

## **Incidence and Distribution of *Tomato Spotted Wilt Tospovirus* (TSWV) in Vegetable Crops in Antalya Province of Turkey**

**Volkan BOZDOGAN\***      **Muharrem Arap KAMBEROGLU\*\***

\* Bayer Crop Science, Antalya, Turkey

\*\* University of Cukurova, Faculty of Agriculture, Department of Plant Protection, 01330 Adana, Turkey

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### **ABSTRACT**

Large scale surveys were conducted during 2007-2009 to monitor the incidence and distribution of *Tomato spotted wilt virus* (TSWV) in tomato (*Solanum lycopersicum* L.), pepper (*Capsicum annuum* L.) and lettuce (*Lactuca sativa*) plants growing greenhouses in Antalya province including Center (Aksu, Kursunlu, Camkoy and Altinova villages), Serik (Center and Cakallik villages) and Kumluca (Center, Mavikent and Beykonak villages) districts by DAS-ELISA tests. In total, 42 tomato, 69 pepper and 12 lettuce commercial greenhouses were surveyed and a total number of 596 samples were collected from 193 tomato, 345 pepper and 58 lettuce plants. TSWV was detected in 526 samples (88%) from 156 tomato (81%), 316 pepper (92%), 54 lettuce (93%) plants.

Three TSWV isolates, coded S1, K1 and A1 were selected from tomato, pepper and lettuce samples respectively for biological characterization. The symptom severity and expression times on indicator plants were varied for each TSWV isolate. The symptoms caused by the lettuce isolate (A1) were observed in 7-10 days whereas for tomato (S1) and pepper (K1) isolates in 20-24 days after inoculation. The S1, K1 and A1 isolates were also additionally confirmed by RT-PCR using two different TSWV specific primers (L1TSWVR (AAT TGC CTT GCA ACC AAT TC)-L2TSWVF (ATC AGT CGA AAT GGT CGG CA) and BR035 (GAA TAT ATG ACA CCA TTG)-PDH006 (CCC AGA GCA ATC AGT GCA)) and the amplicons of the expected sizes, 276 and 514 bp, were obtained.

**Key words:** Tomato spotted wilt virus (TSWV), ELISA, PCR, incidence, vegetables

### **ÖZET**

#### ***Tomato Spotted Wilt Tospovirus* (TSWV)'nin Antalya İlinde (Türkiye) Yetiştirilen Sebzelerde Bulunma Oranı ve Dağılımı**

Domates lekeli solgunluk virüsü (TSWV)'nin, Antalya ilinde, Merkez (Aksu, Kurşunlu, Çamköy ve Altınova köyleri), Serik (Merkez ve Çakallık köyleri) ve Kumluca (Merkez, Mavikent and Beykonak köyleri) ilçeleri sera alanlarında yetiştirilen domates, biber ve marul bitkilerinde bulunma oranı ve dağılımını ELISA yöntemi kullanılarak ortaya koymak amacıyla, 2007- 2009 yılları arasında geniş alanlarda surveyler düzenlendi. Toplamda, ticari üretim yapılan 42 domates, 69 biber ve 12 marul serası gezildi ve bu alanlardan 193 domates, 345 biber ve 58 marul olmak üzere 596 bitki örneği toplandı. TSWV enfeksiyonu 156'sı domates (%81), 316'sı biber (%92) ve 54'ü marul (%93) olmak toplam 526 örnekte (%88) saptandı.

Biyolojik çalışmalarda kullanılmak üzere, S1, K1 ve A1 kodlu üç farklı TSWV izolatu, sırasıyla domates, biber ve marul örneklerinden seçilmiştir. İndikatör bitkiler üzerinde, herbir izolatuın simptom şiddeti ve çıkış zamanları farklılık göstermiştir. Marul izolatu (A1) tarafından meydana getirilen simptomlar 7-10 gün içerisinde gözlenirken, bu süre domates (S1) ve biber (K1) izolatları için 20-24 gün arasında olmuştur. Bunlara ilaveten, S1, K1 ve A1 izolatları, TSWV'ye spesifik iki farklı primer çifti (L1TSWVR (AAT TGC CTT GCA ACC AAT TC)-

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L2TSWVF (ATC AGT CGA AAT GGT CGG CA) and BR035 (GAA TAT ATG ACA CCA TTG)-PDH006 (CCC AGA GCA ATC AGT GCA)) kullanılarak RT-PCR yöntemi ile de doğrulandı ve beklenen büyüklüklere (276 and 514 bp) sahip bandlar gözlemlendi.

