DETERMINATION OF PROPAGATION CHARACTERISTICS OF *Prunus arabica* Oliv. ALMOND SPECIES

Hasan DENİZHAN[1*], Fırat Ege KARAAT[2], Ramazan Bestami KARAHAN[1], Adil GEZER[1]

[1] *Republic of Türkiye Ministry of Agriculture and Forestry, Nuts Research Institute, Adıyaman, Türkiye*

[2] *Adıyaman University, Faculty of Agriculture, Department of Horticulture, Adıyaman, Türkiye*

**Corresponding author: denizhan.hasan@tarimorman.gov.tr*

Geliş (Received): 26.05.2022 **Kabul (Accepted):** 09.10.2022

ABSTRACT

In this study, it was aimed to determine the propagation characteristics of *Prunus arabica* Olivier (*P. arabica*) in terms of seed germination and cutting rooting success. The material of the study consisted of seeds and cuttings taken from seven different genotypes of *P. arabica* species that naturally grow in Kâhta District of Adıyaman Province. The study was conducted under temperature and humidity (25°C and 80 %) controlled greenhouse conditions. The seeds were directly sown to soil in the greenhouse without any pretreatment and germination percentages were recorded. The cuttings were pretreated by different doses of Indole Butyric Acid (0, 1000, 2000 ppm) and Acetylsalicylic Acid (0, 50, 100 ppm) and combinations of 1000 and 2000 ppm IBA with 100 ppm ASA. The pretreated cuttings were taken to perlite medium in the greenhouse and callus formation and rooting success were recorded. In terms of seed germination, seeds of *Prunus dulcis* var. amara were used as control. The germination rate of different *P. arabica* genotypes in the study varied between 55 and 65 % which was higher than *Prunus dulcis* var. amara. While the highest callus formation was obtained from 1000 ppm IBA application with 73.30 %, the lowest callus formation was found in 50 ppm ASA application with 14.25 %. The cuttings of 1000 and 2000 ppm IBA pretreatments were the only rooted cuttings in the study with the 3.30 and 2.70 % rooting percentage, respectively. As a result of the study, it was concluded that *P. arabica* species can be successfully propagated with seeds, but the low clonal propagation success together with high callus formation as a result of the pretreatments which indicate that the rooting success may be improved.

Keywords: *Prunus arabica* Olivier, cutting, hormones, propagation, rootstock

Prunus arabica Oliv. BADEM TÜRÜNÜN ÇOĞALTIM DURUMUNUN BELİRLENMESİ

ÖZET

Bu çalışmada, *Prunus arabica* Oliv. (*P. arabica*) türünün tohum çimlenme ve çelik köklenme başarısı açısından çoğaltma özelliklerinin belirlenmesi amaçlanmıştır. Çalışmanın materyalini Adıyaman ilinin Kâhta ilçesinde doğal olarak yetişen *P. arabica* türünün yedi farklı genotipinden alınan tohum ve çelikler oluşturmuştur. Çalışma sıcaklık ve nem (25 °C ve % 80) kontrollü sera koşullarında yürütülmüştür. Tohumlar herhangi bir ön işleme tabi tutulmadan doğrudan serada toprağa ekilmiş ve çimlenme oranları kaydedilmiştir. Çelikler, farklı dozlarda İndol Bütirik Asit (0, 1000, 2000 ppm), Asetilsalisilik Asit (0, 50, 100 ppm) ve bunların bazı kombinasyonları olan 1000 ve 2000 ppm IBA ile 100 ppm ASA uygulamalarına tabi tutulmuştur. Ön işlem uygulanan ve serada perlit ortamına aktarılan çeliklerin kallus oluşumu ve köklenme başarısı kaydedilmiştir. Tohum çimlenmesi açısından *Prunus dulcis* var. amara kontrol olarak kullanılmıştır. Çalışmada farklı *P. arabica* genotiplerinin çimlenme oranı % 55 ile % 65 arasında gerçekleşmiş ve *Prunus dulcis* var. amara türünden daha yüksek bulunmuştur. En

