# Survival and Reproductive Success of Tobacco Thrips on Three Tomato Spotted Wilt Virus Infected and Noninfected Peanut Cultivars

L. E. Garcia<sup>1\*</sup>, G. G. Kennedy<sup>2</sup>, and R. L. Brandenburg<sup>2</sup>

#### ABSTRACT

A comparison of the survival and reproductive success of *Frankliniella fusca* (Hinds) (Thysanoptera: Thripidae) on tomato spotted wilt virus (Bunyviridae: Tospovirus) (TSWV) infected and uninfected peanut plants was conducted under greenhouse conditions in North Carolina. Three cultivars-NC 9, NC-V11, and NC 12Cadapted to North Carolina production practices were evaluated. A total of 180 individually caged plants, in three replicates, were infested with 20 female F. fusca each. Adult and larval thrips were collected after 30 d on the plants. Final counts were square root transformed and a mixed model analysis of variance conducted. Effects of cultivar and the virus-by-cultivar interaction were not statistically significant. TSWV-infected plants had significantly fewer adult and larval F. fusca than did uninfected plants for adults (P = 0.04) and for larvae (P= 0.01). This study reports on an alternative method of assessing TSWV resistance among peanut cultivars and the trend appears to support the conclusions of a previous field study, which found NC 9 more susceptible to TSWV than either NC-V11 or NC 12C.

Key Words: Frankliniella fusca, TSWV.

Tomato spotted wilt virus (TSWV), a Tospovirus, was first recognized as a significant plant pathogen in Australia in 1916 (Britlebank, 1919). It has since become an important disease of many ornamental and food crops worldwide (Peters et al., 1996). This virus is unusual among plant viruses in that it is a member of a family of viruses, Bunyviridae, that predominantly infect animals. The virus may have adapted independently to thrips species after first originating as a vertebrate pathogen (Mound, 1996). Currently, there are seven confirmed thrips Tospovirus vectors (Mound, 1996), and eight serologically distinct plus four putative species of Tospoviruses (Prins and Goldbach, 1998). In North Carolina, there are three thrips vectors of TSWV including Frankliniella fusca (Hinds), F. occidentalis (Pegrande), and Thrips tabaci Lindeman. Of these, F. fusca, the tobacco thrips, is the most common species associated with peanuts (Barbour and Brandenburg, 1994). Frankliniella occidentalis and T. tabaci are only occasionally found in North Carolina peanut fields.

The association of TSWV with its vector, *F. fusca*, and

the epidemiology of the disease in North Carolina peanuts are not completely understood. It has been recognized, however, that tobacco thrips overwinter in North Carolina (Cho *et al.*, 1995) and that TSWV can be found overwintering in infected tobacco thrips (Garcia *et al.*, 2000a) and in infected winter annual and perennial weed species (R. Groves and G. G. Kennedy, unpub.). Tobacco thrips emerge in the spring from weed hosts and/ or overwintering sites (Barbour and Brandenburg, 1994; Garcia *et al.*, 2000) and migrate to seedling peanut plants as they emerge from the soil surface. Spring-migrating thrips, principally from nearby locations (Garcia *et al.*, 2000a), vector the virus into peanut fields and are the source of primary infection.

Secondary spread of TSWV in peanut fields is not as significant as primary spread and develops principally as a result of intra-field movement of viruliferous tobacco thrips from plant to plant (Camann *et al.*, 1995). For secondary infection to occur, TSWV-infected peanut plants must be able to support the development of immature thrips. Therefore, the extent to which secondary infection occurs is partially dependent upon the attractiveness and suitability of TSWV-infected peanut plants as a host for thrips. Because TSWV is only acquired by immature thrips and transmitted following a 3- to 7-d latent period (Peters *et al.*, 1996) by adults in a persistent fashion (Sakimura, 1962, 1963), it is essential to the spread of this pathogen that the plant virus host also be a suitable thrips host.

The attractiveness and suitability of virus-infected plants and uninfected plants as hosts for thrips varies (Terry, 1997). There is some evidence indicating that F. occidentalis on lettuce and Thrips palmi on cucumber are more attracted to virus-infected plants (Yudin et al., 1987; Culliney, 1990; Terry, 1997). Bautista et al. (1995), also working with F. occidentalis, demonstrated feeding and oviposition preferences for various TSWV-infected hosts over noninfected hosts. However, there is disagreement in the literature concerning the suitability of virus-infected host plants for survival and development of F. occidentalis (Robb, 1989; Wijkamp, 1995). The suitability of TSWV-infected North Carolina peanut cultivars as hosts for *F*. *fusca* larvae and adults as compared to noninfected plants was investigated in this greenhouse study.

