

TOMATO YELLOW VEIN STREAK VIRUS: RELATIONSHIP WITH *BEMISIA TABACI* BIOTYPE B AND HOST RANGE

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ABSTRACT: The Tomato yellow vein streak virus (ToYVSV) is a putative species of begomovirus, which was prevalent on tomato crops in São Paulo State, Brazil, until 2005. The objectives of this study were to evaluate the interaction between ToYVSV and its vector *Bemisia tabaci* biotype B and to identify alternative hosts for the virus. The minimum acquisition and inoculation access periods of ToYVSV by *B. tabaci* were 30 min and 10 min, respectively. Seventy five percent of tomato-test plants were infected when the acquisition and inoculation access periods were 24 h. The latent period of the virus in the insect was 16 h. The ToYVSV was retained by *B. tabaci* until 20 days after acquisition. First generation of adult whiteflies obtained from viruliferous females were virus free as shown by PCR analysis and did not transmit the virus to tomato plants. Out of 34 species of test-plants inoculated with ToYVSV only *Capsicum annuum*, *Chenopodium amaranticolor*, *C. quinoa*, *Datura stramonium*, *Gomphrena globosa*, *Nicotiana clevelandii* and *N. tabacum* cv. TNN were susceptible to infection. *B. tabaci* biotype B was able to acquire the virus from all these susceptible species, transmitting it to tomato plants.

Key words: *Solanum lycopersicon*, geminivirus, begomovirus, aleyrodidae, transmission

TOMATO YELLOW VEIN STREAK VIRUS: INTERAÇÃO COMA *BEMISIA TABACI* BIÓTIPO B E GAMA DE HOSPEDEIROS

RESUMO: O Tomato yellow vein streak virus (ToYVSV) é uma espécie putativa de begomovirus que infecta o tomateiro (*Solanum lycopersicon*) em diversas regiões do Brasil onde se cultiva essa solanácea, sendo a espécie prevalente no estado de São Paulo até 2005. Estudou-se a interação do ToYVSV com a *Bemisia tabaci* biótipo B e identificaram-se hospedeiras alternativas deste vírus. Os períodos de acesso mínimo de aquisição (PAA) e de inoculação (PAI) foram de 30 min e 10 min, respectivamente. A porcentagem de plantas infectadas chegou até cerca de 75% após um PAA e PAI de 24 h. O período de latência do vírus no vetor foi de 16 horas. O ToYVSV foi retido pela *B. tabaci* até 20 dias após a aquisição do vírus. Não foi detectada transmissão do vírus para progênie da *B. tabaci* biótipo B oriundas de insetos virulíferos. De 34 espécies de plantas testadas como hospedeiras somente *Capsicum annuum*, *Chenopodium amaranticolor*, *C. quinoa*, *Datura stramonium*, *Gomphrena globosa*, *Nicotiana clevelandii* e *N. tabacum* cv. TNN foram suscetíveis à infecção com o ToYVSV, por meio de inoculação com a *B. tabaci*. As espécies suscetíveis ao ToYVSV serviram de fonte de inóculo para a transmissão do vírus para tomateiros por meio de *B. tabaci* biótipo B.

Palavras-chave: *Solanum lycopersicon*, geminivirus, begomovirus, aleyrodidae, transmissão

INTRODUCTION

Recent studies on molecular characterization of begomoviruses infecting tomato plants (*Solanum lycopersicon*) indicated the existence of a complex diversity of species in Brazil (Ambrozevícus et al., 2002; Ribeiro et al., 2003), resulting from the introduction and dispersal throughout the country of the effective vec-

tor *Bemisia tabaci* biotype B since the 1990's (Lourenço & Nagai, 1994; França et al., 1996). Currently, this complex comprises four recognized species, at least eight other potential species, including Tomato yellow vein streak virus (ToYVSV) (Colariccio et al., 2007), and some recombinant forms of this virus, which have not been properly characterized (Ambrozevícus et al., 2002; Calegario et al., 2007; Fernandes et al., 2008).

