SCIENTIFIC NOTE

Identification, Geographical Distribution and Host Plants of *Bemisia tabaci* (Genn.) Biotypes (Homoptera: Aleyrodidae) in the State of Paraná, Brazil

Sueli S. Martinez, Alfredo O. R. de Carvalho, Luiz G. Vieira, Liliane M. Nunes and Anésio Bianchini

IAPAR, Instituto Agronômico do Paraná, Caixa postal 481, 86001-970, Londrina, PR, Brasil. e-mail: suemart@onda.com.br

An. Soc. Entomol. Brasil 29(3): 597-603 (2000)

Identificação, Distribuição Geográfica e Plantas Hospedeiras de Biótipos de *Bemisia tabaci* (Genn.) (Homoptera: Aleyrodidae) no Estado do Paraná

RESUMO – O objetivo deste trabalho foi identificar os biótipos de *Bemisia tabaci* (Genn.) presentes no estado do Paraná e determinar sua distribuição geográfica e plantas hospedeiras. Cerca de 50 adultos foram coletados em diversas culturas hospedeiras, plantas daninhas e ornamentais, nas regiões Norte, Noroeste, Nordeste, Oeste e Centro do estado, de janeiro a maio, durante os anos de 1998 e 1999. As espécies foram identificadas por meio de RAPD-PCR, usando-se o primer Operon H-16. Populações de mosca branca foram detectadas a partir do mês de fevereiro em ambos os anos, raramente atingindo níveis superiores a um adulto por folha. O inseto foi encontrado em somente 66% das áreas amostradas e o mosaico dourado do feijoeiro praticamente não foi detectado durante o período. Ambos biótipos A e B de *B. tabaci* foram coletados no estado do Paraná, sendo o biótipo B mais restrito à Região Norte. Apesar de este biótipo ter sido encontrado colonizando uma faixa maior de plantas hospedeiras, ele não ainda não se encontra amplamente distribuído nas diversas regiões e a maior parte dos espécimes coletados foram identificados como biótipo A. Um padrão de bandas distinto daqueles obtidos para os biótipos A e B de *B. tabaci* foi obtido exclusivamente com as populações coletadas em mandioca, indicando a possível presença de um biótipo específico para essa cultura.

PALAVRAS-CHAVE**:** Insecta, mosca branca, RAPD, variabilidade de DNA.

ABSTRACT - This work was carried out in order to identify *Bemisia tabaci* (Genn.) biotypes present in the state of Paraná and to determine their geographical distribution and host plants. About 50 adults were collected in several host crops, weeds and ornamental plants in North, Northwest, Northeast, West and Central areas of the state, from January to May, 1998 and 1999. The species were identified by means of RAPD-PCR, using the primer Operon H-16. Whitefly populations were detected mainly from February on, in both years, seldom achieving more than one adult per leaf. The insect was found in only 66% of the sampled areas. Bean golden mosaic was almost never observed during the pe-

riod. Both biotype A and biotype B of *B. tabaci* were found in the State of Paraná, the last one being more restricted to the north region. Although biotype B was found colonising a wider range of host plants, it has not spread out in all the state and most of the whitefly specimens found are still the biotype A. A banding pattern distinct of those obtained for A and biotype B of *B. tabaci* was obtained exclusively with the populations collected from cassava, thus indicating the possible presence of a biotype specific for this crop.

KEY WORDS: Insecta, silverleaf whitefly, sweetpotato whitefly, RAPD, DNA variability.

The sweetpotato whitefly *Bemisia tabaci* (Genn.) has been reported as an important pest in Brazil for nearly 30 years (Costa *et al.* 1973). The main damage caused by this species is related to gemminivirus transmission (Bedford *et al*. 1994), which can be extremely virulent, like the golden mosaic virus, transmitted to common beans, that can cause near 100% loss during the "dry season" (November to April) (Bianchini *et al.* 1989). However, the occurrence of this species and of the bean golden mosaic virus in the state of Paraná decreased during the last years, possibly due to: a) the reduction of bean planted areas in regions and periods with high incidence of the pest, which followed a governmental policy of providing financial support only for the regions determined in annual research recommendations (Kranz *et al.* 1988); b) the replacement of host crop areas (cotton) with non-host ones (maize); and c) the use of cultivars resistant to the virus, like IAPAR-57/1992, IAPAR-65/1993 e IAPAR-72/1994 (Bianchini 1999).

