# FRUIT CHARACTERISTICS OF PROMISING WALNUT GENOTYPES FROM THE REGION OF EASTERN SERBIA

Svetlana M. PAUNOVIĆ\*, Rade K. MILETIĆ

Fruit Research Institute, Čačak, Republic of Serbia

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This research was conducted to examine the fruit characteristics of selected walnut genotypes from the native population of Eastern Serbia. Over 28 trees that deserve attention were recorded through the selection procedure. By grafting, grouping and cultivation under the same conditions at the Fruit Research Institute in Čačak, seven promising genotypes with the most favourable fruit characteristics were identified. Important physical properties of the fruit, such as fruit weight, kernel weight, fruit dimensions and some kernel and shell properties were investigated. The fruits of the selected genotypes were large or very large. The fruit weight ranged from 13.4 to 17.9 g, whereas the kernel weight varied from 6.22 to 8.92 g. The kernel was light-coloured or yellow, of very good taste. The kernel percentage in the tested genotypes was high, over 50.0% (50.7–55.3%). Pearson's correlation coefficient revealed a significant correlation between fruit and kernel characteristics. PCA analysis showed an adequate grouping of genotypes based on fruit quality. The obtained results suggest that selected promising walnut genotypes showed very good fruit properties, and may contribute to the increase of walnut production in the future.

Keywords: Juglans regia L., genotype, fruit properties

## INTRODUCTION

Walnut production has a long tradition in Serbia, and its wide range of applications makes it one of the most appreciated fruit species. Serbia has favourable climatic and soil conditions for

Corresponding author: Svetlana M. Paunović, Fruit Research Institute, Čačak, Republic of Serbia, Kralja Petra I, 932 000 Čačak, Serbia, Phone: +381 32 321 375, Fax: +381 32 321 391, E-mail: <a href="mailto:svetlana23869@gmail.com">svetlana23869@gmail.com</a>

the intensive production of walnut. The increase in the standard of living has led to increasing demand for walnuts, given their incomparably high nutritional value. However, current walnut production is insufficient to meet the country's needs, as it supplies less than 50% of national requirements (PAUNOVIĆ and MILETIĆ, 2013). The reason for this is the generative propagation of walnut from natural populations over the last several decades, resulting in the presence of populations with a pronounced biotype-specific polymorphism. The very rich and heterogeneous population represents a significant available genetic fond, which presents good opportunities for walnut breeding to obtain new cultivars or to use them in cross-breeding activity. In addition, high variability in walnut populations affects fruit properties by increasing diversity in fruit weight, kernel weight, shell thickness and colour, and other morphological properties (SHARMA and SHARMA, 2001a; OZKAN and KOYUNCA, 2005).

Fruit weight and kernel percentage are important parameters that affect fruit quality. However, fruit weight and kernel percentage are not only effective indicators of the external and internal quality of walnuts but also significant parameters for walnut breeding. Therefore, many studies have been carried out dealing with fruit characteristics of promising genotypes of walnuts grown in different countries (SIMSEK, 2010; COSMULESCU and BOTU, 2012; SHARMA *et al.*, 2014; POLAT *et al.*, 2015; JAĆIMOVIĆ *et al.*, 2020).

With this in mind, the purpose of the investigation was to determine the fruit characteristics of promising walnut genotypes that will be further included in the procedure for the registration of cultivars.

### MATERIALS AND METHODS

The present study was conducted in a collection of walnut genotypes planted at the Fruit Research Institute, Čačak, Western Serbia. All planted genotypes (28) originated from the natural population of Eastern Serbia. Investigated genotypes had been grafted and grown in identical conditions. Seven promising genotypes with the most favourable properties were selected according to fruit characteristics.

The description of fruit characteristics is given based on the descriptor for the walnut from the International Union for the Protection of New Varieties of Plants (UPOV, 1999).

Approximately 50 fruit samples were collected from each genotype and certain fruit parameters were evaluated. Fruit weight and kernel weight were determined on a Mettler precision scale with a readability of 0.01 g. The kernel percentage was determined based on fruit and kernel weight.

#### Statistical analysis

Pearson's correlation coefficient was used to determine the relationship between fruit characteristics. Principal Component Analysis (PCA) was performed to determine the difference in significance between walnut genotypes. Statistical analyses were carried out using Statgraphic Centurion 18 program (Manugistics, Inc., Rockville, MD, USA).

