

EFFECTS OF DIFFERENT PLANT GROWTH REGULATOR CONCENTRATIONS ROOTING OF SP-2 (PRUNUS SPINOSA) CLONAL CANDIDATE ROOTSTOCK IN VITRO CONDITIONS

Farklı Bitki Büyüme Düzenleyici Konsantrasyonlarının SP-2 (*Prunus spinosa*) Klonal Anaç Adayının *In Vitro* Koşullarda Köklendirilmesine Olan Etkileri

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ÖZET

Bu çalışma, farklı bitki büyüme düzenleyicileri konsantrasyonlarının *In Vitro* kültür koşullarında rejenere edilmiş klonal anaç adayı SP-2 (*Prunus spinosa*)'nın köklenebilme olanaklarının araştırılması için 2017 yılında yürütülmüştür. Çalışmada, MS besi ortamı ve bu ortama ilave edilen altı farklı bitki büyüme düzenleyici kombinasyonları MSK-1 (0.5 mg.l⁻¹ IBA), MSK-2 (1 mg.l⁻¹ IBA), MSK-3 (0.1 mg.l⁻¹ IBA, 0.5 mg.l⁻¹ BA), MSK-4 (0.1 mg.l⁻¹ IBA, 0.5 mg.l⁻¹ TDZ), MSK-5 (0.5 mg.l⁻¹ IBA, 0.1 mg.l⁻¹ TDZ, 0.05 mg.l⁻¹ NAA), kontrol olarak ise MSK-0 (Hormonsuz normal MS) kullanılmıştır. Çalışma sonunda sürgün uzunluğunun en yüksek MSK-3 besi ortamında 79.85 mm ile elde edildiği saptanmıştır. Ayrıca köklenme değerleri olarak kök uzunluğunun en yüksek MSK-1 besi ortamından 133.33 mm değeriyle, en fazla kök sayısının ise MSK-0 besi ortamında explant başına 4.83 adet olduğu saptanmıştır. Çalışma sonunda en iyi köklenme değerinin MSK-1 besi ortamı kullanılan materyallerde olduğu, kontrol ortamının da (MSK-0) oldukça olumlu sonuçlar verdiği görülmüştür. Klonal anaç adayı SP-2 (*Prunus spinosa*) sadece besi ortamı kullanılarak köklendirilebileceği kanaati hasıl olmuştur.

Anahtar Kelimeler: Anaç, erik, *in vitro*, *Prunus spinosa*.

ABSTRACT

Rooting possibilities *In vitro* conditions with applied plant growth regulators concentration of SP-2 (*Prunus spinosa*), candidate rootstock that obtained by *in vitro* regenerating programm was determined in this study at 2017 year. Medium, and this medium added six different plant growth regulator combinations MSK-1(0.5 mg.l⁻¹ IBA), MSK-2(1 mg.l⁻¹ IBA), MSK-3(0.1 mg.l⁻¹ IBA, 0.5 mg.l⁻¹ BA), MSK-4(0.1 mg.l⁻¹ IBA, 0.5 mg.l⁻¹ TDZ), MSK-5 (0.5 mg.l⁻¹ IBA, 0.1 mg.l⁻¹ TDZ, 0.05 mg.l⁻¹ NAA), and MSK-0 (normal MS without any hormone) as control was used material. Maximum average shoot length per explant at the MSK-3 medium (79.85 mm) was found. In addition, it was determined that maximum root length per (133.33 mm) and highest root number as 4.83 number of shoot at the control medium MSK-0 was observed in this study. At the end of the study, it was observed that the best rooting value MSK-1 medium and the control medium (MSK-0) gave very positive results was occurred. On the other hand it was decided that clonal rootstock candidate SP-2 (*Prunus spinosa*) may be rooting without using any plant growth regulators.

Key Words: Rootstock, plum, *in vitro*, *Prunus spinosa*.

INTRODUCTION

Prunus spinosa is a wild plum which grows in the south and eastern regions of Anatolia shows a rather dwarf development species of compared to other types of wild and domestic plum varieties and species. The 2-3 years-old plants are often used for interstock and hybrid rootstock breeding studies because they are generally suckery and thorny (Milosevic, 2006). However, it is thought that because of healthy development varieties grafted on *Prunus spinosa* rootstock, compatible with some of the apricot and plum varieties, promising results in uptake plant nutrients will continue to be used in rootstock breeding (Ugur,2017).

SP-2 (*Prunus spinosa*) is a rootstock candidate that compatible with some apricot varieties, does not show sucker, can uptake plant nutrients to the scion, show dwarf growing and can be propagated with tissue culture *in vitro* condition (Ugur ve Paydas Kargi, 2017).

