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ORIGINAL ARTICLE

Comparison of the Effects of Some Applications on Root and Shoot Development of Different *Vitis* Species

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Abstract

Grape varieties, rootstocks and genotypes belonging to the *Vitis* species have been grown for various purposes since ancient times. Different *Vitis* species can be propagated through both grafted and non-grafted methods. There are significant differences in rooting and shoot development among the rootstocks of different *Vitis* species. These differences significantly affect sapling yield and quality. In our study, we used grape varieties belonging to different species and polyploidy levels and the American grapevine rootstocks most commonly used in Türkiye to investigate the effects of five organic and inorganic based products on rooting and shoot development. We used 12 different grape varieties and seven different American grapevine rootstocks as materials. Two of the grape varieties belonging to different *Vitis* species were tetraploid. We investigated the effects of the five different applications on callus formation, root development level, number of roots, root length, number of shoots, shoot length and number of leaves on the shoot (excluding the rootstock). As a result, MAS RAIZ application, which is an organic based product, gave the best results in terms of callus development and shoot number, and 2000 ppm indole butyric acid applications gave the best results in terms of root development level and number of roots. Considering the general averages in terms of root length, shoot length and number of leaves on the shoot, pure water application gave better results than other applications. Growers must choose the most appropriate application, especially considering efficiency and cost. In addition, new products of chemical and organic origin that claim to promote rooting enter the market every year. These new products should be compared with scientific research, and the most accurate information should be provided to both growers and nurseries.

Keywords Interspecies · Polyploidy · Cuttings · Rootstock · Root/shoot Development

Introduction

Good root formation is a very important factor to increase the efficiency of both grafted and non-grafted production methods (Leakey 2014). This is a very important issue in the propagation of different *Vitis* species, and researchers have tried to increase these rates with various applications for adequate rooting and subsequent shoot development (Boeno and Zuffellato-Ribas 2023). Grafting is a very old method used for vegetative propagation and improving plant adaptation to different soil conditions. It is widely used in many horticultural crops, including different grapevine species (Gainza et al. 2015). Especially with the phylloxera pest causing serious problems in vineyard areas, the demand for grafted grapevine saplings increased and production in many countries had to switch from ungrafted to grafted production. However, in many parts of the world, the non-grafted and own-rooted production method is still widely used in light soils that are not contaminated with phylloxera.

The necessity of using rootstocks for unsuitable soils due to the phylloxera pest in viticulture has been understood and many rootstocks with different characteristics have been developed. Depending on the rootstock species used, cuttings may show different rooting properties. The use of difficult-to-root rootstocks in the production of saplings negatively affects growers/nurseries. For this reason, some applications are used to increase the rooting rate and quality of difficult-to-root grapevine rootstocks (Satisha and Adsule 2006). In addition, different *Vitis* species are grown on their own roots in soil types that do not require rootstock, and similar applications are required to help them root.

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Although there have been significant changes over the years in the annual number of grapevine saplings produced in Turkey, an average of 4–5 million grapevine saplings is produced annually. Approximately 75% of these saplings are grafted and 25% are ungrafted grapevine saplings (Durmaz et al. 2019). Rooting is important in the production of both grafted and ungrafted grapevine saplings, and some applications need to be carried out to ensure a high percentage of rooting of both varieties and rootstocks (Waite et al. 2015; Köse et al. 2023).

In the production of saplings of varieties and rootstocks belonging to different *Vitis* species, some have difficulty rooting and this negatively affects both nursery owners and growers. For this reason, many practices such as bottom heating, paraffin covering, soaking in water, cultural practices, rooting medium, planting method, plastic mulch, tunnel and hormone use are applied to increase the rooting rate and quality of difficult-to-root grapevine rootstocks (Barlass and Skene 1980; El Shaima and Saleh 2018). Different plant growth regulating hormones are widely used because they generally help root formation. Today, indole butyric acid (IBA), naphthalene acetic acid (NAA) and indole 3 acetic acid (IAA) are the most commonly used auxins to stimulate and promote rooting in cuttings (Satisha and Adsule 2006; Madhavan et al. 2021). However, in recent years, different preparations of organic and non-organic origin have appeared on the market and started to be used instead (Singh and Chauhan 2020; Jindo et al. 2022).

In this study, callus, root and shoot developments were examined as a result of five different applications on 1-year-old cuttings of 12 grape varieties and seven different American grapevine rootstocks, and the best applications were determined using a range of parameters. Comparisons were made between applications of organic and non-organic based products, both of which can be alternatives to the widely used IBA application and their combination with IBA.

Materials and Methods

Materials

In the winter of 2023, cuttings were taken from the 1-year-old shoots of a total of 12 grape varieties belonging to different species and polyploidy levels from the Vineyard Genetic Resources parcel located at Yalova Atatürk Horticulture Central Research Institute (YAHCRI), Turkey. In addition, cuttings of a total of seven American grapevine rootstocks were used from the Vineyard Genetic Resources parcel of Manisa Viticulture Research Institute (MVRI), Turkey. This was carried out in the greenhouse within the 'Greenhouse Application and Research Unit' of Bursa Uludağ University Faculty of Agriculture, Department of Horticulture. Information on the different *Vitis* species used in the study is given in Table 1. The cuttings of rootstocks and varieties were taken in January during the pruning period and stored in the cold storage of Bursa Uludağ University, Faculty of Agriculture, Department of Horticulture, in an environment of +4 °C and 80% humidity until March.