## RESULTS AND DISCUSSION

In many countries, the selection of walnut is carried out by the method of simple selection out of natural populations with high-fruit quality trees (COSMULESCU and BOTU, 2012). However, the fruit quality in walnut populations is quite non-uniform, which is the result of high

variability. The present data demonstrate that the population was dominated by walnuts with very small (46.7%) and small fruits (32.3%), whereas those with large (4.3%) and very large fruits (2.7%) were in minority (Table 1).

Table 1. Fruits weight in walnut population

Fruit weight (g)	< 8.0	8.1-10.0	10.1-12.0	12.1-14.0	>14.1
Share in walnut	46.7	22.2	14.0	4.3	2.7
population (%)	40.7	32.3	14.0	4.5	2.1

A considerable percentage of trees with small and very small fruits are indicative of population specificity, which is in agreement with the findings of ZENELI *et al.* (2005), who reported that in native populations of walnut, there are trees with great differences in fruit weight. Also, MILETIĆ *et al.* (2010) found that the walnut population is dominated by trees with small fruits (30.1%), while those with very large fruits are the fewest (0.07%).

Kernel percentage is a very significant property and parameter of quality of the population studied, especially from the standpoint of economics. The studied walnut population was dominated by fruits with medium (29.0%), high (28.7%) and low (16.0%) kernel percentage, whereas the share of fruits with extremely low (2.2%) kernel percentage was least evidenced (Table 2).

Table 2. Kernel percentage in walnut fruits

Kernel percentage	<30	30-35	35-40	40-45	45-50	50-55	>55
Share in walnut	2.2	7.2	16.0	20.0	20.7	15.0	2.5
population (%)	2.2	1.2	16.0	29.0	28.7	15.2	3.3

Differences in fruit weight and kernel percentage in the walnut population can be explained by the generative propagation of walnut in natural populations and growing walnuts without human activity and agrotechnics under different soil and climatic conditions (JAĆIMOVIĆ et al., 2020).

By grafting, grouping and cultivation under the same conditions in the Čačak region, seven promising genotypes were selected based on fruit characteristics. The data showed that the selected walnut genotypes were of large and very large fruit and kernel weight. The fruit weight ranged from 13.4 to 17.0 g, while the kernel weight varied from 6.22 g to 8.92 g (Fig. 1). The average fruit and kernel weight was highest in genotype ČA12 and the lowest in genotype ČA5. Also, the high fruit and kernel weight were detected in genotypes ČA4 (16.5 g and 8.53 g, respectively) and ČA2 (16.1 g and 8.22 g, respectively). In the walnut breeding program increasing the weight of kernel is a priority, whereas good kernel quality is a desirable and important property in walnut production (COSMULESCU and BOTU, 2012).

The results on fruit dimensions are shown in Table 3. Fruit length, width, and height of the tested walnut genotypes varied from 37.89 mm (ČA3) to 52.13 mm (ČA2), 28.99 mm (ČA5) to 38.43 mm (ČA4) and 29.67 mm (ČA5) to 38.65 mm (ČA4), respectively.

Kernel percentage is highly influenced by fruit dimensions and weight, which is an important feature in the selection of genotypes with favorable pomological characteristics (COSMULESCU and BOTU, 2012). The presented results showed that the percentage of kernel in total fruit weight in all cases was high (over 50.0%). The highest kernel percentage was determined in genotypes ČA4 (55.3%) and ČA3 (55.2%), whereas the lowest in genotype ČA38 (50.7%). In general, a high kernel percentage is detected in other examined genotypes, ranging from 51.2% to 54.6%. The fruits were mostly elongated or round. The kernel colour was yellow or light, and the taste very good. SHARMA *et al.* (2014) reported that the colour of the kernel is one of the most important pomological characteristics, and that walnut fruits with light kernel colour are considered to be of better quality compared to amber and brown colour.

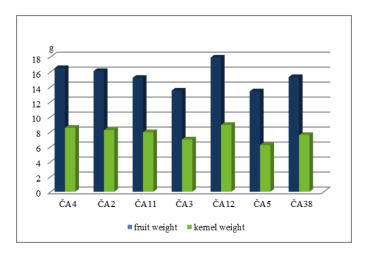


Figure 1. Fruit and kernel weight of selected walnut genotypes

Table 3. Fruit and kernel characteristics of selected walnut genotypes

Selection		Fruit dimensions (	mm)	Kernel	Fruit	Kernel	Kernel
	Length	Width	Thickness	percentage	shape	colour	taste
ČA4	42.78	38.43	38.65	55.3	Round	Yellow	Very good
ČA2	52.13	36.07	37.02	54.6	Round	Yellow	Very good
ČA11	45.23	35.97	34.09	53.5	Elliptical	Light yellow	Good
ČA3	37.89	31.61	32.40	55.2	Oval	Light brown	Excellent
ČA12	48.57	35.43	38.02	52.4	Elliptical	Light yellow	Excellent
ČA5	39.63	28.99	29.67	51.2	Round	Yellow	Very good
ČA38	37.21	32.53	35.77	50.7	Elliptical	Light yellow	Very good

All isolated walnut genotypes had moderately prominent or prominent sutures. The shell surface was smooth or slightly wrinkled, mostly light or light brown, with thin or medium thickness (Table 4).