The first report on ToYVSV infecting tomato plants in Brazil occurred in 1997, in the region of Campinas, State of São Paulo (Faria et al., 1997). In this region, incidence of ToYVSV infected plants ranging from 58% to 100% in 2003 and from 14 to 27% in 2004 were observed (Rezende, J.A.M. data not published). In tomato plants cultivated under plastic greenhouse conditions the incidence of ToYVSV ranged from 4.8% to 69.3%, depending on the planting season (Vecchia et al., 2007). This virus has also been previously reported to infect green pepper (*Capsicum annuum*) (Nozaki, 2006) and potato (*Solanum tuberosum*) (Ribeiro et al., 2006; Souza-Dias et al., 1996).

The acquisition and transmission of begomoviruses by *B. tabaci* has been extensively studied, and these parameters can vary depending on the virus and the aleyrodid biotype (Cohen & Nitzany, 1966; Polston et al., 1990; Zeidan & Czosnek, 1991; Mehta et al., 1994a; Rubinstein & Czosnek, 1997; Ghanim & Czosnek, 2000; Muniyappa et al., 2000). Determining these parameters, together with knowledge on the range of virus hosts, allows to elucidate the epidemiology of different begomovirus diseases, as well as to develop disease management strategies. In Brazil, up to now these parameters have been identified only for the transmission of one begomovirus isolate, taxonomically related to *Tomato rugose mosaic virus* (ToRMV), by *B. tabaci* biotype B (Santos et al., 2003).

This study aimed to investigate the acquisition, retention and transmission of a ToYVSV isolate by *B. tabaci* biotype B, and to identify potential alternative virus hosts.

MATERIAL AND METHODS

ToYVSV isolate and *B. tabaci* biotype B maintenance

The ToYVSV isolate used in this study was maintained in tomato plants by *B. tabaci* transmission. The identity of the virus was confirmed by partial DNA-A nucleotide sequence analysis of a fragment flanking the common region (RC) and the 5' of the coat protein gene (*cp*) (data not shown). Virus-free *B. tabaci* biotype B adults were reared on collard greens (*Brassica oleracea*), soybean (*Glycine max*), and tomato plants, maintained in a greenhouse equipped with insect-proof screen. The *B. tabaci* species/biotype was identified by Dr. Geni L. Villas Boas and Dr. Maria Esther N. F. Boiteux (Embrapa Hortalícias). In addition, the whiteflies induced silvery leaf on *Cucurbita pepo* cv. Caserta, which is characteristic of this biotype.

ToYVSV detection by PCR

DNA extraction from tomato leaves and from *B.*

tabaci biotype B nymphs and adults was achieved according to the methods described by Dellaporta et al. (1983) and Mehta et al. (1994b), respectively. Total DNA was used in a PCR reaction using the degenerate primer pairs PAR1c715/PAL1v1978, which amplifies a DNA A fragment with approximately 1,300 bp, and PBL1v2040/PCR1, which amplifies a DNA B fragment with approximately 500 to 650 bp (Rojas et al., 1993).

Determination of ToYVSV acquisition (AAP) and inoculation access period (IAP) by *B. tabaci* biotype B

Adult, virus-free *B. tabaci* individuals were placed in 50 mL polypropylene vials (*ca* 150 insects) containing a ToYVSV-infected tomato leaf, in order to acquire the virus. Nine virus AAP by the vector were evaluated: 10, 20, and 30 min, 1, 2, 4, 8, 16, and 24 h. After each AAP, the insects were transferred, in groups of 15 individuals, into pots containing two tomato test plants cultivar Santa Cruz Kada Gigante, at the stage when two true leaves were present. Pots were covered with insect-proof cages, and the insects remained there for a 24 h feeding period. To evaluate the IAP, the virus-free insects were initially submitted to a 24 h virus AAP. Groups of 15 insects were transferred to tomato test plants as described above. Nine virus IAP by the vector were evaluated: 10, 20, and 30 min, 1, 2, 4, 8, 16, and 24 h. Ten test plants were used for each AAP and IAP assays. Upon completion of the various AAP and IAP assays, the insects were immediately eliminated by spraying the plants with pyrethroid insecticides and imidacloprid (systemic). The plants were maintained in the greenhouse for periodic symptom evaluations. Infection in all plants was confirmed by PCR for DNA-A detection.