Table 1 Some information about the *Vitis* species used as material in the study

Variety/rootstock	Species	Poliploidy Level	Origin	Seeded/seedless
'Atak 77'	*V. vinifera*	2N	YAHCRI	Seeded
'Crimson Seedless'	*V. vinifera*	2N	YAHCRI	Seedless
'Heukboseok'	Interspecies hybrid	4N	YAHCRI	Seeded
'Isabella'	*V. labrusca*	2N	YAHCRI	Seeded
'Kismish Rozovyi'	*V. vinifera*	2N	YAHCRI	Seedless
'Kyoho'	Interspecies hybrid	4N	YAHCRI	Seeded
'Muscat Bleu'	Interspecies hybrid	2N	YAHCRI	Seeded
'Müşküle'	*V. vinifera*	2N	YAHCRI	Seeded
'Özer Beyazı'	*V. vinifera*	2N	YAHCRI	Seedless
'Regent'	Interspecies hybrid	2N	YAHCRI	Seeded
'Rizpem'	*V. labrusca*	2N	YAHCRI	Seeded
'Superior Seedless'	*V. vinifera*	2N	YAHCRI	Seedless
41 B[a]	Interspecies hybrid	2N	MVRI	–
99 R[a]	Interspecies hybrid	2N	MVRI	–
110 R[a]	Interspecies hybrid	2N	MVRI	–
1103 Paulsen[a]	Interspecies hybrid	2N	MVRI	–
V. rupestris Du Lot[a]	*V. rupestris*	2N	MVRI	–
Kober 5 BB[a]	Interspecies hybrid	2N	MVRI	–
140 R[a]	Interspecies hybrid	2N	MVRI	–

[a]Rootstock

Table 2 Five different applications and doses used in root varieties and rootstocks

Applications		
First application	Control (pure water)	The basal ends of cuttings were immersed in pure water for 5 s
Second application	2000 ppm Indole-3-butyric acid (IBA)	The basal ends of cuttings were immersed in 2000 ppm IBA solution for 5 s
Third application	TONIPLANT (0.43 NAA + 0.018 % NAD) 50 g/11 water	The basal ends of cuttings were immersed in TONIPLANT 500 g/l solution for 5 s
Fourth application	MAS RAIZ 250 ml/11 water	The basal ends of cuttings were immersed were immersed in MAS RAIZ 250 ml/l solution for 5 s
Fifth application	1000 ppm IBA + MAS RAIZ (250 ml/11 water)	The basal ends of cuttings were immersed in 1000 ppm IBA + MAS RAIZ 250 ml/l solution for 5 s

While planning the study, as many different *Vitis* species as possible and also varieties with different characteristics were included. For this reason, when selecting varieties, different varieties and species were selected according to species, different ploidy levels, seeded/seedless and different berry colours.

Applications

In the study, five different applications were made with a total of 19 grape varieties and rootstocks and their effects on rooting and shoot development were investigated (Table 2 and Fig. 1).

In the study, as a control (1st application), the basal parts of the cuttings prepared from woody shoots of different *Vitis* species immersed in pure water for about 5 s. In the 2nd application, the basal parts of the cuttings immersed in 2000 ppm IBA solution, which is the most commonly used application, for about 5 s. The third application

Fig. 1 Images of applications packaged and dissolved in water

Fig. 2 Post-planting images of cuttings of *Vitis* spp. in the greenhouse

Fig. 3 Calluses formed at the bottom of the cuttings and their development level used in scoring (0–3)

was made with TONIPLANT (Genta Tarım, Turkey), which contains 0.43% NAA +0.018% NAD (1-NAPHTHYL ACETAMIDE), and is increasingly used today, by growers as well as nurseries. The basal parts of the cuttings were kept in the solution prepared with pure water for about 5 s. As a 3rd application, the cuttings were immersed in MAS RAIZ solution (Servalesa, Spain), which is suitable for the organic production model, for the same period of time. MAS RAIZ is sold as rooting agent, soluble liquid for foliar or root application. The product contains plant hormones, amino acids, seaweed, biological bio-activators and some nutrients designed to stimulate roots and ensure better development and thickening of shoots. For this reason, we used this product alone (for its suitability for organic sapling production) in the 4th application and its mixture with IBA as the 5th application.

Our aim was to examine an application (TONIPLANT), whose use has rapidly increased in recent years, and an application (MAS RAIZ) suitable for the organic viticulture model, by comparing it with standard and control applications in different *Vitis* species. For each application, six cuttings were used for varieties and rootstocks and the application was applied in three replications. Since five applications were made for each variety, a total of 90 cuttings were used for one variety. Cuttings were labelled with the variety name and application number during planting. Rooting medium was prepared by mixing coco peat and perlite in a 1:1 ratio into the area where the plantings would be made (Fig. 2).

Rooting and Shoot Development Parameters

Callus Level (Callus Formation) (0–3 Scale): While evaluating the callus level of grape cuttings, scales used by Çelik (2000) were taken into consideration and this scale was modified and used. In order to determine the level of callus tissue development at the basal of the cuttings, a score scale between 0 and 3 was used and evaluations were made as follows (Fig. 3):

- 0 refers callus tissue formation was not observed;
- 1 refers 1 and 33% callus formation in the rooting zone;
- 2 refers 34 and 66% callus formation in the rooting zone;
- 3 refers 67 and 99% callus formation in the rooting zone.