**Anahtar Sözcükler:** TSWV, ELİSA, PCR, survey, sebzeler

### INTRODUCTION

Tomato, pepper and lettuce are economically important crops and extensively grown in Antalya province in Turkey. According to datas of Turkish statistical institute, Antalya province produced 66.68% (2,134,374 tons), 53.76% (257,151 tons) and 19.16% (13,027 tons) of total production of tomato, pepper and lettuce in greenhouse in Turkey in 2013, respectively (Anonymous, 2014). There are many factors including plant diseases and pests cause severe economic losses in tomato, pepper and lettuce productions by reducing quantity and quality of yield. *Tomato spotted wilt virus* (TSWV), member of the *Tospovirus* genus within the *Bunyaviridae* family, is one of the most important virus that cause disease in tomato, pepper and lettuce plants (Gnayem, 1995). This virus is distributed worldwide and occurs in 900 plant species including crops and ornamental plants as well as weeds in many parts of the world (Goldback and Peters, 1994). It is transmitted persistently through several species of thrips although western flower thrips (*Frankliniella occidentalis*) is one of the most efficient vectors (Cho et al., 1987; Eckel et al., 1996; German et al., 1992). The symptoms induced by TSWV vary depending on the host, infection time and environmental conditions, often; stunting and wilting of plants, ringspots on leaves and fruits, chlorotic and necrotic lesions, deformation and dieback symptoms on leaves and stems are occur in infected plants (Holguin-Pena and Rueda Puente, 2007).

TSWV was detected first in tobacco plants in Canakkale region with the rate of infection of 80-100% (Azeri, 1981) then it was reported in tomato, pepper, eggplants, lettuce and squash as well as buttercup plants from different parts of Turkey (Azeri, 1994; Guldur et al., 1995; Yurtmen et al., 1998; Arli- Sokmen et al., 2005; Turhan and Korkmaz, 2006; Kucuk and Kamberoglu, 2008; Kamberoglu et al., 2009; Yardimci and Kilic Culal, 2009; Kamberoglu and Alan, 2011).

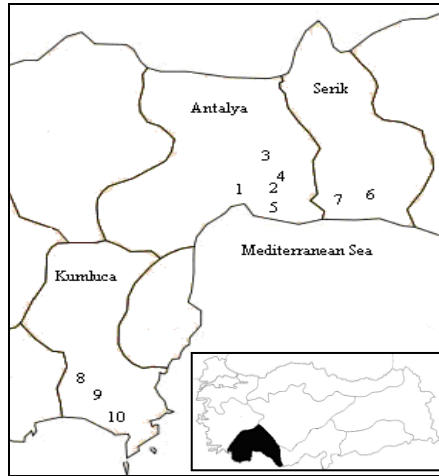
The presence of TSWV and thrips have emerged as important problems in many of the vegetables growth areas in Turkey as well as in the world in recent years. Although Antalya province is an important vegetable production area in Turkey, the presence of TSWV, its incidence and distribution and severity of losses caused by TSWV infection are largely unknown in this region. Because of this, the present study was conducted to detect TSWV and assess its incidence and distribution in tomato, pepper and lettuce growing greenhouses in Antalya province by DAS-ELISA and biological characterization of some isolates by mechanical inoculation.

### MATERIALS and METHODS

#### Surveys and Sample Collection

Surveys were conducted between 2007-2009 in randomly selected tomato, pepper and lettuce growing greenhouses located in Center (Aksu, Kursunlu, Camkoy and Altinova villages), Serik (Center and Cakallik village) and Kumluca (Center and Mavikent and Beykonak villages) districts of Antalya province (Figure 1).

