KSÜ Tarım ve Doğa Derg 23 (6): 1428-1434, 2020
KSU J. Agric Nat 23 (6): 1428-1434, 2020
DOI:10.18016/ksutarimdoga.vi.714642

The Effects of UV Irradiation and Incubation Time on *in vitro* Phenolic Compound Production in 'Karaerik' Grape Cultivar

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ABSTRACT
In this study, the effect of Ultraviolet (UV) irradiation on induction of individual and total phenolics production on callus cultures of 'Karaerik' grape cultivar was investigated. Callus tissues were obtained from the leaves of the cuttings grown in *in vitro* plants. As a culture medium, Gamborg B-5 was utilized with 0.1 mg L^{-1} NAA (Naphthaleneacetic acid) and 0.2 mg L^{-1} Kin (Kinetin). Callus tissues were sub-cultured twice with 21 days intervals. After the second subculture, 12-day-old callus tissues were exposed to 254 nm UV-C light at 10 cm distance from the source for 10 and 15 min by opening covers of the petri dishes in sterile cabin. After the treatment, callus tissues were incubated under dark conditions. Phenolic compounds were measured at 24th, 48th and 72nd hours. Individual phenolic compounds were analyzed by HPLC (High Pressure Liquid Chromatography) and total phenolic compounds were measured by spectrophotometer. As a result of the study, it was found that UV irradiation was effective for induction the production of phenolic compounds in the callus tissues of 'Karaerik' grape cultivar and this effect was closely related to the application time.

Research Article

Article History
Received : 04.04.2020
Accepted : 04.06.2020

Keywords
Grapevine
Analytical methods
Elicitor
Callus culture
Secondary metabolite

'Karaerik' Üzüm Çeşidinde *in vitro* Fenolik Bileşik Üretimi Üzerine UV Işını Uygulaması ve İnkübasyon Sürelerinin Etkisi

ÖZET
Bu çalışmada, Karaerik üzüm çeşidine ait kallus kültürlerinde, bireysel ve toplam fenolik bileşik üretiminin uyarılması üzerine Ultraviyole (UV) ışınının etkisi incelenmiştir. Kallus kültürleri, *in vitro* bitkiciklerin yaprak ayalarından elde edilmiştir. Kültür ortamı olarak, 0.1 mg l^{-1} NAA (Naftalen asetik asit) ve 0.2 mg l^{-1} Kin (Kinetin) eklenmiş Gamborg B-5 ortamı kullanılmıştır. Kalluslar, 21 gün ara ile iki defa alt kültüre alınmıştır. İkinci alt kültürden sonra, 12 gün yaşlı kalluslara, steril kabin içerisinde petri kutularının kapakları açılarak, 10 cm uzaklıktan 10 ve 15 dk süreyle 254 nm dalga boyuna sahip UV-C ışını uygulanmıştır. Uygulamanın ardından kalluslar karanlık koşullarda inkübe edilmiştir. Fenolik bileşik ölçümleri 24., 48. ve 72. saatlerde yapılmıştır. Bireysel fenolik bileşikler HPLC (Yüksek Basınçlı Sıvı Kromatografisi), toplam fenolik bileşikler ise Spektrofotometre ile analiz edilmiştir. Çalışma sonucunda UV ışınının Karaerik üzüm çeşidine ait kallus dokularında fenolik bileşik üretimini uyarmada etkili ve bu etkinin uygulama süresi ile yakından ilişkili olduğu saptanmıştır.

Araştırma Makalesi

Makale Tarihçesi
Geliş Tarihi : 04.04.2020
Kabul Tarihi : 04.06.2020

Anahtar Kelimeler
Asma
Analitik yöntemler
Elisitör
Kallus kültürü
Sekonder metabolit

To Cite : Çelik M, Keskin N, Özdemir AF 2020. The Effects of UV Irradiation and Incubation Time on *in vitro* Phenolic Compound Production in 'Karaerik' Grape Cultivar. KSU J. Agric Nat 23 (6): 1428-1434. DOI: 10.18016/ksutarimdoga.vi. 714642.

