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The Effects of Seed Infestation by *Fusarium proliferatum* on Root and Crown Rot, Plant Growth and Phenolic Compounds in Roots of Some Pumpkin (*Cucurbita pepo* L.) Cultivars

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ABSTRACT

This study investigates the reactions of four summer pumpkin cultivars (cvs. Çağlayan, Mert Bey, Sena Hanım, TG38) to root and crown rot caused by *Fusarium proliferatum* by taking into account criteria such as disease severity, plant growth (number of leaves, height, dry and fresh weight of shoot) and the accumulation of phenolic compounds in the roots. Seeds of each cultivar were inoculated with the pathogen and left to develop for 1 month at 25°C in a controlled climate room. The content of phenolic compounds in ethanolic root extracts was determined using high-performance liquid chromatography (HPLC). Cv. Sena Hanım had the lowest disease severity (4.40%) among the cultivars, followed by cvs. Çağlayan (10.62%) and Mert Bey (11.07%). Plants developed from inoculated seeds of cvs. Çağlayan and Sena Hanım had no decrease in the number of leaves and in length, fresh and dry weight of shoots in comparison to the control (plants from non-inoculated seeds), while cv. Mert Bey demonstrated a decrease at very low rates in shoot fresh and dry weight (2.24% and 0.77%, respectively). The phenolic compound that exhibited the highest increase in root extracts of cv. Sena Hanım compared to the control among the cultivars was *p*-coumaric acid (6.57-fold). This study demonstrates that *p*-coumaric acid can play an important role in the resistance of pumpkin to seed infestation by *F. proliferatum*.

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Introduction

Pumpkin (*Cucurbita pepo* L.) in Cucurbitaceae family, is one of the most significant vegetables for human nutrition. Previously, seed-borne fungi such as *Alternaria burnsii* and *Didymella bryoniae* were shown to cause seedling rot, although the disease severity rate was not recorded (Lee et al., 1984; Paul et al., 2015). In a recent study, it was reported that seed-borne *Fusarium proliferatum* caused root and crown rot in this plant at a rate of 51.07% (Demir et al., 2023). The improvement of eco-friendly control strategies is critical for long-term sustainability of pumpkin production since there is no information on the control of this disease. The majority of research on resistance to fungal diseases in *Cucurbita pepo* has focused on soil-borne fungi like *Fusarium solani* f. sp. *cucurbitae* (Nagao et al., 1994; Ayala-Doñas, et al., 2022) and *Phytophthora capsici* (Padley et al., 2008; Krasnow et al., 2017; Michael et al., 2019; Ayala-Doñas et al., 2022) as well as airborne fungi like *Sphaerotheca fuliginea* (Cohen et al., 1993, Cohen et al., 2003). However, there is no published information on the reactions of pumpkin cultivars to seed-borne fungi. In this context, it is also necessary to investigate some resistance mechanisms in pumpkin against *F. proliferatum*. Phenolic compounds are among the most important and prevalent secondary products in plants, and they have been shown to contribute to plant disease resistance (Nicholson and Hammerschmidt, 1992; Hammerschmidt and Smith-Becker, 1999; Cheynier et al., 2013).The presence of phenolic compounds such as caffeic acid, chlorogenic acid, ferulic acid, gallic acid, *p*-coumaric acid, protocatechuic acid (3,4-dihydroxybenzoic acid), sinapic acid, syringic acid and vanillic acid in pumpkin fruits has been determined, and their importance for human health was underlined (Dragovi-Uzelac et al., 2005; Kulczyński and Gramza-Michałowska, 2019), although there has been no study about the existence of any phenolics in roots of pumpkin. It has been suggested that the total amount of phenolic compounds in pumpkin leaves or fruits infected

with powdery mildew and yellow vein mosaic virus increased as a defense mechanism against these diseases (Jaiswal et al., 2013; Zhang et al., 2021). It has been proposed that foliar sprays of phenolics like chlorogenic acid may enhance resistance at an earlier stage of *Cucumber mosaic virus* infection in squash, which is a *C. pepo* variety, potentially controlling this viral disease (Abdelkhalek et al., 2022). The objectives of this study were to (a) determine disease severity in plants after inoculating seeds of four different pumpkin cultivars with *F. proliferatum*, (b) investigate the effect of pathogen on some plant growth parameters (leaf number, shoot length, shoot fresh and dry weight), and (c) investigate the role of phenolic compounds in roots in protecting different pumpkin cultivars against root and crown rot caused by this pathogen.