yüksek kallus oluşumu % 73,30 ile 1000 ppm IBA uygulamasından, en düşük kallus oluşumu ise % 14,25 ile 50 ppm ASA uygulamasından elde edilmiştir. Çalışmada sadece 1000 ppm ve 2000 ppm IBA uygulamaları yapılan çelikler sırasıyla % 3.30 ve % 2.70 oranında köklenmiştir. Çalışma sonucunda *P. arabica* türünün tohumla başarılı bir şekilde çoğaltılabileceği belirlenmiş, ancak yapılan ön işlemler sonucunda kallus oluşumunun yüksek olmasına rağmen klonal çoğaltma başarısının düşük olduğu belirlenmiş olsa da yapılacak farklı uygulamalarla bu türün köklenme başarısının iyileştirilebileceğini sonucuna varılmıştır.

Anahtar Kelimeler: *Prunus arabica* Olivier, çelik, hormon, çoğaltma, anaç

1. INTRODUCTION

Almond is one of the most important nuts for commercial production. Adaptation to extreme climate conditions combined with the ability to develop a deep and extensive root system allow almonds to exploit a wide variety of ecological conditions. Almond trees were adapted to mild winters and dry, hot summer conditions due to its low chilling requirement, rapid shoot growth and high tolerance to summer heat and drought (Gradziel et al., 2017).

One of the important factors effects the success of fruit growing is the use of the right rootstock. The rootstock selection is as important as cultivar selection and the use of those which are suitable for the soil conditions and cultivation techniques directly affect yield and quality (Hepaksoy, 2017). In modern fruit growing, rootstock use is a necessity for different reasons such as ensuring the resistance of the cultivars against adverse factors, diseases and pests caused by soil and climatic conditions, expanding its adaptability, increasing fruit yield and quality, and dwarfing the tree. Rootstocks to be used should be easily propagated, and have good graft compatibility with the grown cultivars. In addition, there should be no rootstock-scion incompatibility, the cultivars grafted on should bear fruit early, be resistant to important diseases, pests and can be easily adaptable to different climatic and soil conditions (Ikinci and Bolat, 2019).

Wild almond species are naturally distributed throughout the Mediterranean climate which is characterized with hot and dry summers and mild winters. Some wild almond species are especially distributed in arid and semi-arid areas. These species show good resistance to drought and high temperature stresses (Imani et al., 2006; Rahemi et al., 2011). Khalil and Al-Eisawi (2000) reported that rootstocks of wild almond species obtained from any region tolerate adverse climate and heavy soil conditions better than other rootstocks. In addition, Kester & Gradziel (1996) reported that to obtain the best rootstock for a particular region, it is very important to use wild almond species by breeding studies to be done among those species.

P. arabica is one of the species can be found in Saudi Arabia, Jordan, Iraq, Syria and the Southeastern Anatolia Region of Turkey where the trees of the species grow naturally in the form of shrubs at usually altitudes of 150-1200 m (Yılmaz et al., 2020). The species has the ability to successfully adapt to climates with hot and dry summers and mild and rainy winters (Hoseinzadeh and Saeb, 2011). Trees of the species form 1.5-2.5 m tall, shrub-shaped trees. Although the species is a late flowering species, it defoliates in summer because it ripens its fruits very early. Its grown areas are mostly bare limestone rocks and sandstone cliffs, sloping areas in valley beds (Browicz, 1982).

P. arabica is counted to be a potential rootstock for the almond cultivation especially because of the advantages of late flowering, dwarf habitus, extremely high resistance to drought and T-budding success (Atlı et al., 2018). For these reasons, it is important to determine the propagation status which is one of the most important issues in terms of nursery production

for its use as rootstock in cultivation. Contrary to this necessity, there is only one study to determine rooting characteristics of the species cuttings (Abu-Laila, 1995) and a few studies have been conducted on seed germination (Abu-Laila, 1995; Khalil and Al Eisawi, 2000; Atlı et al., 2018; Yilmaz et al., 2020). For those reason, the previous studies done to determine the propagation status of the species is limited.