Leaf and fruit samples were taken from the plants showing typical TSWV symptoms such as wilting, mosaic, vein necrosis, ringspots and leaf deformation (Figure 2). The samples were placed to labelled plastic bags and transferred to the virology laboratory of University of Cukurova in an ice box. The plant tissues were stored at +4°C shortly.



**Figure 1.** Map of surveyed area in Antalya province. Antalya district: 1. Center, 2. Aksu, 3. Kursunlu, 4. Camkoy, 5. Altinova; Serik district: 6. Center, 7. Cakallik; Kumluca district: 8. Center, 9. Beykonak, 10. Mavikent

## DAS-ELISA

The suspected field samples and mechanically inoculated indicator plants were tested with TSWV specific ELISA kit (Bioreba, Switzerland) by DAS-ELISA method. The method was applied according to instruction of manufacturer.

Briefly, plant saps were extracted in extraction buffer and put into ELISA wells previously coated with TSWV specific IgG in carbonate buffer (pH: 9.6). After incubation at +4°C overnight, alkaline phosphatase conjugated antibody in conjugate buffer was loaded to the wells. The plates were incubated at 30°C for 2 hours and p-nitrophenyl phosphate in substrate buffer (1mg/ml) was added to each well. Absorbance values were read in 405 nm by Medispec ESR 200 ELISA reader at 30 and 60 minutes at room temperature. The absorbance values were considered as positive those that twice the negative control's. Virus free plants and buffer alone were served as negative control.

With respect to ELISA results, tomato (S1), pepper (K1) and lettuce (A1) isolates of TSWV were selected and further tested by mechanical inoculation and RT-PCR. Selected TSWV isolates were maintained on mechanically inoculated pepper plants. Five plants were inoculated for each isolates.

## Biological characterization of TSWV isolates

Biological characteristics of TSWV isolates were assessed by mechanical inoculation according to Mandal et al. (2001). Fresh tissues were extracted in cold 0.01M phosphate buffer (pH: 7.0) contains 2% sodium sulphide and the sap was rubbed onto carborundum (500 mesh) dusted leaves of indicator plants at 2-4 leaf stage. After mechanical inoculation, inoculated leaves were washed with tap water to remove debris and plants were kept in the climatic room (25-27°C). Indicator plants were observed daily for symptoms expression and tested by ELISA 30 days after inoculation. Five plants of each species namely *Capsicum annuum* L., *Chenopodium quinoa* Willd., *Nicotiana rustica* L., *N. glutinosa* L., *N. tabacum* L. "Xanthii", *N. tabacum* L. "Xanthii nc" and *N. tabacum* L. "Samsun" were inoculated for each TSWV isolates.

