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Research Article (Araștırma Makalesi)

#### Reactions of Fifteen Onion Cultivars Commonly Cultivated in Turkey to Leek yellow stripe virus (LYSV)\*\*

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#### Article Info

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#### Keywords

Cultivar reactions, LYSV, Onion. Abstract: Onion (Allium cepa L.) cultivars commonly cultivated in Turkey were for the first time examined for their reactions to Leek yellow stripe virus (LYSV). Fifteen onion cultivars consisted of red, white, and yellow bulb cultivars were grown in pots and mechanically inoculated with LYSV-12.6Po, a newly characterized Turkish isolate. The experiment was in completely randomized block design with three replicates, containing 30 plants in each. One replicate was also planted as control. Serological and molecular detection methods, and statistical analysis on five growth parameters were used to evaluate their reactions to LYSV. LYSV infections in KG kırmızı, UG beyaz, Sampiyon, Perama, Seyhan, and Hazar were not detected by DAS-ELISA, but RT-PCR gave positive results to LYSV for all 15 tested cultivars. t-test results showed that LYSV inoculation caused significant reduction in all growth parameters for most cultivars. Duncan's multiple range test found that Anka, UG kahverengi, and KG kahverengi performed badly in all growth parameters. On the other hand, Sampiyon and Perama were not significantly different to each other in all growth parameters, and performed very well in plant weight, plant height, and no. of leaves parameters. Based on results of this study, Şampiyon and Perama were considered as the most tolerant cultivar to LYSV.

## Türkiye'de Yaygın Olarak Üretilen On Beş Soğan Çeşidinin Leek yellow stripe virus (LYSV)'üne Karşı Reaksiyonları\*\*

#### Makale Bilgileri

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#### Anahtar kelimeler

Çeşit reaksiyonları, LYSV, Soğan. Öz: Türkiye'de yaygın olarak yetiştirilen soğan (Allium cepa L.) türlerinin bu çalışma ile Leek yellow stripe virus (LYSV)'üne karşı reaksiyonları ilk olarak araştırılmıştır. Kırmızı, beyaz ve sarı soğan çeşitlerinden oluşan 15 tür saksılarda yetiştirilmiş ve mekaniksel olarak Türkiye'de yeni karakterize edilmiş olan LYSV-12.6Po izolatı ile aşılanmıştır. Denemede rastgele tam blok tasarımıyla 3 tekerrür ve her tekerrürde 30 bitki düzenlenmiştir. Ayrıca 1 tekerrür de kontrol olarak hazırlanmıştır. Serolojik ve moleküler testlerle LYSV'nin bitkilerdeki varlığı test edilmiş ayrıca ele alınan 5 gelişme parametrelerinin belirlenmesi için istatistiksel analizler uygulanmıştır. LYSV enfeksiyonu KG kırmızı, UG beyaz, Şampiyon, Perama, Seyhan ve Hazar çeşitlerinde DAS-ELISA ile belirlenememiştir, ancak RT-PCR testlerinde ele alınan 15 çeşidin tümü de pozitif sonuçlar vermiştir. t-testi sonucuna göre LYSV enfeksiyonu, ele alınan parametrelerde çeşitlerin çoğunda belirgin azalmalara neden olmuştur. Duncan çoklu aralık testine göre, Anka, UG kahverengi ve KG kahverengi tüm gelişme parametreleri açısından çok kötü sonuçlar vermişlerdir. Diğer taraftan Şampiyon ve Perama ele alınan tüm gelişme parametreleri açısından birbirinden çok farklı olmayıp, bitki ağırlığı,

bitki boyu ve yaprak sayısı açısından en iyi sonuçları vermişlerdir. Bu çalışmadan elde edilen sonuçlara göre Şampiyon ve Perama, LYSV enfeksiyonuna en tolerans çeşitler olarak belirlenmişlerdir.

\*\*This research is part of a Doctoral dissertation in Department of Plant Protection, Faculty of Agriculture, Ankara University by Adyatma Irawan SANTOSA.

#### 1. Introduction

Leek yellow stripe virus (LYSV) is one of the most frequently found viruses infecting Allium spp., such as leek (Allium ampeloprasum L.), onion (Allium cepa L.), and garlic (Allium sativum L.). It is a member of family Potyviridae, genus Potyvirus which genome only has one open reading frame (ORF) to encodes a polyprotein, including NIa-Pro (nuclear inclusion a – proteinase), NIb (nuclear inclusion b), VPg (viral protein genome linked) and CP (coat protein) (Adams et al., 2005). Nucleotide sequences of CP gene are useful in phylogenetic analysis of the virus (Vučurović et al., 2016).