In this study, it was aimed to contribute to the literature in terms of the propagation characteristics of *P. arabica* species and to determine the effects of different pretreatments to rooting performance of the species cuttings.

2. MATERIAL AND METHODS

The plant material of the study consisted of seeds and cuttings of *P. arabica* species located in Taşlıca Village of Kahta District of Adıyaman Province (Figure 1). Totally of 140 seeds and 180 one year cuttings were collected from each of 7 different *P. arabica* genotypes (coded as T1 to T7). Both the seeds and cuttings were collected in February 2021. As control of seed germination tests, seeds of *Prunus dulcis* var. amara which were obtained from a commercial seed supplier for seedling growers were used.

Figure 1. *P. arabica* species

Collected seeds were subjected to seed germination test and the cuttings to rooting test including callus formation and rooting percentage. The germination and rooting tests were performed under controlled greenhouse conditions (25 °C temperature and 80 % humidity).

For the seed germination test, the collected seeds were sowed to the soil of the greenhouse without any pretreatment in February 2021. The germination of seeds were followed for 16 weeks and the germination percentage was recorded for each genotype and the control.

For the rooting test of the cuttings, perlite medium which was prepared and moistened in rooting boxes (40 cm in depth). To determine the effects of the pretreatments on rooting, the cuttings were subjected Indole Butric Acid (IBA) and Acetyl Salicylic Acid (ASA) pretreatments as rooting solutions. The pretreatments of 0, 1000 and 2000 ppm IBA and 0, 50 and 100 ppm ASA solution and combination of the IBA doses with 100 ppm ASA were applied. Cuttings were treated in ASA solution for 24 hours as reported by Akbulut and Yiğit (2014), and IBA solution for 10 seconds just before the planting. The cuttings (20 to 25 cm in length and 10 to 20 mm in diameter) were transferred to perlite medium in February 2021. Both callus and the root formations were followed until June 2021.

The study was carried out in three replications for six applications, excluding the control, and 30 cutting were included in each replication. The data obtained were evaluated according to the Duncan Multiple Comparison test at P≤0.05 significance level using the "SPSS for Windows 23.0" (IBM Inc., Chicago, IL, USA) package program.

3. RESULTS AND DISCUSSION

3.1. Seed germination rate

The seed germination percentage results of different *P. arabica* genotypes are presented in Table 1. The highest germination was obtained in T5, T6 and T7 types. The lowest germination was observed in T2 and T4 types. The average seed germination percentage was found as 60 % when the results of all genotypes were calculated together. The germination rate of *Prunus dulcis* var. amara seeds was found to be lower than all other *P. arabica* types.

Khalil & Al-Eisawi (2000) reported that *P. arabica* seeds that were not subjected to any stratification and hormone application did not germinate. Yilmaz et al. (2020) reported that the germination rates of *P. arabica* seeds that were not subjected to different pre-chilling processes were between 2.7-4 %. Atli et al. (2018) reported that the germination rate was 46.7-59.3 % in seeds that were only stratified for 30 days and were not subjected to any hormonal application. However, in this study, the germination percentage in seeds of *P. arabica* that were not subjected to any stratification or hormonal treatment varied between 55 and 65 %. The possible reasons of the various results reported by the previous studies and this current study would be the differences of the used genotypes, the seed collection stage and the germination conditions.

Table 1. Germination rates of seeds of *P. arabica* genotypes and the control

P. arabica types	Germination Rate %
T1	60 ± 6.07 ab
T2	55 ± 5.19 b
T3	60 ± 6.24 ab
T4	55 ± 2.64 b
T5	65 ± 5.29 a
T6	65 ± 4.28 a
T7	65 ± 6.24 a
Prunus dulcis var. amara	33 ± 2.00 c

Statistically significant differences (P≤0.05) obtained between the values were indicated with different letters

3.2. Callus formation and rooting rates in cuttings

Callus formation and rooting rates of cuttings taken from *P. arabica* genotypes are given in Table 2. The highest callus formation and rooting was obtained from cuttings applied only 1000 ppm with 73.30 % and 3.30 %, respectively. The highest callus formation was obtained from 0 ASA + 1000 IBA subjects to 73.30 % and the lowest callus was obtained from 50 ASA + 0 IBA doses. Even though, similar callus formations were obtained from single 100 ppm ASA and 2000 ppm IBA applications, rooting was obtained only from IBA applications.