## Total RNA Extraction

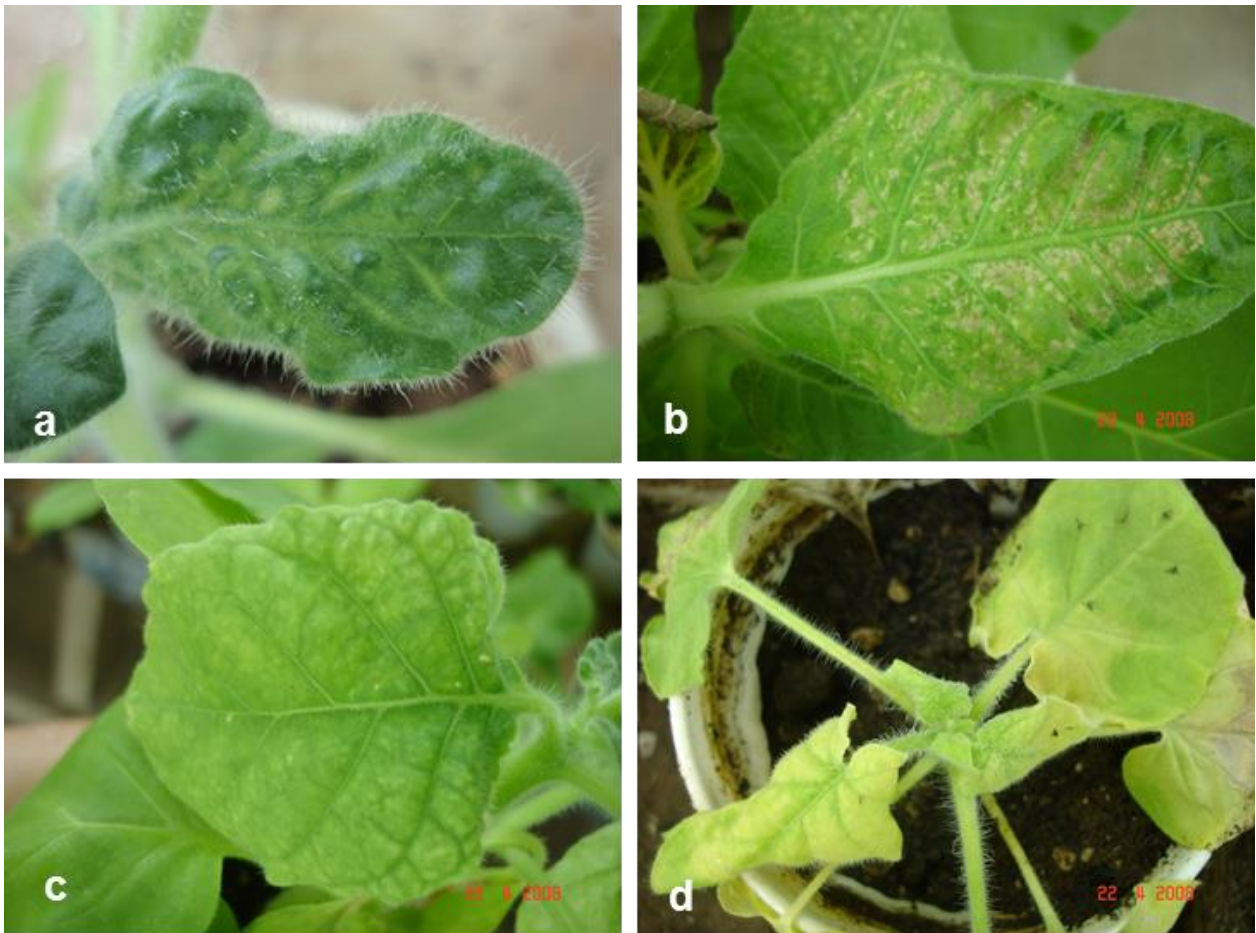
Fresh leaves of TSWV inoculated pepper plants were used for purification of total RNAs and method was applied as recommended by Astruc et al. (1996).

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Briefly, leaves were extracted in TE buffer (100mM Tris-HCl pH 8.0, 50mM EDTA, 500mM sodium chloride and 0.1% 2-mercapthoethanol) and 20% of SDS was added after low speed centrifugation. Then, the tubes were incubated in hot water bath at 65°C for 30 minutes and 5M of potassium acetate added. The liquid parts were transferred to new tubes after centrifugation and 3M sodium acetate and 100% alcohol was added. After incubation at -20°C overnight, the pellets were precipitated by centrifugation and washed with 70% alcohol. Finally, pellets were resuspended in RNase free sterile distilled water.



**Figure 2.** Tomato spotted wilt virus symptoms on different plants a. Necrotic lesions on tomato leaves; b. Necrotic ringspots on tomato fruit; c. Chlorosis and downward curling in pepper leaves; d. Chlorotic ringspots on pepper fruit; e. Stunting in lettuce plant (left, healthy; right, infected); f. Necrotic areas and leaf deformation in lettuce



**Figure 3.** Symptoms of TSWV on indicator plants. a. Leaf deformation, curling, ringspots and chlorosis on *N. rustica* L.; b. Leaf deformation and necrosis on *N. tabacum* L. “Samsun”; c. Chlorosis and leaf deformation on *N. tabacum* L. “Xanthii”; d. Chlorosis and dieback in *Nicotiana glutinosa* L.