LYSV had been detected infecting onion, leek, and garlic cultivated in Iran and New Zealand (Shahraeen et al., 2008; Ward et al., 2009). Other attempts failed to detect LYSV in onion, although they found the virus infecting leek and garlic in the surveyed areas (Dovas and Vovlas, 2003; Vučurović et al., 2017). It was first reported in Turkey in leek planted in Marmara region (Korkmaz and Cevik, 2009). Later on, the virus was shown to infect leek, onion, and garlic grown in East Mediterranean, Amasya province, and Marmara region of Turkey (Fidan and Baloğlu, 2009; Fidan, 2010; Sevik and Akcura, 2013; Tuzlalı, 2018; Sarı et al., 2020).

LYSV was also detected among onion and leek samples collected in Ankara province recently, with infection rate on onion was estimated to be 13.3%. LYSV-12.6Po, one of the newly characterized Ankara isolates, produced moderate symptoms on onion and mild symptoms on leek and garlic when mechanically inoculated to those species. Nucleotide sequence of partial NIb+CP region of LYSV-12.6Po was deposited in NCBI GenBank with accession number MN070127. Host indexing and phylogenetic studies showed that LYSV-12.6Po is biologically and genetically similar to an Argentines isolate, LYSV-L-Arg (AY007693) (Santosa and Ertunç, 2020).

With almost 2 million tons production annually, onion is one of the most economically valuable multipurpose vegetables in Turkey (Güvenç, 2019; Demir, 2020; TUIK, 2020). LYSV, which apparently present in all major *Allium* crops planting regions of Turkey, could pose a significant threat to onion production in the country. LYSV infection was known to lead medium to heavy bulb weight loss in garlics (Lot et al., 1998). Besides that, LYSV is transmissible in non-persistent manner by several aphid species (Brunt et al., 1996; Lunello et al., 2002); potentially making it spreads over long distance further to other regions of world and Turkey. Therefore, control methods against the virus, such as the use of resistant/tolerant varieties, need to be established.

Currently, only little information was known regarding reactions of *Allium* spp. varieties to LYSV infection. Messidrome was observed to have less severe bulb weight loss (17%) than Germidour (26%) and Printanor (54%) in a study to determine reactions of the three French garlic cultivars against LYSV (Lot et al., 1998). A fertile garlic clone named clone 211 was found to have a high level of resistance to LYSV according to another French study (Lot et al., 2001). During a field survey of garlic viruses that was conducted in Czech Republic, all of 87 Vekan variety samples were tested to be LYSV-free, which could signify its resistance to the virus (Klukáčková et al., 2007). Bulb size of Taşköprü 56, a garlic cultivar widely planted in Turkey, was reported to be significantly reduced upon either natural or mechanical inoculation with LYSV. However, this finding was very limited since it only involved one garlic cultivar, and genome sequence of the LYSV isolate used in the study was not available (Fidan, 2010).

Screening for resistance/tolerance to LYSV presented in this study was for the first time performed on onion cultivars commonly cultivated in Turkey. Furthermore, there were 15 cultivars tested against LYSV-12.6Po, a well characterized Turkish isolate. So, the obtained results could be important in the strategy to control LYSV spread and minimize onion yield loss in the country.

## 2. Material and Methods

Screening of 15 onion cultivars from three seed companies (Table 1) for their reactions to LYSV-12.6Po was performed in a greenhouse at Plant Protection Department, Ankara University during November 22, 2019 - April 27, 2020. Temperature inside greenhouse was kept between 24 - 28°C.

Table 1. Onion cultivars used in this study.

No.	Cultivars name*	Company	Characteristics					
1.	Anka	Aka ziraii ürünleri Ltd. Şti., Bursa	White bulb with dark brown outer skin.					
2.	KG kahverengi	Bayram Tohum, Ankara	White bulb with light brown outer skin.					
3.	KG kırmızı	Bayram Tohum, Ankara	Red bulb with red outer skin.					
4.	UG kahverengi	Bayram Tohum, Ankara	White bulb with light brown outer skin.					
5.	UG kırmızı	Bayram Tohum, Ankara	Red bulb with red outer skin.					
6.	UG beyaz	Bayram Tohum, Ankara	White bulb with white brown outer skin.					
7.	Şampiyon	Tancepa tarım ürünleri Ltd. Şti., Bandırma, Balıkesir	White bulb with yellow-brown outer skin.					
8.	Gence	Tancepa tarım ürünleri Ltd. Şti., Bandırma, Balıkesir	White bulb with dark brown outer skin.					
9.	İnci	Tancepa tarım ürünleri Ltd. Şti., Bandırma, Balıkesir	White bulb with dark brown outer skin.					
10.	Naz	Tancepa tarım ürünleri Ltd. Şti., Bandırma, Balıkesir	White bulb with yellow-brown outer skin.					
11.	Perama	Tancepa tarım ürünleri Ltd. Şti., Bandırma, Balıkesir	White bulb with yellow-brown outer skin.					
12.	Taraz	Tancepa tarım ürünleri Ltd. Şti., Bandırma, Balıkesir	White bulb with dark brown outer skin.					
13.	Seyhan	Tancepa tarım ürünleri Ltd. Şti., Bandırma, Balıkesir	White bulb with yellow-brown outer skin.					
14.	Burgaz	Tancepa tarım ürünleri Ltd. Şti., Bandırma, Balıkesir	Red bulb with red outer skin.					
15.	Hazar	Tancepa tarım ürünleri Ltd. Şti., Bandırma, Balıkesir	White bulb with yellow-brown outer skin.					

