

## Tomato spotted wilt disease in Palestine#

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### Abstract

Tomato is one of the top popular crops in Palestine; infected with several pathogens where viruses are among them. Recently, fruit spotted symptoms were noticed on tomatoes fruits that drastically affected crop's productivity. This study was conducted to detect *Tomato spotted wilt virus* (TSWV) as the main known cause of fruit spotted disease. Field surveys were carried through the growing seasons of 2017-2018 in Palestinian tomatoes fields. Symptoms were reported on tomato plants that varied from fruits' yellow and brown rings and/ or irregular spots patterns. In some cases, plants' leaves expressed reduction in growth; mottling and/or wilting. Serological tests using DAS-ELISA technique was applied to detect TSWV on tomato plants which revealed an infection prevalence of 1.27%. This study is considered the first report of TSWV in the country. Knowing that the virus possibility of being transmitted by thrips; increased incidence due to this vector, as well as transmissibility through infected propagating materials could raise up its threat on tomato industry. The use of virus resistant tomato varieties is discussed in the study.

**Keywords:** Tomato, TSWV, ELISA

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## Introduction

Tomato (*Lycopersicon esculentum*, Miller), and also known as (*Solanum lycopersicon*, Linnaeus); belongs to the family Solanaceae, is the most popular crop in Palestine. Tomatoes are cultivated in more than 1200 hectares, constitute 1.5% of total agricultural land cultivated in the West Bank, where Jenin and Tubas governorates are the highest producer of tomatoes (ARIJ, 2015).

Tomatoes were subjected to several viral infections that affect their productivity worldwide such as: *Tomato yellow leaf curl virus* (TYLCV), *Tomato yellow spot virus* (TYSV), *Tomato chlorotic mottle virus* (TCMV), *Tomato rugose mosaic virus* (TRMV), *Tomato yellow vein streak virus* (TYVSV); *Tomato bushy stunt virus* (TBSV) and many others (Damicone & Brandenberger, 2017; Hanssen *et al.*, 2010). The most reported tomatoes infecting viruses in Palestine were *Tomato yellow leaf curl virus* (TYLCV) which reached up to 93% in Jenin and Tubas; followed by *Cucumber mosaic virus* (CMV), that was up to 51% (Sawalha, 2009; Amro, *et al.*, 2014). Meanwhile *Tomato mosaic virus* (ToMV) and *Potato Virus Y* (PVY) were found in very low percent (Sawalha, 2011). In addition to that, "spotted wilt" disease was reported on tomatoes worldwide caused mainly by *Tomato spotted wilt virus* (TSWV) (genus *Tospovirus*; family *Bunyaviridae*) and recently *Tomato Brown rugose fruit virus* (ToBRFV) was reported on tomatoes in different countries (Salem, *et al.* 2016; Luria, *et al.* 2017; Alkowni, *et al.* 2019).

The broadest host range in the genus *Tospovirus* mainly is *Tomato spotted wilt virus* (TSWV) (Tsompana & Moyer, 2008). It has the most economical impact of all members of the genus *Tospovirus* (Goldbach & Kuo, 1996; Goldbach & Peters, 1994). TSWV-infected tomato plants can be identified by the presence of yellow, brown ring or other line pattern on the fruits, reduction in the growth of tomato plant and consisted of

systemic chlorotic and necrotic spots on leaves or tip dieback (French, *et al.* 2015; Sherwood, *et al.* 2009). It is worth to mention that variation in the virus disease symptoms and severity of infection depend on the host genotype, time of infection, environmental conditions, pathogen strain and host developmental stage (Mehraban, *et al.* 2005; Sherwood, *et al.* 2009; Rosello, *et al.* 1996; Sevik & Arli-Sokmen, 2012).

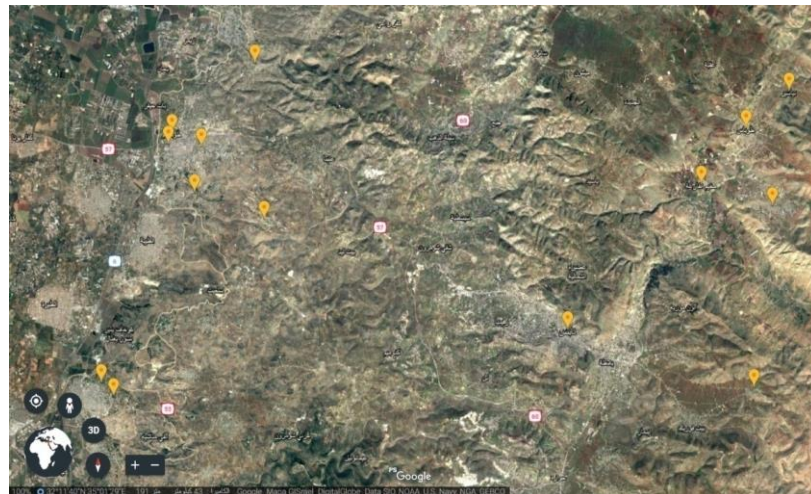
Tospoviruses are exclusively transferred by number of vegetative thrips which are the vector for transmission of TSWV (Riley, *et al.* 2011).