It is determining that Kabaasi, Hasanbey and Hacıhaliloglu apricot varieties grafted on this rootstock more dwarf than Myrobolan 29C and GF rootstocks and 56% and 51% respectively (Ugur, 2017). Also, the SP-2 (*Prunus spinosa*) to be resistant to the *Meloidogyne incognita* (rac-1) and *Meloidogyne javanica* (rac-1) was found according to the egg set reaction scale by 0.0 ± 0.0 (Gürkan ve ark., 2017). It has been reported that positive results have been obtained using oxin hormones with cytokine hormone as BA, BAP and TDZ on the propagations of *Prunus* rootstocks and varieties by tissue culture methods *in vitro* conditions (Fotopoulos and Sotiropoulos, 2005; Andreu and Marin, 2005; Sadeghi et al., 2015; Petri and Scorza, 2009; Zou, 2010; Nas et al., 2010). In addition, the use of indole-butyric acid (IBA), indole-acetic acid (IAA) and naphthalenacetic acid (NAA) as the oxin form is more common (Silveira et al., 2002). That rooting success of rootstocks varies *in vitro* conditions according to the type of explant material used together with the concentrations of hormones to be used can be said (Perez-Toreno et al., 2000). It is also reported at the end of some research studies that the changes in mineral content and use rates in media are effective in rooting success (Dimassi-Theriou, 1995).

Regeneration and rooting possibility of this rootstock candidate propagating by tissue culture *in vitro* conditions using various plant growth regulator concentrations and with more less using media was aimed in this study.

MATERIAL AND METHOD

In vitro conditions obtained that 8 weeks old and 10 mm tall micro cuttings as source of explants were used in the study. Each explant, 60 x 100 mm magenta, approximated about 70 ml, that plant growth regulating concentrations were added in different proportions (Table 1) MS medium (Murashige & Skoog, 1962) was transmitted at the 15.05.2017 and the results were taken at the 03.08.2017. The medium was sterilized at 121°C for a period of 15 minutes after adjusting the pH to 5.8 for 6 g l⁻¹ (agar for microbiology agar, Fluka). Magenta seedlings were kept 16 hours at day under fluorescence light with lamps of TLD 36 W / 84 (45 $\mu\text{mol m}^{-2} \text{s}^{-1}$, 400-700 nm) at $\pm 21^\circ \text{C}$ after the lid was closed. After approximately 50 days, the number and length of the shoot and root per explant were measured and statistically analyzed by taking the arithmetic mean of each root and its values.

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Table 1. Different hormone concentrations used in rooting studies.

Concentrations	IBA	BA	TDZ	NAA
MSK-0	0	0	0	0
MSK-1	0.5	0	0	0
MSK-2	1.0	0	0	0
MSK-3	0.1	0.5	0	0
MSK-4	0.1	0	0.5	0
MSK-5	0.5	0	0.1	0.05
MSK-6	0.5	0.1	0	0.05

The experiment was established in randomized plot test, three magenta with each parcel and five explant in each magenta. LSD test (Armitage & Herbert, 1996) was used in multiple comparisons after the variance analysis (ANOVA) obtained data.

DISCUSSION

Regeneration situation of the SP-2 (*Prunus spinosa*) rootstock candidate, which has been established in seven different plant growth regulatory concentrations, is given in Table 2. The lengths of the

shoots were between 46.25 mm and 79.85 mm, and the differences were statistically significant at 5%.

The longest shoot development at 79.85 mm in MSK-3 was measured followed by the values of MSK-6 and MSK-0 at 68.43 mm and 68.34 mm respectively with the three sites being statistically located in the same group. The MSK-4 has been the concentration that the lowest shoot length value is observed.

Table 2. Regeneration situations of the SP-2 (*Prunus spinosa*) rootstock candidate, which has been established in different concentration environments.

Concentrations	Length of shoot (mm)	Number of shoot
MSK-0	68.34 ^{ab}	1.00
MSK-1	59.03 ^{bc}	2.33
MSK-2	54.67 ^{bc}	1.33
MSK-3	79.85 ^a	2.00
MSK-4	46.25 ^c	2.33
MSK-5	56.52 ^{bc}	1.00
MSK-6	68.43 ^{ab}	1.33
LSD 0.05	18.76*	N.S.

The results of the study which lasted for 50 days, are quite different when compared to Fotopoulos and Sotiropoulos (2005) for 30 days. Although the shoot length values obtained in this study were statistically different from 46.25 mm and 79.85 mm respectively, Fotopoulos and Sotiropoulos (2005) reported that the shoot length values obtained in their study were distributed between 8.1 mm and 15.88 mm, and also the plant growth regulating concentrations values were not statistically significant. It is estimated this situation that due to combination differences used in both studies and application time. However, Sadeghi et al. (2015), three different doses of BA in two different media that vegetative propagation *in vitro* condition of semi dwarf Tetra (*Prunus domestica*) were used in their study. They were indicated which without any other plant growth regulator the length of the shoot that varies from 14.5 to 20.30 mm and that this difference was statistically significant the result of this study. This situation was indicated that, despite the increase in combination of plant growth regulators, an increase shoot lengths can not be achieved. However, the same situation was not seen in the number of shoot. The number of shoot that our study were lower than Sadeghi et al. and Fotopoulos and Sotiropoulos 2005's study. But Petri and Scorza 2010's investigations were similar with our study and there was no statistical difference in this study too. This can be explained by the fact that a lower number of plant growth regulating concentrations can be more effective in breeding. It can be said that the combination of the hormone concentration differences in the development of shoots is 0.5 BA mg 1-1 and 0.1 IBA mg 1-1. However, it was found interesting to note that the control application values were high in control.