Virus latency period (LP) determination in the vector

The ToYVSV acquisition and transmission processes by *B. tabaci* biotype B were identical to those described previously. After a virus AAP of 1 h the insects were transferred to healthy tomato test plants. Fifteen insects were used per each two plants. Virus IAPs of 7, 9, 11, 13, 15, and 23 h were adopted. After each period, the insects were immediately eliminated as previously described. Ten test plants were used for each IAP. Periodical symptom evaluations were conducted, and plant infection was confirmed by DNA-A detection by PCR.

Determination of virus retention period by the vector

Approximately 1000 *B. tabaci* biotype B adults had a ToYVSV AAP of 48 h as previously described. The insects were then transferred to a ToYVSV-resistant

collard greens plant (*Brassica oleraceae*), maintained in an insect-proof cage inside a greenhouse. Fifteen days later, the insects were transferred to a new collard greens plant to prevent that first-generation adults would perhaps become mixed with previously-transferred virus-bearing insects. Periodically, groups of ten adults were transferred to two tomato test plants for a 48-h virus IAP, while another group of five insects was used for viral DNA-A detection by PCR. This procedure was repeated eight times over a period from 15 to 20 days, until no more insects were found.

ToYVSV detection in the *B. tabaci* progeny

B. tabaci biotype B adults had a ToYVSV AAP of 24 h as previously described. Approximately 50 insects were immediately transferred into four tomato test plants, for a 24-h IAP in order to later confirm insect infectivity. Another 50 insects were transferred to a ToYVSV-resistant eggplant individual (*Solanum melongena*), placed in an insect-proof cage inside a greenhouse. The insects remained in the eggplants for 12 days to oviposit and were then eliminated manually. Fifty fourth-instar nymphs were collected from the eggplant leaves for viral DNA-A detection by PCR. The nymphs were analyzed in groups of 10. Adults emerged from the eggplant were transferred to tomato test plants for a virus IAP of 24 h. Twenty to 30 insects were confined in four test plants. Some of those adults were later collected for viral DNA-A detection by PCR.

Identification of ToYVSV host plants

The plant species inoculated with the ToYVSV isolate by means of virus-bearing *B. tabaci* biotype B adults are listed in table 5. In two assays the insects had 24-h virus AAPs and were then transferred to test plants of various species for a 48-h virus IAP, and were then eliminated as previously described. Twenty insects were confined into groups of three test plants of each species. In the third assay, the test plants of the various species were confined in an insect-proof cage containing a virus-bearing colony of the aleyrodid. The test plants remained exposed to the virus-bearing insects for 48 h, and were then removed and sprayed

with systemic insecticide (imidacloprid). All test plants inoculated were maintained in the greenhouse and were evaluated periodically for the expression of symptoms. Leaf samples of symptomatic and asymptomatic plants were collected 30 and 90 days after inoculation for DNA-A and DNA-B detection by PCR.

RESULTS AND DISCUSSION

Interaction between ToYVSV and *B. tabaci* biotype B

The minimum access periods for ToYVSV acquisition and inoculation by *B. tabaci* biotype B were 30 and 10 minutes, respectively. Infected plant means of 20 and 35% were obtained, respectively. A gradual increase occurred in number of infected plants as virus AAP and IAP increased, reaching 70% and 75% at an AAP and IAP of 24 h, respectively (Table 1). The PCR analysis for DNA-A detection confirmed that the plants were infected (data not shown).