Root Growth Level (0–4): Scoring was made according to the rooting status of the cuttings based on the scale below (Fig. 4);

- 0 point refers no rooting was observed;
- 1 point refers roots between 0.5 and 1.0 cm were observed from one side;
- 2 point refers roots > 1.0 cm were observed on one side;
- 3 point refers rooting was observed from both sides in the rooting area;
- 4 point refers complete rooting of the bottom part of the cutting was observed.

Number of Roots: While evaluating the root number, root length, shoot number and number of leaves of grape cuttings, the results were evaluated according to the method used by İsfendiyaroğlu and Kacar (2019). The main roots

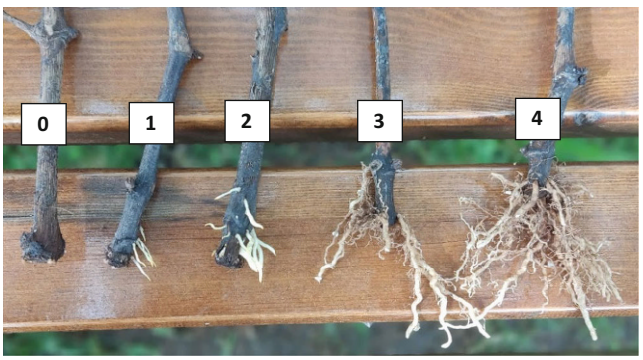

Fig. 4 Different root development levels of cuttings and their scores (0–4)

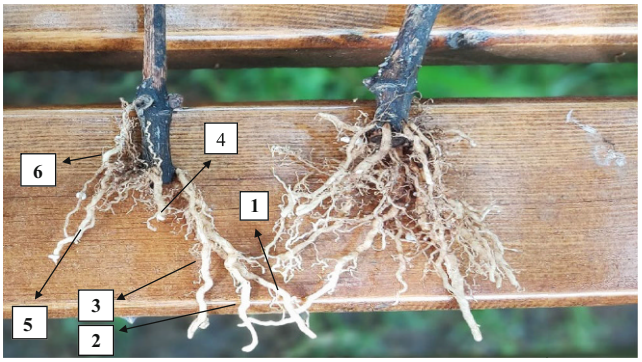

Fig. 5 Primary roots emerging from the bottom of the cuttings and their counting. The numbers in the figure show how the main roots in the cutting are counted and indicate that the cutting in the figure has a total of 6 main roots

growing at the bottom of the cuttings were determined by counting them one by one (Fig. 5).

Root Length: The lengths of the roots emerging from the bottom of the rooted cuttings were measured with the help of a ruler.

Number of Shoots: Shoots coming from buds were counted.

Shoot Length: In cuttings, the length between the bud where the shoot emerged and the tip of the shoot was measured with a ruler.

Number of Leaves on the Shoot: The leaves on the strongest developing shoot were counted. While this measurement was made only on varieties, it was not made on American grapevine rootstocks because it is not of practical importance.

Statistical Analysis

The experiment was set up according to the randomized parcel design with three replications. The data obtained were subjected to variance analysis in the JPM-16 package program (JMP Statistical Discovery LLC, Cary, NC, USA), and the differences between the applications were statistically evaluated using the LSD test at a significance level of 0.05. The differences were determined at a 5% confidence interval ($P \leq 0.05$).

Results

As a result of this study conducted with different *Vitis* species, the applications had varying effects on rooting and shoot development. In the data obtained, variety-application averages, the effects of applications on the parameters and the results within the varieties were evaluated.

According to the average results obtained on a variety basis from five applications made to each variety, 'Heukboseok (4n)' was the variety that formed the most callus in terms of callus level. In terms of root development, the interspecies 'Muscat Bleu' (Interspecies) variety was the variety with the best root development level. Considering the results obtained from the number of roots, it

Table 3 Effects of application averages on root and shoot development parameters in varieties/rootstocks[z]