INTRODUCTION
Grapes play an important role in human nutrition and synthesize phenolic compounds that one of the leading compounds of secondary metabolites. Phenolic compounds are secondary metabolites containing at least one aromatic ring and one hydroxyl group

attached and exhibit antioxidant activity due to their easily oxidizable properties. It is known that phenolic compounds are responsible for color, taste, and aroma attributed to grapes. These compounds are the most important components of quality and also have supportive effects on human nutrition and health (Kunter et al. 2013, Gökçen et al. 2017, Canturk et al. 2018). Although sugars and organic acids are the main quality components of grapes, phenolic compounds are also considered important constituents.

It is known that the presence and the ratio of certain phenolic substances in the composition of the grape is primarily a genetically controlled characteristic in varieties. In recent years, phenolic compounds were asserted as an important quality criterion for grape cultivars, and studies related with the determination of differences between them in terms of production capacity become popular (Rodríguez-Montealegre et al. 2006; Lutz et al. 2011; Perestrelo et al. 2012; Atak and Göksel 2019; Castro-Lopez et al. 2019; Colombo et al. 2019; Di Lorenzo et al. 2019). However, the content is characterized by depending on the climate, soil effects, maturity stage and cultural practices in the cultivation area (Ribéreau-Gayon et al. 2000). Therefore, it is necessary to benefit from controlled conditions to evaluate for obtaining the greater phenolic compounds per unit area. At the present time, many phenolic compounds are produced by *in vitro* cultures. Thus, callus and cell suspension cultures are widely used for such purpose (Bonello et al. 2019). Biotic and abiotic elicitors are used to increase the production and concentrations of *in vitro* phenolic compounds (Barz et al., 1988). Ultraviolet (UV) light is one of the most effective abiotic elicitors (Keskin, 2019). Furthermore, UV-C irradiation is an important factor that can act as a switch, controlling expression of specific genes involved in cell growth and secondary metabolism of plants (Versari et al., 2001).

Although phenolic compounds can be classified in different ways, they are generally categorized into two groups as flavonoids and non-flavonoids. Flavonoids include flavan-3-ols (tannins), flavonols, and anthocyanins. Non-flavonoids consist of phenolic acids and stilbenes (López-Vélez et al. 2003).

In this study *in vitro* phenolic compounds production capacity and the effect of UV irradiation on phenolic compounds via callus culture were determined in *Vitis vinifera* L. cv. Karaerik.

Thus, the total and some individual phenolic compounds production potential of 'Karaerik' grape cultivar was demonstrated and the effect of UV light on increasing this potential was evaluated in terms of irradiation and incubation times.

MATERIAL and METHODS

In the study, one-year old canes of 'Karaerik' grape cultivar were used as the primary plant material for the callus culture. The canes obtained from the pruning period of 2016 and 2017 were maintained in a climate chamber at 25 °C and 8/16 h photoperiod conditions.

In vitro shoot regeneration

At the beginning of the *in vitro* studies, single node cuttings were obtained from the canes and forced to grow in a climate chamber. Single node explants were cultured to obtain shoots which were the source of the *in vitro* leaf explants. Shoot proliferation studies were given as follow: Explants were disinfected for 15 min by using 10% sodium hypochlorite solution (0.5% NaOCl) with 1-2 drops 0.01% Tween 20. After disinfection, the materials were rinsed 3 times with sterile distilled water for at least 5 min. MS (Murashige and Skoog 1962) nutrient medium was used to obtain *in vitro* shoots. The pH of the prepared medium was set to 5.7 by adding 4.4 g L^{-1} MS medium. As a growth regulator, 1.0 mg L^{-1} BAP (6-benzylaminopurine) was added to increase shoot growth. The nutrient medium with sucrose (3%) and agar (6%) were sterilized in an autoclave (20 min at 121 °C). Cultured single node explants were incubated at 8/16 h photoperiod and 25 °C. *In vitro* shoots were obtained by sub-culturing process with 21 days interval until reaching a sufficient number of leaf explants. Then these leaf explants used for callus culture.