The minimum ToYVSV acquisition and inoculation periods by *B. tabaci* are slightly different from those found for the transmission of a begomovirus isolate taxonomically related to ToRMV by the same insect in Brazil (Santos et al., 2003), which were of 15 and 30 min, respectively. Results similar to the ones observed in the present study were obtained in the transmission of *Cotton leaf curl virus* from India (CLCuKV), *Squash leaf curl virus* (SLCV), *Tomato leaf curl Bangalore virus - C* (ToLCBV-C) (sin. *Tomato leaf curl virus* from Bangalore, India - ToLCV-Ban4), and *Tomato yellow leaf curl virus* (TYLCV-EG) isolate from Egypt by *B. tabaci* biotype B (Cohen et al., 1983; Nateshan et al., 1996; Muniyappa et al., 2000; Mehta et al., 1994a). A TYLCV isolate from Jordan had minimum acquisition and inoculation access periods of 60 and 30 minutes, respectively (Mansour & Al-Musa, 1992). Longer acquisition and inoculation access periods, of 1 h and 2 h, respectively, were observed in an interaction between *Chino del tomate virus* (CdTV) and *B. tabaci* biotype B (Brown & Nelson, 1988).

Table 1 - Transmission of Tomato yellow vein streak virus (ToYVSV) by *Bemisia tabaci* biotype B after different acquisition access (AAP) and inoculation access periods (IAP).

| Time | | 10 min | 20 min | 30 min | 1 h | 2 h | 4 h | 8 h | 16 h | 24 h |
|-------|------------------------------|--------|--------|--------|------|------|------|------|------|------|
| AAP* | No. of infected/ | Exp. 1 | 0/10 | 0/10 | 2/10 | 2/10 | 4/10 | 2/10 | 8/10 | 8/10 |
| | Inoculated plants | Exp. 2 | 0/10 | 0/10 | 2/10 | 3/10 | 5/10 | 3/10 | 5/10 | 6/10 |
| | Means of infected plants (%) | | 0 | 0 | 20 | 25 | 35 | 25 | 65 | 70 |
| IAP** | No. of infected/ | Exp. 1 | 4/10 | 5/10 | 4/10 | 4/8 | 4/8 | 4/10 | 4/10 | 6/10 |
| | Inoculated plants | Exp. 2 | 3/10 | 4/10 | 4/10 | 5/10 | 5/10 | 4/10 | 4/10 | 7/10 |
| | Means of infected plants (%) | | 35 | 45 | 40 | 50 | 50 | 40 | 40 | 75 |

*24 h access to ToYVSV inoculation; **24 h access to ToYVSV acquisition

The ToYVSV latency period (LP) in *B. tabaci* biotype B adults was 16 h as determined in two independent experiments (Table 2). This result was identical to the LP reported for a ToRMV isolate from Brasília, Brazil (Santos et al., 2003) and similar to the LP for CdTV, which was approximately 17 h (Brown & Nelson, 1988). The SLCV LP in *B. tabaci* biotype B was 19 h, while the LP for the TYLCV isolate from Jordan was 20 - 24 h (Cohen et al., 1983; Mansour & Al-Musa, 1992), a little higher than the value obtained for ToYVSV. A LP longer than 24 h was found for the TYLCV isolate from Egypt (Mehta et al., 1994a), while a 6-h LP was reported for ToLCBV-C (Muniyappa et al., 2000).

B. tabaci biotype B adults that had a ToYVSV AAP of 48 h and were then maintained in a plant immune to the virus were capable of transmitting the virus for up to 20 days after acquisition. The virus presence in the insects was confirmed by PCR (Table 3). In both experiments, the number of infected plants decreased gradually, although the virus was detected in the insects until 25 days after acquisition. This reduction in

Table 2 - Transmission of Tomato yellow vein streak virus (ToYVSV) by *Bemisia tabaci* biotype B after a virus acquisition access period (AAP) of 1 hour and different inoculation access periods (IAP).

| IAP (h) | Latency period (h)* | Number of infected/inoculated plants | |
|---------|---------------------|--------------------------------------|--------------|
| | | Experiment 1 | Experiment 2 |
| 7 | 8 | 0/12 | 0/12 |
| 9 | 10 | 0/12 | 0/12 |
| 11 | 12 | 0/12 | 0/12 |
| 13 | 14 | 0/12 | 0/12 |
| 15 | 16 | 1/12 | 1/12 |
| 23 | 24 | 2/12 | 1/12 |

*Latency period = IAP+ AAP of 1 h.

the number of infected plants might have occurred because ToYVSV concentration decreased in *B. tabaci* biotype B, since the relationship of begomoviruses with this insect is of a persistent-circulative nature, without virus replication in the insects. A TYLCV quantitative study in virus-bearing *B. tabaci* biotype B adults indicated a reduction of 9% per day in virus genome detection in insects maintained in a virus-immune host (Mason et al., 2008).