## Materials and Methods

**Experimental Design**. The relative suitability of TSWVinfected and noninfected peanut cultivars as hosts for adult *F. fusca* survival and larval development was compared on three commercially available peanut cultivars—NC 9, NC-V11, and NC 12C. The experiment was conducted under a regime of 14:10 h (light/dark) in a greenhouse on the campus of North Carolina State Univ. in Raleigh during the summer of 1998. Temperatures in the greenhouse over the

<sup>&</sup>lt;sup>1</sup>North Carolina Dept. of Agric. and Consumer Serv., Raleigh, NC 27611.

<sup>&</sup>lt;sup>2</sup>Dept. of Entomology, North Carolina State Univ., Raleigh, NC 27695. \*Corresponding author (email: lloyd.garcia@ncmail.net).

study period fluctuated from an average daily low of 22.8 C to an average daily high of 39.7 C.

Forty seeds from each cultivar, at a rate of two seeds/pot, were sown on each of three treatment periods (21 April, 14 May, and 15 May 1998). Sterile growing media was used (Scott's® Metro Mix® 220 Growing Medium, Scotts-Sierra, Marysville, MD). Plants in each pot were covered with a cylindrical thrips-proof cage constructed of clear plastic sheeting, "Vivak" (AIN Plastics of North Carolina, Greensboro, NC) which was extended into the potting media along the edge of the pot to a depth of 10.2 cm. The seams of the cylindrical cages were sealed with a silicone caulking compound. A thrips-proof screen (BugBed123, Greenthumb Group, Downers Grove, IL) covered the top opening of the cage and also was sealed with silicone caulk. Cages were approximately 25.40 cm in diameter and 41.6 cm tall.

TSWV Inoculation. At 9 d post-planting, the peanut seedlings were transferred to a dark environment at approximately 22.2 C. On day 10 post-planting, half of the peanut plants from each cultivar (20 plants per cultivar and two plants per pot were mechanically inoculated with TSWV. Two leaves on each plant were inoculated with extract from infected leaves ground in inoculation buffer (0.01 M Tris, 0.01 M Na<sub>2</sub>S<sub>3</sub>, and 0.1% Cystine HCL, pH 7.8). The inoculum, designated as the GT isolate of TSWV, originated in tomatoes from Georgia (GA) and was obtained from Dr. J. Moyer, North Carolina State Univ., Dept. of Plant Pathology. This isolate was thrips-transmitted to Emilia sonchifolia (L.) and mechanically transferred to Nicotiana benthamiana Domin from which it was extracted to inoculate the peanut plants. The plants were returned to the greenhouse 24 hr after inoculation. Twelve days later, as foliar symptoms became visible, the plants were thinned to one plant per pot to minimize variation in plant size. Foliage samples were taken from the plants and tested by enzyme-linked immunoabsorbent assay (ELISA) (Agdia, Elkhart, IN). All mechanically inoculated plants used in the experiments tested positive for the presence of TSWV. A plant was considered positive if its optical density (OD) value was greater than the mean plus three times the standard deviation of the OD value of five predesignated healthy control wells on the same microtiter plate.

**Thrips Infestations**. Twenty adult female *F. fusca* were added to each caged plant 13 d after inoculation, or 23 d post-planting. Thrips of uniform age were selected from cultures where thrips were allowed to feed and oviposit on green bean pods, *Phaseolus* spp., for 2 d before the bean pods were replaced. These bean pods were used to initiate colonies of similar aged thrips. Thrips were aspirated into screened Pasteur pipettes and held for transfer to the caged potted peanut plants. The Pasteur pipettes were stuck into the potting media and the screen plug removed to allow the thrips to escape into the individual cages.

**Data Collection and Analysis**. Thirty days after the thrips were added to the caged peanut plants, the plants were cut off at soil level with scissors and quickly transferred to plastic buckets,  $16 \times 20$  cm (ht × diam) with tight fitting lids and screened tops and bottoms. A single green bean pod (*Phaseolus* spp.) was added to each bucket as an alternative food source for the thrips. Cut plants were placed in an air-conditioned room with fans for 3 d of drying time in the buckets before the thrips were collected, using an aspirator, for counting. All adult thrips from each caged plant were counted and larvae from the last two replications

were counted. Thrips counts were recorded according to cultivar as adults or larvae, presence or absence of virus, and replication. Data were analyzed using SAS 6.12 General Linear Models Procedure and tests of hypotheses for Mixed Model ANOVA with square-root transformed data. Because some of the plant samples contained no thrips, data were analyzed with and without the zero count data.