However, a new whitefly biotype, the silverleaf whitefly, also known as *B. argentifolii* Bellows & Perring (Perring *et al.* 1993) was recorded in 1991 in the State of São Paulo on ornamental plants, mainly *Chrysanthemum morifolium* and poinsettia, and on vegetables, such as tomato, brocoli, egg-plant and squash (Lourenção & Nagai 1994). Subsequently the species has spread out to many other states in Brazil and to other hosts and presently it causes severe losses in Northeast and Southeast regions. *B. argentifolli* attacks a wider range of host plants and, besides transmitting viruses as *B. tabaci*, it causes striking plant disorders, resulting from the phloem suction, such as: squash silverleaf (Yokomi *et al*. 1990), tomato irregular ripening (Lourenção & Nagai 1994), stem blenching in cole crops, poinsettia and so on. Furthermore, this species is reported as resistant to most of the insecticides registered for use against whiteflies (Vilas-Boas *et al*. 1997).

Most of the host crops of the silverleaf whitefly are grown in Paraná, thus bringing into concern the potential damage of this species to the agriculture of the state. This paper is a preliminary report of a three-year project, which began in 1998, designed to provide the identification of the whitefly biotypes present in the area, their geographical distribution and the host plants colonised by the pest, in order to evaluate the actual situation of the species in the state.

Whiteflies are normally identified by means of pupal characteristics, but this stage has no specific characteristics to reliably distinguish the different biotypes (Mound apud De Barro & Driver 1997). The only faithful methods indicated for this purpose are those based on the variability in proteins or DNA (De Barro & Driver 1997) and the RAPD-PCR (randomly amplified polymorphic DNA – polimerase chain reaction) has the advantage of using small amount of fresh or frozen material.

Adults of whitefly were surveyed in sev-

eral of their most important host crops: soybean (*Glycine max*), dry beans (*Phaseolus vulgaris*), peanuts (*Arachys hypogaea*), cotton (*Gossypium hirsutum*), sweetpotato (*Ipomoea batatas*), cassava (*Manihot esculenta*), squash (*Cucurbita moschata*), on the ornamentals poinsettia (*Euphorbia pulcherrima*) and *Crossandra* sp., and on some weeds, like *Euphorbia heterophylla*, *Ipomoea acuminata*, *Sida rhombifolia*, *Leonurus sibiricus* and *Bidens pilosa*. Evaluations were made from January to May, in 1998 and 1999, in North, Northeast, Northwest and Central regions of Paraná State, identified as the most favourable period and areas for *B. tabaci* development (Hohmann & Carvalho 1989, Hohmann & Martinez 2000). About 50 adults were collected in each crop by means of a suction apparatus, transferred to a vial containing 70% alcohol and brought to the laboratory where they were maintained in the refrigerator, at 4ºC until DNA extraction. Five replications were used for each crop and five adults were used from each sample. The DNA extraction method was adapted from Cheung *et al.* (1993) and the homogenisation of the adults in microcentrifuge tubes under liquid nitrogen was included. The DNA quantification was performed by fluorimetry and the samples, containing more than 15 ng DNA/ml, were used for PCR-RAPD. DNA quality for each tube containing 1000 ng was determined by eletrophoresis, at 60V in an agarosis gel at 0.8%, stained with ethidium bromide at 0.05% and photographed by means of a Polaroid camera using Polaroid film 667, or a digital camera. PCR-RAPD was developed according to Gawel & Bartlett (1993), using the primer OP H-16, as suggested by De Barro & Driver (1997). The presence/absence of bands between 300 and 500 pb on the profiles was used to distinguish the whitefly biotypes (De Barro *et al.* 1997), in comparison to standard profiles performed for A and Biotype B of *B. tabaci* (Fig. 1). The same photographic cam-