The rooting process of *Amygdalus* species has been reported as very difficult, and accordingly, various hormones have been studied at different doses (Cebetas et al., 2017; Kasim et al., 2009; Reighard et al., 1990; Kester and Sartori, 1966). Qurnfleh et al. (1997) investigated the in vitro rooting status of *P. arabica* cuttings under different hormonal

applications and rooting mediums. They reported that the highest rooting was obtained with 86.7% of 0.01 mg/l IBA, 0.2 mg/L 6-Benzylaminopurine, 30 g/l sucrose and 6 g/l agar MS medium and 250 and 500 ppm NAA applications. Akbulut & Yiğit (2014) reported that callus formation was higher in different almond species with 50 and 100 ppm ASA + IAA doses. In this study, the rooting percentage of ASA + IBA combinations was found to be lower compared to single applications. It was thought that this situation was caused by factors such as the type of hormone used, the dose of application and the difference in plant species and genotypes. Abu-Laila (1995) applied 2000, 3000 and 4000 ppm IBA to *P. arabica* soft-cutting cuttings collected in November, January and May and reported that the highest rooting was obtained from 2000 ppm IBA treatment with 27.5 % was applied in January. However, in this study, while 3.30 % and 2.70 % rooting were obtained in 1000 and 2000 ppm single doses of IBA, respectively, no rooting was observed in other applications (Table 2). This can be related to the different cutting collection times, the genotypes, the ecological conditions, and the rooting conditions.

Table 2. The callus formation and rooting percentages of different ASA and IBA doses

Treatments (ppm)	Callus Formation Percentage (%)	Rooting Percentage (%)
Control	28.10 ± 1.57 e	0
50 ASA + 0 IBA	14.25 ± 0.91 f	0
100 ASA + 0 IBA	59.50 ± 3.85 b	0
0 ASA + 1000 IBA	73.30 ± 4.58 a	3.30 ± 0.35 a
0 ASA + 2000 IBA	59.45 ± 2.18 b	2.70 ± 0.39 a
100 ASA + 1000 IBA	46.15 ± 3.67 c	0
100 ASA + 2000 IBA	38.45 ± 2.60 d	0

Statistically significant differences ($P \leq 0.05$) obtained between the values were indicated with different letters

4. CONCLUSIONS

When the seed germination test results in the study are evaluated, although there is no stratification and pre-cooling application *P. arabica* was successfully germinated and showed higher performance compared to the *Prunus dulcis* var. amara seeds. Based on the obtained results, *P. arabica* species was concluded to be propagated successfully with the seeds. Furthermore, pre-cooling and hormonal applications can increase the germination percentage which would be beneficial to be determined in the future studies. The callus formation of *P. arabica* cuttings was increased with different doses of IBA and ASA. However, the desired rooting success could not be obtained in any applications. These results indicated low clonal propagation success together with an improvement potential based on the higher callus formation percentages obtained in the pretreatments. On the other hand, when the results were compared with the previous studies it was concluded that the cutting collection time and rooting conditions would be another important factor on rooting success. For these reasons in the future studies, in order to increase the success of rooting, the effects of different doses of hormonal applications and rooting conditions should be detected on *P. arabica* cuttings taken at different times.

REFERENCES

Abu-Laila, K. M. A., 1995. Propagation of *Amygdalus arabica* Oliv. by Stem Cuttings and Seeds. Agris FAO, (Unpublished) Master Thesis, Amman, Jordan, 91 pp.