### Results

**TSWV Inoculations**. All mechanically inoculated plants exhibited symptoms of systemic infection, such as concentric ring spots with purple discoloration and twisted petioles. Noninfected plants showed no TSWV symptoms. The mean optical density reading of the mechanically inoculated plants was 1.05 nm compared to a mean optical density reading of 0.023 nm for the noninoculated plants.

Thrips Counts. Thrips in approximately 30% of the cages either failed to establish or to survive the 30-d evaluation period. Failure to establish did not differ significantly among cultivars (mean NC 9 = 73.5%; mean NC-V11 = 73%; mean NC 12C = 68.5%) or between infected and uninfected caged plants (mean infected = 70%; mean uninfected = 73%). Cultivar main effect did not have a significant effect on the number of thrips adults or larvae that developed during the study (Table 1). A significant main effect was observed in both adult and larval thrips counts between virus-infected and uninfected plants (Table 2). Plants which were not mechanically inoculated with TSWV tended to have more adults and larval thrips at the end of the 30-d period than the TSWV-inoculated plants. The difference was not statistically significant for adults (P = 0.10) but was for immatures (P = 0.05). Reanalysis of the data with the zero counts removed resulted in more pronounced differences between virus-infected and noninfected plants (adult P = 0.04 and larvae P = 0.01).

Although the cultivar-by-virus interaction was not statistically significant for either adult or larval counts, the larval thrips population on infected NC 9 and NC-V11 tended to be lower than on uninfected plants. For NC 9, the mean counts on infected and uninfected plants

Table 1. Mean F. fusca adult and larvae counts and SEM across TSWV-infected and uninfected caged plants by cultivar and by plants with and without zero counts, 1998.<sup>4</sup>

		Adı	ılt co	ount	Larval count					
		All	]	nfested		All	Iı	nfested		
	ł	plants⁵		plants <sup>c</sup>		plants	plants			
Cultivar	n	Count	n	Count	n	Count	n	Count		
NC 9	60	<b>31.6±5</b> .3	44	43.1±6.5	40	17.3±3.5	29	23.9±4.2		
NC-V11	60	$32.9\pm5.7$	44	$44.8 \pm 7.0$	40	$22.7 \pm 5.2$	29	$31.3\pm6.5$		
NC 12C	60	25.7±4.0	41	37.7±4.8	40	$16.4 \pm 3.2$	27	24.2±3.9		

<sup>a</sup>Means of three cultivars with three replicates, each with 10 TSWV-infected and 10 uninfected plants.

<sup>b</sup>Means of tobacco thrips counts by cultivars were not significantly different at P = 0.05.

<sup>c</sup>Infested plants indicate only those plants where thrips were present at the conclusion of the study.

Table 2. Mean *F. fusca* adult and larvae counts and SEM from TSWV-infected and uninfected caged plants including and excluding caged plants with zero counts, 1998.<sup>\*</sup>

		Adult	: cou	int	Larval count				
	All Infested					All		Infested	
Culti-		plants <sup>b</sup>		plants <sup>c</sup>		plants	plants		
var	n	Count	n	Count	n	Count	n	Count	
Pos	90	$23.7 \pm 3.5*$	63	33.9±4.4**	60	$10.6 \pm 2.4 **$	40	15.9±3.2**	
Neg	90	36.4±4.6*	66	$49.6 \pm 5.6 * *$	60	27.0±3.7**	45	36.0±4.2**	

"Means of three replications representing 10 TSWV-infected and 10 uninfected plants in three peanut cultivars.

<sup>b</sup>Counts are significant at \*P = 0.10 and \*\*P = 0.05.

<sup>c</sup>Infested plants indicate only those plants where thrips were present at the conclusion of the study.

was 16.5 and 29.8, respectively, and for NC-V11 plants the mean counts were 7.8 on infected and 53.2 on uninfected. On NC 12C plants, the larval populations were similar on both infected and uninfected plants, 23.9 and 24.5, respectively.