Establishment of callus cultures

Leaf blades obtained from *in vitro* plantlets were used as callus initial explants. Gamborg B-5 (Gamborg et al., 1968) culture media was prepared by was 3.2 g L^{-1} concentration with pure water. The pH of the nutrient medium was adjusted to 5.7. As growth regulators, 0.1 mg L^{-1} NAA and 0.2 mg L^{-1} Kin were added (Oğuz et al. 2020). Then, the nutrient medium with sucrose (30%) and agar (0.8%) were sterilized by autoclaving for 20 min at 121 °C. The cultures were incubated at 8/16 photoperiod and 25 °C and the calli were sub-cultivated twice with 21 day-intervals.

Elicitor treatment

In the study, as an elicitor, the effect of short wavelength UV-C light (254 nm) was investigated. Overall, 12-day-old cultures were exposed to UV irradiation for 10 and 15 minutes at a distance of 10 cm by opening the lid of the petri dishes in a sterile cabinet (Keskin and Kunter 2007, 2008, 2009, 2010).

The callus cultures performed UV irradiation were incubated at 25 °C in dark conditions for 24, 48 and 72 h. At the end of incubation, callus was weighed as 1 g, wrapped in aluminum foils and stored at -80 ºC until analysis. Controls were taken from 12-day-old cultures

as 1 g from each and samples were stored in aluminum foil at -80 ºC until analysis.

Determination of phenolic compounds and contents

Both total and individual phenolic compounds were determined in the study. For total phenolic compound analysis, 1 g of callus tissue was homogenized with 1 ml of methanol (Ika Ultra-Turrax T20 Basic, Germany) at the medium speed for 2 min and then stored under dark conditions for 30 min at room temperature. The samples were filtered through filter paper and taken into Eppendorf tubes and stored at -80 ºC until analysis. Total phenolic content was determined by reading the absorbance of the solutions in a spectrophotometer (Varian Bio 100, Australia) at 725 nm wavelength by Folin-Ciocaltaeu colorimetric method (Swain and Hillis 1959). Total phenolic content was expressed as mg g^{-1} gallic acid equivalent (GAE).

For the separation of individual phenolic compounds (protocatechuic acid, gallic acid, chlorogenic acid, *p*-coumaric acid, ferulic acid, *q*-coumaric acid, vanillic acid, rutin, syringic acid, phloridzin and *trans*-resveratrol) by HPLC, the method determined by Rodriguez-Delgado et al. (2001) was used and phenolic content was expressed as µg g^{-1} Fresh Weight (FW). Overall, 1 g of callus tissue was homogenized, 4 ml of methanol was added and centrifuged at 15000 rpm for 15 min. The supernatant was filtered through a 0.45 µm millipore filter and stored in brown sample bottles. Chromatographic separation was performed on the Agilent 1100 HPLC system using a Diode Array detector (DAD) and a 250*4.6 mm 4µm octadecylsiloxane (ODS) column. Solvent A Methanol-acetic acid-water (10:2:88) and solvent B Methanol-acetic acid-water (90:2:8) were used in the mobile phase. HPLC conditions for the separation of individual phenolic compounds were determined at 254 and 280 nm wavelength, the flow rate at 1 mL min^{-1} and injection volume at 10 µL.

Statistical analysis

In the study, descriptive statistics of total and individual phenolic compounds were expressed as "Mean ± Standard error of mean". Factorial Repeated Measured Analysis of Variance was performed to determine any differences between the irradiation time (10 and 15 min) and incubation time (24, 48 and 72 h). Following the analysis of variance, groups means were compared by Duncan's Multiple Range Test by SPSS at $P \leq 0.5$.

RESULTS and DISCUSSION

In this study, total and individual phenolic compounds obtained from the treatment of two UV irradiations (10 and 15 min) and three incubation times (24, 48 and 72 h) at 12-day-old calli of 'Karaerik' grape cultivar. Results indicated that there was a significant interaction of "irradiation time x incubation time" ($P < 0.05$) in the study.