### RT-PCR

The RT-PCR studies were carried out in a Veriti 96 well thermalcycler (Applied Biosystem, USA) using TSWV specific primers, L1TSWVR (AAT TGC CTT GCA ACC AAT TC); L2TSWVF (ATC AGT CGA AAT GGT CGG CA) and BR035 (GAA TAT ATG ACA CCA TTG); PDH006 (CCC AGA GCA ATC AGT, GCA) for amplification of 276 bp and 514 bp amplicon sizes, respectively (Mumford et al., 1994; Eiras et al., 2001).

For the complementary DNA (cDNA) synthesis, 1µl of total RNA was incubated with 1µl of reverse primer (10mM) at 70°C for 5 minutes and then chilled on ice for 90 sec. Then other components, 16.6µl of sterile distilled water, 5µl of reverse transcriptase buffer (5X), 1µl of dNTPs (10mM), 0.3µl of RNase inhibitor (40U/µl) and 0.1µl of MMLV-RT (200U/µl) were added to the reaction and tubes were incubated at 42°C for 60 minutes.

PCR was carried out by mixing 1µl of cDNA, 16.8µl of sterile distilled water, 2.5µl of 10X Taq polymerase buffer, 1.5µl of MgCl<sub>2</sub>, 1µl of dNTPs, 0.2µl of Taq DNA polymerase (5U/µl), 1µl of forward primer (10mM) and 1µl of reverse primer (10mM) mixture. PCR conditions were 2 min at 94°C followed by 35 cycles at 94°C for 30 sec, 52°C for 30 sec and 72°C at 2 min and final extension step at 72°C for 10 min. The PCR products were analyzed by electrophoresis on 1% agarose gel (Gallitelli and Minafra, 1994).

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## RESULTS

### Surveys and Sample Collection

A total of 380.25 da with 123 greenhouses producing tomato, pepper and lettuce were surveyed in Antalya province. A total of 596 samples (193 tomatoes, 345 peppers and 58 lettuce) were taken from the plants showing a range of TSWV symptoms in the leaves and fruits. In surveys; 331 samples (98 tomatoes, 175 pepper and 58 lettuce) from 69 greenhouses (22 tomatoes, 35 pepper and 12 lettuce, 200.25 da; Table 1) in Center, 150 samples (70 tomatoes and 80 pepper) from 30 greenhouses (14 tomatoes and 16 pepper, 75.25 da; Table 2) in Serik and 115 samples (25 tomatoes and 90 pepper) from 24 greenhouses (6 tomatoes and 18 pepper, 104.75 da; Table 3) in Kumluca were tested by DAS- ELISA tests.

### Frequency of TSWV

ELISA results showed that 526 out of 596 plant samples (88.25%) were infected by TSWV in Antalya province. In the study, 156 of 193 tomato samples, 316 of 345 pepper samples and 54 of 58 lettuce samples were found to be infected by TSWV with infection rates of 80.83%, 91.6 % and 93.1%, respectively.

In Center district, a total of 331 samples were collected and 289 samples were infected with TSWV (87.31%). Among the samples, 77 tomato (78.57%), 158 pepper (90.28%) and 54 lettuce samples (93.10%) were infected with TSWV. The highest rates of infection were detected 92.72%, 92.30%, and 100% for pepper, tomato and lettuce samples in Altinova village, respectively. The lowest infection rate (68%) for tomato was detected in Aksu village, and for pepper and lettuce were in Kursunlu with the rates of 88% and 90%, respectively (Table1).