Materials and Methods

Pathogen and Cultivars

AYFA Tarım Limited Company (Bursa/Turkey) and the Trakya Agricultural Research Institute (Edirne/Turkey) provided seeds of a permitted (cv. TG38) and three registered (cvs. Çağlayan, Mert Bey, Sena Hanım) summer pumpkin (*Cucurbita pepo* L.) cultivars, respectively. A *Fusarium proliferatum* (FusP9) isolate was obtained from naturally infected pumpkin seeds. In a previous study, this isolate caused the highest disease severity (51.07%) in summer pumpkin seedlings (Demir et al., 2023). The isolate was cultured for 10 days on potato dextrose agar (PDA) at 25±2°C in the dark.

Pathogen Inoculation, Assessment of Disease Severity and Plant Growth Parameters

First, seeds of each cultivar were sterilized by immersing in a 2% NaClO solution for 7 minutes, subsequently washed three times with sterile distilled water, and air dried for 30 minutes on sterile paper towels. These seeds were pre-germinated in 9 cm petri dishes with filter paper moistened with sterile distilled water for three days at 25°C. Pre-germinated seeds (Erginbas-Orakci vd., 2016; Terna et al., 2022) were then inoculated with FusP9 isolate by immersing them in a suspension of conidia (1×10^6 conidia/ml) containing 0.1% (v/v) Tween 20 for 1 h at 25°C (Aslam et al., 2021; Demir et al., 2023). Sterile distilled water-treated seeds were used as control. Inoculated and non-inoculated seeds were sown separately in pots (12.5 × 10 cm, width x height, with 400 g mixture volume) filled with a mixture of peat (Klasman-Deihmann, Germany) and sand (3:1), which was sterilized in the autoclave. The plants were grown under a photoperiod of 12 h light (25°C) and 12 h darkness in a controlled room for 30 days. The treatments were set up in a randomized plot design with five replications (each pot had eight seeds).

Thirty days after inoculation, plants of the cultivars were gently uprooted from their respective pots and data were recorded in each replication regarding disease severity and some parameters for plant growth such as leaf number, length, fresh weight and dry weight (after drying 72 hours at 50°C) of shoots. Because varied symptoms were detected in developing plants, the severity of the disease was assessed using a 0-4 scale (Figure 1) based on a modified version of the scales reported by Jamiołkowska et al. (2012), Seo and Kim (2017) and Reyad et al. (2021): [0: Healthy, 1: Browning in roots, no browning of stem, 2: Browning in both roots and stems, 3: Post emergence damping-off, 4: Pre-emergence damping off]. The following formula (Townsend and Heuberger, 1943) was used to determine disease severity.

$$\mathrm{DS} = \frac{\sum(\mathrm{DL} \times \mathrm{NPDL})}{\mathrm{TNP} \times \mathrm{HSL}}$$

DS : Disease severity (%)
DL : Disease level
NPDL: Number of plants showing disease at that level
TNP : Total number of plants
HSL : The highest severity level

The pathogen was re-isolated from diseased plant areas and diagnosed using morphological characterization. The decreases (%) in root length, shoot length, shoot fresh and dry weight of each inoculated cultivar (C_p) with FusP were calculated relative to the average values for the non-inoculated (C) respective cultivar by following equation (Alisaac et al., 2018; Cong et al., 2018).

$$\text{The decrease in growth parameter (\%)} = \frac{\mathrm{C} - \mathrm{C_p}}{\mathrm{C}} \times 100$$

Figure 1. The evaluation of disease severity using 0-4 scale for the pumpkin cultivars inoculated with *Fusarium proliferatum*.
0: Healthy 1: Browning in roots, no browning of stem, 2: Browning in both roots and stems, 3: Post-emergence damping-off, 4: Pre-emergence damping-off

