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TURKISH HAZEL (*CORYLUS COLURNA* L.) OFFSPRING VARIABILITY AS A FOUNDATION FOR GRAFTING ROOTSTOCK PRODUCTION

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Abstract

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The production of Hazel planting material grafted on Turkish Hazel rootstocks in the Faculty of Agriculture nursery Rimski Sancevi had commenced in 1989. Preliminary research in the selection of Turkish Hazel parent trees for seed production and collection dates back to 1983. Quality seeds are a prerequisite for the successful production of seedlings. In the agro-ecological conditions of the nursery, monitoring of the one- and two-year-old Turkish Hazel seedling growth indicators demonstrated that one-year-old plants were not viable rootstocks. However, two-year-old seedlings, based on the development of the root neck and the achieved plant height, showed potential for grafting Hazel on Turkish Hazel rootstocks. In the first ten days of April 2010, tongue grafting was performed. The grafting success ranged from 53.10% on the rootstock A_1 for the decorative form *Corylus avellana* 'Atropurpurea', to 92.60% on the rootstock B_9 and *Tonda Gentile Romana* variety.

Key words: Turkish Hazel, nuts, seedling, grafting, nursery production

Introduction

At the Faculty of Agriculture in Novi Sad, in the Department of Fruit Growing, Viticulture, Horticulture and Landscape Architecture, the work on selection, breeding and production of Hazel planting material is being carried out. In 1983, the program for Turkish Hazel (*Corylus colurna* L.) genotype selection commenced, whereby trees from natural population as well as those sourced from Novi Sad green areas were used as source material. The selection included 45 Turkish Hazel samples from which several genotypes were identified. Genotypes are characterised by alternative yield, quality morphological, technological and physiological nut traits, as well as excellent health condition (Ninić-Todorović, 1990). The Turkish Hazel seedling growth parameters are currently understudied. In the controlled conditions, Harris et al. (1995, 2001) examined impact of stress and timing of replanting on the development intensity of the seedlings, both above and below ground, as well as the size of the Turkish Hazel habit.

Turkish Hazel seed material was collected from the parent trees growing in Futoski park, located at 45°15'6" N latitude, 19°49'39" E longitude and 79 m altitude. Sowing was conducted at the Faculty of Agriculture nursery Rimski Sancevi, on the plot located at 45°20'18" N latitude, 19°50'31" E longitude and 80 m altitude. The selected parent tree cultivation and the Turkish Hazel (*Corylus colurna* L.) planting material production were conducted under same ecological conditions.

The seedling development was monitored in the first and second year in the Rimski Sancevi fruit tree nursery. Hazel cultivar grafting was performed on the two-year-old Turkish Hazel seedlings by tongue grafting method. The results indicated excellent compatibility and grafting success rate. Seedlings grafted at

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a greater height are suitable for plantations where Hazel is grown as a tree, thus allowing full application of mechanisation and adequate protection. Hazel cultivar grafting methods using rootstocks of different varieties, as well as Turkish Hazel produced by generative means, was reported by Lagerstedt (1969, 1971, 1975).

In Serbia, Hazel cultivar grafting on the Turkish Hazel rootstocks has been successfully performed since 1989, as reported by Ninić-Todorović et al. (1994, 2003, 2006, 2007, 2011), Korać et al. (1995, 1996), Cerović et al. (2007) and other authors.

Matherials and Methods

Turkish Hazel fruits were collected now of the cupule stem physiological independence from the parent tree. The maturity indicator is the yellow-green cupule colour, combined with the light brown colour of the visible part of the pericarp (Ninić-Todorović, 1990). Nut sowing was conducted mid-October 2008, in 200 m-long rows, 1.20 m apart, intended for mechanized cultivation. The growth indicator studies were carried out on 30 one- and two-year-old seedlings each, in four repetitions. Tree height and root length were measured. For the determination of root system morphological characteristics, image analysis software package "ImageJ" was used (Wayne Rasband, National Institute of Health, USA). Seedling root was prepared using the Bouma et al. (2000) method, cleaned meticulously and rinsed with distilled water, after which it was scanned with 400 dpi resolution in 8-bit grey scale, using Canon CanoScan 4400F scanner. Image conversion into binary mode suitable for analysis was performed by the ImageJ software, using triangle method.

Root neck thickness was determined using micrometer with 0.01 mm precision. Presence of I and II order roots was detected. Tree and root mass were measured using technical scales with 0.01 g precision.