\*KG = Kısa gün (short day); UG = Uzun gün (long day)

### 2.1. Experiment design and statistical analyses

Screening experiment was conducted in completely randomized block design with 3 replicates containing 30 plants in each. One replicate was also used as control. Ten onion seedlings were planted on each pot. The pots (16 cm in height and 16 cm in diameter) were sterilized by washing with 5% sodium hypochlorite bleach (clorox), and then filled with sterilized soil mix of perlite and peat moss, 1:3, respectively (Al-Shahwan et al., 2018). Onion plants were germinated from seeds since LYSV is not a seed-borne virus (Brunt et al., 1996).

Treated plants were dusted with abrasive-celite then mechanically inoculated with LYSV-12.6Po at 3-4 true leaf stage. Control plants were rubbed with Potassium phosphate buffer (pH 7) only, as described by Santosa and Ertunç (2020). Twelve weeks post-inoculation, onion plants were carefully harvested then evaluated based on two LYSV detection parameters: serological by ELISA, and molecular by RT-PCR; and five growth parameters: plant weight, plant height, root length, bulb diameter, and number of leaves. Statistical analyses on growth parameters using t-test and ANOVA continued with Duncan's Multiple Range Test (DMRT) were performed on Statistix ver. 8.1 (Analytical Software, USA).

### 2.2. LYSV detection in treated plants by ELISA and RT-PCR

Since there are total of 1350 treated plants, it is impractical to identify LYSV infection on each of them. Thus, ELISA and RT-PCR were used only to confirm tested cultivars susceptibility to LYSV. A small piece of 25 mg leaf was taken from each of 90 treated plants of each cultivar, then they were put together to create a composite sample of each cultivar to be tested in ELISA and RT-PCR.

Serological test using LYSV specific ELISA kit was performed according to the manufacturer instructions (Bioreba, Switzerland). The results were evaluated using criteria and equipment that were described by Santosa and Ertunç (2020).

Tris-EDTA buffer based procedures were applied to extract total nucleic acids from composite samples (Presting et al., 1995). The obtained nucleic acids were then used in two steps RT-PCR (Santosa and Ertunç, 2020). A pair of specific primer, F-5' TCACTGCATATGCGCACCAT 3' and R-5' GCACCATACAGTGAATTGAG 3' was applied in the PCR to amplify 1020 bp fragment of LYSV partial Nib+CP region (Fajardo et al., 2001).

# 3. Results

Only nine cultivars were tested positive for LYSV infection by ELISA. On the other hand, RT-PCR gave positive reactions to LYSV for all 15 tested cultivars (Figure 1 and Table 2).



Figure 1. RT-PCR amplified 1020 bp fragment of partial LYSV NIb+CP region on 15 cultivars, thus confirmed LYSV infection on all tested cultivars. a. Anka, b. KG kahverengi, c. KG kırmızı, d. UG kahverengi, e. UG kırmızı, f. UG beyaz, g. Şampiyon, h. Gence, i. İnci, j. Naz, k. Perama, l. Taraz, m. Seyhan, n. Burgaz, o. Hazar, p. Negative control (healthy onion), M. 100 bp marker (GeneDirex, Taiwan)

LYSV inoculation caused 12.4 - 51.7% plant weight, 4.2 - 46.9% plant height, 2.7 – 47.3% root length, 16.5 - 42.2% bulb diameter, and 1.2 – 26.8% no. of leaves reductions in all cultivars except in Seyhan (bulb diameter) (Table 2 and Figure 2). According to t-test, all of cultivars showed significant reduction on their weights upon inoculation by LYSV. All cultivars also showed significant differences with their respective controls in four other parameters, except KG kırmızı (plant height), UG kırmızı (root length), Şampiyon (root length and no. of leaves), Gence (root length), and Seyhan (bulb diameter) (Table 2).

