al. 2012), *B. tabaci* MED was detected in Florida in 2011 (7 detections), 2013 (2), and 2014 (3) primarily on poinsettia (11) with a single detection on hibiscus. All *B. tabaci* MED detections were from nursery or greenhouse environments (McKenzie unpublished data). In 2016, landscapers and pest control operators in Palm Beach County, Florida, began experiencing problems controlling extremely high populations of *Bemisia* whiteflies on hibiscus plantings in multiple

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Table 1. Sample date, location by city and zip code, host plant, electrophoresis and sequencing results, and National Center for Biotechnology Information (NCBI) accession number for residential landscape and open field detections of *Bemisia tabaci* Mediterranean (MED).

Sample date	Location by city and zip code	Host plant	Electrophoresis results MEAM1:MED[a]	Uni-tab MED sequence results	NCBI accession number
25 Apr 2016	North Palm Beach, FL 33408	Hibiscus	0:44	Eastern MED	KY073617
4 May 2016	North Palm Beach, FL 33408	Hibiscus	0:4	Eastern MED	KY073618
4 May 2016	North Palm Beach, FL 33408	Lantana	0:20	Eastern MED	KY073619
4 May 2016	North Palm Beach, FL 33408	Ficus / Hibiscus	0:12	Eastern MED	KY073620
12 May 2016	Palm Beach, FL 33480	Hibiscus	0:20	Eastern MED	KY073621
13 May 2016	Boca Raton, FL 33496	Hibiscus	0:20	Eastern MED	KY073622
20 May 2016	Boynton Beach, FL 33437	Hibiscus	0:20	Eastern MED	KY073623
24 May 2016	Palm Beach, FL 33480	Hibiscus	0:1	Eastern MED	KY073624
7 Jun 2016	Boca Raton, FL 33496	Firecracker flower	0:14	Eastern MED	KY073625
25 Jul 2016	Tequesta, FL 33469	Porterweed	0:18	Eastern MED	KY073626
25 Jul 2016	Tequesta, FL 33469	Hibiscus	0:18	Eastern MED	KY073627
	Field Detections				
17 Aug 2016	Elkton, FL 32033	Sweet Potato	15:1	Eastern MED	KY073628
23 Aug 2016	Elkton, FL 32033	Sweet Potato	138:0	no MED data	Not applicable
25 Aug 2016	Boynton Beach, FL 33472	Morning glory on fallow vegetable field border	1:16	Eastern MED	KY073629
16 Sept 2016	Boynton Beach, FL 33472	Morning glory on fallow vegetable field border	2:3	Eastern MED	KY131960

[a]MEAM1 = Middle Eastern Asia Minor 1.

residential neighborhoods. In total, 10 residential landscapes were identified and subsequently determined to be infested by eastern *B. tabaci* MED (Table 1). *Bemisia tabaci* MED detected at 1 location was traced back to a wholesale nursery, and the infestation was the direct result of a new planting of hibiscus. Before it was contained, this infestation spread to lantana and a ficus hedge located on 2 neighboring properties. *Bemisia tabaci* MED was also detected on single family residences with established hibiscus (not new plantings), firecracker flower or mixed ficus hedges that did not have any apparent connection and were kilometers apart from one another. The original source of these infestations remains unknown. *Bemisia tabaci* MED was detected in Florida as far north as Tequesta, as far east as Palm Beach Island, as far south as Boca Raton, and as far west as Boynton Beach with a few detections in the center of Palm Beach County. In all of these residential landscape collections, *B. tabaci* MED comprised the entire sample. Three residential landscape collections resulted in 100% *B. tabaci* MEAM1 (data not shown). All residential detections were in very affluent well-manicured landscapes.

Florida vegetable fields have been heavily sampled to determine *B. tabaci* species composition for well over a decade and especially in tomato (McKenzie et al. 2004, 2009, 2012; Schuster et al. 2010; Caballero et al. 2014; Smith et al. 2016) with only *B. tabaci* MEAM1 being detected. In 2016, we processed 551 whiteflies from 28 vegetable field samples including tomato (9), cucumber (2), potato (1), watermelon (6), eggplant (3), and sweet potato (7); all were *B. tabaci* MEAM1. In Aug 2016, *B. tabaci* MED was detected in a field of sweet potato in Elkton, St. John County (Table 1), at a very low ratio (15:1 MEAM1: MED). This collection was totally comprised of immature nymphs removed from a sweet potato leaf from an isolated open field planted 90 d prior indicating that, because whitefly immatures are not mobile after the 1st instar, *B. tabaci* MED had oviposited on sweet potato in the field. However, approximately 1 wk after the initial collection, additional whitefly samples were taken from the same field, and only *B. tabaci* MEAM1 was detected (138:0 MEAM1: MED). *Bemisia tabaci* MED was also detected (1:16 MEAM1: MED) on morning glory weeds growing on the border of a fallow vegetable field ready to go into production in Palm Beach County and was confirmed 3 wk later with another sample (2:3 MEAM1: MED). This field was 800 m west of a wholesale nursery known to have *B. tabaci* MED.

