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Authors: Avila, Yolanda, Stavisky, Julianne, Hague, Sara, Funderburk, Joe, Reitz, Stuart, et al.

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EVALUATION OF *FRANKLINIELLA BISPINOSA* (THYSANOPTERA: THRIPIDAE) AS A VECTOR OF THE *TOMATO SPOTTED WILT VIRUS* IN PEPPER

YOLANDA AVILA[1], JULIANNE STAVISKY[1], SARA HAGUE[1], JOE FUNDERBURK[1], STUART REITZ[2] AND TIM MOMOL[1]
[1]North Florida Research and Education Center, University of Florida, 155 Research Road, Quincy, FL 32351

[2]USDA ARS CMAVE, 6383 Mahan Drive, Tallahassee, FL 32308

ABSTRACT

Frankliniella occidentalis is the key vector responsible for the emergence of *Tomato spotted wilt virus* as a global threat to agriculture. *Frankliniella bispinosa* is a common thrips in Florida, the Bahamas, and Bermuda, but the role of *F. bispinosa* in the epidemiology of the virus is not known. The purpose of this study was to determine the ability of *F. bispinosa* to acquire and transmit *Tomato spotted wilt virus* in pepper. In laboratory experiments, the number of larvae produced per *F. bispinosa* female was less than the number of larvae produced per *F. occidentalis* female. The larvae of *F. bispinosa* successfully acquired *Tomato spotted wilt virus*, although at a lower percentage than *F. occidentalis*. Viruliferous adults of both species transmitted the virus to pepper. Our results confirm the competence of *F. bispinosa* as a vector of *Tomato spotted wilt virus*.

Key Words: *Frankliniella occidentalis*, Tospovirus, vector competence, viral acquisition, viral transmission, *Capsicum annuum*

RESUMEN

El trips, *Frankliniella occidentales*, es un vector clave y responsable para la emergencia del virus de la marchitez manchada del tomate como una amenaza global para la agricultura. Un otro especie de trips común en Florida, Bahamas y Bermuda es *Frankliniella bispinosa*, pero su papel en la epidemiología del virus de la marchitez manchada del tomate no es conocido. El propósito de este estudio fue para evaluar la habilidad de *F. bispinosa* para adquirir y transmitir el virus de la marchitez manchada del tomate al chile. Las larvas de *F. bispinosa* adquirieron con buen éxito el virus de la marchitez manchada del tomate, aunque a un porcentaje menor que en *F. occidentalis*. Adultos viruliferos de las dos especies transmitieron el virus a chile. En experimentos del laboratorio, el número de larvas producidas por hembra de *F. bispinosa* fue menor que el número de larvas producidas por la hembra de *F. occidentalis.* Nuestros resultados confirman la capacidad de *F. bispinosa* como un vector del virus de la marchitez manchada del tomate.

In addition to damaging plant tissues while feeding, some species of thrips vector *Tomato spotted wilt virus* (TSWV), a tospovirus transmitted through the saliva of thrips during feeding (Hunter & Ullman 1992; Ullman et al. 1997). Tomato spotted wilt was first observed in 1915 in Australia and described as the "spotted wilt" of tomatoes (Brittlebank 1919) that later was associated with transmission by thrips (Pittman 1927). The viral etiology was reported by Samuel et al. (1930).

TSWV causes economic loss in many agricultural crops. The virus has a broad host range, infecting over 1000 plant species, and causing an estimated crop loss of one billion US dollars per year throughout its host range (Prins & Goldbach 1998). Tomato, tobacco, lettuce, pepper, papaya, eggplant, green beans, artichokes, broad beans, celery, some ornamental plants, and other plants experience severe losses due to the virus (Rosella et al. 1996).

Ullman et al. (1997) reviewed the relevant scientific literature involving the relationship between TSWV and its thrips vectors. Only first and second instars of vector thrips species acquire TSWV during feeding upon an infected host, and the virus survives molting, pupation, and the replacement of tissues during the prepupal and pupal stages of thrips development. Adults are unable to acquire TSWV. Infected adults are responsible for transmission and spread.