ANOVA which continued with DMRT showed that, in responses to LYSV inoculation, Şampiyon, Perama, and Hazar did not show significant differences to each other in all five growth parameters. UG kahverengi and KG kahverengi which both produce brown colored bulbs were not significantly different to each other in all growth parameters except no. of leaves. Şampiyon, UG beyaz, Perama, and Gence were not significantly different to each other and the best performers in plant height and no. of leaves (Table 2).

15 onion cultivars	
LYSV-12.6Po to	
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Table 2. Effec	

ы П П	A R C P	+	+	+	+ +	+ +	+	+	+	+ +	+ +	+	+ +	+	+ +	+	un
	R (%)	24.7	20.3	5.3	15	11.3 -	5.9	1.2	2.4	- 22.5	16.5 -	3.7	26.8 -	10.3	14.4	13.1	in colu
No. of leaves*	Ι	$5.8\pm0.4^{g}$	$6.3\pm0.3^{g}$	$7.1\pm0.1^{de}$	$6.8\pm0.2^{\rm ef}$	$7.1\pm0.2^{cde}$	$8\pm0.1^{a}$	<b>8±0.1</b> <sup>a</sup>	$8\pm0.2^{\rm abc}$	$6.9\pm0.2^{def}$	$7.1\pm0.1^{de}$	$7.8{\pm}0.1^{\rm ab}$	$6\pm0.2^{g}$	$7\pm0.1^{\rm def}$	$7.1{\pm}0.1^{\rm de}$	$7.3\pm0.3^{bcd}$	e letter with
	С	7.7±0.5	7.9±0.6	7.5±0.7	$8{\pm}0.5$	$8{\pm}0.5$	$8.5{\pm}0.5$	<b>8.1±0.3</b>	$8.2{\pm}0.4$	$8.9{\pm}0.6$	$8.5{\pm}0.8$	$8.1{\pm}0.3$	$8.2{\pm}0.4$	$7.8 \pm 0.4$	8.3±0.5	$8.4{\pm}0.7$	s with sam
b diameter (mm)*	R (%)	37	42.2	27.6	34.7	31.6	31.8	20.5	23.8	35.8	16.5	22.6	23.2	ŀ	27.4	21.2	Mean
	Ι	$5.8\pm0.2^{g}$	6.7±0.3 <sup>cdef</sup>	$8.4{\pm}0.6^{a}$	$6.2\pm0.1^{efg}$	$6.5\pm0.2^{cdef}$	7.3±0.2 <sup>bc</sup>	$7\pm0.3^{bcd}$	$6.1{\pm}0.5^{\rm efg}$	$6.1{\pm}0.2^{\rm fg}$	$7.1\pm0.3^{\rm bc}$	7.2±0.2 <sup>bc</sup>	$6.3\pm0.3^{defg}$	<b>7.6±0.2</b> <sup>ab</sup>	6.9±0.3 <sup>bcde</sup>	$6.7\pm0.2^{cdef}$	(5) (n = 100);
Bu	С	9.2±0.9	$11.6\pm 2.4$	$11.6 \pm 1.8$	9.5±1.3	9.5±0.8	$10.7 \pm 1.5$	$8.8\pm1$	$8{\pm}1.2$	9.5±0.7	8.5±0.7	9.3±0.8	8.2±0.4	<b>7.1±0.7</b>	9.5±1.3	8.5±1.6	/alue > 0.0
	R (%)	5.8	38.5	43.5	31.3	3.2	16.7	10.3	2.7	37.2	12	20.5	47.3	38.6	15	9.2	est (p v
Root length (cm)*	I	$4.9\pm1^{\mathrm{efg}}$	$4.8\pm0.2^{\rm efg}$	$5.2\pm0.2^{def}$	$4.6\pm0.2^{\mathrm{fg}}$	<b>6.1±0.2</b> <sup>bcd</sup>	$7.5\pm0.3^{a}$	5.2±0.2 <sup>def</sup>	<b>7.3±0.3</b> <sup>ab</sup>	$4.9\pm0.7^{defg}$	7.3±0.6 <sup>ab</sup>	5.8±0.8 <sup>cde</sup>	3.9±0.4 <sup>g</sup>	$5.1\pm0.1^{\text{def}}$	$6.8\pm0.3^{\rm abc}$	5.9±0.1 <sup>cde</sup>	ifferent in t-t
	C	5.2±2.6	7.8±1.5	$9.2 \pm 1$	$6.7{\pm}1.5$	$6.3\pm 2$	9-0∓6	5.8±1.5	7.5±1.4	7.8±1.7	$8.3{\pm}0.9$	$7.3\pm1.4$	74±1.2	$8.3{\pm}1.8$	$8{\pm}1.5$	$6.5 \pm 1.4$	nificantly d
	R (%)	24.5	25.2	4.2	24.7	30.7	20	10.5	19.9	46.9	19.2	17.7	31.4	16	24.3	27.1	not sig
ant height (cm)*	Ι	$26.5\pm1^{defg}$	$25.2\pm1.1^{\mathrm{fg}}$	<b>32±0.7</b> <sup>a</sup>	$26.2{\pm}0.1^{\rm efg}$	$25.7\pm0.4^{fg}$	31.2±0.6 <sup>abc</sup>	$31.5\pm0.7^{ab}$	28.6±1.4 <sup>cde</sup>	$25\pm0.9^{\mathrm{fg}}$	$26.5{\pm}1.6^{def}$	$28.9\pm0.9^{bcd}$	$24.3\pm1.4^{g}$	$27.2\pm1^{def}$	$26.8{\pm}0.4^{\rm def}$	$28.3\pm1.3^{de}$	in bold are
Ы	С	35.1±5.1	33.7±4.5	33.4±5	$34.8\pm3.3$	37.1±2.9	39±5	35.2±4.4	35.7±5.1	47.1±3.6	32.8±3.8	$35.1 \pm 3.2$	35.4±2.1	32.4±2.5	35.4±4.6	38.8±7.4	ans printed
Plant weight (gram)*	R (%)	30.3	26.4	26	26.7	38.9	33.9	12.4	30.3	51.7	29.2	20.2	45.9	29.6	31.3	29.9	on; Me
	I	$6.2\pm0.2^{g}$	$6.4{\pm}0.1^{\mathrm{fg}}$	7.4±0.2 <sup>de</sup>	$6.6\pm0.1^{\mathrm{fg}}$	$6.6\pm0.2^{\mathrm{fg}}$	8.4±0.1 <sup>cde</sup>	$10.6\pm 1.2^{a}$	8.3±0.5 <sup>cde</sup>	$7\pm0.3^{\rm fg}$	$8.5\pm0.5^{bcd}$	$10.3\pm0.7^{a}$	$7.2\pm0.3^{\rm ef}$	$8.8\pm0.2^{\mathrm{bc}}$	$8.8\pm0.4^{\mathrm{bc}}$	9.6±0.7 <sup>ab</sup>	R =Reducti
	C	$8.9{\pm}1.1$	$8.7{\pm}1$	$10\pm 1.3$	$9{\pm}1.2$	$10.8 \pm 0.8$	12.7±1.7	12.1±1.1	$11.9\pm 1.6$	14.5±1.3	$12\pm 2.3$	12.9±1.5	$13.3\pm0.6$	12.5±1.5	12.8±1.5	$13.7\pm 1$	inoculated;
Cultivar name*		Anka	KG kahverengi	KG kırmızı	UG kahverengi	UG kırmızı	UG beyaz	Şampiyon	Gence	İnci	Naz	Perama	Taraz	Seyhan	Burgaz	Hazar	*C = control; I = ]