Akbulut, G. B, Yiğit E., 2014. Effects of acetlysalicylic acid with indole-3-acetic acid on rooting and pigmentation in Amygdalus L. Cumhuriyet Science Journal, 35(2): 1-10.

Atli, H. S., Kayra R., Yavic A., 2018. Determination of propagation statue of *Amygdalus arabıca* Oliv. almond species grown in Siirt province. Fresenius Environmental Bulletin, 27(12A/2018): 9345-9351.

Browicz, K. 1982. Chorology of Trees and Shrubs in South-West Asia and Adjacent Regions. Polish Scientific Publishers, Poznan, 172 pp.

Gradziel, T. M., Curtis R., Socias i Company R., 2017. "Production and Growing Regions, 70-86". In: Almonds: Botanys, Production and Uses. (Eds. Socias i Company, R., Gradziel, T. M.), Boston, USA, 494 pp.

Hepaksoy, S. 2017. Propagation of GF 677 (P. amygdalus x P. persica) Clone Rootstock in Tissue Culture by Shoot-tip Techique. Journal of Agriculture Faculty of Ege University, 54 (4): 447-451.

Hosenzadeh, J., Saeb K., 2011. Morphological diversity of *Amygdalus arabica* Oliv. in natural forests of Ilam province, Iran. Journal of Biodiversity and Ecological Sciences, 1: 245–248.

Imani, A., Hassani D., Rahemi A., 2006. Following almond footprints in Iran. Scripta Horticulturae, 4: 71-79.

İkinci, A., Bolat İ., 2019. "Rootstock Use in Fruit Growing, 278-283". I. International Harran Multidisciplinary Studies Congress (8-10 Mart 2019, Şanlıurfa), 1611 pp.

Kasim, N. E., Abou Rayya, M. S., Shaheen, M., Yehia, T. A., Ali, E. L. 2009. Effect of different collection times and some treatments on rooting and chemical internal constituents of bitter almond hardwood cuttings. Research Journal of Agriculture and Biological Sciences, 5: 116-122

Kester, D. E., Gradziel T. M., 1996. "Almonds, 1-97". In: Fruit Breeding Nuts. 3rd ed. (Eds. J. Janick, Moore J. N.), New York, USA, 288 pp.

Kester, D. E., Sartori, E., 1966. Rooting of cuttings in populations of peach (*Prunus persica* L.) almond (*Prunus amygdalus* Batsch) and their F1 hybrids. American Society for Horticultural Science, 88: 219-223.

Khalil, R. Y., Al-Eisawi D. M., 1998. "Seed germination of-Amygdalus arabica Oliv. as influenced by stratification and certain plant bioregulators, 21-30". XXV. International Horticultural Congress Part 7: Quality of Horticultural Products, (2-7 August 1998, Brussels), 292 pp.

Qrunfleh, M. M., Al-Eisawi, D. M., Moh'd I, H. 1997. Rooting Ability of Microcuttings of Desert Almond (*Amygdalus arabica* Oliv.). Horticultural Science, 32(3): 532D-532.

Rahemi, A., Taghavi T., Fatahi R., Ebadi A., Hassani D., Chaparro J., Gradziel T., 2011. Seed germination and seedling establishment of some wild almond species. African Journal of Biotechnology, 10 (40): 7780-7786. https://doi.org/10.5897/AJB10.1064.

Reighard, G., Cain, D., Newall, W. 1990. Rooting and survival potential of hardwood cuttings of 406 species, cultivars and hybrids of *Prunus*. Horticultural Sciences, 25: 517-518.

Rubio-Cabetas, M. J., Antonio Felipe J., Bordas M., 2017. "Propagation Techniques, 228-239". In: Almonds: Botany, Production and Uses. (Eds. R. Socias i Company, T. M. Gradziel), Boston, USA, 494 pp.

Yılmaz, M., Kalkan M., Demirbag H., 2020. Seed characteristics of Amygdalus arabica in Adiyaman region of Turkey. Dendrobiology, 84: 49-57. https://doi.org/10.12657/denbio.084.005.