#### Discussion

In this study, the results indicate that survival and reproductive success of *F. fusca* is lower on TSWVinfected peanut plants than on uninfected plants of the three tested cultivars. The cause of the decrease in survival rate is unknown, but may be attributable to a reduction in the nutritional quality of the virus-infected plants as suggested by DeAngelis *et al.* (1993) for *F. occidentalis* on impatiens necrotic spot wilt virus-infected plants of *Lobelia erinus* L. Alternatively, it could reflect an adverse effect of the virus directly on the thrips. The results also indicate that the negative effect on thrips survival caused by TSWV-infected plants may help to limit secondary spread in peanut fields by reducing the number of infective thrips.

A preliminary field study in 1995 (Garcia, 1999) found higher thrips counts but a lower TSWV incidence on NC-V11 than on NC 9. A subsequent field study, in which TSWV infection ranged from 5-7% (Garcia et al., 2000), produced similar findings. In the present investigation conducted on potted plants growing in a greenhouse, there were no significant differences among cultivars in the number of adult and larval thrips, but populations of both immatures and adult thrips were lower on TSWVinfected than on uninfected plants. This study suggests that uninfected NC-V11 may be a better host than NC 9 and that infection with TSWV may reduce the host suitability of both NC 9 and NC-V11, with the reduction in suitability being greater in NC-V11 than in NC 9 (Table 3). R. Groves (pers. commun.) observed a similar effect of TSWV infection on F. fusca populations in tobacco. A greater survival rate of tobacco thrips on infected NC 9 plants may increase the likelihood of a higher incidence of secondary infection among NC 9 plants than among NC-V11 plants.

Adult and larval counts for uninfected NC 12C plants tended to be lower than for the other two cultivars. However, the highest larval counts among the infected plants were on TSWV-infected NC 12C plants (Table 3). This suggests that, when infected, NC 12C may be a comparatively good host for tobacco thrips, and that uninfected NC 12C plants may not be as suitable as a host for F. fusca as uninfected NC 9 and NC-V11. However, further experiments are required to verify this conclusion. Uninfected NC 12C, being a poor thrips host, could result in low rates of infection early in the season as compared to NC 9 and NC-V11 plants, and a field study (Garcia et al., 2000) with these three cultivars confirmed this conclusion. From a management and production perspective, growing cultivars which are poor thrips hosts would help lessen the damage caused by TSWV because infection would occur later in the season when disease has less impact on peanut yield than when TSWV occurs early in the season.

Additional greenhouse and field experiments should be conducted with additional peanut cultivars to investigate the survival and reproductive success of tobacco thrips on TSWV-infected and uninfected plants. This information will be useful in helping to identify TSWV-

Гable 3.	Mean F.	fusca i	adult and	larvae	counts	and SEM	1 from	caged	plants	by y	virus	and c	ultivar	, 1998.'	H

Virus		Adult count					Larval count				
	Cultivar	Al	l plants <sup>b</sup>	Infe	sted plants <sup>c</sup>	All plants		Infested plants			
		n	Count	n	Count	n	Count	n	Count		
Pos	NC 9	30	$26.5 \pm 7.1$	20	39.8±9.3	20	$10.8 \pm 4.4$	13	16.5±6.2		
Pos	NC-V11	30	$22.8 \pm 5.4$	22	$31.1 \pm 6.6$	20	$5.5 \pm 2.0$	14	$7.8 \pm 2.7$		
Pos	NC 12C	30	$21.9 \pm 5.5$	21	$31.3\pm7.0$	20	$15.6 \pm 5.1$	13	23.9±6.8		
Neg	NC 9	30	36.7±8.0	24	$45.8 \pm 9.1$	20	23.9±5.0	16	29.8±5.3		
Neg	NC-V11	30	$42.9 \pm 9.8$	22	$58.5 \pm 11.7$	20	$39.9 \pm 8.6$	15	53.2±9.2		
Neg	NC 12C	30	$29.6 \pm 5.7$	20	44.4±6.3	20	$17.2 \pm 4.0$	14	$24.5 \pm 4.4$		

<sup>a</sup>Means of three replications of three cultivars, each with 10 TSWV-infected plants and 10 uninfected plants.

<sup>b</sup>Means of adult and larval counts by TSWV-infected or uninfected and cultivar were not significantly different at P = 0.05.

<sup>c</sup>Infested plants indicate only those plants where thrips were present at the conclusion of the study.

resistant cultivars and in designing new TSWV management techniques.