Variety/rootstock	Callus level (0–3)	Root growth level (0–4)	Number of roots	Root length (cm)	Number of shoots	Shoot length (cm)	Number of the leaves on the shoot
'Atak 77'	1.58^{c}	1.87^{ef}	$5.26^{g–ı}$	4.97^{fg}	1.64^{bc}	5.82^{gh}	5.40^{cd}
'Superior'	0.84^{ef}	1.94^{e}	$12.04^{d–g}$	4.85^{fg}	1.81^{b}	$6.98^{f–h}$	7.01^{a}
'K. Rozovyi'	0.76^{ef}	$2.33^{c–e}$	28.40^{ab}	$6.27^{d–f}$	1.62^{bc}	15.59^{cd}	7.11^{a}
'Ö. Beyazı'	1.28^{cd}	$2.20^{c–d}$	32.32^{a}	5.75^{ef}	1.13^{de}	12.20^{de}	4.45^{de}
'Heukboseok'	2.73^{a}	2.10^{de}	$10.45^{e–h}$	5.60^{ef}	1.04^{ef}	$9.55^{e–g}$	2.67^{f}
'Müşküle'	1.63^{bc}	2.91^{ab}	23.86^{bc}	10.04^{a}	$1.55^{b–d}$	$9.88^{e–g}$	5.82^{bc}
'Kyoho'	0.73^{ef}	1.30^{fg}	$8.11^{e–ı}$	3.92^{g}	1.00^{ef}	$6.58^{f–h}$	1.98^{f}
'Regent'	$0.67^{e–g}$	$2.58^{a–d}$	32.64^{a}	$6.53^{c–e}$	$1.29^{c–e}$	12.77^{de}	5.47^{cd}
'Rizpem'	1.48^{c}	2.11^{de}	$10.69^{d–h}$	8.07^{bc}	1.63^{bc}	$9.71^{e–g}$	4.55^{de}
'Muscat Bleu'	0.99^{de}	2.97^{a}	32.10^{a}	$7.61^{b–d}$	1.55^{bd}	35.03^{a}	6.69^{ab}
'Isabella'	1.33^{cd}	$2.66^{a–d}$	$13.26^{d–f}$	8.73^{ab}	1.05^{ef}	20.03^{bc}	4.21^{e}
'Crimson'	2.06^{b}	$2.53^{a–d}$	15.55^{de}	$7.51^{b–d}$	1.99^{b}	$7.44^{f–h}$	$5.29^{c–e}$
110 R	0.43^{fg}	0.26^{h}	$1.07^{ı}$	1.48^{h}	1.00^{ef}	3.32^{h}	–
1103 *P*	0.74^{ef}	0.81^{gh}	$7.69^{f–ı}$	2.35^{h}	$0.94^{e–g}$	$9.38^{e–g}$	–
99 R	0.30^{g}	0.39^{h}	$3.12^{hı}$	1.85^{h}	0.51^{g}	5.91^{gh}	–
41 B	1.68^{bc}	$2.72^{a–c}$	18.24^{cd}	8.40^{b}	1.90^{b}	23.58^{b}	–
140 R	0.56^{fg}	0.58^{h}	$4.44^{g–ı}$	2.05^{h}	0.62^{fg}	10.75^{ef}	–
V.R. Du Lot	0.48^{fg}	$2.62^{a–d}$	27.20^{ab}	5.83^{ef}	2.74^{a}	31.19^{a}	–
Kober 5 BB	1.68^{bc}	$2.39^{b–e}$	32.13^{a}	$6.21^{d–f}$	1.73^{b}	19.73^{bc}	–

[z]Letters after the numerals within the columns show significant difference at $p \leq 0.05$ according to Duncan's multiple range test

was observed that 'Regent' (Interspecies), 'Özer Beyazı' (*V. vinifera* + seedless), 'Muscat Bleu' (Interspecies) varieties and Kober 5BB rootstock had the highest number of roots. The best result in root length was obtained from 'Müşküle' (*V. vinifera*) variety, and the best result in shoot number was obtained from *Vitis rupestris* Du Lot American grapevine rootstock. According to the evaluation made according to shoot length, 'Muscat Bleu' (Interspecies) and *Vitis rupestris* Du Lot were the ones that gave the best results. In terms of the number of leaves on the shoot, 'Superior Seedless' and 'Kismish Rozovyi' varieties belonging to the *V. vinifera* species were determined as the varieties with the highest number of leaves on the shoot (Table 3).

Considering the average results from the applications, the 3rd, 4th and 5th applications were the ones that gave the best results in the callus status parameter. The second application was determined as the most successful application for root development level and number of roots. While the 1st application (control) stood out for root length, the 4th application was chosen as the best for shoot number. In terms of shoot length, the 1st application (control) was slightly more prominent than the other applications. Considering the number of leaves on the shoot, very similar results were obtained between the treatments and no statistical difference was found (Table 4).

Considering the effects of a total of five different applications on different *Vitis* species one by one, the results obtained can be summarized as follows: Although there was a numerical difference between callus level (0–3), root development level (0–4), number of roots, shoot length, number of shoots and number of leaves on the shoot in 'Atak 77' variety, this difference was not found to be statistically significant. The 5th application was statistically determined as the best application only in terms of root length.

In the 'Superior Seedless' variety, the 3rd and 4th applications stood out in terms of callus condition. It was determined that the 2nd application stood out in terms of root development level, root length, number of roots and shoot length. The 4th application stood out in terms of the number of leaves on the shoot. There was no statistical difference in the number of shoots.

When callus status, shoot length and number of leaves on the shoot were examined, no statistical difference was found in the 'Kismish Rozovyi' variety. The 1st, 3rd and 5th applications gave the best results in terms of root development level; the 1st and 3rd applications in terms of number of roots; the 1st and 5th applications in terms of root length; while in terms of the number of shoots, the 5th application gave the best results. Considering all applications, the 1st and 3rd applications (Control and Toniplant) gave better results than the others.

The results obtained from the 'Özer Beyazı' variety were as follows: the 3rd application in terms of callus condition; the 1st and 3rd applications in terms of root development level. Considering the number of roots, root length, shoot length, number of shoots and number of leaves on the shoot, the 1st application gave the best result.

The 1st and 4th applications gave the best results in terms of callus status in 'Heukboseok (4n)' variety; considering the root development level, number of root and root length, the 2nd application; 4th and 5th applications in terms of number of shoots; considering the shoot length and the number of leaves on the shoot, the 3rd application gave the best results.

In the 'Müşküle' variety, the third application stood out in terms of callus status; the 2nd and 4th applications at the root development level; the 2nd application in terms of number of roots, number of shoots and leaves on the shoot. In terms of root and shoot lengths, all applications except the 5th application gave the best results.