Outbreaks of tomato spotted wilt are difficult to manage. Growing seedlings under cover, avoiding sequential plantings, removing acquisition hosts for the larvae, rotating with non-susceptible crops, and use of UV-reflective mulch are sometimes useful as management tactics (Cho et al. 1998; Kucharek 1990; Momol et al. 2004; Rosella et al. 1996). Tomato hybrids were developed with a single-gene dominant resistance trait, but this resistance was overcome by strains of the virus

(Rosella et al. 1996). Attempts to regulate vector populations with insecticides have not been successful, and populations of thrips developed resistance to broad-spectrum insecticides (Brodsgaard 1994; Immaraju et al. 1992). Further, primary spread of TSWV is not prevented by insecticides because insecticide-exposed viruliferous adults successfully transmitted the virus before death (Momol et al. 2004).

Thrips species known to transmit TSWV are *Thrips tabaci* (Lindeman), *Thrips setosus* (Moulton), *Frankliniella occidentalis* (Pergande), *Frankliniella schultzei* (Trybom), *Frankliniella fusca* (Hinds), and *Frankliniella intonsa* (Trybom) (Sherwood et al. 2001). *Frankliniella occidentalis* is the primary vector of TSWV due to its increasingly global distribution (Wijkamp et al. 1995). *Frankliniella bispinosa* (Morgan), which is distributed in parts of the southeastern US, Bermuda, and the Bahamas (Nakahara 1997), has been suspected, but not proven, as a vector of TSWV (Tsai et al. 1996; Webb et al. 1998).

The plants on which adult thrips can be collected have been cited in the literature as host plants (Mound and Teulon 1995), but adults frequently inhabit flowers that are not reproductive hosts. Adults of *F. bispinosa* are abundant in the flowers of bell pepper, *Capsicum annuum* L., in Florida along with adults of other species, including *F. occidentalis* (Funderburk et al. 2000; Hansen et al. 2003; Reitz et al. 2003). The suitability of pepper as a reproductive host of *F. bispinosa* has not been determined, and the possible role of *F. bispinosa* in TSWV epidemics is unknown. The purpose of our research was to determine the competence of *F. bispinosa* as a vector of TSWV in pepper. An experiment was conducted to determine the ability of *F. bispinosa* to reproduce and acquire the virus on pepper compared to the key vector, *F. occidentalis*. Another experiment was conducted to verify that *F. bispinosa* adults are able to transmit the TSWV to uninfected pepper.

Materials and Methods

Pepper Establishment and Maintenance

Individual 'Camelot X3R' bell peppers were transplanted into a 16 x 16 cm pot containing soil mixture (Fafard 3B Mix, Agawam, MA) and about 100 were maintained under greenhouse conditions. Plants were fertilized with Peat-Lite special 15-16-17 fertilizer (Scotts-Sierra Horticultural Products Company, Marysville, OH) and Miracle-Gro Bloom Booster 10-52-10 fertilizer (Miracle-Gro, Marysville, OH). Virus acquisition and transmission experiments were conducted in growth rooms at 23 to 25°C under a photoperiod of 14 h light, 10 h darkness.

Virus Acquisition Experiment

Plants for use in TSWV acquisition trials were mechanically inoculated with TSWV isolates collected from naturally infected pepper plants at the North Florida Research and Education Center. Using a mortar and pestle, we homogenized TSWV infected leaf tissue in 5% sodium sulfite solution containing diatomaceous earth in order to prepare an inoculum. Cheesecloth was used to apply inoculum to 3 or 4 leaves per experimental plant. Seven to 10 days after mechanical inoculum, experimental plants were tested for TSWV infection by a commercially available double antibody sandwich enzyme-linked immunosorbent assay (ELISA) kit (Agdia, Elkhart, IN). Glass tubes and polystyrene balls (Precision Plastic Ball Co., Chicago, IL) were used in place of a microplate. Samples were scored for the presence of a colorimetric reaction indicating TSWV infection.

Individual TSWV infected pepper plants between 8 and 10 weeks of age were covered by a polyethylene cylindrical cage (35 cm × 15 cm) (n = 23 and 25 for *F. bispinosa* and *F. occidentalis*, respectively). The opening at the top was covered with fine mesh to prevent thrips escape, and there were two side openings (2.5 cm × 2.5 cm) covered with mesh. Ten females of *F. occidentalis* or *F. bispinosa* were introduced into the cage containing one infected plant. The adults were removed after 6 d. The plants were visually inspected for larvae at 6, 8, and 10 d after initial infestation with adult thrips. Each larva when found was transferred to green bean pods. After developing into adult, each was tested with an indirect ELISA to detect for the presence of the NSs protein encoded by TSWV RNA (Bandla et al. 1994). The NSs protein is present in thrips cells as a result of TSWV replication, demonstrating that the thrips is a host for TSWV.