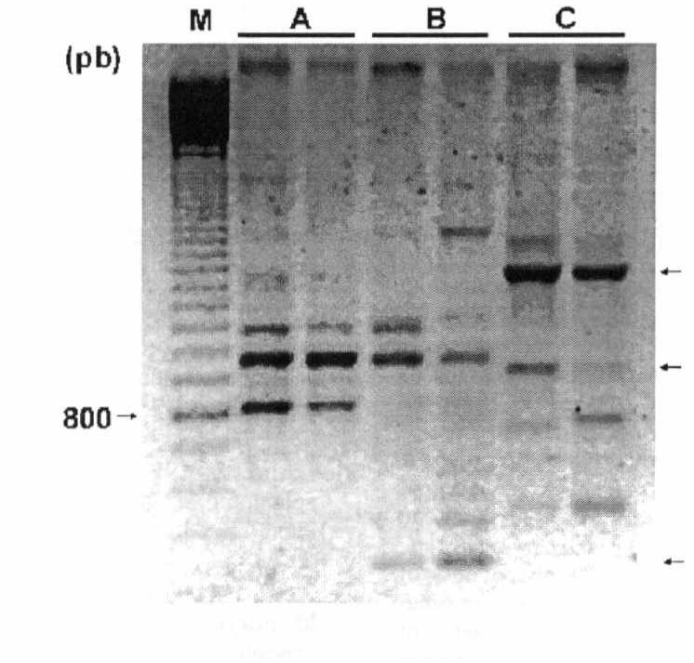

Figure 1. RAPD profiles of *B. tabaci*. M – Ladder 100 marker; A – Biotype A; B – Biotype B; C – cassava biotype. (setae indicate the specific bands)

era described above was used to record these profiles.

Whitefly populations started to be detected mainly in February, in both years (1998/9), seldom achieving more then one adult per leaf. The species was found in only 66% of the sampled areas and mostly after the main crops had already been harvested; populations were found in the weeds which remained in these areas. The bean golden mosaic was almost rarely observed in bean plants during the period.

Both A and biotype B of *B. tabaci* were found in different crops in Paraná. Although it was expected that the biotype B had totally replaced the former one due to its higher range of hosts (Bedford *et al*. 1994), this was not proved true in Paraná, since the biotype A was observed in most of the regions evaluated.

To the moment, the biotype B was more often found in the North region (Fig. 2). There is only one record of this species in the Central area, which is close to the North region. It was found once in the West region, on the ornamental poinsettia, considered the major disseminator of this biotype (Byrne & Devonshire 1993, Vilas Boas *et al*. 1997). North of Paraná is the region closest to the State of São Paulo, where this biotype was first detected (Lourenção & Nagai 1994) and where very high populations were observed mainly on soybean areas in 1996 (D.R. Sosa-Gómez – unpublished data). It is possible that this contiguity and the presence of large areas with host species in both states have favoured the introduction of the biotype B in Paraná.

Both biotypes were found on dry beans and on *E. heterophylla*, weed that remained in the field after dry beans or soybean crops were harvested (Table 1). Only the biotype A was found in peanuts and only the biotype B was collected in sweetpotato, squash, and in the ornamental poinsettia and *Crossandra* sp., thus confirming the wider number of hosts of the biotype B and its presence also in ornamental plants. The simultaneous presence of these two species was not observed in the same crop area.

A banding pattern distinct of those obtained for biotype A and B of *B. tabaci* was obtained exclusively with the populations collected from cassava, in North and Northwest regions (Fig. 1). The two bands with about 920 pb and 1300 pb exclusively found in the cassava biotype profile and the presence of the single band of about 460 pb, also observed in biotype A but not in biotype B of *B. tabaci*, represent a consistent reproducible pattern which is unique in the cassava biotype, when the Primer Operon16 is used. It seems that an adaptation of this biotype to cassava crop has occurred, since it was not found in any other crop and no other *B. tabaci* biotypes were found in cassava.

It is possible to conclude that there are several biotypes of *B. tabaci* in the state of Paraná, all of them detected with the primer Operon 16. Also, although the biotype B of *B. tabaci* was found colonising different crops, it has not spread throughout the state and the biotype A is still found in most of the regions surveyed. These studies will be continued during the next years, in larger areas and higher variety of host species in order to map the regions of occurrence of the different *Bemisia* species and evaluate the movement of the populations.

Literature Cited

Bedford, I.D., R.W. Briddon, J.K. Brown, R.C. Rosell & P.G. Markham. 1994. Geminivirus transmission and biological characterization of *Bemisia tabaci* (Gennadius) biotypes from different geographic regions. Ann. Appl. Biol. 125: 311-325.

Bianchini, A. 1999. Resistance to bean golden mosaic virus in bean genotypes. Plant Dis. 83: 615-620

Bianchini, A., J.R. Menezes. & A.C. Maringoni. 1989. Doenças e seu controle, p. 189-216, In IAPAR. O Feijão no Paraná, IAPAR, Circular Técnica 63, Londrina, 303 p.