Extraction of Phenolic Compounds from Roots and HPLC Analysis

To assess the phenolic component content in roots, 1 g of sample was taken from each replicate and extracted with 32 mL of ethanol (99%) using an ultrasonic water bath for 30 minutes (Kulczyński and Gramza-Michałowska, 2019) and then for 24 hours in the dark. The extracts were sterilized using a 0.22 µm pore size membrane filter (Millipore, Millipore Co., Billerica, Ma, USA). The extracts directly were analyzed through the conditions used by Kulczyński and Gramza-Michałowska (2019) with High-Performance Liquid Chromatography (HPLC) for content of phenolic compounds such as cafeic acid, chlorogenic acid, 3,4-dihydroxybenzoic acid, gallic acid, *p*-coumaric acid, sinapic acid, syringic acid and vanillic

acid. The amounts of phenolic compounds in the roots of inoculated cultivars were compared to their respective controls. The increase (as fold) in each phenolic compound of each cultivar was calculated by dividing the amount of phenolic compound in each replicate of inoculated cultivar by the average amount of its non-inoculated control (Abdelrahman et al., 2016; Özer et al., 2021; Piasecka et al., 2022).

Statistical Analysis

The homogeneity of variances and normality of distribution were initially evaluated with SPSS version 23 (IBM SPSS Statistics for Windows, IBM Corp., Armonk, NY, USA) using Levene's and Shapiro-Wilk tests, respectively. The severity of disease in plants, the amounts of chlorogenic acid, vanillic acid, syringic acid and *p*-coumaric acids in the roots of inoculated and non-inoculated plants, and the increase in *p*-coumaric acid completely satisfied the assumptions of two-tailed student T test and the ANOVA test. The two-tailed student T test and two-tailed Wilcoxon Ranks test (at $P < 0.05$ and $P < 0.01$) were used to make comparisons for the amounts of phenolics in roots of inoculated and non-inoculated plants for each cultivar exhibiting parametric or non-parametric data, respectively. Significant differences in disease severity and *p*-coumaric acid increase of the cultivars were assessed using the Tukey HSD test at $P < 0.05$ with Software JMP Version Pro 17 for Windows (Cary, North Carolina, USA). Significant differences for the other non-parametric data such as decreases in plant growth parameters and increases in other phenolics, were analyzed with the Friedman Test ($P < 0.05$)), and the Wilcoxon rank-sum test was utilized for post-hoc analysis with IBM-SPSS. The means based on the decrease (%) in each growth parameter and the increase (fold) in each phenolic compound were compared with disease severity using Pearson's correlation coefficient at $P = 0.05$.

Results and Discussion

Disease Severity of Cultivars and Reductions in Plant Growth by Fusarium Proliferatum

The disease severity varied greatly between cultivars (Figure 2). The cv. Sena Hanım exhibited lowest disease severity (4.40%), and the differences between the disease severity of this cultivar and other cultivars were statistically significant. The highest disease severity with 14.17% was determined in cv. TG38, while the disease severities of cvs. Çağlayan (10.62%) and Mert Bey (11.07%) were in the same statistical group as cv. TG38. Considering the decreases in plant growth parameters measured in this study, the decreases in the number of leaves, shoot length, and the fresh and dry weights of shoots were considerably higher in inoculated TG38 than in other cultivars (Table 1). Although the decreased rates in fresh and dry weights of shoots in cv. Mert Bey were low, they were statistically significant as compared to cvs. Çağlayan and Sena Hanım. In previous studies, pumpkin cultivars or genotypes were only screened for resistance to *F. solani* f. sp. *cucurbitae* (Nagao et al., 1994; Ayala-Doñas et al., 2022), *P. capsici* (Padley et al., 2008; Krasnow et al., 2017; Michael et al., 2019; Ayala-Doñas et al., 2022) and *Sphaerotheca fuliginea* (Cohen et al., 1993, Cohen et al., 2003). It was recently found that the FusP9 isolate isolates used in the present study caused root and crown rot of 51.07% at 10 days after inoculation on seeds of the pumpkin cultivar TG22 (*C. pepo*) in petri dishes by blotter method. (Demir et al., 2023). In the current study, the pumpkin cultivars were tested for their susceptibility to *F. proliferatum* (FusP9 isolate) by seed infestation under pot conditions and the isolate exhibited disease severity percentages of 10.62%, 11.07%, 4.40%, and 14.17% on cvs. Çağlayan, Mert Bey, Sena Hanım and TG38, respectively.

