Incidence of *Tomato spotted wilt virus* on tomato (*Lycopersicon esculentum*) varieties infected by natural field infection of thrips

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Abstract

***Tomato spotted wilt virus* (TSWV) is a major constraint to tomato production in Kenya. Symptoms of tomato spotted wilt differ among hosts and can be variable in a single host species. Common symptoms of TSWV infection include stunting, which is more severe when young plants are infected, and chlorotic or necrotic rings on the leaves and fruits. Management of the primary vectors of the virus disease, the western flower thrips (*Frankliniella occidentalis* Pergande) and tobacco thrips (*F.fusca* and *F.bispinosa*) is the main factor underlying management of the disease. A study was conducted with the objective to**

screen five tomato varieties (Cal J, Marglobe, Money Maker, Roma, and Riogrande) for resistance to TSWV by thrips-mediated infection in the field. Infection lowered yield and fruit quality. Variety Roma was the most tolerance under natural field infection. Metasystox insecticide significantly suppressed thrip populations under field condition.

Keywords: *Tomato spotted wilt virus, thrips infestation, tomato varieties.*

Introduction

Since 2001,Tomato spotted wilt virus (TSWV), has become a major limiting factor and most severe problem for many tomato growers in Kenya with yield lossesof up to 60% and 100% (Wangai et al., 2001). Tomato spotted wilt virusis a member of the genus *Tospovirus* in the family *Bunyaviridae*. The vectors have a polyphagous feeding behaviour and secretive habits of hiding within flowers and buds (Rapando *et al*., 2009). In addition, the virus and its thrips vector have a wide host range that includes both dicotyledonous and monocotyledonous which makes control of TSWV challenging (Coutts and Jones 2005; Ramkat *et al*., 2006). TSWV is persistently transmitted by at least eight species of adult thrips. First instar larvae acquire the virus by feeding on infected plants. In Kenya, the western flower thrip (*Frankliniella occidentalis* Pergande) is considered the most important TSWV vector (Wangai et al., 2001). Control of TSWV can be achieved by combining several methods (Boiteux,

1995) but the use of resistant cultivars and control of vectors are the most efficient approaches in integrated management programs (Wells et al., 2002).

Material and methods

Field experiments were conducted at Kenya Agricultural Research Institute Njoro, Kenya. Healthy tomato seedlings of varieties Cal J, Marglobe, Money Maker, Roma, and Riogrande were germinated and transplanted at a 4-leaf development stage. Field trials were carried out over two years from January – June 2004 and from November 2005 – April 2006 with test plants being exposed to thrip infestation with no insecticide treatment. The control treatments were sprayed with metasystox (oxyd emeton-methyl) insecticide at two week intervals. The experimental design was a split plot replicated three times in a randomized complete block design. The main plots treatments were those exposed to natural infestation and those protected with insecticide (control).

Visual examination for disease symptoms was used to determine infection. A scale of 0 to 5 was used to score the intensity of symptom expression, where 0 represent no symptom and 5 a dead test plant. A direct enzyme-linked immunosorbent assay (ELISA) was used to detect TSWV in plant samples (EPPO, 2002). In the year 2013 (November-December) and 2014 (January to April), a market survey was carried out in Nakuru to check for the presence of the diseased fruits.