Table 4. Shell characteristics of selected walnut genotypes

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Selection	Suture	Shell	Shell	Shell thickness
	prominence	surface	colour	
ČA4	Moderately prominent	Slightly wrinkled	Light	Thin (3)
ČA2	Prominent	Smooth	Light	Thin (3)
ČA11	Prominent	Smooth	Light	Medium (5)
ČA3	Moderately prominent	Smooth	Light brown	Medium (5)
ČA12	Moderately prominent	Slightly wrinkled	Light	Thin (3)
ČA5	Prominent	Smooth	Light	Thin (3)
ČA38	Moderately prominent	Slightly wrinkled	Light	Thin (3)

These results can be supported by the findings of MILETIĆ *et al.* (2003), MITROVIĆ and MILETIĆ (2007) and CEROVIĆ *et al.* (2014), who presented similar results in terms of the physical properties of fruits and kernels of walnut genotypes investigated from the natural population of Serbia. Many studies have shown similar fruit characteristics in walnut trees from natural populations in different countries. JAĆIMOVIĆ *et al.* (2020) indicated that walnut selections from Montenegro have fruit weight between 8.43 and 13.84 g, kernel weight between 4.20 and 6.54 g and kernel percentage between 39.20 and 52.25%. Also, the result of research by SKENDER *et al.* (2020) on the fruits and kernel properties in Bosnia and Herzegovina reported that the average nut thickness ranged from 23.27 to 37.15 mm, nut width from 22.47 to 31.18 mm, nut height from 24.71 to 43.30 mm, nut weight5.86 to 16.25 g, kernel weight from 1.66 to 5.07 g and kernel percentage from 26.96% to 48.25%. Furthermore, SIMSEK (2010) determined in Turkey that fruit weight, fruit length, kernel weight and kernel ratio varied from 14.31 to 9.63 g, from 44.93 to 34.00 mm, from 6.99 to 5.38 g and from 62.16 to 44.06%, respectively.

Table 5. Correlations between fruit and kernel characteristics

	FW	FL	FWID	FT	KW	KP
FW	1	0.523	0.655*	0.673*	0.722*	-0.235
FL		1	0.635*	0.710*	0.676*	-0.157
FWID			1	0.688*	0.713*	0.120
FT				1	0.653*	-0.237
KW					1	-0.325
KP						1

<sup>\*</sup> FW – fruit weight; FL – fruit length; FWID – fruit width; FT – fruit thickness;

KW-kernel weight; KP-kernel percentage.

In this study, a significant correlation was observed between fruit and kernel characteristics (Table 5). The highest correlation was found between fruit weight and kernel weight (r = 0.722), which is in agreement with the findings of ESKANDARI *et al.* (2005), who recorded a positive correlation between kernel weight and fruit weight. Correlation coefficients among fruit characteristics revealed a significant positive correlation of fruit weight with fruit length (r = 0.523), fruit width (r = 0.655) and fruit thickness (r = 0.673). Moreover, fruit dimensions were significantly correlated with each other (r = 0.635-0.710). A high correlation was also perceived between kernel weight and fruit dimensions (r = 0.653-0.713), confirming the previously established finding by MAHMOODI *et al.* (2016) and GHANBARI *et al.* (2018), who reported significant correlations among different fruit characteristics such as fruit weight, fruit dimensions and kernel weight. ARZANI *et al.* (2008) found a few significant correlations between fruit weight and kernel weight, as well as between fruit weight and fruit dimensions.

Unlike previous characteristics, a low negative correlation was found between kernel percentage and fruit weight, as well as kernel weight and fruit dimensions (length, width, thickness), which is comparable to the research by SHARMA and SHARMA (2001b), who observed that the negative correlation between kernel percentage and fruit weight, and fruit size was a reflection of higher weight of endocarp. In general, numerous studies have recorded significant correlations between fruit weight and kernel weight, and also between fruit weight and fruit dimensions (AMIRI *et al.*, 2010; MOSIVAND *et al.*, 2013; TOOLIR and MOZAFFARI, 2020).