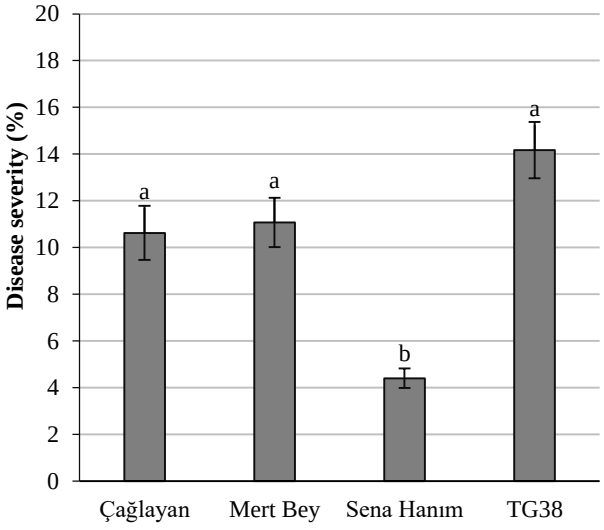

Figure 2. Disease severity caused by *Fusarium proliferatum* (FusP9 isolate) in different pumpkin cultivars. Each value represents the mean and standard error of five replications.
Bars topped by a different letter differ significantly according to the Tukey HSD test at $P<0.05$. $F_{3,16}= 16.4568$, $P<0.0001$

Phenolic Compounds in Roots of the Cultivars

Syringic acid and *p*-coumaric acid were detected in the roots of all cultivars (Figure 3). However, 3, 4-dihydrobenzoic acid was only present in the roots of cv. Çağlayan, exhibiting significantly higher amounts in the roots of inoculated plants (1.72 ppm) than those of non-inoculated plants (1.37 ppm, $P < 0.05$) (data not presented). The presence of chlorogenic acid and vanillic acid varied among cultivars. While the amount of chlorogenic acid (3.51 ppm), syringic acid (25.99 ppm) and *p*-coumaric acid (4.40 ppm) in inoculated cv. Sena Hanım was considerably greater than the control (1.34 ppm, 9.94 ppm and 0.67 ppm, respectively). The amounts of the same phenolic compounds, except syringic acid, in inoculated cv. TG38 were significantly lower (1.43 ppm and 0.82 ppm for chlorogenic acid and *p*-coumaric acid respectively) than its control (2.05 ppm and 1.20 ppm for the same phenolics, respectively). Interestingly, syringic acid also accumulated at higher rate (16.86 ppm) in the roots of inoculated cv. TG38 than in the control (14.55 ppm). Caffeic acid, gallic acid, and sinapic acid, which were detected in pumpkin fruit (Dragovi-Uzelac et al., 2005; Kulczyński and Gramza-Michałowska, 2019), were not found in the roots of the pumpkin cultivars examined in this study.