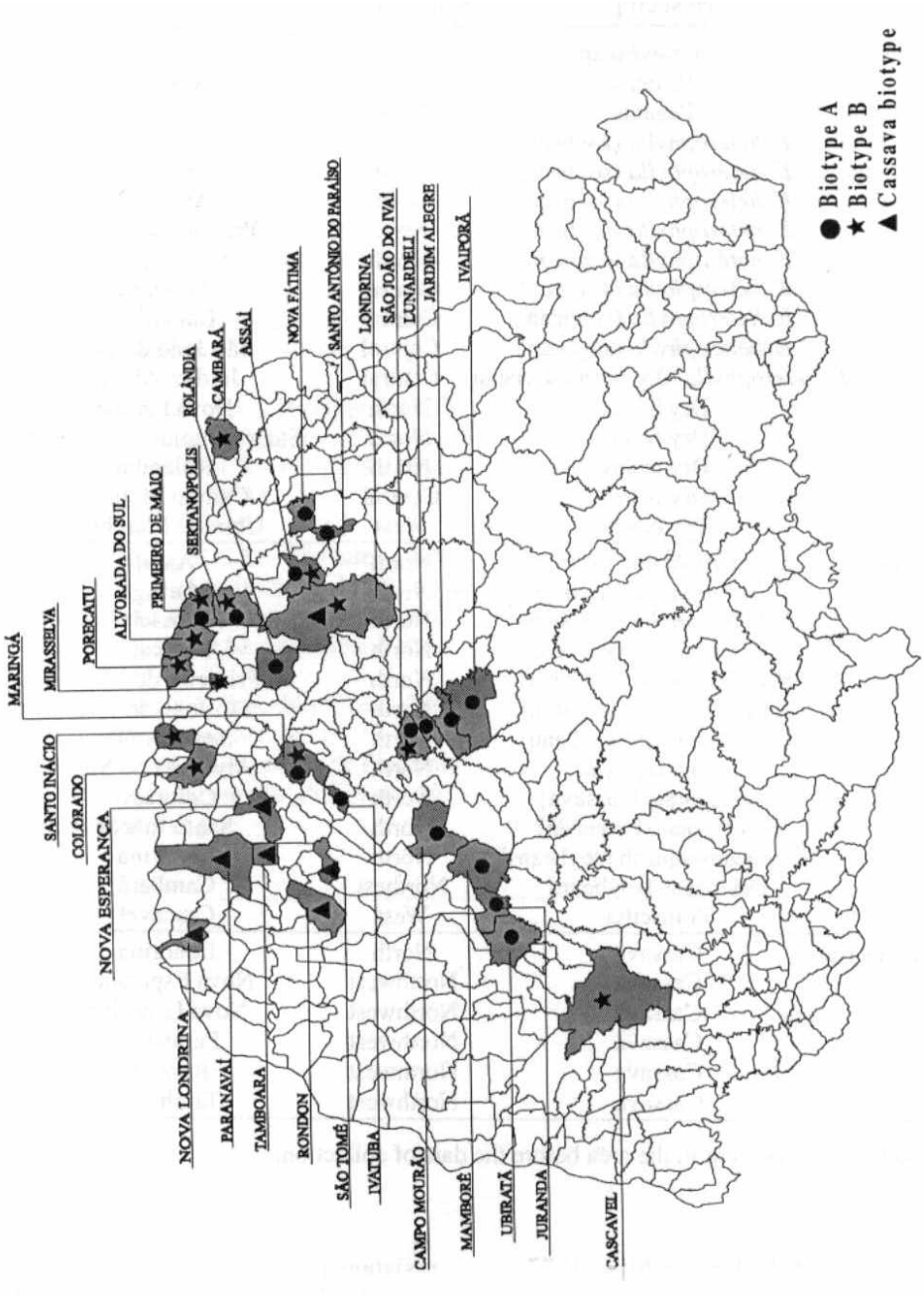

Figure 2. Geographical distribution of *B. tabaci* biotypes in Paraná State, Brazil. (January to May, 1998, 1999).

Table 1. *B. tabaci* biotypes collected in different crops in several regions of the Parana State, during summer seasons of 1998 and 1999.