The effects of UV irradiation and incubation times on total phenolic content are presented in Table 1. The value of 546.58 mg g^{-1} obtained in the control calli increased by 1.6 times to 885.75 mg g^{-1} after 24 h following 10 min of UV light induction and reached the highest value of 970.75 mg g^{-1} after 48 h. After 72 h, the total phenolic compounds decreased up to 804.08 mg g^{-1}. The difference between the incubation times was not significant (Table 1).

Total phenolic content, measured as 992.417 mg g^{-1} at 24th h for 15 min of UV irradiations, increased to 1009.08 mg g^{-1} at 48th h and decreased to 879.08 mg g^{-1} at 72nd h. The highest total phenolic content in callus was measured at 48th h and the lowest one was measured at 24th h of incubation. The differences between the incubation times were not significant (Table 1).

It was found that the total phenolic content obtained after 10 and 15 min UV irradiation were always higher than those of the control group and the difference was statistically significant ($p<0.05$). As compared the UV irradiation times, although the difference was not statistically significant, the irradiation of 15 min resulted in higher phenolic compound production (992.42 mg g^{-1}, 1009.08 mg g^{-1}, 879.08 mg g^{-1}; respectively), the highest value was obtained at 48th h with 15 min UV irradiation time.

The effect of irradiation and incubation times on individual phenolic content is presented in Table 2.

Table 1. Effect of UV irradiation and incubation times on total phenolic compound content (mg g^{-1})
Çizelge 1. UV ışını uygulama ve inkübasyon sürelerinin toplam fenolik bileşik içeriğine (mg g^{-1}) etkisi

UV Irradiation Time (min) *UV Uygulama Süresi (dk)*	Incubation Time (h) (*İnkübasyon Süresi (sa)*)		
	24	48	72
	Mean ± SEM *Ortalama±St. Hata*	Mean ± SEM *Ortalama±St. Hata*	Mean ± SEM *Ortalama±St. Hata*
10	885.75* ± 82.17	970.75* ± 94.95	804.08* ± 51.20
15	992.42* ± 101.35	1009.08* ± 106.68	879.08* ± 82.98
Mean of the control group: *Kontrol grubu ortalaması:*	546.58 ±19.65 *546.58±19.65*		

* The difference from the control is statistically significant ($p<0.05$). SEM: Standard error of mean

Table 2. Effect of UV irradiation and incubation times on individual phenolic compound content
Çizelge 2. UV ışını uygulama ve inkübasyon sürelerinin bireysel fenolik bileşik içeriğine etkisi

Individual Phenolics (μg g^{-1}) *Bireysel Fenolikler (μg g^{-1})*	Incubation Time (h) *İnkübasyon Süresi (sa)*	UV Irradiation Time (min) (*UV Işını Uygulama Süresi (dk)*)	
		10	15
		Mean ± SEM *Ortalama±St. Hata*	Mean ± SEM *Ortalama±St. Hata*
Protocatechuic acid	Control	7.47 ± 0.42	
	24	5.28 b * ± 0.65	6.59 * ± 0.14
	48	7.66 a ± 0.53	8.64 ± 0.97
	72	6.88 ab ± 0.20	7.84 ± 1.22
Vanillic acid	Control	13.37 ± 1.56	
	24	9.82 * ± 2.180	12.48 ± 0.98
	48	15.78 * ± 3.93	14.10 ± 0.51
	72	13.49 ± 1.95	11.91 ± 2.40
Rutin	Control	1.73 ± 0.35	
	24	2.29 * ± 0.08	2.38 * ± 0.94
	48	1.71 ± 0.291	2.18 ± 0.83
	72	1.75 ± 0.19	1.69 ± 0.33
Gallic acid	Control	0.81 ± 0.15	
	24	0.74 ± 0.13	1.31 ± 0.24
	48	0.99 ± 0.18	1.31 ± 0.32
	72	0.85 ± 0.11	0.84 ± 0.14
Chlorogenic acid	Control	4.57 ± 0.60	
	24	6.04 ± 1.25	10.14 * ± 3.30
	48	14.56 * ± 6.31	10.67 * ± 2.94
	72	6.88 ± 1.24	9.07 * ± 1.14
Syringic acid	Control	0.47 ± 0.06	
	24	0.31 ± 0.09	0.23 * ± 0.03
	48	0.30 * ± 0.04	0.33 ± 0.18
	72	0.35 ± 0.06	0.33 ± 0.08
p-coumaric acid	Control	0.13 ± 0.06	
	24	0.36 ± 0.15	0.71 * ± 0.17
	48	0.55 ± 0.29	0.82 * ± 0.33
	72	0.09 ± 0.04	0.37 * ± 0.18
Ferulic acid	Control	0.16 ± 0.09	
	24	0.41 b *± 0.14	1.27 * ± 0.60
	48	1.09 a * ± 0.17	1.28 * ± 0.46
	72	0.39 b * ± 0.07	1.12 * ± 0.50
q-coumaric acid	Control	0.07 ± 0.05	
	24	0.12 ± 0.02	0.09 ± 0.02
	48	0.17 ± 0.04	0.09 ± 0.01
	72	0.08 ± 0.03	0.08 ± 0.05
Phloridzin	Control	1.94 ± 0.98	
	24	2.37 ± 0.89	3.54 * ± 1.70
	48	3.05 ± 0.38	4.16 * ± 2.16
	72	2.17 ± 0.94	2.98 ± 0.46
trans-resveratrol	Control	2.77 ± 0.87	
	24	4.39 * ± 0.86	6.66 * ± 2.13
	48	5.42 * ± 1.39	7.09 * ± 2.84
	72	3.26 ± 0.54	4.74 * ± 1.93