The retention period of different begomoviruses in *B. tabaci* biotype B adults is also quite variable, from four to seven days in the case of CdTV (Brown & Nelson, 1988), nine days for *Tomato leaf curl Sinaloa virus* (ToLCSinV) (sin. *Sinaloa tomato leaf curl virus* - STLCV) (Idris & Brown, 1998), 11 to 12 days for ToLCBV-C (Muniyappa et al. 2000), 11 to 20 days for TYLCV isolate from Jordan (Cohen & Nitzany, 1966; Mansour & Al-Musa, 1992), and 26 days for SLCV (Cohen et al., 1983). These differences in virus AAP, IAP, LP, and period of retention within the insect are expected because, besides the variations in the conditions of the various experiments, especially with respect to insect numbers and the manner by which those insects were handled, other variables, such as begomovirus species, geographic origin of the virus isolate and of the *B. tabaci* biotype B population might affect these parameters, as reported in studies involving other begomoviruses (Picó et al., 1996; Muniyappa et al., 2000).

The ToYVSV did not pass onto the *B. tabaci* biotype B progeny, whose adults initially acquired the virus from infected tomato plants. Aleyrodid adult samples from each of the five independent experiments conducted were able to transmit ToYVSV right after acquisition, and thus proved to be viruliferous. Nevertheless, PCR analyses of total DNA extracted from first-generation nymphs and adults did not detect ToYVSV DNA-A. No tomato test plant became

Table 3 - Detection of Tomato yellow vein streak virus (ToYVSV) in *Bemisia tabaci* biotype B adults by PCR and number of plants infected by transmission with insects until 25 days after virus acquisition.

| Days after virus acquisition | Positive PCR Samples/No. samples tested* | | No. of infected plants/No. of inoculated plants** | |
|------------------------------|--|--------|---|--------|
| | Exp. 1 | Exp. 2 | Exp. 1 | Exp. 2 |
| 0 | 3/4 | 3/4 | 4/4 | 4/4 |
| 3 | 3/4 | 4/4 | 3/4 | 3/4 |
| 6 | 2/4 | 4/4 | 2/2 | 2/3 |
| 9 | 4/4 | 4/4 | 2/4 | 2/2 |
| 12 | 4/4 | 4/4 | 1/3 | 1/4 |
| 15 | 4/4 | 4/4 | 1/3 | 1/4 |
| 20 | 3/4 | 3/4 | 1/4 | 0/4 |
| 25 | 4/4 | NT | 0/4 | NT |

*Each sample consisted of five *B. tabaci* adults; ** Infection confirmed by PCR; NT: Not tested due to insect death.

infected when inoculated with first-generation adults (data not shown). Evidence for the lack of transmission of other begomoviruses into virus-bearing *B. tabaci* biotype B progenies has been reported previously (Costa, 1976; Idris & Brown, 1998). On the other hand, the nucleic acid from the Brasília ToRMV isolate was detected by PCR in all nymph stages and adults of *B. tabaci* biotype B from a virus-bearing colony, although adults were not capable of transmitting the virus to tomato plants (Santos et al., 2003). For TYLCV, however, Ghanim et al. (1998) showed

that the virus can be transmitted through the eggs of *B. tabaci* biotype B for at least two generations.

ToYVSV host species

Among the 34 species tested, only *C. annuum*, *C. amaranticolor*, *C. quinoa*, *D. stramonium*, *G. globosa*, *N. clevelandii* and *N. tabacum* cv. TNN were identified as susceptible to ToYVSV (Table 4). *C. amaranticolor*, *C. quinoa* and *G. globosa* were asymptomatic, while *C. annuum*, *D. stramonium*, *N. clevelandii* and *N. tabacum* cv. TNN exhibited mild systemic invasion symptoms.