Table 3. Rooting situations of the SP-2 (*Prunus spinosa*) rootstock candidate, which has been established in different concentration environments.

Concentrations	Length of root (mm)	Number of root
MSK-0	120.33 ^a	4.83 ^a
MSK-1	133.33 ^a	1.00 ^{bc}
MSK-2	0.00 ^d	0.00 ^c
MSK-3	0.00 ^d	0.00 ^c
MSK-4	0.00 ^d	0.00 ^c
MSK-5	101.72 ^b	4.33 ^a
MSK-6	46.50 ^c	1.33 ^b
LSD 0.05	18,95**	1.18**

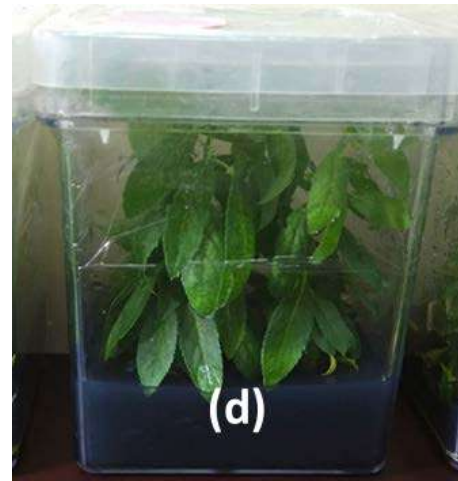
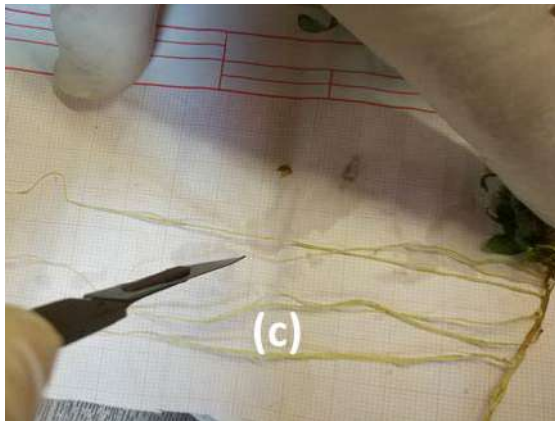
The rooting situation of the SP-2 (*Prunus spinosa*) rootstock candidate which has been established in seven different plant growth regulatory concentrations is given in Table 3. The root length values between 46.50 mm and 133.33 mm, on the other hand the differences statistically significant at 5% were determined. The highest root length values MSK-1 and MSK-0 plant growth regulatory concentrations 133.33 and 120.33 mm respectively that statistically identical in the same group were found. It can also be said that there is an important root development with 101.72 mm in the MSK-5 and there was no root growth in MSK-2, MSK-3 and MSK-4 plant growth regulatory concentrations. In addition to the root values best root number values MSK-0 with MSK-5 were found, MSK-1 and MSK-6 plant growth regulatory concentrations did not show statistically significant differences that ranging from 1.00 to 1.33. When viewed to the root development values it can be said that there is no statistically significant relationship between rooting capabilities and the hormone concentrations used. The increase in root count and root length values in the plants cultivated in the MSK-0 plant growth regulatory concentrations that SP-2 (*Prunus spinosa*) does not have a positive effect on the hormones used for rooting purposes.

Sadeghi et al.2015 found that in their study propagation of Tetra (*Prunus domestica*) rootstock that belonging to the plum species different IBA concentrations that used *in vitro* studies, a decrease in root length values, despite the increased IBA dose in three different medias similarly to our study and this values have been statistically significant. However, Fotopoulos and Sotiropoulos 2005 observed that average root length values in the PR204 / 84 (*Prunus persica x Prunus amygdalus*) hybrid rootstock was increase with the increased IBA dose. It is thought that this difference in rootstock species can be due to differences in some of the internal factors. It is noteworthy that the results obtained by the close relationship of the Tetra (*Prunus domestica*) which investigated by Sadeghi et al.2015 and SP-2 (*Prunus spinosa*) used in our study are also similar.

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RESULT

As a result, it was determined that MSK-0 and MSK-1 plant growth regulatory concentrations have been elevated from the other different hormone concentrations that used in rooting the SP-2 (*Prunus spinosa*) rootstock candidate *in vitro* were found. It has been observed that some of the plant growth regulatory concentrations have a positive effect on the rooting values of this candidate rootstock. However, positive results have also been obtained in the MS media with non plant growth regulatory concentrations. This result can be considered as a pretty good result when considered commercially. However, the probability that different concentrations can produce better results should always be considered.



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Şekil 1: (a),(b): An overview of rooted plants, (c): Root measurement and counting operations, (d): A healthy plant that has completed its development *in vitro*.

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