The mean numbers of *F. occidentalis* larvae and *F. bispinosa* larvae recovered per cage after 10 days were compared with a two sample t-test [PROC TTEST in SAS System Software (SAS Institute 1999)]. The percent virus acquisition of *F. occidentalis* and *F. bispinosa* larvae also was compared by a two-sample t-test.

Virus Transmission Experiment

Transmission trials were conducted to confirm that the adults of *F. bispinosa* and *F. occidentalis* adults transmit TSWV to pepper. Cohorts of about 50 *F. bispinosa* and *F. occidentalis* larvae were allowed to feed on infected tomato fruit until developing into pupae. After developing into an adult, 5 to 10 of each species from each cohort were tested to verify TSWV acquisition with the indirect ELISA method described previously (Bandla et al. 1994). Twenty putatively viruliferous *F. occidentalis* or *F. bispinosa* adults from co-

horts that tested positive were introduced into a polyethylene cage (55-cm-long ×x 30-cm-wide × 48-cm-high) containing 4 healthy pepper plants of 8 weeks old. There were 19 and 8 cages established for *F. occidentalis* and *F. bispinosa*, respectively. The peppers were tested after 21 days for TSWV by ELISA as described above. Transmission by *F. occidentalis* and *F. bispinosa* was compared by a chi-square test.

Results

Pepper was a suitable reproductive host for *F. occidentalis* and *F. bispinosa* in our study. The mean total number (+ SEM) of larvae recovered over 10 d when introducing 10 *F. occidentalis* or *F. bispinosa* adult females on individually caged pepper plants infected with TSWV was 47.7 (±7.2) and 15.3 (±2.5), respectively. The difference was significant (t = -4.08, df = 58, $P < 0.0001$).

A higher percentage of *F. occidentalis* acquired the virus versus *F. bispinosa* (t = -2.07, df = 53, $P < 0.05$).The mean percent acquisition (±SEM) of TSWV by *F. occidentalis* and *F. bispinosa* larvae feeding on infected pepper plants, as determined by an indirect ELISA to detect for the presence of the NSs protein encoded by the virus RNA, was 21.9 (±3.1) and 14.6 (±2.9), respectively.

The adults of *F. bispinosa* and *F. occidentalis* successfully transmitted TSWV to pepper. In the virus transmission experiments, pepper plants exposed to TSWV-infected adults of *F. bispinosa* were ELISA positive in 4 out of 8 replicates. Pepper plants exposed to viruliferous adults of *F. occidentalis* were ELISA positive in 6 out of 19 replicates. The difference in transmission between the two species was not significant (χ^2 = 0.8; df = 3).

Discussion

The results from the acquisition experiment indicate that under laboratory conditions *F. occidentalis* is more likely to acquire the virus, and thus may be a more effective vector in pepper than *F. bispinosa*. The number of larvae of *F. occidentalis* produced per female was 3.1-fold greater, indicating that *F. occidentalis* has a greater intrinsic capacity than *F. bispinosa* to increase on pepper. The greater the intrinsic capacity of increase of a vector species on a host plant the greater the potential for acquisition and spread of TSWV (Peters et al. 1996). Differences in feeding preferences and host suitability between *F. occidentalis* and *F. bispinosa* may result in varying abilities of each species to acquire TSWV depending on the host plant. Webb et al. (1998) observed in laboratory studies higher rates of acquisition of TSWV for *F. bispinosa* than for *F. occidentalis* when the larvae fed on *Datura stramonium* L.

Van de Wetering et al. (1999) analyzed 14 populations of *F. occidentalis*. Each population acquired and transmitted TSWV, but there were marked differences in their efficiency, expressed as the percentage of transmitting adults. Laboratory experiments also do not account for ecological factors that influence thrips populations under field conditions, such as parasitism or predation that may reduce thrips populations, nor does it account for differences in mobility and other behaviors between the two thrips species that might affect the spread of TSWV in field peppers. Ramachandran et al. (2001) showed that the adults of *F. bispinosa* moved more rapidly in field peppers than the adults of *F. occidentalis*. This behavior allowed the adults of *F. bispinosa* to more frequently escape predation of *Orius insidiosus*.