a,b: The difference is statistically significant ($p < 0.05$). SEM: Standard error of mean
* The difference from the control is statistically significant ($p < 0.05$).

Protocatechuic acid content was measured as 7.47 μg g^{-1} (Table 2). This value decreased to 5.28 μg g^{-1} at the 24th h of 10 min UV irradiation and increased again at the 48th h. This was slightly above the control (7.66 μg g^{-1}) and decreased again at the 72th h (6.88 μg g^{-1}). The difference of the 24th h from the control and differences among the incubation times were found to be statistically significant. Similarly, to 10 min irradiation, content of protocatechuic acid in callus decreased (6.59 μg g^{-1}) within 24 h after 15 min of UV irradiation, increased again at 48th h and reached the maximum value of its group (8.64 μg g^{-1}) and decreased again at 72th h (7.84 μg g^{-1}). When both UV irradiation time were compared, it was determined that the

difference between irradiation times was not significant, however, the 15 min irradiation resulted in higher accumulation than the 10 min irradiation (Table 2). For both irradiation times, the highest protocatechuic acid content was observed at the 48th h.

Vanillic acid content of the callus was measured as 13.37 µg g^{-1} in the control group and this value decreased within 24 h for both UV irradiations (10 min: 9.82 µg g^{-1}; 15 min: 12.48 µg g^{-1}). Effect of the irradiation times were appeared at the 48th h and increased to 15.78 µg g^{-1} with the 10 min irradiation and to 14.10 µg $g^{-1}$with the 15 min irradiation. At the end of the 72 h incubation (10 min:13.49 µg g^{-1}; 15 min:11.91 µg g^{-1}), a decrease was observed (Table 2). At the 48th h, the highest vanillic acid production was obtained for both 10 min and 15 min UV irradiation times and 10 min irradiation was found more effective than 15 min irradiation.

The rutin content was recorded as 1.73 µg g^{-1} in control callus however, 10 min UV irradiation was effective at 24th h and the value increased 1.3 times (2.29 µg g^{-1}). For rutin content, interestingly a reduction was occurred (1.71 µg g^{-1}) even below the value of control at 48th h, however the value slightly increased to 1.75 µg g^{-1} at 72nd h. Similarly, after 15 min of UV irradiation and 24 h of incubation, the rutin content in the callus was measured as 2.38 µg g^{-1}. This value decreased to 2.18 µg g^{-1} after 48 h of incubation. At 72nd h, reduction was below those of the control group (Table 2). As compared both UV irradiation times for rutin, it was determined that the 10 min irradiation showed a higher accumulation than 15 min irradiation (Table 2). The highest rutin content was observed at 24th h in both irradiation times.