One- and two-year-old seedlings were harvested during the dormant vegetative period in 2009 and 2010, respectively.

Seedling morphological characteristics measurement results were tabulated and subsequently subjected to statistical analysis. In order to determine trait variability, for each characteristic examined, coefficient of variation (CV) was shown. Statistical significance of differences amongst genotypes was tested using Duncan test included in the statistical software package STATISTICA 11 (StatSoft, Inc., Tulsa, OK, USA).

The pH value of the soil was determined in the suspension of soil and H_2O, by METREL, MA 3657 pH meter, whereas $CaCO_3$ content was established volumetrically, using Scheibler calcimeter. The total N content was determined by CHNS analyzer (ELEMENTAR, Vario EL, Elementar Analysensysteme GmbH, Hanau, Germany). The humus content was measured through oxidization of organic matter by potassium bichromate (Simakov, Tsyplemkov, 1969). Finally, available P and K contents were determined using AL method (Enger et al., 1960).

Results and Discussion

In the plant nursery, sowing seeds for fruit variety rootstock production, including Turkish Hazel, is common practice, due to the lack of qualified permanent staff with relevant expertise. Consequently, most of the work is performed in autumn, when the soil is of favourable composition, well prepared and irrigated. Nuts were sawn 7 cm apart at the 6-8 cm depth. The seed interspacing of 7 cm is optimal for Turkish Hazel seedling development in the first two vegetative periods in the nursery. The seeds sprout hypogenically, cotyledones are easily separated from the pericarp, and the first seedlings appear in May.

In the 2009 vegetative period, the care measures applied to the one-year-old seedlings included destruction of crust, hoeing, weeding and watering using drip system. As seedlings proved resistant to pests and diseases, the use of protective measures has been omitted.

Seedling production must satisfy fundamental criteria, such as high quality, high yield per unit area and economical viability of the production process. These goals can only be met if the seedling production is based on the use of high quality reproductive material, is performed in the conditions suitable for the given environment, and respects economic laws implicit in any type of production. European directive EU 2092/91 stipulates that plants used as seed material in biological production must be cultivated for the minimum of two years, so that the seeds can be used for rootstock production (Lammerts van Beuren et al., 2004).

Turkish Hazel seedling material production meets all the cost-effectiveness criteria and aims. In addition to the necessary maintenance, grafting is performed *in situ*, without the application of protective measures, which confirms the economic viability of the three-year production cycle.

From the nursery production perspective, the tree and root development biological characteristics are mostly exhibited in the first two years of seedling growth. Measurements of one-year-old plant tree height, root length and other characteristics are shown in Table 1.

The analysis of one-year-old Turkish Hazel (*Corylus colurna* L.) seedlings indicated statistically significant and very important differences amongst examined characteristics of different genotypes (Table 1). The plant height varied from 13.48 cm to 17.82 cm, with the highest coefficient of variation in this parameter found in genotype B_9 (16.31%). The genotype B_2 is also singled out with the highest mean values of root length (35.13 cm), number of first order roots (49.36) and root mass (4.18 g). This genotype, however, does not show significant deviation in plant height, root neck thickness and tree mass, compared to other genotypes. The lowest mean root length values (30.81 cm), number of first order roots (32.30) and root mass (2.93 g) were found in B_9 genotype, for which the highest values of coefficient of variation in measured parameters were also found. The root neck thickness values varied from 5.66 mm (B_4) to 5.89 mm (A_1) and no statistically significant differences amongst examined genotypes were found. The root mass values do not show significant variation amongst selected samples either. The coefficient of variation values ranging from 45.17% (A_1) to 56.43% (B_9) indicate that the tree mass is the most variable one-year-old Turkish Hazel plant characteristic.

Seed material characteristics, external environmental factors and implemented technological nursery

Table 1

One-year-old Turkish Hazel (*Corylus colurna* L.) plant growth indicators in 2009