In addition to the residential and open field detections, *B. tabaci* MED has been detected in 8 wholesale nurseries from 4 counties and 17 retail nurseries from 8 counties in Florida (Table 2). *Bemisia tabaci* MED was detected on multiple sampling dates from several of the nurseries. Hibiscus was the driving host plant in nursery (wholesale and retail) infestations and accounted for 78% of *B. tabaci* MED detections followed by firecracker flower and eggplant each with 9% and a single lantana detection. Sixty-five percent of the *B. tabaci* MED positive samples (21) were 100% *B. tabaci* MED and 35% (11) were mixed populations of *B. tabaci* MEAM1 and MED. One hundred samples from retail and wholesale nurseries were 100% *B. tabaci* MEAM1 and 1 sample from a retail nursery sampled from mint was 5:1 MEAM1:NW (National Center for Biotechnology Information accession number: KY131961) and represents the first detection of *B. tabaci* NW in Florida since it was displaced by *B. tabaci* MEAM1 in the mid-1980s (McKenzie et al. 2004, 2009, 2012).

We do not know why *B. tabaci* MED (Q biotype) emerged as a pest in Florida landscapes in 2016. *Bemisia tabaci* is not usually a problem in the landscape due to the multiple host plants and the presence of a complex of natural enemies that keep whiteflies in check. In addition, commercial producers of ornamental plants have successfully managed *B. tabaci* MED since its first detection in Florida years ago.

Several factors may explain the detection of *B. tabaci* MED in Florida landscapes in 2016. First, environmental conditions may have been favorable for the buildup of *B. tabaci* in the landscape. It is also possible that growers have gotten complacent and less vigilant in implementing solid whitefly management strategies. In addition, due to recent publicity about potential impacts on pollinators, some large retail garden centers have been pressuring ornamental producers to stop using neonicotinoid insecticides. Certain neonicotinoids are among the few insecticides that are highly effective against *B. tabaci* MED. As a result, some growers may have relied on older, less effective chemistries to which *B. tabaci* MED is resistant. These include pyrethroids, organophosphates, carbamates, insect growth regulators, and some neonicotinoids (Nauen et al. 2002, Horowitz et al. 2005, Nauen & Denholm, 2005).

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Table 2. Sample date, host plant, nursery environment, Florida county, and biotype specific primer results for wholesale and retail nursery detections of *Bemisia tabaci* Middle Eastern Asia Minor 1 (MEAM1; biotype B) and Mediterranean (MED; biotype Q).

				Biotype specific primer	
Date received	Host plant	Nursery environment[a]	Florida county	MEAM1 (B)	MED (Q)
11 May 2016	Hibiscus	Wholesale[1]	Palm Beach	0	16
1 Jun 2016	Hibiscus	Wholesale[2]	Highlands	0	23
12 Jun 2016	Hibiscus	Retail[1]	Martin	1	1
13 Jun 2016	Hibiscus	Wholesale[2]	Highlands	1	7
13 Jun 2016	Hibiscus	Wholesale[3]	Palm Beach	0	1
14 Jun 2016	Hibiscus	Retail[2]	Seminole	0	5
14 Jun 2016	Hibiscus	Retail[2]	Seminole	0	16
17 Jun 2016	Hibiscus	Retail[3]	Palm Beach	0	20
17 Jun 2016	Hibiscus	Retail[3]	Palm Beach	1	8
17 Jun 2016	Hibiscus	Retail[4]	Duval	0	20
22 Jun 2016	Hibiscus	Retail[5]	Broward	0	2
22 Jun 2016	Firecracker flower	Retail[6]	Pinellas	0	6
23 Jun 2016	Hibiscus	Retail[7]	Palm Beach	0	1
23 Jun 2016	Hibiscus	Retail[8]	Martin	0	2
27 Jun 2016	Eggplant	Retail[9]	Hillsborough	14	1
27 Jun 2016	Hibiscus	Retail[10]	Palm Beach	0	1
6 Jul 2016	Eggplant	Retail[11]	Seminole	10	1
6 Jul 2016	Hibiscus	Wholesale[4]	Miami-Dade	6	1
6 Jul 2016	Hibiscus	Retail[12]	Palm Beach	0	18
18 Jul 2016	Hibiscus	Wholesale[5]	Palm Beach	0	6
18 Jul 2016	Hibiscus	Retail[13]	Palm Beach	0	4
18 Jul 2016	Lantana	Wholesale[6]	Hillsborough	15	2
18 Jul 2016	Hibiscus	Wholesale[1]	Palm Beach	0	13
18 Jul 2016	Hibiscus	Wholesale[1]	Palm Beach	1	3
22 Jul 2016	Hibiscus	Retail[14]	Palm Beach	0	7
27 Jul 2016	Firecracker flower	Wholesale[7]	Hillsborough	13	3
27 Jul 2016	Hibiscus	Wholesale[8]	Miami-Dade	0	16
29 Jul 2016	Firecracker flower	Wholesale[7]	Hillsborough	0	8
12 Aug 2016	Eggplant	Retail[15]	St. Lucie	15	1
22 Aug 2016	Hibiscus	Retail[16]	Martin	0	3
8 Sept 2016	Hibiscus	Retail[17]	Palm Beach	0	3
8 Sept 2016	Hibiscus	Wholesale[8]	Miami Dade	6	10