In this research, surveys for detection the presence of *Tomato spotted wilt virus* (TSWV) were carried in the country for the first time and virus presence was confirmed using serological detection tools.

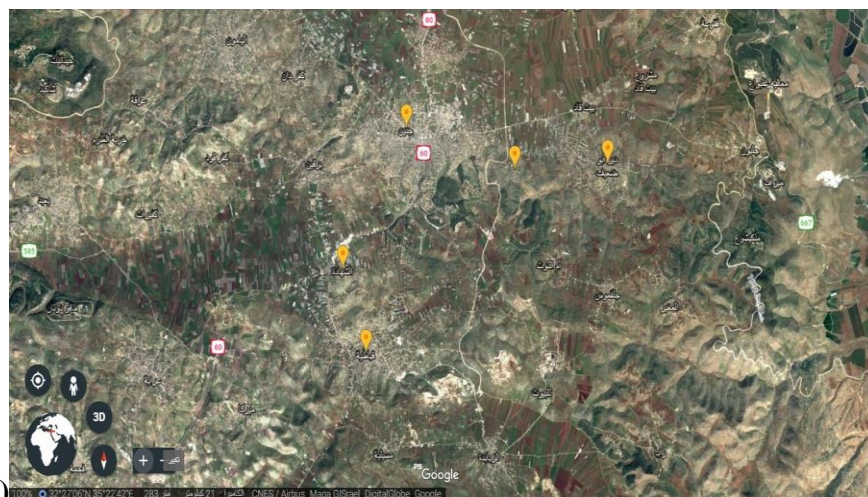
## **Materials and Methods**

### ***Field Survey and Sample Collection***

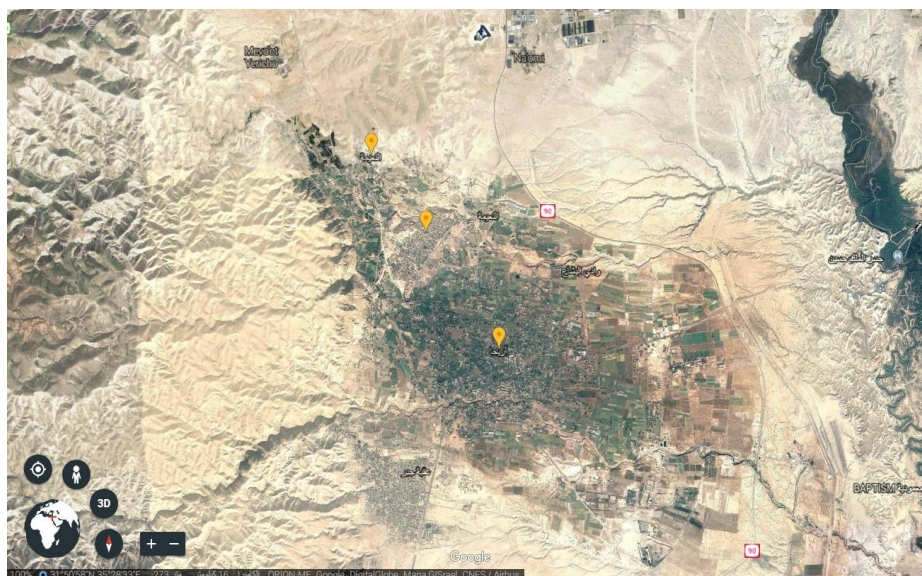
Tomato fields in six districts of Palestine (Jericho, Jenin, Tubas, Tulkarem, Nablus and Qalqelia) were surveyed (Figure 1). Symptoms of viral cause were inspected with particular emphasis on tomato spotted wilt one. More than 300 of tomato leaf samples were collected randomly in each field/greenhouse from symptomatic and symptomless plants covering more than 25 different sites. The collected samples were stored in plastic bags and conserved in cold chambers at 4°C for later serological and /or molecular tests.



(A)



(B)



(C)

**Figure (1):** The locations of the surveyed fields in the sampling areas in Tubas, Nablus, Tulkarem, and Qalqelia (A); Jenin district (B), and Jericho. (C).

Samples were included more than 20 different tomato cultivars (Ikram; Nukran; Izmir; Cherry; Shekram; Balahya; Princess; 593; 554; AX64-782F1; Granada; Gory; Izmuna; 888; 026; Just; & Mayla) (Table 1).