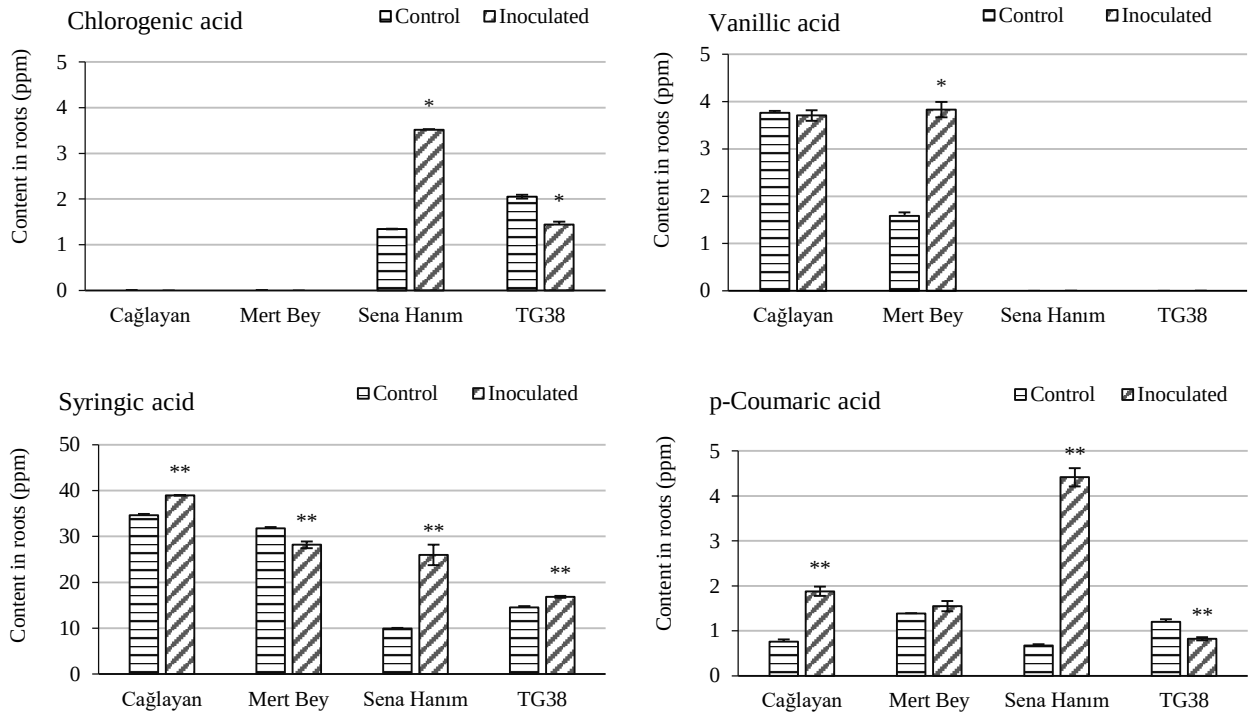

Figure 3. The phenolic compounds and their concentrations (ppm) in root extracts from seeds of different pumpkin cultivars inoculated with *Fusarium proliferatum* (FusP9 isolate) compared to mock-inoculated (control).
Each value indicates the mean and standard error of five replications. Significant differences between control and inoculated plants for each cultivar are indicated with the symbols "*" ($P < 0.05$) "**" ($P < 0.01$).

Table 1. Decreases (%) in some growth parameters of the cultivars from seeds inoculated with *Fusarium proliferatum* (FusP9 isolate) versus plants from non-inoculated seeds

Cultivars	Decrease (%) in[2]			
	Leaf number	Shoot length	Shoot fresh weight	Shoot dry weight
Çağlayan (Ça)	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
Mert Bey (MB)	0.00±0.00	0.00±0.00	2.24±0.18	0.77±0.06
Sena Hanım (SH)	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
TG38	13.13±2.35	5.18±0.42	13.95±3.06	20.45±2.53
P[1]	0.002	0.002	0.002	0.002
Pairwise	Significance of cultivar pairwise[3]			
Ça-MB	ns	ns	*	*
Ça-SH	ns	ns	ns	ns
Ça-TG38	*	*	*	*
MB-SH	ns	ns	*	*
MB-TG38	*	*	*	*
SH-TG38	*	*	*	*

[1]Friedman test; [2]Each value indicates the mean±standard error (SE) of five replications; [3]Based on Wilcoxon rank-sum test, significant differences between cultivar pairwise comparisons for each growth parameter are indicated with the symbol "*" ($P < 0.05$); ns: not-significant

Based on the increases in syringic and *p*-coumaric acid, which were found in the roots of all treated cultivars, versus roots from the control, the highest increase was *p*-coumaric acid (6.57-fold) in the roots of cv. Sena Hanım, followed by cv. Çağlayan (Table 2). An increase in *p*-coumaric acid was not detected in cv. TG38. The increase in syringic acid in the roots of cv. Sena Hanım was considerably greater than in the other cultivars, but it was also significantly higher in the roots of cv. TG38 having the disease severity than in those of cv. Mert Bey. The increases in chlorogenic and vanillic acid were significantly higher in cvs Sena Hanım ($P < 0.05$) and Mert Bey ($P < 0.05$), respectively, than in the other cultivars (TG38 and Çağlayan, respectively), which contain these phenolics. Phenolic compounds serve an important role in plant defense against diseases. They are also induced in response to injury and may play a role as precursors for lignin and suberin production (Matern and Kneusel, 1988; Nicholson and Hammerschmidt, 1992; Dixon and Paiva, 1995; Hammerschmidt and Smith-Becker, 1999). A previous report (Huang and Backhouse, 2005) indicated that total phenolics in roots of sorghum seedlings inoculated with *F. proliferatum* were increased as a defense response. Increase in *p*-coumaric acid only showed significant negative correlation with disease severity ($r = -0.985$, $P < 0.05$). As a result, the increased amount of *p*-coumaric acid in the diseased roots of cv. Sena Hanım, which has the least disease severity, appears to contribute to resistance against *F. proliferatum.* To the best our knowledge, this is the first resistance study of root and crown rot occurred after seed infestation by *F. proliferatum* in summer pumpkin (*C. pepo*) cultivars.