Genotype	Seedling height, cm		Seedling root lenght, cm		Root neck thickness, mm		Number of first order roots		Tree mass, g		Root mass, g	
	$\bar{x}$	C_V(%)	$\bar{x}$	C_V(%)	$\bar{x}$	C_V(%)	$\bar{x}$	C_V (%)	$\bar{x}$	C_V(%)	$\bar{x}$	C_V(%)
A_1	13.48a	11.56	34.23ab	16.87	5.89a	17.75	42.76ab	20.27	2.32b	45.17	3.72a	32.56
B_2	16.01b	13.63	35.13b	21.91	5.84a	19.44	49.36b	25.02	2.53b	47.65	4.18a	40.52
B_4	16.82ab	13.71	33.13ab	18.48	5.66a	20.04	40.28a	18.12	2.00a	48.21	3.75a	36.19
B_5	17.82a	14.39	32.18ab	18.83	5.67a	18.94	40.51a	18.31	2.81c	50.19	4.09a	37.65
B_9	16.84ab	16.31	30.81a	29.7	5.71a	20.43	32.30c	24.05	1.80a	56.43	2.93a	49.37

Differences in values within columns that are not followed by the same letter are statistically significant at a 0,05 significance level

Table 2

Two-year-old Turkish Hazel (Corylus colurna L.) plant growth indicators in 2010

Genotype	Seedling height, cm		Seedling root lenght, cm		Root neck thickness, mm		Number of second order roots		Tree mass, g		Root mass, g	
	$\bar{x}$	C_V(%)	$\bar{x}$	C_V(%)	$\bar{x}$	C_V(%)	$\bar{x}$	C_V (%)	$\bar{x}$	C_V(%)	$\bar{x}$	C_V(%)
A_1	42.53a	31.04	46.83a	13.27	12.38a	34.98	19.56b	30.44	12.56a	48.05	11.00a	41.57
B_2	46.82c	26.75	48.13ab	23.8	9.74b	20.22	22.63e	30.79	9.16b	44.85	14.29b	36.97
B_4	47.63e	31.67	51.01b	18.56	11.11d	25.32	16.36a	23.88	10.66d	28.95	15.28c	43.57
B_5	47.34d	35.41	48.80ab	19.79	10.76c	21.03	19.70c	31.41	12.56a	33.48	17.50e	44.51
B_9	46.63b	24.68	49.66ab	17.15	9.36a	24.46	21.50d	26.92	9.36c	38.49	15.69d	31.75

Differences in values within columns that are not followed by the same letter are statistically significant at a 0,05 significance level

processes played a significant role in the formation of one-year-old Turkish Hazel seedlings. Development of Turkish Hazel seedlings in second vegetative year is shown in Picture 1.

In the second year of the study, in addition to the aforementioned growth indicators, presence of second order roots was noted. The two-year-old Turkish Hazel (*Corylus colurna* L.) seedling growth parameter values indicate significant variability amongst selected samples (Table 2). Based on the analysis results, the B_4 genotype can be singled out, due to the highest plant height (47.63 cm) and root length (51.01 cm) mean values. The lowest mean values of these parameters were noted in genotype A_1, for which the highest root neck thickness (12.38 mm) and the number of second order roots (12.56) was recorded. The root length was the characteristic with the lowest variability, i.e. the examined genotypes demonstrated considerable similarity in this respect. The coefficient of variation for this parameter ranged from 13.27% (A_1) to 23.80% (B_2). The greatest number of first order roots (22.63), yet the smallest number of second order roots (9.16), was recorded for B_2 genotype. Mean tree mass values ranged from 11.00 g (A_1) to 17.50 g (B_5), whereas root mass varied from 22.75 g (A_1) to 29.27 g (B_9). These two parameters demonstrated significant variation in two-year-old Turkish Hazel seedlings. The tree mass coefficient of variation values ranged from 31.75% (B_9) to 44.51% (B_5), whereas high values — 24.37% (B_9) to 41,00% (A_1) — were also noted for root mass.

The comparison of the of the most significant growth parameters in one- and two-year-old Turkish Hazel seedlings indicated marked differences in growth trends. The most significant difference was observed in the heights

Picture 1. Development of Turkish Hazel (*Corylus colurna* L.) seedlings in second vegetative year

of the examined plants, with the two- to three-fold increase in height in the second year, depending on the genotype. Similarly, the root length followed the tree growth intensity. However, smaller variation in root length growth parameters between one- and two-year-old plants was recorded, compared to the corresponding tree height measurements.

The successful seedling development in the nursery can also be attributed to the favourable regional ecological conditions. Of particular importance is the high quality soil of calcareous chernozem type. Basic chemical characteristics of the soil are presented in Table 3.

The soil is characterised by the neutral reactivity, medium available phosphorus content and optimal available potassium content. The humus substances (2.70%) adequately supply the top soil (0 – 30 cm), which is beneficial for the root system development in the first year of seedling growth. At the end of the first vegetative period, the mean root length ranged from 30.81 cm (B_9) to 35.13 cm (B_2). The one-year-old Turkish Hazel plant root system is of taproot form, with numerous fibrous first order roots. The one-year-old seedlings, however, are not viable fruit growing rootstock material due to their insufficient development.