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Figure 2. Reactions of some cultivars to LYSV-12.6Po inoculation. A. Control (C) and inoculated (I) plants of Inci and UG kırmızı at 8 weeks post-inoculation. B. Heights of control (C) and inoculated (I) plants of Inci were measured at 12 weeks post-inoculation. C. Bulb diameters of control (C) and inoculated (I) plants of Inci were measured at 12 weeks post-inoculation.

#### 4. Discussion and Conclusion

Currently, at least 167 species infecting a wide variety of monocots and dicots belong to *Potyvirus*, making it the largest plant virus genus. Members of *Potyvirus* are transmissible mechanically, by grafting, and also non-persistently by 200 species of aphids, which allow them to spread to every corner of tropical and subtropical regions (Wylie et al., 2017). Considerable yield losses on various crops caused by *Potyvirus* had been recorded. Output reduction by *Bean common mosaic virus* (BCMV), a prevalent virus on beans (*Phaseolus vulgaris* L.), reached up to 100% (Worrall et al., 2015). LYSV lessened garlic bulb weight and perimeter up to 28% and 9%, respectively, under greenhouse conditions. In the field experiment of the same study, the virus decreased bulb weight and perimeter up to 36% and 13%, respectively (Lunello et al., 2007).

Approaches for *Potyvirus* management mainly fall into three major categories: control of aphids vector, cross protection, and resistant/tolerant varieties. Aphids vector management are not economically viable to be employed in large farming areas. Besides that, the method usually involves synthetic insecticides which could contaminate fields and produces. Cross protection which utilizes mild strain of a virus to induce plant resistance against more virulent strain cannot be applied in all

plant species and against all members of *Potyvirus*. Even if cross protection was successfully developed, the difficulty to provide sufficient mild strain inoculated seedlings to farmers potentially increases production costs. Because of these reasons, plant resistance is considered the most suitable option in *Potyvirus* diseases management (Sharma et al., 2013).