Results and Discussion

The disease symptoms were observed two weeks after inoculation and included; leaf curl, leaf yellowing, chlorosis, necrotic lesions and stunted growth. In addition, market survey to check for the presence of TSWV on fruits between November to December 2013 and April 2014 showed disease symptoms an indicator that the disease is causing yield loss again. Symptoms observed on the fruits in the market included necrotic ring spots with uneven colouration. Presence of TSWV in the field was confirmed by ELISA in symptomatic plants. Based on disease scores there were significant differences among the treatments each year with the plants inoculated by natural infestation of thrips having the highest disease scores and the insecticide protected control having low disease scores (Table 1). There were significant variety differences in disease scores each year. Naturally infested plants of variety Cal J and Riogrande had the highest disease scores in 2004 and 2006 (Table 1). Variety Riogrande in the year 2004 had a significantly higher disease incidence on the plants inoculated by natural infestation compared to its protected controls while the disease level in other naturally infected varieties were not significantly different from protected controls. In the year 2006 naturally infected plants of variety Cal J had disease scores which were significantly higher than those of Money Maker and Roma. Variety Roma was found to have disease scores each year that were consistently significantly lower from other varieties (Table 1). These results are similar to those of Riley and Pappu (2000) and Pappu *et al.* (2000) where use of the insecticide Imidacloprid (Chloronicotinyl) and Actigard (Acibenzolar - S – Methyl) showed significantly low disease score between treated and untreated controls. Branch and Brenneman (2003) observed that disease scores in some cultivars of peanut that received insecticide control against TSWV, showed significant difference from untreated while other treated cultivars showed no difference from untreated cultivars.

Thrips were physically counted on field tomato plants after every three weeks starting two weeks after transplanting by tapping top, middle and bottom of each plant on a piece of white paper. The data obtained were used to compute the pest population expressed as an average number of thrips per plant. Metasystox insecticide was effective in reducing the thrips population. Thrip population varied with more thrips in 2006 than 2004. In both the years, plants naturally infested had thrips numbers which were significantly higher than protected controls (Fig 1 and 2). The control of the vectors and the disease by the use of insecticides has been reported by Castellane *et al.* (1995), Pappu *et al.* (2000) and Riley and Pappu

(2000) who observed significant reduction of thrip population and TSWV disease in tomatoes between insecticide treated and non-treated control plots. There was no significant correlation between number of thrips and disease scores in the year 2004, but the population of thrips was significantly positively correlated (r = 0.5) with disease scores in the year 2006. Thrip populations do not necessarily directly correlate with disease severity (Parisi *et al.,* 1998; Mcpherson *et al.,* 1999). Momol (2002) suggest that the adults persistently transmit TSWV and their control with insecticide does not prevent successful transmission due to the short time of feeding necessary for infection to occur. At the same time the species *Frankliniella occidentalis* that was common in this study is very effective in transmission. However in the year 2006 the significant correlation between disease and thrip population indicate that the high disease ratings resulted from high thrips population which caused both direct mechanical and indirect effect of spreading TSWV on plants, an association that has been observed by Jarosik *et al.* (1997) on cucumber plants following infestation by thrips. There were no significant differences among varieties in both 2004 and 2006 for thrips population. However varieties Roma, Riogrande and Cal J were leading in thrips infestations while Money Maker and Marglobe were lower each year. The high numbers of thrips on Roma, Riogrande, and Cal J could be due to their bushy canopy because they were not pruned while Money Maker and Marglobe were pruned. Saunyama and Knapp (2003) observed that pruning and trellising of tomatoes resulted in better mite management and disease incidence. These has also been reported by Agboh-Noameshie *et al*. (1997) who observed that changes in canopy structure by pruning creates a micro-environment that reduces the population of flower thrips more than in non-pruned plants of cassava. Variety Roma had high vegetative growth in spite of the higher thrips population.

Fruits were harvested, weighed and graded for marketability. Unmarketable fruits had TSWV symptoms including discoloration of the skin, sunken finger like depressions, and malformation. Marketable fruits were without disease symptoms. Naturally infested plants of all the five tomato varieties tested produced fruit weights that were similar from those of equivalent protected controls in both 2004 and 2006. However tomato variety had a significant effect on fruit weight in both 2004 and 2006 with protected controls of variety Money Maker having significantly higher yields than other varieties except Roma in 2004 while in 2006 they were different from those of Cal J only. These results differ from those of Castellane *et al.* (1995), Pappu *et al.* (2000) and Riley and Pappu (2000) who reported increased yields of tomatoes with the use of insecticide which differed significantly from un-protected controls because of reduced vector and disease symptoms. Use of insecticide caused non statistically different increase in marketable yield when compared with the natural infestation. Variety Money Maker had the highest marketable yields both in 2004 and 2006. Its yields differed from those of the other varieties in 2004 except Roma while in 2006 they only differed from those of Cal J (Table 1). Riley and Pappu (2000) demonstrated a significant reduction in TSWV using tolerant variety Stevens. However its benefits did not result in acceptable levels of marketable yield. Use of the insecticide, Imidacloprid (Chloronicotinyl) increased marketable yield but not significantly when compared to untreated controls. The use of Actigard (Acibenzolar - S – Methyl) which is a plant activator that activates plant defence, lowered disease symptoms leading to significantly higher marketable yields compared to untreated controls (Pappu et al., 2000). Costa *et al*. (1999) observed varietals differences towards total and marketable yields and attributed this to the combined effect of vegetative development of plants and the mechanism of tolerance that result in acceptable marketable yields even when thrip population cannot be totally controlled.