[a]Samples under nursery environment with the same superscript number are confirmatory (2 or more collections at the same wholesale or retail nursery from different sampling dates).

Because we have detected *B. tabaci* MED in open field crops in the United States for the first time, and this pest is known to attack vegetables and cotton in other countries, there is a risk that unmanaged populations of *B. tabaci* MED could move from protected ornamental greenhouse production to open agriculture. In some cases, vegetable transplants and an array of ornamental plants are grown together in a greenhouse, which further increases the risk of introducing *B. tabaci* MED to field plantings of vegetables. There is also a concern that increased pesticide resistance will evolve in *B. tabaci* MEAM1.

To decrease the risk of *B. tabaci* MED and insecticide resistant *B. tabaci* MEAM1 spreading from ornamentals to field-grown vegetables and cotton, it is critical for producers and managers of ornamental plants to practice sound integrated pest management. This includes the use of cultural control, biopesticides, natural products (oils, soaps), and biological control. These management practices can provide some control of whiteflies under low pest pressure. However, under higher pest pressure, it is critical for growers to select targeted chemistry with known activity against *B. tabaci* MED and, if necessary, destroy infested crops.

This research was partially funded by the Floriculture and Nursery Research Initiative of the United States Department of Agriculture, Agricultural Research Service.

Summary

For the first time in the United States, *Bemisia tabaci* MED (Q biotype of *B. tabaci* [Gennadius]; Hemiptera: Aleyrodidae) was detected outside of greenhouse or nursery environments when it was collected from 10 residential landscape and 2 open field environments in Florida. *Bemisia tabaci* MED was also detected in 8 wholesale nurseries from 4 counties and 17 retail nurseries from 8 counties in Florida. Hibiscus was the host plant driver for *B. tabaci* MED whitefly infestations in retail and wholesale nurseries and in Florida residential landscapes. One mint sample from a retail nursery contained a single New World whitefly and represents the first detection of New World *B. tabaci* in Florida since it was displaced by *B. tabaci* Middle Eastern Asia Minor 1 in the mid-1980s.

Key Words: MEAM1; Middle Eastern Asia Minor 1; biotype B; NW; New World; whitefly

Sumario

Por primera vez en los Estados Unidos, se detectó *Bemisia tabaci* MED (biotipo Q de *B. tabaci* [Gennadius]; Hemiptera: Aleyrodidae)

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fuera del ambiente de invernadero o de vivero cuando fue recolectada de 10 ambientes residenciales y 2 ambientes de campo abierto en la Florida. La MED también fue detectada en 8 viveros al por mayor de 4 condados y 17 viveros comerciales de 8 condados en la Florida. *Hibiscus* fue la planta hospedera que impulsó las infestaciones de la mosca blanca del MED en los viveros al por menor y comerciales y en los ambientes residenciales de la Florida. Una muestra de menta de un vivero comercial tenía una mosca blanca Nuevo Mundo del complejo de *Bemisia* y representa la primera detección del Nuevo Mundo *B. tabaci* en la Florida desde que fue desplazada por MEAM1 a mediados de los años ochenta.

Palabras Clave: MEAM1; Oriente Medio Asia Menor 1; biotipo B; noreste; nuevo mundo; mosca blanca

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