## Acknowledgments

We thank R. Groves for technical assistance, the use of thrips cages, and the supply of thrips adults; C. Berger, S. Costa, and V. Covington for advice and technical help; T. Isleib for the peanut seed; and J. Moyer for TSWV inoculum (North Carolina State Univ.). C. Browning provided statistical analysis. This research was made possible and supported in part by the North Carolina Dept. of Agric. and Consumer Serv. Partial funding for this project was provided by the North Carolina Peanut Grower's Assoc., Inc. and USAID Peanut CRSP (Grant No. DPN-4048-G-00-0041-00).

## Literature Cited

- Barbour, J.D., and R.L. Brandenburg. 1994. Vernal infusion of thrips into North Carolina peanut fields. J. Econ. Entomol. 87: 446-451.
- Bautista, R.C., R.F.L. Mau, J.J. Cho, and D.M. Custer. 1995. Potential of tomato spotted wilt tospovirus plant hosts in Hawaii as virus reservoirs for transmission by Frankliniella occidentalis

- (Thysanoptera: Thripidae). Phytopathology 85:953-958.
  Brittlebank, C.C. 1919. Tomato diseases. J. Agric., Victoria 27:231.
  Camann, M.A., A.K. Culbreath, J. Pickering, J.W. Todd, and J.W. Demski. 1995. Spatial and temporal patterns of spotted wilt epidemics in peanut. Phytopathology 85:879-885. Cho, K., C.S. Eckel, J.F. Walgenbach, and G.G. Kennedy. 1995.
- Overwintering of thrips (Thysanoptera: Thripidae) in North Carolina. Environ. Entomol. 24:58-67.
- Culliney, T.W. 1990. Population performance of Thrips palmi (Thysanoptera: Thripidae) on cucumber infected with a mosaic virus. Proc. Hawaiian Entomol. Soc. 30:85-90.

- DeAngelis, J.D., D.M. Sether, and P.A. Rossignol. 1993. Survival, development, and reproduction in western flower thrips (Thysanoptera: Thripidae) exposed to impatiens necrotic spot virus. Environ. Entomol. 22:1308-1312.
- Garcia, L.E. 1999. Epidemiology of tomato spotted wilt virus. Ph.D. Diss., Univ. of North Carolina, Raleigh.
- Garcia, L.E., R.L. Brandenburg, and J.E. Bailey. 2000. Incidence of tomato spotted wilt virus (Bunyviridae) and tobacco thrips in virginia-type peanuts in North Carolina. Plant Dis. 84: 459-464.
- Mound, L.A. 1996. The Thysanoptera vector species of tospoviruses. Acta Hort. 431:298-309.
- Peters, D., F. Wijkamp, F. van de Wetering, and R. Goldbach. 1996. Vector relations in the transmission and epidemiology of tospoviruses. Acta Hort. 431:29-43.
- Prins, M., and R. Goldbach. 1998. The emerging problem of tospovirus infection and nonconventional methods of control. Trends in Microbiol. 6:31-35.
- Robb, K. 1989. Analysis of Frankliniella occidentalis (Pegrande) as a pest of floricultural crops in California greenhouses. Ph.D. Diss., Univ. of California, Riverside. Diss. Abst. Int. AAI9002637.
- Sakimura, S. 1962. Frankliniella occidentalis (Thysanoptera: Thirpidae), a vector of tomato spotted wilt virus, with special reference to color forms. Ann. Ent. Soc. Amer. 55:387-389.
- Sakimura, S. 1963. Frankliniella fusca, an additional vector for the tomato spotted wilt virus, with notes on Thrips tabaci, another vector. Phytopathology 53:421-415.
- SAS Institute. 1985. SAS User's Guide: Statistics. SAS Inst., Cary, NC.
- Terry, L.I. 1997. Host selection, communication and reproductive behavior, pp. 65-118. In T. Lewis (ed.) Thrips as Crop Pests. CAB International, New York.
- Wijkamp, I. 1995. Virus-vector interaction of tomato spotted wilt virus in Frankliniella occidentalis is neither accompanied by pathological effects nor by transovarial transmission. Ph.D. Diss., Wageningen Agric. Univ., The Netherlands.
- Yudin, L.S., W.C. Mitchell, and J.J. Cho. 1987. Color preference of thrips (Thysanoptera: Thripidae) with reference aphids (Homoptera: Aphididae) and leaf-miners in Hawaiian lettuce farms. J. Econ. Ent. 80:51-55.