Principal Component Analysis (PCA) was conducted between fruit and kernel characteristics, and the results were explained with the three principal components (Table 6). The first principal component concentrated 54.1% of the total variance, the second 21.7%, and the third 12.6%.

Table 6. Principal component loadings of some fruit and kernel characteristics

Variable	PC1	PC2	PC3
fruit weight	0.967*	0.046	0.140
fruit length	0.830*	0.268	-0.344
fruit width	0.778*	-0.047	0.034
fruit thickness	0.748*	0.247	0.279
kernel weight	0.994*	0.079	0.201
kernel percentage	0.445	0.887*	0.450
Eigenvalue	5.952	2.392	1.395
Variance (%)	54.1	21.7	12.7
Cumulative (%)	54.1.5	75.9	88.5

Variables with a correlation coefficient larger than 0.70 (absolute value) were determined in PC1, and had a high contribution for fruit weight, fruit dimensions (length, width and thickness) and kernel weight, while PC2 had a high loading for kernel percentage.

The relationship of the analysed walnut genotypes can be explained by a Scatterplot of the variables (Fig 2.). A grouping of walnut genotypes shows that the separation was done according to the fruit and kernel characteristics. Walnut genotypes formed four separate clusters in different zones of the sample map. The first group was composed of ČA11 and ČA12

selections represented in the positive part of PC1, separated according to the excellent fruit and kernel properties. The second and third groups included selections ČA2, ČA4 and ČA3, which represent a moderately distinct group in terms of fruit and kernel characteristics compared to genotypes from the positive part of PC1. Genotypes ČA5 and ČA38 from the fourth group showed lower values of the tested parameters compared to other genotypes.

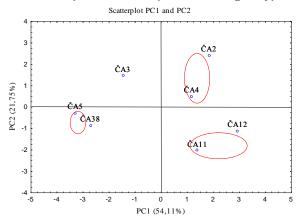


Figure 2. Scatterplot based on characteristics of fruit and kernel in promising walnut genotypes

#### **CONCLUSION**

The tested walnut selections singled out from the populations of Serbia exhibited excellent characteristics of the fruit. Fruits of these genotypes are large and very large, elongated or roundish, with a large kernel percentage. Pearson's correlation coefficient revealed a significant correlation between fruit characteristics, especially between fruit weight and kernel weight. Also, PCA showed a clear separation of walnut genotypes in four groups based on fruit and kernel characteristics. The genotypes ČA11 and ČA12 particularly stood out with the highest fruit and kernel weight, as well as a satisfactory colour of the kernel. Given the good fruit characteristics, all stated genotypes suggest the necessity for further testing and introduction in production, and can be used in breeding programs in the future for their positive fruit properties.

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## OSOBINE PLODA PERSPEKTIVNIH GENOTIPOVA ORAHA IZ REGIONA ISTOČNE SRBLIE

Svetlana M. PAUNOVIĆ, Rade K. MILETIĆ

Institut za voćarstvo, Čačak, Republika Srbija

#### Izvod

Istraživanje je sprovedeno u cilju ispitivanja karakteristike plodova odabranih genotipova oraha iz autohtone populacije istočne Srbije. U postupku selekcije evidentirano je više od 28 stabala koja zaslužuju pažnju. Kalemljenjem, grupisanjem i gajenjem u istim uslovima, u Institutu za voćarstvo, Čačak, identifikovano je sedam perspektivnih genotipova sa najpovoljnijim osobinama ploda. Ispitivane su važne fizičke osobine ploda, poput mase ploda, mase jezgre, dimenzije ploda i nekih osobina jezgre i ljuske. Plodovi odabranih genotipova su bili krupni ili veoma krupni. Masa ploda se kretala od 13.4 do 17.9 g, dok je masa jezgre varirala od 6.22 do 8.92 g. Boja jezgre je bila svetla ili žuta, veoma dobrog ukusa. Randman jezgre kod ispitivanih genotipova bio je visok, preko 50.0% (50.7–55.3%). Pearsonov koeficijent korelacije je otkrio značajnu korelaciju između karakteristike ploda i jezgre. PCA analiza je pokazala adekvatno grupisanje genotipova na osnovu kvaliteta ploda. Dobijeni rezultati ukazuju da su odabrani perspektivni genotipovi oraha pokazali veoma dobre osobine ploda, i da mogu doprineti povećanju proizvodnje oraha u budućnosti.

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