Table 4 - Reaction of different plant species inoculated with Tomato yellow vein streak virus (ToYVSV) by means of *Bemisia tabaci* biotype B.

| Species tested | No. of positive PCR plants/No. of inoculated plants* | | | |
|-------------------------------------|--|--------|--------|------------|
| | Exp. 1 | Exp. 2 | Exp. 3 | Symptoms** |
| <i>Abelmoschus esculentus</i> | 0/6 | 0/6 | 0/6 | - |
| <i>Amaranthus viridis</i> | 0/6 | 0/6 | 0/6 | - |
| <i>Capsicum annuum</i> cv. Magali R | 3/6 | 3/6 | 4/6 | M |
| <i>C. baccatum</i> | 0/6 | 0/6 | NT | - |
| <i>Chenopodium amaranticolor</i> | 2/6 | 1/6 | 2/6 | NS |
| <i>C. quinoa</i> | 1/6 | 1/6 | 1/6 | NS |
| <i>Commelina benghalensis</i> | 0/6 | 0/6 | 0/6 | - |
| <i>Curcurbita pepo</i> cv. Caserta | 0/6 | 0/6 | 0/6 | - |
| <i>Datura metel</i> | 0/6 | 0/6 | NT | - |
| <i>D. stramonium</i> | 3/6 | 2/6 | 3/6 | M |
| <i>Euphorbia heterophylla</i> | 0/6 | 0/6 | 0/6 | - |
| <i>Glycine max</i> cv. Conquista | 0/6 | 0/6 | 0/6 | - |
| <i>Gomphrena globosa</i> | 1/6 | 1/6 | 1/6 | NS |
| <i>Gossypium hirsutum</i> | 0/6 | 0/6 | 0/6 | - |
| <i>Ipomoea grandifolia</i> | 0/6 | 0/6 | 0/6 | - |
| <i>Nicotiana benthamiana</i> | 0/6 | 0/6 | 0/6 | - |
| <i>N. clevelandii</i> | 3/6 | 1/6 | 2/6 | VC, RL |
| <i>N. edwardson</i> | 0/6 | 0/6 | NT | - |
| <i>N. rustica</i> | 0/6 | 0/6 | NT | - |
| <i>N. tabacum</i> cv. Havana | 0/6 | 0/6 | 0/6 | - |
| <i>N. tabacum</i> cv. TNN | 2/6 | 1/6 | 3/6 | VC, RL |
| <i>N. tabacum</i> cv. Turkish | 0/6 | 0/6 | 0/6 | - |
| <i>N. tabacum</i> cv. Xanthi | 0/6 | 0/6 | NT | - |
| <i>Nicandra physaloides</i> | 0/6 | 0/6 | 0/6 | - |
| <i>Oxalis latifolia</i> | 0/6 | 0/6 | 0/6 | - |
| <i>Phaseolus vulgaris</i> | 0/6 | 0/6 | 0/6 | - |
| <i>Physalis floridana</i> | 0/6 | 0/6 | 0/6 | - |
| <i>Raphanus raphanistrum</i> | 0/6 | 0/6 | 0/6 | - |
| <i>Senna obtusifolia</i> | 0/6 | 0/6 | 0/6 | - |
| <i>Sida rhombifolia</i> | 0/6 | 0/6 | 0/6 | - |
| <i>Solanum americanum</i> | 0/6 | 0/6 | 0/6 | - |
| <i>S. sisymbriifolium</i> | 0/6 | 0/6 | 0/6 | - |
| <i>S. tuberosum</i> cv. Cupido | 0/6 | 0/6 | NT | - |
| <i>Sonchus oleraceus</i> | 0/6 | 0/6 | 0/6 | - |

*Exp. 1 and 2: conducted by confining virus-bearing insects in individual cages; Exp. 3: conducted by exposing the plants to the virus-bearing colony. **M: Mosaic; VC: Vein clearing; RL: Rugose leaf; NS: No symptoms; NT: Not tested; -: Lack of infection.

Symptoms were observed approximately 20 days after inoculation and gradually attenuated as plants grew, becoming imperceptible 90 days after inoculation. Even after that period, extracts from plants identified as susceptible to ToYVSV still gave positive reactions to the presence of DNA A, but not to DNA B (data not presented), which was also not detected in the previous analysis, when plants were younger.