The abundance of *F. bispinosa* and *F. occidentalis* in certain geographical regions also should be considered when assessing these species as a potential threat as a vector of TSWV in field pepper. Hansen et al. (2003) found *F. bispinosa* in much greater abundance than *F. occidentalis* in central Florida, while both species were abundant in northern Florida.

In this study, we have shown that under laboratory conditions *F. bispinosa* is a competent vector of TSWV in pepper. The species reproduced and acquired TSWV from infected pepper plants. Viruliferous adults of *F. bispinosa* transmitted TSWV to pepper. Reproduction of *F. occidentalis* on pepper and virus acquisition by the larvae feeding on pepper was greater than that by *F. bispinosa*; however, species-specific attributes may play a role in the ability of both vectors to vector TSWV in field conditions. The adults of *F. bispinosa* are more mobile within pepper fields than the adults of *F. occidentalis* (Ramachandran et al. 2001). TSWV epidemics occur on field pepper in our region (Gataitis et al. 1998), but the role of each species in disease epidemiology under field conditions is not understood.

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References Cited

Bandla, M. D., L. R. Campbell, D. E. Ullman, and J. L. Sherwood. 1994. Interaction of tomato spotted wilt virus tospovirus (TSWV) glycoproteins with a thrips midgut protein, a potential cellular receptor for TSWV. Phytopathology 88: 98-104.

Brittlebank, C. C. 1919. Tomato diseases. J. Dept. Agric., Victoria 7: 231-235.

Brodsgaard, H. F. 1994. Insecticide resistance in European and African strains of western flower thrips (Thysanoptera: Thripidae) tested in a new residue-on-glass test. J. Econ. Entomol. 87: 1141-1146.

CHO, J. J., R. F. L. MAU, S.-Z. PANG, M. WANG, D. GONSALVES, J. WATTERSON, D., M. CUSTER, AND D. GONSALVES. 1998. Approaches for controlling tomato spotted wilt virus, pp. 547-564 *In* A. Hadidi, R. K. Khetarpal, and H. Koganezawa [eds.], Plant Virus Disease Control. APS Press, St. Paul, MN. 704 pp.

FUNDERBURK, J., J. STAVISKY, AND S. OLSON. 2000. Predation of *Frankliniella occidentalis* (Thysanoptera: Thripidae) in field peppers by *Orius insidiosus* (Hemiptera: Anthocoridae). Environ. Entomol. 29: 376-382.

GATAITIS, R. D., C. C. DOWLER, AND R. B. CHALFANT. 1998. Epidemiology of tomato spotted wilt in southern Georgia. Plant Dis. 82: 752-756.

HANSEN, E. A., J. E. FUNDERBURK, S. R. REITZ, S. RAMACHANDRAN, J. E. EGER, AND H. MCAUSLANE. 2003. Within-plant distribution of *Frankliniella* species (Thysanoptera: Thripidae) and *Orius insidiosus* (Heteroptera: Anthocoridae) in field pepper. Environ. Entomol. 32: 1035-1044.

HUNTER, W. B., AND D. E. ULLMAN. 1992. Anatomy and ultrastructure of the piercing-sucking mouthparts and paraglossal sensilla of *Frankliniella occidentalis* (Pergande) (Thysanoptera: Thripidae). Intern. J. Insect Morphol. Embryol. 21: 17-35.

IMMARAJU, J. A., T. D. PAINE, J. A. BETHKE, K. L. ROBB, AND J. P. NEWMAN. 1992. Western flower thrips (Thysanoptera: Thripidae) resistance to insecticides in coastal California greenhouses. J. Econ. Entomol. 85: 9-14.

KUCHAREK, T., L. BROWN, F. JOHNSON, AND J. FUNDERBURK. 1990. Tomato Spotted Wilt Virus of Agronomic, Vegetable, and Ornamental Crops. Circ-914. Fla. Coop. Ext. Serv., University of Florida, Gainesville.