In the 'Kyoho (4n)' variety, there was no statistical difference in terms of callus status. The 2nd and 4th applications gave the best results in terms of root development level and root length; the 2nd application in terms of number of roots, shoot length and number of leaves on the shoot; considering the root length, the 2nd and 4th applications; while the 4th application gave the best results in the number of shoots.

While the 5th application gave the best results in callus status and shoot number in the 'Regent' variety, no statistical difference was found when looking at the root development level, number of roots, root length, shoot length and number of leaves on the shoot.

In the 'Rizpem' variety, the 4th application gave the best results in terms of root development level, while the 2nd application gave the best results in terms of root number. All

Table 4 Statistical analysis table of average applications of all varieties/rootstocks[z]

Application	Callus level (0–3)	Root growth level (0–4)	Number of roots	Root length (cm)	Number of shoots	Shoot length (cm)	Number of leaves on the shoot
Pure water	1.15^{ab}	2.03^{ab}	18.36^{b}	6.38^{a}	1.42^{ab}	14.64^{a}	3.35
2000 ppm IBA	0.95^{b}	2.28^{a}	22.28^{a}	6.20^{ab}	1.30^{b}	13.17^{ab}	3.31
TONIPLANT	1.18^{a}	1.88^{bc}	15.82^{bc}	5.50^{bc}	1.36^{ab}	13.46^{ab}	3.14
MAS RAIZ	1.25^{a}	1.92^{bc}	14.50^{bc}	5.41^{bc}	1.55^{a}	12.05^{b}	3.13
MAS RAIZ +IBA	1.24^{a}	1.69^{c}	12.88^{c}	4.94^{c}	1.41^{ab}	13.90^{ab}	3.02

[z]Letters after the numerals within the columns show significant difference at $p \leq 0.05$ according to Duncan's multiple range test

applications except the 5th application were found to be successful in terms of root length. No statistical difference was found in terms of callus status, number of shoots, shoot length, and number of leaves on the shoot.

Considering the callus status of the 'Muscat Bleu' variety, the 3rd and 5th applications gave the best results, while at the root development level, other applications were successful except the 3rd application. The 2nd application gave the best results in terms of number of roots; the 1st application in root length; the 5th application in terms of shoot length; while the 1st application gave the best result in terms of the number of leaves on the shoot. No statistical difference was found in the number of shoots.

Considering the root development level and number of roots in the 'Isabella' variety, the second application gave the best results. The best results in root length were obtained from the 1st and 3rd applications. No statistical difference was found when looking at callus status, number of shoots, shoot length and number of leaves on the shoot.