Current plant breeding process has been accelerated by the advent of DNA marker techniques. Random amplified polymorphic DNA (RAPD) and restriction fragment length polymorphism (RFLP) provided "Marker-assisted selection (MAS)" that greatly helps in making accurate selection of viral resistance genotypes in plant species (Collard and Mackill, 2008; Akçalı Giachino and İnan, 2019). Further application of genetic engineering techniques developed SunUp and Rainbow, two papaya (*Carica papaya* L.) varieties highly resistant to *Papaya ringspot virus* (PRSV), a *Potyvirus*, and widely planted in Hawaii, USA (Gonsalves et al., 2010). However, the use of such transgenic varieties is mostly confined only in certain areas due to concern and regulations regarding environmental issues and food safety (Azad et al., 2014).

Two Potyviruses: LYSV and *Onion yellow dwarf virus* (OYDV) are infecting and causing sizeable damage on *Allium* crops worldwide (Lot et al., 1998; Shahraeen et al., 2008; Ward et al., 2009; Elnagar et al., 2011), but resistance against them is still severely lack studied. All 20 leek varieties were found to be susceptible, but Autumn Giant Triumphator, Ligina, Siegfried Frost, and Winta are the most tolerant against LYSV according to a study in Denmark (Paludan et al., 1980). All of tested onion cultivars in this study were also proven by ELISA and RT-PCR to be susceptible to LYSV infection through mechanical inoculation. However, it also should be noticed that LYSV infection in KG kirmizi, UG beyaz, Şampiyon, Perama, Seyhan, and Hazar could only be detected by RT-PCR, which indicated low concentration of LYSV in these cultivars. PCR had been demonstrated as a more sensitive means for *Allium* viruses detection than ELISA (Dovas et al., 2001).

These results also provided further evidences that low LYSV infection rate among onions in Ankara province (13.3%) was mainly due to absence of aphids vector (Santosa and Ertunç, 2020), not resistance of onion varieties. LYSV was not widespread among susceptible onion varieties probably due to the fact that although it has been repeatedly shown to be mechanically transmitted through sap in experimental conditions (including by this study), the virus seems rather difficult to be mechanically transmitted by mere onion plant contacts in nature. LYSV is locally spread mainly by aphids, and transmission over long distance was thought mostly done by infected propagative materials (Lunello et al., 2002).

LYSV inoculation was shown to affect onion root length less than other growth parameters, since there are three cultivars that had no significant means differences to their respective control in root length, according to t-test. However, when percentage of means reductions were also put into consideration, LYSV inoculation seems to harm all growth parameters (more than 40% reduction in some cultivars), except no. of leaves (maximum reduction was only 26.8% on Taraz).

There are likely correlations among cultivars that were produced by same company in their reactions to LYSV. Şampiyon and Perama (both yield white bulb with yellow-brown outer skin and were produced by Tancepa Tarım Ürünleri Ltd. Şti.) performed better in plant weight, plant height, and no. of leaves than most other cultivars, according to DMRT. On the other hand, UG kahverengi and KG kahverengi (white bulb with dark brown outer skin, produced by Bayram Tohum) demonstrated worse reactions than most other cultivars in plant weight, plant height, root length, and no. of leaves based on DMRT calculation. UG kahverengi and KG kahverengi also showed considerable reduction in plant weight, plant height, root length, and no. of leaves, respectively, upon LYSV inoculation. Anka (white bulb with dark brown outer skin, produced by Aka Ziraii Ürünleri Ltd. Şti.) was the worst performer in plant weight, onion diameter, and no. of leaves. It is also worth mentioned that Taraz (white bulb with dark brown outer skin, produced by Tancepa Tarım Ürünleri Ltd. Şti.) was the worst performer in plant weight and root length, according to DMRT, and had >40% reductions in the both growth parameters.

In general, four cultivars that produced white bulb with dark brown outer skin (Anka, Taraz, UG kahverengi, and KG kahverengi) performed badly in all growth parameters. Some cultivars that produced white bulb with yellow-brown outer skin (Şampiyon, Perama, and Hazar) performed very well in plant weight, plant height, and no. of leaves. Burgaz, a cultivar that produced red bulb with red outer skin, performed at average level in all growth parameters after LYSV inoculation. KG kırmızı and UG kırmızı which both yield red bulb with red outer skin, and were produced by Bayram Tohum

were only not significantly different to each other in root length and no. of leaves. These results indicated that there are rather large genetic variations to LYSV resistance among germplasm collections belong to different seed companies in Turkey.

Based on this study's findings, it can be suggested that Şampiyon and Perama had the best reactions to mechanical inoculation of LYSV under greenhouse conditions. Among all tested cultivars, both are considered as the most tolerant to LYSV since the virus infection on them was only detected by sensitive RT-PCR, they had low means reduction in all growth parameters, and t-test and DMRT results showed that they performed better than most other cultivars in three growth parameters.