Table 2. Increase in accumulation of phenolic compounds in roots from seeds inoculated with *Fusarium proliferatum* (FusP9 isolate) versus roots from non-inoculated seeds

Cultivars	Increase (fold) in[2]				
	3,4-Dihydrobenzoic acid	Chlorogenic acid	Vanillic acid	Syringic acid	*p*-Coumaric acid
Çağlayan (Ça)	1.25±0.05	-[3]	0.00±0.00	1.12±0.01	2.49±0.14 b[5]
Mert Bey (MB)	-	-	2.42±0.10	0.00±0.00	1.12±0.08 c
Sena Hanım (SH)	-	2.62±0.01	-	2.61±0.22	6.57±0.30 a
TG38	-	0.00±0.00	-	1.16±0.01	0.00±0.00 d
P[1]	-	0.025	0.025	0.003	
Pairwise	-	Significance of cultivar pairwise[4]			
Ça-MB	-	-	*	*	
Ça-SH	-	-	-	*	
Ça-TG38	-	-	-	ns	
MB-SH	-	-	-	*	
MB-TG38	-	-	-	*	
SH-TG38	-	*	-	*	

[1]Friedman test; [2]Each value indicates the mean±standard error (SE) of five replications; [3]The compound is not present; [4]Based on Wilcoxon rank-sum test, significant differences between cultivar pairwise comparisons for each phenolic are indicated with the symbol "*" ($P < 0.05$). [5]The means with a different letter in the column differ significantly according to the Tukey HSD test at $P<0.05$ $F_{3,16}= 280.9147$, $P<0.0001$; ns: not-significant.

Conclusion

The results of our study suggest the importance of the measurement of phenolic compounds and plant growth parameters in resistance tests of pumpkin cultivars, in addition to disease severity. All cultivars of the current study demonstrated relatively low disease severity after seed infestation by *F. proliferatum*. However, in the cultivar with the lowest disease severity, we did not find any decrease in plant growth, number of leaves, shoot length, and fresh and dry weights of shoots. Moreover, *p*-coumaric acid among phenolic compounds increased at the highest amount in this cultivar. These finding may be help establish strategies to develop resistant pumpkin cultivars to *F. proliferatum*. In the future, it will be required to investigate the antifungal action of *p*-coumaric acid on this pathogen.

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Authors'Contributions

This article is a part of Master Science Thesis by Ebru Sevinç. Nuray Özer is a supervisor.

References

Abdelhalek A, Király L, Al-Mansori AA, Younes HA, Zeid A, Elsharkawy MM, Behiry S. 2022. Defense responses and metabolic changes involving phenylpropanoid pathway and PR genes in squash (*Cucurbita pepo* L.) following *Cucumber mosaic virus* infection. Plants, 11: 1908. doi: 10.3390/plants11151908

Abdelrahman M, Abdel-Motaal, F, El-Sayed M, Jogaiah S, Shigyoa M, Ito SI, Tran LSP. 2016. Dissection of *Trichoderma longibrachiatum*-induced defense in onion (*Allium cepa* L.) against *Fusarium oxysporum* f. sp. *cepa* by target metabolite profiling. Plant Science, 246: 128-138. doi: 10.1016/j.plantsci.2016.02.008

Alisaac E, Behmann J, Kuska MT, Dehne HW, Mahlein AK. 2018. Hyperspectral quantification of wheat resistance to *Fusarium* head blight: comparison of two *Fusarium* species. European Journal of Plant Pathology, 152: 869-884. doi: 10.1007/s10658-018-1505-9