**Table 1.** The results of TSWV surveys conducted in the Center (Antalya) of district

Species	Villages	No. of field surveyed	Total area surveyed (da)	No. of samples collected	No. of samples TSWV positive	Infection rates (%)
Tomato	Kursunlu	7	29	35	26	74.28
	Altinova	6	16.5	26	24	92.30
	Aksu	5	14.5	25	17	68.00
	Camköy	4	11	12	10	83.33
<b>Total</b>		<b>22</b>	<b>71</b>	<b>98</b>	<b>77</b>	<b>78.57</b>
Pepper	Kursunlu	10	27	50	44	88.00
	Altinova	11	34	55	51	92.72
	Camköy	14	42	70	63	90.00
<b>Total</b>		<b>35</b>	<b>103</b>	<b>175</b>	<b>158</b>	<b>90.28</b>
Lettuce	Kursunlu	4	8	20	18	90.00
	Altinova	1	0.25	5	5	100.0
	Kircami	7	19	33	31	93.93
<b>Total</b>		<b>12</b>	<b>27.25</b>	<b>58</b>	<b>54</b>	<b>93.10</b>
<b>The overall total</b>		<b>69</b>	<b>200.25</b>	<b>331</b>	<b>289</b>	<b>87.31</b>

According to Table 2, 130 out of the 150 samples tested, were found to be infected by TSWV in Serik district (86.66%). TSWV was detected in 58 of 70 tomatoes (82.85 %) and 72 of 80 pepper samples (90%). The highest TSWV incidences were detected in Cakallık village with the rates of 85% and 92.5% for tomatoes and pepper, respectively. The lowest disease incidences were detected in the Centre of district with the rate of 82% for tomatoes and 87.5% for peppers.

**Table 2.** The results of TSWV surveys conducted in Serik district

Species	Villages	No. of field surveyed	Total area surveyed (da)	No. of samples collected	No. of samples TSWV positive	Infection rates (%)
Tomato	Center of district	10	37	50	41	82.00
	Cakallik	4	5.75	20	17	85.00
<b>Total</b>		<b>14</b>	<b>42.75</b>	<b>70</b>	<b>58</b>	<b>82.85</b>
Pepper	Center of district	8	18.25	40	35	87.5
	Cakallik	8	14.25	40	37	92.5
<b>Total</b>		<b>16</b>	<b>32.50</b>	<b>80</b>	<b>72</b>	<b>90.00</b>
<b>The overall total</b>		<b>30</b>	<b>75.25</b>	<b>150</b>	<b>130</b>	<b>86.66</b>

In Kumluca district, 107 of 115 samples (93.04%) were infected by TSWV. Among the samples, 21 of 25 tomato (84%) and 86 of 90 pepper samples (95.55%) were infected with TSWV. The infection rates were detected by 96.92% and 92% in peppers in Mavikent and Beykonak, respectively whereas the rate was 100% in tomatoes in both villages (Table 3).

**Table 3.** The results of TSWV surveys conducted in Kumluca district

Species	Villages	No. of field surveyed	Total area surveyed (da)	No. of samples collected	No. of samples TSWV positive	Infection rates (%)
Tomato	Center of district	4	13	18	14	77.77
	Mavikent	1	0.75	2	2	100.0
	Beykonak	1	3	5	5	100.0
<b>Total</b>		<b>6</b>	<b>16.75</b>	<b>25</b>	<b>21</b>	<b>84.00</b>
Pepper	Mavikent	13	60	65	63	96.92
	Beykonak	5	28	25	23	92.00
<b>Total</b>		<b>18</b>	<b>88</b>	<b>90</b>	<b>86</b>	<b>95.55</b>
<b>The overall total</b>		<b>24</b>	<b>104.75</b>	<b>115</b>	<b>107</b>	<b>93.04</b>

### Host range studies

Characteristic TSWV symptoms were observed on indicator plants inoculated by A1, S1 and K1 isolates. These isolates caused leaf curl, deformation, stunting and chlorosis symptoms in *N. tabacum* “Xanthii” and *N. tabacum* “Samsun” plants. In addition to these symptoms, ringspots were observed in *Capsicum annum*, *N. tabacum*. “Xanthii nc” and *N. rustica* plants. Only, necrotic local lesions was developed in inoculated *Chenopodium quinoa* Willd. plants. A1 isolate which was different then other isolates showed chlorosis and stunting, as well as dieback and necrotic local lesions in *N. glutinosa* plants. Whereas only chlorosis and stunting symptoms were observed on S1 and K1 inoculated *N. glutinosa* plants (Figure 3; Table 4).