All of tested onion cultivars need to be further examined for their reactions to LYSV under field conditions, and under mix infection with other viruses. Resistance genes to *Potyvirus*, which probably could cover both LYSV and OYDV, in onion and other *Allium* spp. cultivars in Turkey also need to be mapped using DNA marker techniques. To our knowledge, such screening study using advanced genetic technology has never been done before on *Allium* spp.

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### References

- Adams, M. J., Antoniw, J. F., & Beaudoin, F. (2005). Overview and analysis of the polyprotein cleavage sites in the family *Potyviridae*. *Mol. Plant Pathol.*, *6*, 471-487.
- Akçalı Giachino, R. R., & İnan, D. (2019). Assessment of genetic diversity in Safflower (Carthamus tinctorius L.) using RAPD markers. Yuzuncu Yil University Journal of Agricultural Sciences, 29(2), 300-308.
- Al-Shahwan, I. M., Santosa, A. I., Abdalla, O. A., & Al-Saleh, M. A. (2018). Behavior of watermelon cultivars after inoculation with *Watermelon mosaic virus* in Saudi Arabia. *Journal of Agricultural Science and Technology A*, 8, 306-314.
- Azad, M. A. K., Amin, L., & Sidik, N. M. (2014). Gene technology for *Papaya ringspot virus* disease management. *The Scientific World Journal*, 2014, 1-11.
- Brunt, A. A., Crabtree, K., Dallwitz, M. J., Gibbs, A. J., & Watson, L. (1996). Plant viruses online: Description and lists from the VIDE database. Leek yellow stripe *potyvirus*. <u>http://bio-mirror.im.ac.cn/mirrors/pvo/vide/descr437.htm</u>. Accessed date: 02.05.2020.
- Collard, B. C. Y., & Mackill, D. J. (2008). Marker-assisted selection: an approach for precision plant breeding in the twenty-first century. *Phil. Trans. R. Soc. B*, 63(1491), 557-572.
- Demir, İ. (2020). An Ethnobotanical study of medicinal plants used in Hizan District (Bitlis-Turkey). *Yuzuncu Yil University Journal of Agricultural Sciences*, 30(4), 732-741.
- Dovas, C. I., Hatzibukas, E., Salomon, R., Barg, E., Shiboleth, Y. M., & Katis, N. I. (2001). Comparison of methods for virus detection in *Allium* species. *J Phytopathol*, 149, 731-737. Dovas, C. I., & Vovlas, C. (2003). Viruses infecting *Allium* spp. in Southern Italy. *Journal of Plant Pathology*, 85(2), 135.
- Elnagar, S., El-Sheikh, M. A., & Abd El-Wahab, A. S. E. (2011). Effect of natural infection with *Onion yellow dwarf virus* (OYDV) on yield of onion and garlic crops in Egypt. *Journal of Life Sciences*, *5*, 634-638.
- Fajardo, T. V. M., Nishijima, M., Buso, J. A., Torres, A. C., Ávila, A. C., & Resende, R. O. (2001). Garlic viral complex: identification of Potyviruses and Carlavirus in central Brazil. *Fitopatologia Brasileira*, 26, 619-626.
- Fidan, H. (2010). Sarımsak, soğan ve pırasadaki virüs hastalıklarını saptanması ve Taşköprü 56 sarımsak tipinin en yaygın virüse karşı reaksiyonunun belirlenmesi (in Turkish). (PhD), Çukurova University, Adana, Turkey.
- Fidan, H., & Baloglu, S. (2009). First report of *Onion yellow dwarf virus* and *Leek yellow stripe virus* in garlic in Turkey. *Plant Dis.*, 93(6), 672.
- Gonsalves, D., Tripathi, S., Carr, J. B., & Suzuki, J. Y. (2010). Papaya ringspot virus. The Plant Health Instructor.