References

Agboh – Noameshie, A., Jackai, L.E.N., Agboola, A.A., Ezumah, H.C. 1997. Tropical Agriculture (Trinidad and Tabago) 74: 210-215.

Bendahmane M., Koo M., Karrer E., Beachy R. M., 1999. Display of epitopes on the surface of tobacco mosaic virus: impact of charge and iso electric point of the epitope on virus host. Journal of Molecular Biology 290:1-20.

Boiteux L. S., 1995. Allellic relationships between genes for resistance to tomato Spotted wilt tospovirus in *Capsicum chinese*. Theoretical and Applied Genetics 90:146-149.

Branch W. D., Brenneman T. B., 2003. Field resistance to cylindrocladium black rot and tomato spotted wilt virus among advanced runner type peanut breeding lines. Crop Protection 19: 349-354.

Castellane P. D., Avaujo J. A., Bortoli S. A., Gabarra P. A., De-Araujo J. A., De-Bortoli S. A., 1995. Influence of mulching and pest control on the yield and arthropods in tomato cv. Rio grande. Clentifica-jaboticabal 23: 343-354.

Costa J., Lacasa A., Catala M. S., 1999. Tomato production under mesh reduces crop loss to tomato spotted wilt virus in some cultivars. Hort Science 34 (4): 634-637.

Coutts B. A., Jones R. A. C., 2005. Suppressing spread of tomato spotted wilt virus by drenching infected source or healthy recipient plants with neonicotinoid insecticides to control thrips vectors. Annals of applied biology 146: 95-103.

EPPO., 2002. Review of EPPO/CAB 1997C. Tomato spotted wilt tospovirus. In Quarantine pests for Europe. (2nd ed) pp 1379-1387. CAB International Walling Ford, UK: CAB

Fraser R. S. S., 1987. Biochemistry of virus infected plants. Research studies press limited. England.

Golnaraghi A. R., Sharaeen N., Pourrahim R., Ghorbani S., Farzadfar S., 2001. First report of tomato spotted wilt virus on soy bean in Iran. Plant Disease 85:1290.

Goodman R. N., Zoltan K., Wood K. R., 1986. The biochemistry and physiology of plant disease. Columbia, University of Missouri Press.

Jarosik V., Kolias M., Lapchin L., Rochat J., Dixon A. F. G., 1997. Seasonal trends in the population increase of *Frankliniella occidentalis* (Thysanoptera: Thripidae) on cucumber. Entomological Research 87: 498-498.

Lucas W. J., and Gilbertson R. L., 1994. Plasmodesmata in relation to viral movement within leaf tissues. Annual Review of Phytopathology 32: 387-411.

Mandal B., Pappu H. R., Cullbreath A. K., Holbrook C. C., Gorbert D. W., Todd J. W., 2002. Differential response of selected peanut (*Arachis hypogae*) genotypes to mechanical inoculation by tomato spotted wilt virus. Plant Disease 86: 939-944.

Mavric I., and Ravnikar M., 2001. First report of tomato spotted wilt virus and impatients necrotic spot virus in Slovenia. Plant Disease 85: 1288.

Mcpherson R. M., Pappu H. R., Jones D. C., 1999. Occurrence of five thrips species on flue cured tobacco and impact on spotted wilt disease incidence in Georgia. Plant Diseases83: 765-767.