Species	Host crop[1]	State Region	Local
Biotype A	*Crossandra* sp.	North	Londrina
	Peanuts	North	Maringá
	Peanuts	West	Juranda
	E. heterophylla (soybean)	North	Assaí
	E. heterophylla (soybean)	North	Ivatuba
	E. heterophylla (soybean)	North	Maringá
	E. heterophylla (soybean)	North	Primeiro de Maio
	E. heterophylla (soybean)	North	Sertanópolis
	E. heterophylla (soybean)	Central	Ivaiporã
	E. heterophylla (soybean)	Central	Lunardeli
	E. heterophylla (soybean)	Central	São João do Ivaí
	E. heterophylla (dry beans/soybean)	Central	Jardim Alegre
	Dry beans	North	Nova Fátima
	Dry beans	North	Santo Antonio do Paraíso
	Dry beans	North	Rolândia
	Dry beans	Central	Campo Mourão
	Dry beans	West	Ubiratã/ Mamborê
Biotype B	*E. heterophylla* (soybean)	North	Assaí
	E. heterophylla (soybean)	North	Maringá
	E. heterophylla (soybean)	North	Mirasselva
	E. heterophylla (soybean)	North	Porecatu
	E. heterophylla (soybean)	North	Primeiro de Maio
	E. heterophylla (soybean)	North	São João do Ivaí
	E. heterophylla (soybean)	North	Sertanópolis
	E. heterophylla	North	Alvorada do Sul
	Sweetpotato [cassava]	North	Colorado
	Sweetpotato [soybean]	North	Santo Inácio
	Dry beans/squash [soybean]	North	Londrina
	Dry beans [soybean]	Northest	Cambará
	Poinsettia	West	Cascavel
Cassava biotype	Cassava	North	Londrina
	Cassava	Northwest	Nova Esperança
	Cassava	Northwest	Nova Londrina
	Cassava	Northwest	Paranavaí
	Cassava	Northwest	Rondon
	Cassava	Northwest	Tamboara

[1]In brackets – crop existent in the area before the data of collection.

Byrne, F.J. & A.L. Devonshire. 1993. Insensitive acetylcholinesterase and esterase polymorphism in susceptible and resistant populations of the tobacco whitefly *Bemisia tabaci* (Genn.). Pest. Biochem. Physiol. 45: 34-42.

Cheung, W.Y., N. Hubert & B.S. Landry. 1993. A simple and rapid DNA microextraction method for plant, animal, and insect suitable for RAPD and other PCR analyses**.** PCR Meth. Appl. 3:60-70.

Costa, A.S., C.L. Costa & H.F.G. Sauer. 1973. Surto de mosca branca em culturas do Paraná e São Paulo. An. Soc. Entomol. Brasil 2: 20-30.

De Barro, P. J. & F. Driver. 1997. Use of RAPD PCR to distinguish the B Biotype from other biotypes of *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae). Austr. J. Entomol. 36: 149-152.

Gawel, N. J. & A.C. Bartlett. 1993. Characterization of differences between whiteflies using RAPD-PCR. Insect Molec. Biol. 2: 33-38.

Hohmann, C.L. & S.M. Carvalho. 1989. Pragas e seu controle, p. 217-246. In IAPAR. O Feijão no Paraná, IAPAR, Circular Técnica 63, Londrina, 303 p.

Hohmann, C.L. & S.S. Martinez. 2000. Pragas e seu controle. p. 77-82. In IAPAR. Feijão. Tecnologia de Produção, IAPAR, Informe da Pesquisa 135, Londrina, 115 p.

Kranz, W.M., A. Bianchini, M.A. Lollato, V. Moda-Cirino, L. Oliari & P.G.F. Ribeiro. 1988. Aptidão fitotécnica e zoneamento de cultivares de feijoeiro no Paraná. IAPAR, Londrina, Informe da Pesquisa, 16 p.

Lourenção, A.L. & H. Nagai. 1994. Surtos populacionais de *Bemisia tabaci* no Estado de São Paulo. Bragantia 53: 53-59.

Perring, T.M., A. Cooper, R.J. Rodriguez, C.A Farrar & T.S. Bellows Jr. 1993. Identification of a whitefly species by genomic and behavioural studies. Science 259: 74-77.

Villas Bôas, G.L., F.H. França, A.C. Ávila & I.C. Bezerra. 1997. Manejo integrado da mosca-branca *Bemisia argentifolii*. Embrapa, Circular Técnica 9 - Embrapa Hortaliças, 11 p.

Yokomi, R.K., K.A. Hoelmer & L.S. Osborne. 1990. Relationships between the sweetpotato whitefly and squash silverleaf disorder. Amer. Phytopatol. Soc. 10: 895-899.

Accepted 30/VI/2000.