When Table 2 was examined in terms of gallic acid, the content was determined as 0.81 µg g^{-1} in the control callus, decreased to 0.74 µg g^{-1} at the end of the 24 h incubation with 10 min UV induction an increased to 0.99 µg g^{-1} after 48 h incubation. The content was recorded as 0.85 µg g^{-1} in final incubation (72 h). For 15 min UV irradiation, gallic acid content increased at 24th h, unchanged at 48th h and decreased to about that of control at 72nd h (0.84 µg g^{-1}). The best irradiation and incubation time in terms of gallic acid were the 24th and 48th h incubation times after the 15 min UV irradiation (Table 2).

Chlorogenic acid contents are presented in Table 2. When the callus were induced with UV for 10 min, the initial value of 4.57 µg g^{-1} reached 6.04 µg g^{-1} at the first 24 h incubation time, and reached the maximum value (14.56 µg g^{-1}) with an increase of 2.4 times after the second 48 h incubation time, and decreased rapidly to 6.88 µg g^{-1} after the final 72 h incubation time.

After 15 min UV irradiation, UV irradiation was effective immediately at 24 and 48 h, and chlorogenic acid content reached to 10.14 µg g^{-1} and 10.67 µg g^{-1}, respectively, with an approximate 2.3 times increased and then decreased to 9.07 µg g^{-1} at 72nd h. The best UV irradiation time was 10 min, while the best incubation time was 48 h (Table 2).

As shown in Table 2, syringic acid concentration was measured as 0.47 µg g^{-1} in the control callus. This value decreased to 0.31 µg g^{-1} after 24 h, and to 0.30 µg g^{-1} after 48 h of UV irradiation for 10 min. At 72nd h, the value (0.35 µg g^{-1}) was lower than control. The results of 15 min UV irradiation were similar that of 10 min UV irradiation. While the syringic acid amount decreased to 0.23 µg g^{-1} at 24 h, the value increased 1.4 times (0.33 µg g^{-1}) at 48 and 72 h (Table 2).

As seen in Table 2, after UV irradiation for 10 min, the *p*-coumaric acid content was observed as 0.13 µg g^{-1} in control callus and increased to 0.36 µg g^{-1} at 24th h and to 0.55 µg g^{-1} at 48th h. At 72nd h, the content of *p*-coumaric acid decreased to 0.09 µg g^{-1}. As a result of 15 min UV irradiation, the amount of *p*-coumaric acid (0.71 µg g^{-1}) started to increase at the first 24th h and reached to the highest value (0.82 µg g^{-1}) at 48th h, and the value decreased to 0.37 µg g^{-1} at 72nd h (Table 2). For *p*-coumaric acid, the best UV irradiation time was 15 min and the best incubation time was 48 h.

Ferulic acid was measured as 0.16 µg g^{-1} in the control callus. For 10 min UV irradiation, this value reached to 0.41 µg g^{-1} at 24th h, and to 1.09 µg g^{-1} at 48th h. At 72nd h, the content decreased to 0.39 µg g^{-1}. For 15 min UV irradiation, ferulic acid content was determined as 1.27 µg g^{-1} at 24th h. This value increased slightly (1.28 µg g^{-1}) at 48th h and decreased to 1.12 µg g^{-1} at 72nd h. For ferulic acid, the highest efficiency was obtained at the 48th h and 15 min UV irradiation.

The *q*-coumaric acid value was 0.07 µg g^{-1} in the control callus. This content increased 1.7 times and reached to 0.12 µg g^{-1} at 24th h for 10 min UV irradiation. The maximum value was obtained as 0.17 µg g^{-1} at 48th h. The content decreased to 0.08 µg g^{-1} at 72nd h. Similarly, for 15 min UV irradiation, *q*-coumaric acid concentration increased within 48th h (0.09 µg g^{-1} at 24 and 48 h) and decreased to 0.08 µg g^{-1} at 72nd h. The best combination was 10 min UV irradiation and 48 h incubation time for *q*-coumaric acid (Table 2).