- Güvenç, İ. (2019). Türkiye'de soğan üretimi, fiyat değişimi, dış ticareti ve rekabet gücü (in Turkish). *Yuzuncu Yil University Journal of Agricultural Sciences*, 29(4), 634-640.
- Klukáčková, J., Navrátil, M., & Duchoslav, M. (2007). Natural infection of garlic (*Allium sativum* L.) by viruses in the Czech Republic. *J Plant Dis Protect*, 144(3), 97-100.
- Korkmaz, S, & Cevik, B. (2009). *Leek yellow stripe virus* newly reported in Turkey. *Plant Pathol.*, 58, 787.
- Lot, H., Chovelon, V., Souche, S., & Delecolle, B. (1998). Effects of onion yellow dwarf and leek yellow stripe viruses on symptomatology and yield loss of three French garlic cultivars. *Plant Dis.*, 82, 1381-1385.
- Lot, H., Chovelon, V., Souche, S., Delecolle, B., Etoh, T., & Messiaen, C. M. (2001). Resistance to *Onion yellow dwarf virus* and *Leek yellow stripe virus* found in a fertile garlic clone. *Acta Horticulturae*, 555, 243-246.
- Lunello, P., Di Rienzo, J., & Conci, V. C. (2007). Yield loss in garlic caused by *Leek yellow stripe* virus Argentinean isolate. *Plant Dis.*, 91, 153-158.
- Lunello, P., Ducasse, D. A., Helguera, M., Nome, S. F., & Conci, V. C. (2002). An Argentinean isolate of *Leek yellow stripe virus* from leek can be transmitted to garlic. *Journal of Plant Pathology*, 84(1), 11-17.
- Paludan, N. (1980). Virus attack an leek: Survey, diagnosis, tolerance of varieties and winter hardiness. *Institut for Plantepatologi*, 1518, 371-385.
- Presting, G. G., Smith, O. P., & Brown, C. R. (1995). Resistance to *Potato leafroll virus* in potato plants transformed with the coat protein gene or with vector control constructs. *Phytopathology*, 85, 436-442.
- Santosa, A. I., & Ertunç, F. (2020). Serological and molecular detection of *Leek yellow stripe virus* infecting onion (*Allium cepa* L.) and leek (*Allium ampeloprasum* L.) in Ankara, Turkey. *Bitki Koruma Bülteni*, 60(1), 49-56.
- Sarı, M., Karanfil, A., & Korkmaz, S. (2020). Çanakkale ilinde Leek yellow stripe virus enfeksiyonunun güncel durumu ve iki farklı gen bölgesine göre kısmi moleküler karakterizasyonu (in Turkish). Çanakkale Onsekiz Mart University Journal of Graduate School of Natural and Applied Sciences, 6(1), 39-48.
- Sevik, M. A., & Akcura, C. (2013). Viruses occurring in onion crop in Amasya province, the major onion producing region in Turkey. *Indian Journal of Virology*, 24(1), 78-81.
- Shahraeen, N., Lesemann, D. E., & Ghotbi, T. (2008). Survey for viruses infecting onion, garlic and leek crops in Iran. *Bulletin OEPP/EPPO Bulletin, 38*, 131-135.
- Sharma, P., Sahu, A. K., Verma, R. K., Mishra, R., Choudhary, D. K., & Gaur, R. K. (2013). Current status of *Potyvirus* in India. *Arch Phytopathology Plant Protect*, 47(8), 906-918.
- Tuzlalı, H. T. (2018). Güney Marmara Bölgesi'nde Allium cinsi bitkilerde Potyvirüslerin tanılanmasi ve karakterizasyonu (in Turkish). (PhD), Çanakkale Onsekiz Mart University, Graduate School of Natural and Applied Sciences, Çanakkale, Turkey.
- TUIK (Türkiye İstatistik Kurumu/Turkish Statistical Institute). (2020). <u>http://www.tuik.gov.tr/Start.do</u> Accessed date: 11.05.2020.
- Vučurović, I., Nikolić, D., Radović, N., Vučurović, A., Ristić, D., Krstić, B., & Stanković, I. (2017). Incidence and distribution of *Leek yellow stripe virus* in *Allium* crops in Serbia. *Pesticides and Phytomedicine (Belgrade)*, 32(3-4), 145–155.
- Vučurović, I., Vučurović, A., Nikolić, D., Bulajić, A., Milošević, D., Krstić, B., & Stanković, I. (2016). First report of *Leek yellow stripe virus* in leek in Serbia. *Plant Dis.*, 100(1), 230.
- Ward, L. I., Perez-Egusquiza, Z., Fletcher, J. D., & Clover, G. R. G. (2009). A survey of viral diseases of *Allium* crops in New Zealand. *Australas Plant Pathol.*, *38*, 533-539.
- Worrall, E. A., Wamonje, F. O., Mukeshimana, G., Harvey, J. J. W., Carr, J. P., & Mitter, N. (2015). Chapter One - *Bean common mosaic virus* and Bean common mosaic necrosis virus: Relationships, Biology, and Prospects for Control. *Adv Virus Res*, 93, 1-46.
- Wylie, S. J., Adams, M., Chalam, C., Kreuze, J., Lopez-Moya, J. J., Ohshima, K., Praveen, S., Rabenstein, F., Stenger, D., Wang, A., & Murilo Zerbini, F. (2017). ICTV Virus Taxonomy Profile: *Potyviridae*. J. Gen. Virol., 98(3), 352–354.