MOMOL, M. T., S. M. OLSON, J. E. FUNDERBURK, J. STAVISKY, AND J. J. MAROIS. 2004. Integrated management of tomato spotted wilt on field grown tomatoes. Plant Dis. 88: 882-890.

MOUND, L. A., AND D. A. J. TEULON. 1995. Thysanoptera as phytophagous opportunists, pp. 3-19 *In* B. L. Parker, M. Skinner, and T. Lewis [eds.], Thrips Biology and Management. Plenum Press, NY. 636 pp.

NAKAHARA, S. 1997. Annotated list of the *Frankliniella* species of the world (Thysanoptera: Thripidae). Contributions on Entomology, International 2: 355-389.

PETERS, D., W. F. VAN DE WETERING, AND R. GOLDBACH. 1996. Vector relations in the transmission and epidemiology of tospoviruses. Acta Hortic. 431: 29-43.

PITTMAN, H. A. 1927. Spotted wilt of tomatoes. Preliminary note concerning the transmission of the 'spotted wilt' of tomatoes by an insect vector (*Thrips tabaci* Lind.). Australian J. Council for Sci. Industrial Res. 1: 74-77.

PRINS, M., AND R. GOLDBACH. 1998. The emerging problem of tospovirus infection and nonconventional methods of control. Trends Microbiol. 6: 31-35.

RAMACHANDRAN, S., J. FUNDERBURK, J. STAVISKY, AND S. OLSON. 2001. Population abundance and movement of *Frankliniella* species and *Orius insidiosus* in field pepper. Agric. For. Entomol. 3: 129-137.

REITZ, S. R., E. L. YEARBY, J. E. FUNDERBURK, J. STAVISKY, M. T. MOMOL, AND S. M. OLSON. 2003. Integrated management tactics for *Frankliniella* thrips (Thysanoptera: Thripidae) in field-grown pepper. J. Econ. Entomol. 96: 1201-1214.

ROSELLA, S., M. JOSE DIEZ, AND F. NUEZ. 1996. Viral diseases causing the greatest economic losses to the tomato crop. I. The tomato spotted wilt virus- a review. Scientia Hort. 67: 117-150.

SAMUEL, G., J. G. BALD, AND H. A. PITTMAN. 1930. Investigations on 'spotted wilt' of tomatoes in Australia. Commonwealth of Australia, Counc. Sci. Ind. Res. Bull. 44: 8-11.

SAS INSTITUTE, INC. 1999. SAS/STAT User's Guide, Version 8. SAS Institute, Cary, NC.

SHERWOOD, J. L., T. L. GERMAN, A. E. WHITFIELD, J. W. MOYER, AND D. E. ULLMAN. 2001. Tospoviruses, pp. 1034-1040 *In* O. C. Maloy and T. D. Murray [eds.], Encyclopedia of Plant Pathology. John Wiley and Sons, New York. 1346 pp.

TSAI, J. H., B. S. YUE, J. E. FUNDERBURK, AND S. E. WEBB. 1996. Effect of plant pollen on growth and reproduction of *Frankliniella bispinosa*. Acta Hortic. 431: 535-541.

ULLMAN, D. E., J. L. SHERWOOD, AND T. L. GERMAN. 1997. Thrips as vectors of plant pathogens, pp. 439-565 *In* T. Lewis [ed.], Thrips as Crop Pests. CAB, Wallingford, Oxon, UK. 740 pp.

VAN DE WETERING, F., M. VAN DER HOEK, R. GOLDBACH, C. MOLLEMA, AND D. PETERS. 1999. Variation in tospovirus transmission between populations of *Frankliniella occidentalis* (Thysanoptera: Thripidae). Bull. Entomol. Res. 89: 579-588.

WEBB, S., J. TSAI, AND F. MITCHELL. 1998. Bionomics of *Frankliniella bispinosa* and its transmission of tomato spotted wilt virus. Abstract: Fourth International Symposium on tospoviruses and thrips in floral and vegetable crops. Wageningen, The Netherlands, p. 67.

WIJKAMP, I., N. ALMAZARA, R. GOLDBACH, AND D. PETERS. 1995. Distinct levels of specificity in thrips transmission of *tospoviruses*. Phytopathology 85: 1069-1074.