Aslam HMU, Ali S, Aslam S, Ali MA, Khan NA, Zhai Y, Gleason ML. 2021. First report of leaf spot of bottle gourd (*Lagenaria siceraria*) caused by *Fusarium equiseti* in Pakistan. Journal of Plant Pathology, 103: 397-398. doi: 10.1007/s42161-020-00730-z

Ayala-Doñas A, Gómez P, de Cara-García M. 2022. Tolerance screening for *Phytophthora capsici* and *Fusarium solani* f. sp. *cucurbitae* in *Cucurbita* spp. and gene expression responses in mutant families. Horticulturae, 8: 191. doi: 10.3390/horticulturae8030191

Cheynier V, Comte G, Davies KM, Lattanzio V, Martens S (2013). Plant phenolics: Recent advances on their biosynthesis, genetics, and ecophysiology. Plant Physiology and Biochemistry, 72: 1-20. doi: 10.1016/j.plaphy.2013.05.009

Cohen R, Hanan A, Paris, HS. 2003. Single-gene resistance to powdery mildew in zucchini squash (*Cucurbita pepo*). Euphytica, 130: 433-441. doi: 10.1023/A:1023082612420

Cohen R, Leibovich G, Shtienberg D, Paris HS. 1993. Variability in the reaction of squash (*Cucurbita pepo*) to inoculation with *Sphaerotheca fuliginea* and methodology of breeding for resistance. Plant Pathology, 42: 510-516. doi: 10.1111/j.1365-3059.1993.tb01530.x

Cong LL, Sun Y, Wang Z, Kang JM, Zhang TJ, Biligetu B, Yang QC. 2018. A rapid screening method for evaluating resistance of alfalfa (*Medicago sativa* L.) to *Fusarium* root rot, Canadian Journal of Plant Pathology, 40: 61-69, doi: 10.1080/07060661.2017.1402822

Demir E, Özer N, Bayraktar H. 2023. Identification of seed-borne fungi in summer (*Cucurbita pepo*) and winter (*Cucurbita moschata*) pumpkins of Turkey. Journal of Plant Pathology, 105: 1087-1101. doi: 10.1007/s42161-023-01451-9

Dixon RA, Paiva NL. 1995. Stress-induced phenylpropanoid metabolism. Plant Cell, 7: 1085-1097. doi: 10.1105/tpc.7.7.1085

Dragovi-Uzelac V, Delonga K, Levaj B, Djakovic S, Pospisil J. 2005. Phenolic profiles of raw apricots, pumpkins, and their purees in the evaluation of apricot nectar and jam authenticity. Journal of Agricultural and Food Chemistry, 53: 4836-4842. doi: 10.1021/jf040494+

Erginbas-Orakci G, Poole G, Nicol JM, Paulitz T, Dababat AA, Campbell K. 2016. Assessment of inoculation methods to identify resistance to *Fusarium* crown rot in wheat. Journal of Plant Disease and Protection, 123: 19-27. doi: 10.1007/s41348-016-0001-8

Hammerschmidt R, Smith-Becker JA. 1999. The Role of Salicylic Acid in Disease Resistance. In Agrawal AA, Tuzun S, Bent E (editors). Induced Plant Defenses Against Pathogens and Herbivores: Biochemistry, Ecology, and Agriculture. Minnesota: American Phytopathological Society. pp. 19-36. ISBN: 978-0890542422 (Print).

Huang LD, Backhouse D. 2005. Induction of defence responses in roots and mesocotyls of sorghum seedlings by inoculation with *Fusarium thapsinum* and *F. proliferatum*, wounding and light. Journal of Phytopathology, 153: 522-529. doi: 10.1111/j.1439-0434.2005.01013.x

Jaiswal N, Singh M, Dubey RS, Venkataramanappa V, Datta D. 2013. Phytochemicals and antioxidative enzymes defence mechanism on occurrence of yellow vein mosaic disease of pumpkin (*Cucurbita moschata*). 3 Biotech, 3: 287-295. doi: 10.1007/s13205-012-0100-6

Jamiołkowska A, Wagner A, Sawicki K. 2012. Fungi colonizing roots of zucchini (*Cucurbita pepo* L. var. *giromontiina*) plants and pathogenicity of *Fusarium spp*. to zucchini seedlings. Acta Agrobotanica, 64: 73-78. doi: 10.5586/aa.2011.009