The symptom expression time and virulence were significantly different in indicator plants for these isolates. The symptoms were appeared in 7-10 days with A1 isolate, however with S1 and K1 isolates took much longer to appear in 20-24 days. The symptoms were very similar on indicator plants caused by all isolates, but A1 isolate caused much more severe symptoms.

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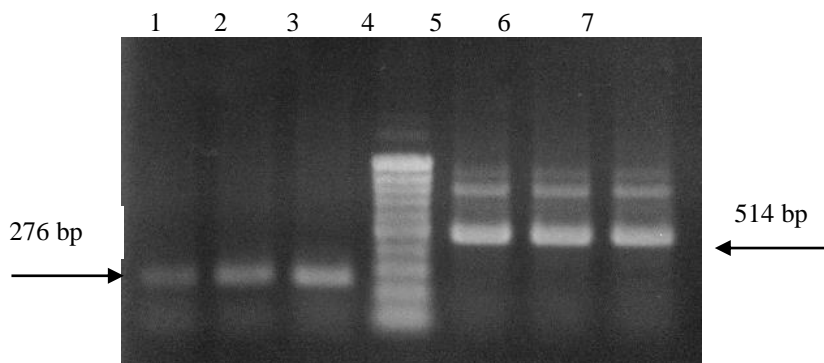
**Table 4.** The symptoms of TSWV isolates on indicator plants

<i>Indicator plants</i>	<i>A1 isolate</i>	<i>K1/ S1 isolates</i>
<i>Chenopodium annuum</i> L.	LC, LD, S, C, Rs	LC, LD, S, C, Rs
<i>Chenopodium quinoa</i> Willd.	NLL	NLL
<i>Nicotiana glutinosa</i> L.	C, S, D, NLL	C, S
<i>Nicotiana tabacum</i> L. “Xanthii”	LC, LD, S, C	LC, LD, S, C
<i>Neotianaitabacum</i> L. “Xanthii nc”	LC, LD, S, C, Rs	LC, LD, S, C, Rs
<i>Nicotiana tabacum</i> L. “Samsun”	LC, LD, S, C	LC, LD, S, C
<i>Nicotiana rustica</i> L.	LC, LD, S, C, Rs	LC, LD, S, C, Rs

C: chlorosis, D: dieback, LC: leaf curling, LD: leaf deformation, NLL: necrotic local lesion, Rs: ringspots, S: stunting

### RT-PCR

The presence of TSWV in symptomatic tomato, pepper and lettuce samples was confirmed by RT-PCR using TSWV (L1TSWVR and L2TSWVF) and tospovirus group (BR035 and PDH006) specific primer pairs, as well as in inoculated indicator plants. The expected size of DNA fragments, 276 and 514 bp, were observed on 1% agarose gel (Figure 4).



**Figure 4.** RT-PCR results conducted with primer pairs, L1TSWVR (AAT TGC CTT GCA ACC AAT TC)-L2TSWVF (ATC AGT CGA AAT GGT CGG CA) and BR035 (GAA TAT ATG ACA CCA TTG)-PDH006 (CCC AGA GCA ATC AGT GCA) and analyzed by electrophoresis on a 1% agarose gel and stained with ethidium bromide. (1. A1 isolate; 2. S1 isolate; 3. K1 isolate; 4. DNA Marker [Fermentas]; 5. A1 isolate; 6. S1 isolate; 7. K1 isolate)

### DISCUSSION

TSWV is one of the most important plant virus, causes serious economic losses in numerous crops worldwide. In this study, the presence and prevalence of *Tomato spotted wilt virus* (TSWV) in tomato, pepper and lettuce plants growing greenhouses in districts of Antalya province were determined. In total, 42 tomato, 69 pepper and 12 lettuce growing commercial greenhouses were surveyed and 156 tomato, 316 pepper and 54 lettuce samples with a range of leaf and fruit symptoms including mosaic, necrosis, ringspots, dieback and leaf deformations were found to be infected by TSWV by ELISA. According to results, TSWV was widespread in tomato, pepper and lettuce growing greenhouses in Antalya province and the average rate of TSWV infection in collected samples was considerably high (88.25%). We assume that in addition to easily transmission of TSWV by mechanically, high population of TSWV vectors, *Frankliniella occidentalis* and *Thrips tabaci* L. and increasing effectiveness of their vectors capabilities in the greenhouse conditions could be the reasons of the high TSWV infection rates in greenhouses. Broadbent and Allen (1995) reported that environmental factors is one of the important issues that