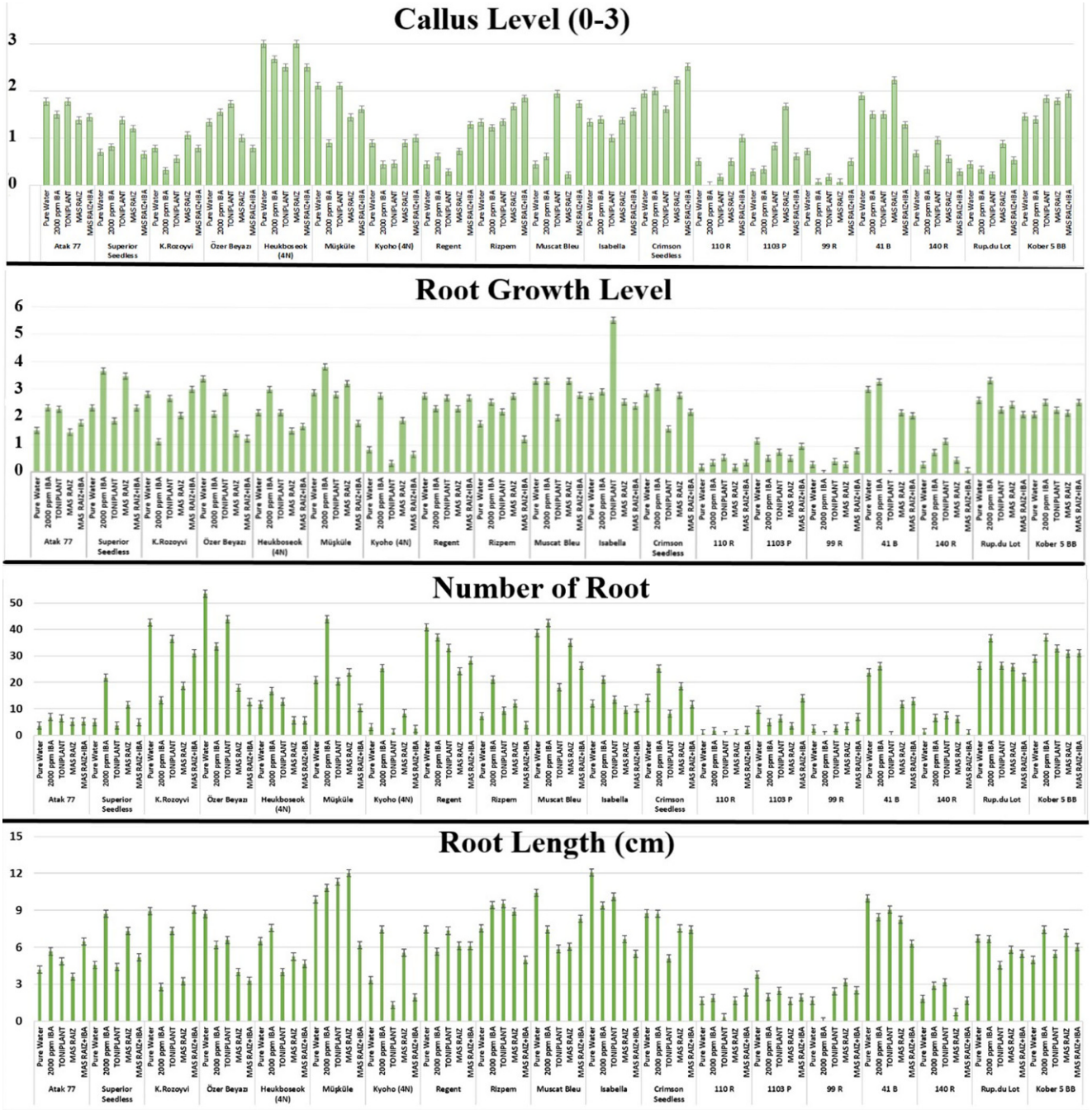

Fig. 6 Graphical representation of all applications separately for varieties/rootstocks

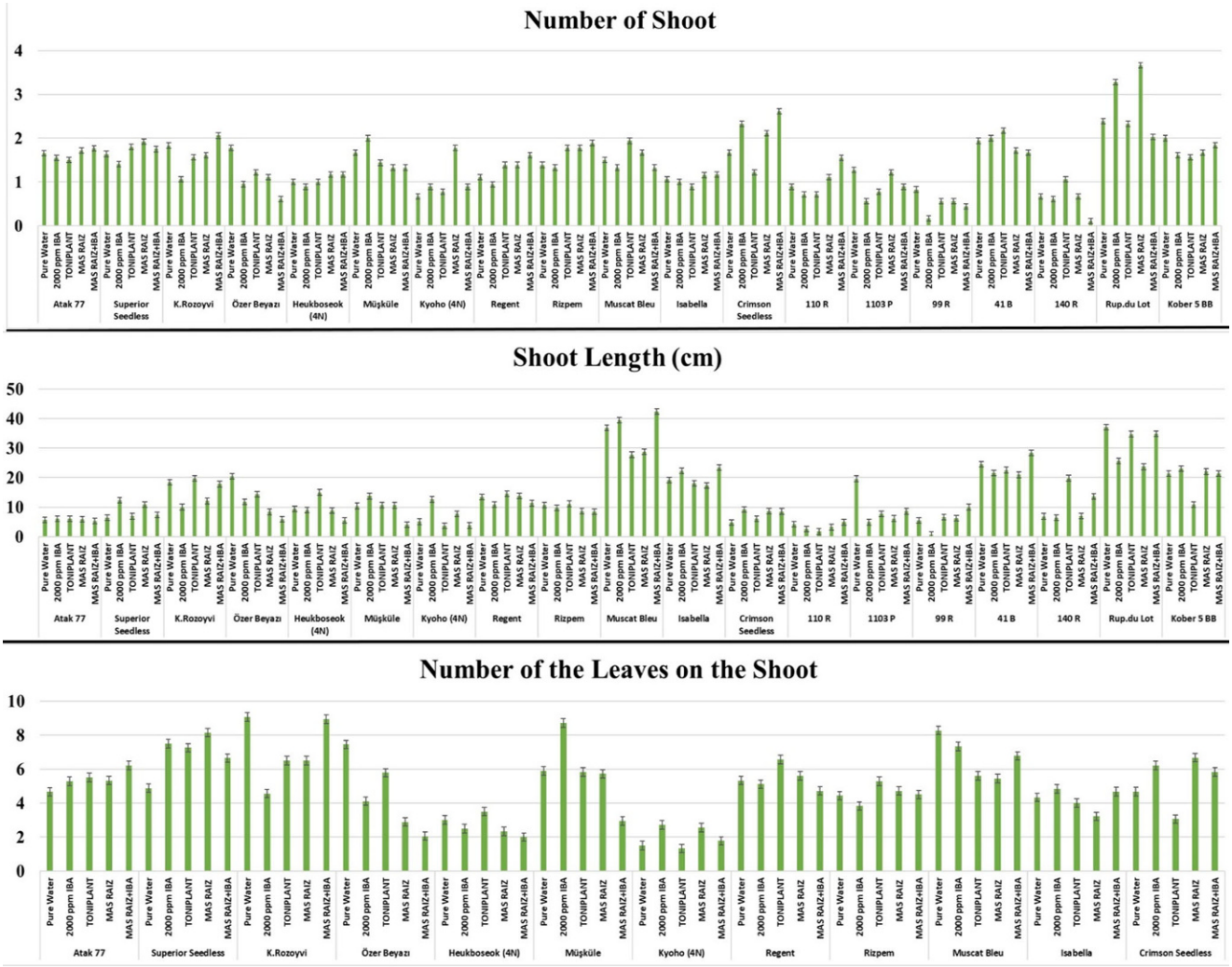

Fig. 6 (Continued)

There was no statistical difference in callus status and shoot length in 'Crimson Seedless', one of the seedless varieties. Applications that give the best results: the 2nd application in terms of root development level and number of roots; the 1st and 2nd applications in terms of root length; the 2nd and 5th applications in terms of number of shoots; the 2nd, 4th and 5th applications in terms of the number of leaves on the shoot (Fig. 6).