Krasnow CS, Hammerschmidt R, Hausbeck MK. 2017. Characteristics of resistance to *Phytophthora* root and crown rot in *Cucurbita pepo*. Plant Disease, 101: 659-665. doi: 10.1094/PDIS-06-16-0867-RE

Kulczyński B, Gramza-Michałowska A. 2019. The profile of secondary metabolites and other bioactive compounds in *Cucurbita pepo* L. and *Cucurbita moschata* pumpkin cultivars. Molecules, 24: 2945. doi: 10.3390/molecules24162945

Lee DH, Mathur SB. Neergaard P. 1984. Detection and location of seed-borne inoculum of *Didymella bryoniae* and its transmission in seedlings of cucumber and pumpkin. Phytopathologische Zeitschrift, 109: 301-308. doi: 10.1111/j.1439-0434.1984.tb00723.x

Matern U, Kneusel RE. 1988. Phenolic compounds in plant disease resistance. Phytoparasitica, 16: 153-170. doi: 10.1007/BF02980469

Michael NV, Fu Y, Meru G. 2019. Inheritance of resistance to *Phytophthora* crown rot in *Cucurbita pepo*. HortScience, 54: 1156-1158. doi: 10.21273/HORTSCI14021-19

Nagao H, Sato K, Ogiwara S. 1994. Susceptibility of *Cucurbita* spp. to the cucurbit root-rot fungus, *Fusarium solani* f. sp. *cucurbitae* race 1. Agronomie, 14: 95-102. doi: 10.1051/agro:19940204

Nicholson P, Hammerschmidt R. 1992. Phenolic compounds and their role in disease resistance. Annual Review of Phytopathology, 30: 369-389. doi: 10.1146/annurev.py.30.090192.002101

Özer N, Coşkuntuna A, Şabudak T. 2021. *Trichoderma harzianum*-induced defence in sunfower (*Helianthus annuus* L.) against *Plasmopara halstedii* with changes in metabolite profling of roots. BioControl Science and Technology, 31: 1403-1418. doi: 10.1080/09583157.2021.1963417

Padley LD, Kabelka EA, Roberts PD, French R. 2008. Evaluation of *Cucurbita pepo* accessions for crown rot resistance to isolates of *Phytophthora capsici*. HortScience, 43: 1996-1999. doi: 10.21273/HORTSCI.43.7.1996

Paul NC, Deng JX, Lee HB, Yu SH. 2015. Characterization and pathogenicity of *Alternaria burnsii* from seeds of *Cucurbita maxima* (Cucurbitaceae) in Bangladesh. Mycobiology, 43: 384-391. doi: 10.5941/MYCO.2015.43.4.384

Piasecka A, Sawikowska A, Witaszak N, Waśkiewicz A, Kańczurzewska M, Kaczmarek J, Lalak-Kańczugowska J. 2022. Metabolomic aspects of conservative and resistance-related elements of response to *Fusarium culmorum* in the grass family. Cells, 11: 3213. doi: 10.3390/ cells11203213

Reyad NEA, El-Sayed SF, Azoz SN. 2021. Evaluation of grafting using cucurbit interspecific hybrids to control *Fusarium* wilt in cucumber. Plant Cell Biotechnology and Molecular Biology, 22: 50-63. doi: 10.56557/pcbmb/2021/v22i37-386486

Seo Y, Kim YH. 2017. Potential reasons for prevalence of *Fusarium* wilt in oriental melon in Korea. Plant Pathology Journal, 33: 249-263. doi: 10.5423/PPJ.OA.02.2017.0026

Terna TP, Mohamed Nor NMI, Zakaria L. 2022. Histopathology of corn plants infected by endophytic fungi. Biology, 11: 641. doi: 10.3390/biology11050641

Townsend GR, Heuberger JW. 1943. Methods for estimating losses caused by diseases in fungicide experiments. Plant Disease Reporter, 27: 340-343

Zhang S, Liu J, Xu B, Zhou J. 2021. Differential responses of *Cucurbita pepo* to *Podosphaera xanthii* reveal the mechanism of powdery mildew disease resistance in pumpkin. Frontiers in Plant Science, 12: 633221. doi: 10.3389/fpls.2021.633221