Mollema C., 1992. Western flower thrips in cucumber: resistant genotypes keep pests under control. Entomology experimentalis et Aplicata 84: 259 – 264.

Momol M. T., Funderburk J. E., Olson S., Stavisky J., 2002. Management of tomato spotted wilt tospovirus (TSWV) on tomatoes with UV – reflective mulch and Acibenzolar – S – methyl. Proceedings of the 7 th international symposium of thysonoptera, Australian national insect collection, canaberra. Pp. 111 – 116.

Pappu H. R., Csinos A. S., Mcpherson R. M., Jones D. C., Stephenson M. G., 2000. Effect of acibenzolar-S-methyl and imidacloprid on suppression of tomato spotted wilt tospovirus in flue-cured tobacco. Crop Protection 19: 349-354.

Parisi B., Roggero P., Grassi G., Masenga V., 1998. Tospovirus (TSWV) on Aubergine in protected cultivation. Informative- Agrario 54: 45-48.

Pourrahim R., Farzadfa S. H., Moini A. A., Shahraneen N., Ahoonmanesh A., 2001. First report of tomato spotted wilt virus in potatoes in Iran. Plant Disease 85:442.

Ramkat, R.C., Wangai, A.W., Ouma, J.P., Rapando, P.N., Lelgut, D.K. 2008. Cropping system influences Tomato spotted wilt virus disease development, thrips population and yield of tomato (Lycopersicon esculentum L.). Annals of Applied Biology 153 373-380.

Rapando, P., Wangai, A., Tabu, I., Ramkat, R. 2009. Variety, mulch and stage of inoculation effects on incidence of Tomato spotted wilt virus disease in cucumber (Cucumis sativus L.). Archives of Phytopathology and Plant Protection 42: 579-586

Riley D. G, Pappu H. R., 2000. Evaluation of tactics for management of thrips vectored tomato spotted wilt virus in tomato. Plant Disease 84: 47- 852.

Saunyama, I. G. M., Knapp, M. 2003. Effect of pruning and trellising of tomatoes on red spider mite incidence and crop yield in Zimbabwe. African Crop Science Journal. 4: 269-277.

Soler S., Diez M. J., Nuez F., 1998. Effect of temperature regime and growth stage interaction patterns of virus presence in TSWV-resistant accessions of *Capsicum chinese*. Plant Disease 82: 1199-1204.

Thomas C. M. and Jones R. A. C., 2003. Selection, biological properties and fitness of resistance breaking strains of tomato spotted wilt virus in pepper. Annals of Applied Biology 142: 235-243.

Wangai A., Mandal B. J., Pappu H. R., Kilonzo S., 2001. First report of tomato spotted wilt virus in Kenya. Plant Disease Journal 85: 1123.

Wells M. L., Culbreath A. K., Todd J. W., Brown S. L., Gorbet D.W., 2002. A regression approach for comparing field resistance of peanut cultivars to tomato spotted wilt tospovirus. Crop Protection 21: 467-474.

Wetering F.V., 1999. Effects of thrips feeding on tospovirus transmission in chrysanthemum. WAU Dissertation No 2606. Agriculture University Wageningen. Netherlands.

Xu P., Blancaflor E. B., Roossinck M. J., 2003. In spite of induced multiple defence responses, tomato plants infected with cucumber mosaic virus and D-satellite RNA succumb to systemic necrosis. Molecular Plant Microbe Interactions16: 467-476.

Appendices

Table 1: Mean values of TSWV disease scores, fruit weights and marketable yield of five Kenyan tomato varieties grown under field conditions at KARI – Njoro in the year 2004 and 2006. Means within a column followed by the same letter are not significantly different at 0.05 level of probability. (+) denotes natural infestation and (-) insecticide protected. Values are means of three replicates, four plants per plot.