Phloridzin content of in the control group was measured as 1.94 µg g^{-1}. This content reached to 2.37 µg g^{-1} at 24th h, and to 3.05 µg g^{-1} at 48th h for 10 min UV irradiation. At 72nd h, it decreased to 1.17 µg g^{-1}. For 15 min UV irradiation, an increase occurred at 48th h and a decrease at 72nd h. The phloridzin content was 3.54 µg g^{-1} in the first 24 h. The highest content was 4.16 48 µg g^{-1} at 48th h and this value decreased to 2.98 µg g^{-1} at 72nd h (Table 2). The best combination was 15 min UV irradiation and 48 h incubation time for phloridzin (Table 2).

The *trans*-resveratrol value was 2.77 µg g^{-1} in the control callus. For 10 min UV irradiation, this content

increased approximately 1.6 times and reached to 4.39 µg g^{-1} at 24th h. This increase continued to 5.42 µg g^{-1} at 48th h. Then decreased to 3.26 µg g^{-1} at 72 h. For 15 min UV irradiation, *trans*-resveratrol content was found 6.66 µg g^{-1} at 24th h, it increased to 7.09 µg g^{-1} which was the maximum value at 48th h, then decreased to 4.74 µg g^{-1} at 72nd h (Table 2). The best combination was 15 min UV irradiation and 48 h incubation time for *trans*-resveratrol.

In the study, UV irradiation was found to be an effective elicitor for callus cultures and phenolic compound production. This finding is in line with Keskin and Kunter, (2007; 2008; 2009; 2010) and Çetin et al. (2011).

In addition to total phenolic content, the amount of 11 individual phenolic compounds were investigated in the study. All the compounds, except syringic acid, were positively affected by UV irradiation. As compared to the control, an increase was observed in the first 24 h, the maximum value was obtained at 48th h, and the contents decreased at 72nd h. Unlike other individual phenolics, the highest rutin content was observed within 24 h while gallic acid remained the same value at 24th and 48th h. On the other hand, as compared to the control, syringic acid value decreased during the incubation time.

The positive effect of UV irradiation is closely related to the irradiation time (Keskin 2019). As in many studies of the effect of UV irradiation time on the induction of phenolic compounds production (Keskin and Kunter, (2007, 2008, 2009, 2010); Çetin et al., 2011), the success achieved in the irradiation time with the incubation time. As a result of 10 and 15 min irradiations, it was determined that phenolic compounds can be produced successfully.

In this study the highest phenolic content was obtained at 48th h for both irradiation times (10 and 15 min). Bais et al. (2000), Takayanagi et al. (2004), Keskin and Kunter (2007, 2008, 2009, 2010); Lui et al. (2010); Keskin and Kunter (2017) reported that the 48th h was the best incubation time, which is consistent with the results of the study. The decrease in the content was realized at 72 h is an expected result. In the study, the values measured at this hour were found lower than control values in some combinations (Table 1). Keskin and Kunter (2007, 2008, 2009, 2010) explained this with the decreasing in the production of phenolic compounds due to the aging of callus. In addition, this can be due to the short-term persistence of secondary metabolites and the loss of their structure by enzymes after a certain stage of incubation (Charlwood et al. 1990).

CONCLUSION

In this study, the production of phenolic compounds in 'Karaerik' cultivar was induced and determined by the tissue culture method by using the induction effect of ultraviolet (UV) light. The study provided a biotechnological approach to increase the production of phenolic compounds in grapevines. As a result of the study, it can be concluded that UV irradiation was effective in induction the production of phenolic compounds in callus tissues of 'Karaerik' and this effect was closely related to the time. In the future studies, based on the successful results obtained in the method, local genotypes should be screened more broadly in *in vitro* conditions. Thus, it will be possible to introduce the country's native genotypes internationally.

ACKNOWLEDGEMENTS

We are grateful for the financial support provided for this research by Van Yüzüncü Yıl University Scientific Research Project Coordination (FYL-2018-7285).

Statement of Conflict of Interest

Authors have declared no conflict of interest.

Author's Contributions

The contribution of the authors is equal.

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