The marked root system growth trend in the second vegetative year has allowed for the development of second order roots, increased root neck thickness, as well as the tree height, which reflected in the greater tree and root mass. Thus, two-year-old seedlings are viable rootstock material, as root neck thickness and plant height are the key indicators of grafting success. Hazelnut cultivar grafting in the fruit tree nursery Rimski Sancevi is therefore conducted on two-year-old Turkish Hazel seedlings by tongue grafting method. Grafting is performed *in situ* in the first ten days of April, as in that period the Turkish Hazel rootstock starts running juices and the first leaves appear. The Hazel cultivar scions are kept in cold storage, at 2 − 3°C and 95% average air humidity, so that, at the time of grafting, they are in the dormant phase. The grafting is performed at heights of 20 to 30 cm from the base. The grafting success rate is high, ranging from 53.1% for A_1 rootstock for decorative form *Corylus avellana* 'Atropurpurea' to 92.6% on rootstock B_9 and *Tonda Gentile Romana* variety. The grafting success is affected by the rootstock, variety, scion storage method and meteorological conditions during and after grafting.

At the end of the vegetative period, the grafted hazel plants are sufficiently developed for extraction. Plants are separated by quality into first and second class, thus enabling distribution.

Grafted hazel plant production is economically viable even on three-year-old rootstocks cultivated *in situ*, whereby tongue grafting is performed on three-year-old Turkish Hazel at the height 60 to 80 cm from the tree base.

Production of hazel variety planting material grafted on Turkish Hazel rootstocks meets the production process economic viability criteria in the favourable ecological conditions of the nursery near Novi Sad. Regular application of technological nursery processes is conducted in parallel with the production of planting material for other fruit varieties. As, since 1989, insect had not affected Turkish Hazel seedlings or pathogen attacks, there had been no need for any protective measures. Ecological production of hazel seedlings is viable when planted in gardens or as a part of intensive cultivation systems. In comparison to other fruit cultures, hazel requires reduced protection. Thus, rootstock production aimed at biological cultivation of planting material is possible, albeit at the higher cost of planting material.

Conclusion

The seeds for rootstock production are harvested from parent plants grown without protection or fertilisation.

Table 3
Basic chemical characteristics of the soil in Rimski Šančevi nursery

Depth, cm	pH, KCl	pH, H_2O	$CaCO_3$, %	Humus, %	N total, %	Al-P_2O_5, mg 100g-1	Al-K_2O, mg 100g-1
0-30	7	8.33	0.83	2.7	0.18	9.63	23.06
30-60	7.31	8.42	0.69	2.58	0.17	1.48	17.63

Given the sturdiness of the plant material and the potential for cultivation without applying protective measures, the shift towards biological rootstock production in Rimski Sancevi is possible in the forthcoming period.

Plants developed from seeds *in situ* in the first year have axial roots with fibrous veins. In the second year, the root system proliferates, spreading across surprisingly large soil volume. In naturally occurring plants, the tree-root mass ratio ranges from 1:1 to 2:1 (Stein, 1978, cited in Stilinović et al., 1980). At the time of replanting, in many varieties, that ratio increases in favour of the above-ground part of the plant. The generative Turkish Hazel reproduction method implemented in Rimski Šančevi nursery yielded similar findings. After two-year-long cultivation period in the nursery, Turkish Hazel can be used in fruit growing, as a rootstock for hazel grafting. Four varieties were grafted with the following grafting success rates: *Tonda Gentile Romana* 92.60%, *Rimski* 91.98%, *Cosford* 83.82%, *Istarski dugi* 82.31% and the decorative cultivar *Corylus avellana* 'Atropurpurea' 53.10%. Excellent *Corylus colurna* L. rootstock compatibility with hazel scion varieties was established. The scions were secured with polyethylene tapes and the scion cut protected by grafting wax.

Based on the growth indicators presented here, it can be determined that the first class seedlings should meet the following criteria: plant height of 17 – 20 cm (genotype B_5), root length 34 – 38 cm (genotypes A_1 and B_2) and root neck diameter of 5 – 7 mm. For first class two-year-old plants, these criteria are 45 – 50 cm, 50 – 60 cm and 9 – 12 mm, respectively.

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