Tomato Varieties	Treatments	Visual disease scores		Fruit weights $Mg\,ha^{-1}$		Marketable yield $Mg\,ha^{-1}$	
		2004	2006	2004	2006	2004	2006
Cal J	+	2.73 b	2.53 a	28.64 c	27.82 b	6.49 e	4.28 c
	-	2.57 bc	1.76 b	31.99 bc	36.58ab	11.68 de	13.16 bc
Marglobe	+	2.22 cde	1.73 ab	36.55 bc	33.32 ab	14.21 cde	14.15abc
	-	2.05 def	1.93 ab	44.56 bc	43.94 ab	17.83 bcd	20.10 abc
MoneyMaker	+	2.14 de	1.63 b	69.29 a	46.89 a	33.88a	19.19 abc
	-	1.85 ef	1.43 bc	55.02 ab	32.69 ab	30.42 ab	21.03 abc
Roma	+	1.69 fg	1.63 b	37.00 bc	30.04 ab	25.12 abc	27.26 ab
	-	1.44 g	1.13 c	46.41 abc	39.38 ab	28.35 ab	28.19 a
Riogrande	+	3.12 a	2.46 a	40.00 bc	42.71 ab	15.93 cde	17.80 abc
	-	2.50 bcd	1.63 b	35.35 bc	45.03 ab	11.98 de	14.92 abc

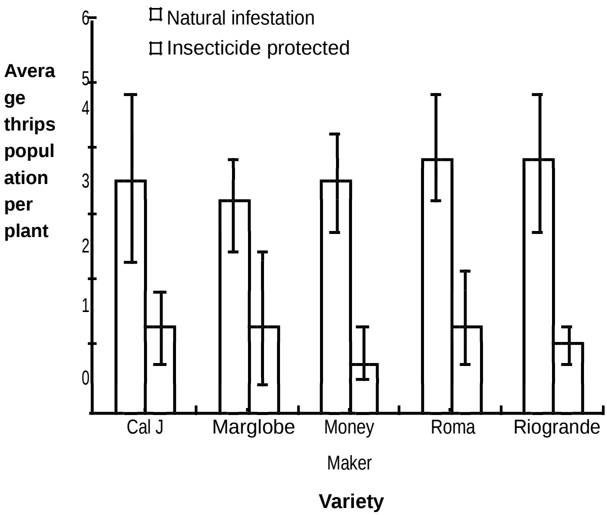

Fig 1: Thrips population on five tomato varieties naturally infested with those protected by insecticide metasystox grown in the year 2004.

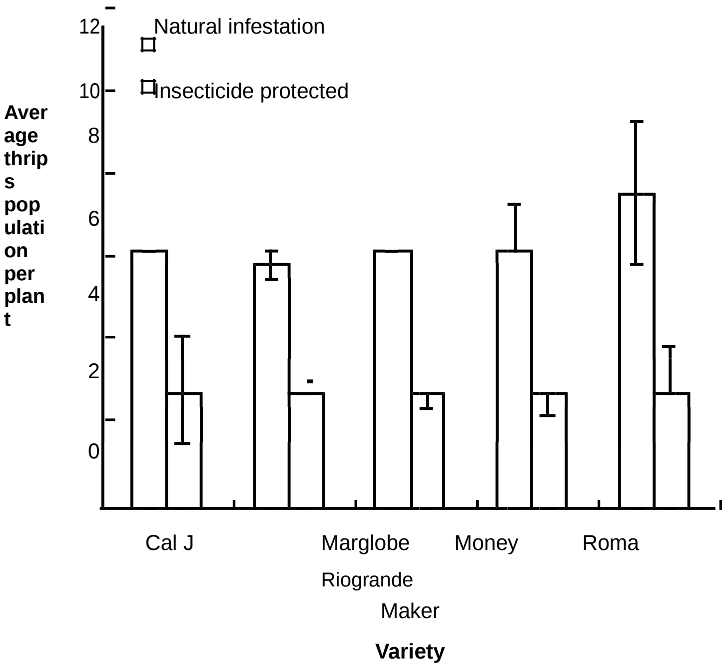

Fig 2: Thrips population on five tomato varieties naturally infested with those protected by insecticide metasystox grown in the year 2006.