The results obtained from different applications to American grapevine rootstock cuttings can be summarized as follows: The 5th application gave the best results in terms of callus status and number of shoots on 110R rootstock. No statistical difference was found when looking at the root development level, number of roots, root length and shoot length.

The 4th application gave the best result in terms of callus condition on 1103 Paulsen rootstock. Considering the root length and shoot length, it was seen that the 1st application was successful. No statistical difference was found between the treatments in the root development level, number of roots and number of shoots.

No statistical difference was found in 99R rootstock in terms of callus status, root development level, number of roots, root length and shoot length. The 1st application gave the best result in the number of shoots.

No statistical difference was found when callus status, number of shoots and shoot length were examined in 41 B rootstock. The second application gave the best results in terms of root development level and number of roots. The 1st, 2nd and 3rd applications gave the best results in root length.

No statistical difference was found in the callus status, number of roots, root length and shoot length in the 140R rootstock. The 3rd application gave the best results in terms of root development level and number of shoots.

The 4th application gave the best results in the case of callus on *V. Rupestris* Du Lot rootstock; considering the root development level and number of roots, the 2nd application.

Parameters	Callus Level (0-3)	Root Growth Level (0-4)	Number of Root	Root Length	Number of Shoot	Shoot lenght	Number of leaves on the shoot
Callus Level (0-3)	**1.00**	-0.44	-0.04	0.30	-0.41	0.28	0.38
Root Growth Level (0-4)	-0.44	**1.00**	-0.03	-0.72	0.96	-0.69	-0.90
Number of Root	-0.04	-0.03	**1.00**	0.66	-0.30	0.26	0.43
Root Length	0.30	-0.72	0.66	**1.00**	-0.85	0.71	0.94
Number of Shoot	-0.41	0.96	-0.30	-0.85	**1.00**	-0.72	-0.98
Shoot lenght	0.28	-0.69	0.26	0.71	-0.72	**1.00**	0.73
Num. of leaves on the shoot	0.38	-0.90	0.43	0.94	-0.98	0.73	**1.00**

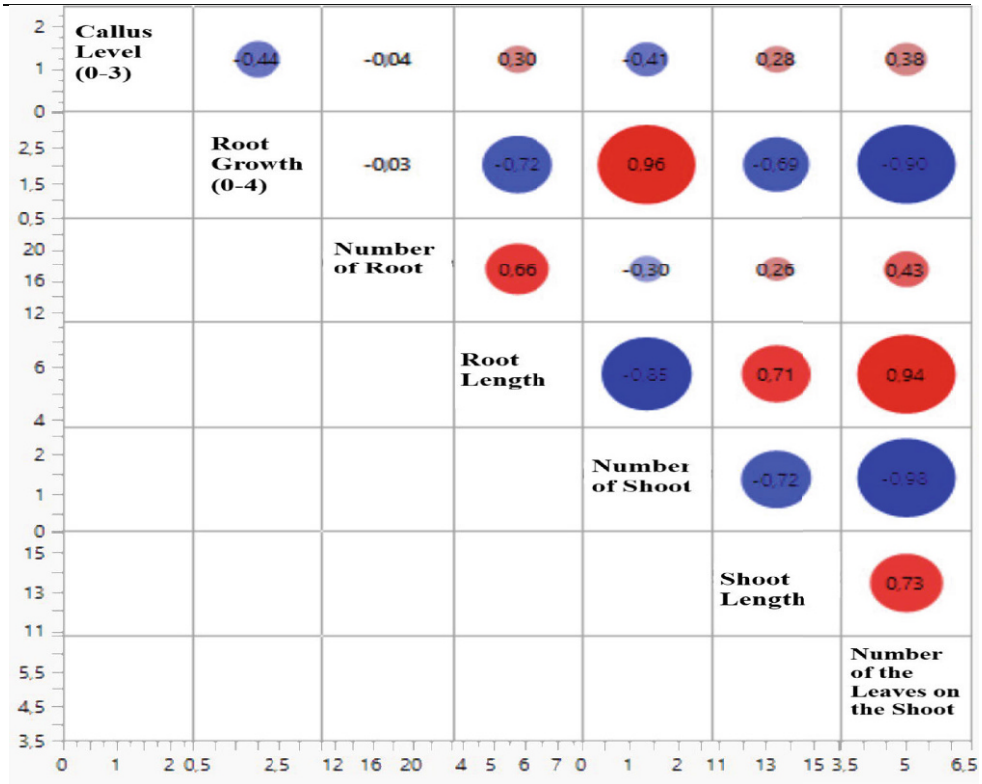

Fig. 7 Multivariate correlations and scatterplot matrix for the parameters of results (the correlations are estimated by the row-wise method)

The 1st application gave the best results in terms of shoot length. No statistical difference was found when looking at root length and number of shoots.

No statistical difference was found in Kober 5 BB rootstock when looking at callus status, root development level, number of roots, root length and number of shoots. The 1st, 2nd and 4th applications gave the best results in shoot length.

Multivariate correlations and scatterplot matrix evaluations of seven different parameters evaluated in the study are given in Fig. 7. In these correlation tables, a very high (–0.98) negative correlation emerged between the number of shoots and the number of leaves on the shoot, while a very high positive correlation (0.96) was detected between the number of shoots and root growth. Similarly, a very high positive correlation (0.94) was determined between the number of leaves on the shoot and root length.