affect the epidemiology of TSWV in a greenhouse and the temperature is effective on susceptibility of plant species, population dynamics and overwintering ability of thrips thus virus transmission and consequently spread of TSWV. Sherwood et al. (2003) reported that TSWV has more than ten species of thrips as vectors and the population of viruliferous thrips, due to overwinter in the field, is important in disease development and epidemiology. Kamberoglu and Alan (2011) reported that pepper and tomato plantations in Cukurova region becomes highly infected with TSWV after the thrips populations start to increase due to temperature rise in spring time. Their observations suggest correlations between mean number of thrips and TSWV incidence.

In the studies made on TSWV in Turkey and different countries of the world, various results were obtained. Ozdemir et al. (2009) reported the presence of TSWV infection in greenhouse-grown tomatoes and in some weeds with the rate of 60,6% in Denizli province although the percentages of TSWV were detected as 3.8%, in pepper in Samsun (Arli-Sokmen et al., 2005), 4.5% in tomato in Canakkale (Turhan and Korkmaz, 2006) and 1.2%, in lettuce in Cukurova (Kamberoglu and Alan, 2011) provinces of Turkey. While, Hajiabadi et al. (2012) detected TSWV in tomato in Alborz and Abiyek regions in Iran with the rate of 4.4 % 3.57% respectively, Abu-Shirbi et al. (2012) reported that TSWV was dominant viral disease in tomato fields in Jordan and about 17.7% of the samples were found to be infected with TSWV. Although, occurrence of TSWV infection on lettuce in Iran firstly reported by Soleimani et al (2011) and in Jordan by Salem et al. (2012), Moreno et al. (2004) reported that TSWV is one of the major virus problems in lettuce fields in the Murcia region in Spain. In addition to survey studies, Sevik and Arli-Sokmen (2012) reported that TSWV caused 42.1% and 95.5% reduction in yield and marketable value of tomato, respectively and Deligoz et al. (2014) reported the resistance breaking strain of TSWV on sweet pepper in Turkey, recently.

Characteristic TSWV symptoms, leaf curl, deformation, stunting, ringspots, necrotic local lesions and chlorosis developed on inoculated plants. The results of host range studies were similar to the reported studies of Antignus et al. (1997) and Mohammadi et al. (2000). They reported that, in general, leaf deformation, yellowing, ringspots and chlorotic and necrotic lesions as well as stunting and death plants symptoms were appeared in TSWV inoculated similar indicator plants. However, A1 isolate, obtained from lettuce plants, was able to cause dieback and necrotic local lesions on *N. glutinosa* plants. It suggested that A1 isolate may have different biological properties from other isolates. Furthermore, A1 isolate caused severe symptoms on indicator plants and symptom expression was observed in 7-10 days whereas S1 and K1 isolates caused milder symptoms and expression time was 20-24 days after inoculation. Based on these results (i.e., considering symptom appearance and severity), A1 isolate was accepted to be much more aggressive than the other isolates.

The results of RT-PCR tests were consistent with ELISA tests and host range studies. Together, these results indicated that leaf deformation, ringspots, chlorosis, necrotic local lesions, stunting and dieback of plants symptoms were caused by A1, S1 and K1 isolates of TSWV in tested crops and indicator plants.

We believed that this present study will then serve as a basis for future studies. Some further studies on large scale surveys, molecular characterization (e.g., determine genome sequence) of TSWV isolates in Turkey, identification of its inoculum sources as well as effective integrated pest management (IPM) strategies to control of the virus and its vectors are lacking and needed to be planned to better understand and control of this disease in economically important crops grown in Turkey.

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