**Table (1):** The number of tomato varieties at each sampling area.

Tomato Varieties	District (Number of sample)						Total
	Nablus	Tulkarem	Tubas	Jenin	Qalqelia	Jericho	
Cluster (Ikram)	33	9	12	15	9	18	96
Cluster (Nukran)	—	—	—	—	6	3	9
Izmir	18	6	6	13	—	4	47
Cherry	—	—	9	—	—	—	9
Shekram	—	—	9	—	—	—	9
Balahya	—	—	3	—	—	—	3
Princess	—	—	3	—	—	—	3
593	—	7	6	—	—	—	13
554	—	—	—	—	9	—	9
AX64-782F <sub>1</sub>	—	6	—	—	—	—	6
Granada	—	—	—	12	—	—	12
Gory	—	—	—	10	—	12	22
Izmuna	—	—	3	3	—	—	6
888	—	9	—	—	—	—	9
026	—	6	—	—	—	6	12
Just	—	—	—	—	6	—	6
Mayla	—	—	—	—	3	15	18

***Serological Detection***

Double Antibody Sandwich (DAS) ELISA was used for detection of the virus in tomato leaf samples. Briefly, about 0.35-0.4 gm of the leaves were ground in sterile mortar and pestil using the grinding buffer. Plant sap was added to the polystyrene plate previously coated with trapping antibodies according to the DAS-ELISA Kit instructions (Agdia, Inc. 52642 County Road 1 Elkhart, IN 46514). After overnight incubation at 4°C; and three times washing with phosphate buffered saline with tween-20 (PBST), the conjugated enzyme antibodies were applied to the plate wells and incubated for 2 hrs. Plate wells then were washed with PBST before the 100 µl of PNP substrate were dispensed into each well; and the plate was covered with aluminum foil and incubated for 60 minutes. The

result was examined by eyes and measured through the plate ELISA reader at 405 nm every 15 min and up to 2 hrs.

## Results and Discussion

The estimated total annual production of tomato is 124,445 tons of all surveyed crops in the West Bank, and representing 23% of the annual total vegetables production in Palestine (ARIJ, 2015). Due to climate changes, this production was facing challenges from management to the increased risks of pathogen and pest attack (Amro, *et al.* 2014; Skendži'c, *et al.* 2021). Viral infections were the most notable factor that limit crop production (Sawalha, 2011); as serious damages and large economical losses on tomato crops frequently occurred by viral infection. The magnitude of damages and economical losses depends on the strain of the virus, the infection time, the variety of tomato and other factors (Amro, *et al.* 2014).

Surveys of tomato fields revealed the existence of viral symptoms only in Qabatya-Jenin District fields. The infected plants exhibit symptoms comparable to those of TSWV (Figure 2). These symptoms were characterized by the regular and steady ring spots on tomato fruits and leaves. These notably TSWV related symptoms were observed mainly in tomato growing fields in Qabatya-Jenin district and reached up to 23.5% of planted tomatoes there. Other observed symptoms were ranged from brown line pattern on tomato stems, to brown ring spots on tomato fruits, and chlorosis, necrotic spots and malformation on tomato leaves in other fields. As a total; the symptoms were noticed on 1.72% of surveyed plants in all other sampling areas. Surprisingly, the symptoms were noticed only on two tomato varieties "Granada" and "Cluster (Ikram)"; the famous two tomato greenhouses grown varieties in Palestine. Thus, these two varieties can be considered more sensitive to the viral infection rather than other varieties tested varieties (Table1).





**Figure (2):** Viral symptoms observed during the field surveys, where brown line pattern on tomato veins; brown ring spots on tomato fruits; and chlorotic, necrotic spots and malformation were noticed on tomato leaves.



Using Double Antibody Sandwich Enzyme-Linked Immunosorbent Assay (DAS-ELISA) Kit (Agdia, Inc., IN, USA) for the TSWV, confirmed in symptomatic samples from these fields of Qabatya region only.

TSWV was found in more than 60 countries all over the world (Karavina & Gubba, 2017). This study reported TSWV with relatively low incidence (1.29%) in Northern region of the West bank of Palestine. According to our knowledge, this was the first report for the presence of *Tomato spotted wilt virus* (TSWV) on tomato in West Bank- Palestine. TSWV was reported on lettuce in Jordan (Salem, *et al.* 2012, Anfoka, *et al.* 2006); where symptoms had been observed on the crops in Jordan Valley; close boarder to Palestine, and later on it was detected on tomato in 2016.

The virus spread in most cases depending on factors such as; climate change which sometimes in favor of the presence of vector. In Palestine these varieties of tomato plants including 554, Just, 026, 593, and Ax64-782fi; were recommended by Ministry of Agriculture as virus resistant ones (Personal communication, Ministry of Agriculture). Further studies on their resistance against TSWV could be plausible.

There are four species of thrips: *Frankliniella occidentalis* (western flower thrips); *F. schultzei*, *F. fusca* (tobacco thrips) and *Thrips tabaci* (onion thrips), that are defined as the most important vector of TSWV because of the wide distribution and the relation between these species and TSWV (Sherwood, *et al.* 2009). Further investigations were recommended to determine the transmissibility of local thrips to the virus in Palestine, since vector genetics appears to be a factor that influences dynamics of disease epidemics and their evolution of insect-vectored viruses (Jacobson & Kennedy, 2013). Even though; the detected tomato samples were believed to be nursery infected seedlings; thus further investigation on distributed propagating materials would be advisable. Except Granada and cluster (Ikram) tomato cultivars, it is recommended to cultivate tomato varieties which did not exhibit any viral symptoms as previously mentioned in this research study.

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