The results for the species susceptible to the ToYVSV isolate used in this study differ partially from those obtained by Souza-Dias et al. (1996), who identified *D. stramonium*, *Physalis* sp., *N. tabacum* cv. Turkish, *P. vulgaris* cv. Preto, *Sida micrantha*, *S. rhombifolia* and *S. tuberosum* cvs. Bintje, Achat, Monalisa, Spunta, Baraka, Atlantic, and Itararé as susceptible to an isolate of this begomovirus from Sumaré, SP, Brazil, transmitted by *B. tabaci* biotype B, but not by mechanical inoculation. *N. benthamiana* and *N. physaloides* were other species identified as susceptible to a ToYVSV isolate obtained from potato in the State of Rio Grande do Sul, Brazil, during a transmission test with this aleyrodid (Ribeiro et al., 2006). Although these two isolates were only transmitted by *B. tabaci* biotype B, Colariccio et al. (2007) reported the mechanical transmission of a ToYVSV isolate from Monte Mor, SP, Brazil, from tomato to tomato cv. Carmem. Attempts to accomplish the mechanical transmission of the ToYVSV isolate used in the present study into the seven virus-susceptible species failed (data not presented).

The observation of a restricted range of ToYVSV-susceptible species, with predominance of species in the family Solanaceae, the variation in the expression of symptoms between susceptible species, and their variability with regard to susceptibility to different isolates of the virus are in accordance with reports for other begomovirus species that infect tomato plants, such as ToLCV (Stonor et al., 2003), ToLCSinV (Idris & Brown, 1998), CdTV (Brown & Nelson, 1988) and TYLCV (Picó et al., 1996).

Table 5 - Transmission of Tomato yellow vein streak virus (ToYVSV) to tomato plants 48 h after virus acquisition by *Bemisia tabaci* biotype B in different virus-susceptible species.

| ToYVSV source plant | No. of infected plants/No. of inoculated plants* | | Mean infected plants (%) |
|---------------------------|--|--------|--------------------------|
| | Exp. 1 | Exp. 2 | |
| <i>C. annum</i> | 1/10 | 6/10 | 30 |
| <i>C. amaranticolor</i> | 0/10 | 4/10 | 20 |
| <i>C. quinoa</i> | 1/10 | 2/10 | 15 |
| <i>D. stramonium</i> | 1/10 | 3/10 | 20 |
| <i>G. globosa</i> | 2/10 | 5/10 | 35 |
| <i>N. clevelandii</i> | 1/10 | 4/10 | 25 |
| <i>N. tabacum</i> cv. TNN | 1/10 | 3/10 | 20 |

*15 insects per tomato test plant. Inoculation access period 48 h.

Tests conducted to verify whether plants identified as susceptible to ToYVSV can serve as a source of inoculum for virus acquisition by *B. tabaci* biotype B and later transmission to tomato plants gave positive results (Table 5). ToYVSV presence was also confirmed in groups of aleyrodids that fed on the various plant species used as sources of inoculum (data not presented). These results demonstrate the importance of eliminating these plant species from the surroundings of new crops, because they could serve as reservoirs of the virus not only for tomato, but also for potato and green pepper crops, where ToYVSV has been previously found (Souza-Dias et al., 1996; Nozaki, 2006; Ribeiro et al., 2006).

Although monitoring and controlling *B. tabaci* biotype B populations are trivial practices for the control of ToYVSV and other begomoviruses in tomato crops in Brazil, they can be ineffective if virus and vector sources exist in the surroundings. The short period of time (10 min) required for ToYVSV inoculation by *B. tabaci* biotype B can be sufficient to allow inoculation of the virus into some plants before the insecticide can act upon the insect. Sometimes, the small yield increase obtained with a number of insecticidal sprays for the control of this aleyrodid in order to control the virus disease might not be justified economically and ecologically. Therefore, an integrated management for disease control, which includes the use of available resistant varieties, elimination of tomato plants from old crops as well as weeds that can harbor the virus and the vector before starting the new crop, rational chemical control of vector based on monitoring insect population should be adopted.

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