Discussion

In the propagation of grafted saplings, when growing on rootstocks or on their own roots, cuttings of *Vitis* species are treated with different doses of plant growth regulators to help rooting. While it has been reported that IBA, one of these growth regulators, is especially effective, the 2000 ppm dose used in our study gave the best results in many studies (Satisha and Adsule 2006; Şen and Yağcı 2016; Rolaniya et al. 2018).

In a similar study conducted by İşlek et al. (2021) with cuttings of a local grape variety, the best result was obtained from the application of 1000 and 2000 ppm IBA in the average shoot length and number of leaves. They also reported that 1000 and 2000 ppm IBA applications were found to be more successful than IAA applications in terms of average leaf number and shoot length. In our study, although it varies depending on species and variety, better results were obtained in those treated with IBA compared to the control group. Apart from these studies, many other studies conducted with grape cuttings reported that 2000 ppm IBA application is the best IBA dose in terms of many parameters, and a very limited number of studies have been conducted with alternative growth regulators and other applications (Roberto et al. 2004; Rolaniya et al. 2018; Shahzad et al. 2019; Madhavan et al. 2021). However, it has been reported that the *Vitis* species used, the rooting medium, rooting conditions and some other factors, rather than the hormone and its appropriate dose alone, can affect the rooting rate and shoot development (Jaleta and Sulaiman 2019; Boeno and Zuffellato-Ribas 2023). Similarly, in our study, the most appropriate rooting application in different *Vitis* species differed on a parameter basis. While this result shows that IBA has positive effects on rooting, NAA or developments with similar content may be more effective in terms of other effects. A similar study was conducted by Kim et al. (2023) on the 'Shine Muscat' grape variety under in vitro conditions and found that NAA inhibited shoot growth and induced callus formation, while IBA and indole-3-acetic acid inhibited shoot growth. It has been reported that IAA generally gives better results in terms of root formation.

Additionally, the authors reported that the addition of exogenous auxin led to the initiation of new primordia formation and specifically promoted the development of the 'xylem pole' vasculature. It has also been reported that the establishment of this special vascular structure plays an important role in facilitating directional auxin transport for new root formation (Ikeuchi et al. 2019). The differences observed in root formation among auxins in our study are consistent with findings from other studies on woody plants, including *Vitis* spp (Smart et al. 2002; Zhao et al. 2022). The selection of appropriate auxin for root induction may vary between different plant species, with IBA being more preferred due to its more consistent and reliable rooting response (Frick and Strader 2018). Our study is also compatible with studies reporting the negative effects of NAA-containing applications on shoot and root induction, toxicity in some plant species, and problems in callus formation (Matsuoka and Hinata 1979).

In different studies, it has been reported that the presence of leaves is an important factor in the rooting of cuttings and that there is a positive relationship between leaves and cuttings showing good rooting (Goode Junior et al. 1982; Goode Junior and Lane 1983; Botelho et al. 2009). As a matter of fact, in our study, a very high correlation was determined between the number of shoots and leaves and rooting (Fig. 7).

El Shaima and Saleh (2018) evaluated the effects of seaweed and licorice which are natural extracts and compared them with IBA on rooting in a grapevine rootstock (Dog Ridge) cuttings. The results showed that both treatments improved the rooting quality and increased the number of roots/number of cuttings, plant height, fresh and dry weight and number of leaves/number of plants. Seaweed increased root length and leaf area more than IBA application. Researchers have reported that these natural extracts are effective in rooting and they can be use an alternative to IBA with more affordable costs, especially in organic production. In a similar study, Astha et al. (2022) compared different preparations of organic origin with different doses of IBA, and the best results were obtained from a preparation of organic origin. Similarly, in our MAS RAIZ application, which is of organic origin and has a rich content, results close to IBA were obtained in some varieties/rootstocks.

Based on our results, it may be concluded that instead of inorganic plant growth regulating hormones, which are often unintentionally used in excessive amounts, organic products can contribute sufficiently to both rooting and plant development, at least for some species and varieties; hence, they can be used as alternative products.

Conclusion

There are many varieties of the *Vitis* species, which is one of the most produced species in the world, but new varieties continue to be added to this list with breeding studies in line with consumer demands. In order to produce these existing and new varieties as grafted/non-grafted, it is necessary to help them root, otherwise the yield of saplings may decrease significantly. In this study, some applications that could be alternatives to the commonly used method for rootstocks/varieties belonging to different *Vitis* species (especially by including an application that can be used in organic production) were compared. As a result of the study, it was seen that different results could be obtained depending on

the variety and species, and it was also seen that some applications could be used as an alternative to the widely used and expensive IBA applications. In subsequent studies, testing different products (nanoparticles, organic preparations, etc.) and tissue culture applications may yield more efficient results, especially for newly developed varieties.

In addition, the most suitable products of organic or inorganic origin developed in recent years can be selected by testing, and the rooting efficiency of saplings can be increased in many fruit species, especially grapevine. Also positive contributions can be made to shoot development with these products.

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Author Contribution A. Atak: Oversaw the experiment, literature review, planned research, performed data analysis, supervised the study and wrote the manuscript. K. Çorak: Literature review, planned research, organised experiments, statistical analyses.

Conflict of interest A. Atak and K. Çorak declare that they have no competing interests.

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