

Qualitative pomological traits of the sweet chestnut (*Castanea sativa* Mill.) in the area of Bosanska Krajina

Bosanska Krajina bölgesindeki kestane ağaçlarının (*Castanea sativa* Mill.) kalitatif pomolojik özellikleri

Aida Tuğ¹ , Mirzeta Memišević Hodžić² , Dalibor Ballian² , Amra Kazić³ 

¹Ege University, Institute of Natural and Applied Science, İzmir, Turkey

²University of Sarajevo, Faculty of Forestry, Sarajevo, Bosnia and Herzegovina

³University of Sarajevo, Institute for Genetic Engineering and Biotechnology, Sarajevo, Bosnia and Herzegovina

ABSTRACT

In Bosnia and Herzegovina, sweet chestnut (*Castanea sativa* Mill.) stands are spread over an approximately 7,000-ha belt, ranging from 150 to 700 m above sea level. The largest population of sweet chestnuts in Bosnia and Herzegovina can be found in the northwest of the country in the Bosanska Krajina area. This study aimed to determine the quality of the 16 subpopulations selected from a single large sweet chestnut population in Bosanska Krajina based on the morphological traits of fruits to determine the future selection and breeding of this promising species. We studied qualitative traits of fruits from 3 locations and 16 subpopulations of a sweet chestnut population in Bosanska Krajina. The observed traits were fruit shape, fruit embryony, and penetration of seed coat into embryo. The results showed that the most common shape of fruit in all subpopulations (except B9) was cylindrical, with a total of 41% of all tested fruits sharing that shape. Out of all subpopulations, 99.4% of the tested fruits were monoembryonic. The penetration of seed coat into embryo was absent in 74.4% of the tested fruits. This type of research of variability within a population can act as a starting point for further research on the quality of sweet chestnut fruits in Bosnia and Herzegovina. The results of such studies can serve as a good basis for selecting high-quality stands, breeding, and preserving genetic diversity.

Keywords: Sweet chestnut, qualitative traits

ÖZ

Bosna Hersek'te kestane (*Castanea sativa* Mill.) meşcereleri, deniz seviyesinden 150 ila 700 m yükseklikte yaklaşık 7000 hektarlık bir kuşak üzerinde bulunmaktadır. Bosna Hersek'teki en büyük tatlı kestane popülasyonu, ülkenin kuzeybatısında Bosanska Krajina bölgesindedir. Bu çalışma, Bosanska Krajina'daki tek bir büyük kestane meşceresinden seçilen 16 alt popülasyonun kalitesini, meyvelerin morfolojik özelliklerine dayanarak belirleyerek, türün gelecekteki seçimini ve yetiştirme koşullarının belirlenmesini amaçlamaktadır. Bosanska Krajina'daki kestane meşcerelerinin bulunduğu 3 lokasyonda ve 16 alt popülasyondaki meyvelerin kalitatif özellikleri incelenmiştir. İncelenen özellikler, meyve şekli, meyve embriyonu ve çenekteki tohum zarfı penetrasyonudur. Sonuçlar, tüm alt popülasyonlarda (B9 hariç) en yaygın meyve şeklinin silindirik olduğunu ve test edilen tüm meyvelerin toplam% 41'inin bu şekilde olduğunu göstermiştir. Tüm alt popülasyonların dışında, test edilen meyvelerin %99,4'ü monoembryoniktir. Test edilen meyvelerin %74,4'ünde çenekteki tohum zarfı penetrasyonu bulunmamaktadır. Bu çalışmada yapılan tür değişkenlik araştırması, Bosna Hersek'teki kestane meyvelerinin kalitesi hakkında daha fazla araştırma yapılması için bir başlangıç noktası olabileceği düşünülmektedir. Bu tür çalışmaların sonuçları, yüksek kaliteli meşcerelerin seçilmesi, yetiştirilmesi ve genetik çeşitliliğin korunması için iyi bir temel oluşturabilir.

Anahtar Kelimeler: Kestane, kalitatif özellikler

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Corresponding author:

Mirzeta Memišević Hodžić
e-mail:
mirzeta.memisevichodzic@gmail.com;
m.memisevic-hodzic@sfsa.unsa.ba

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INTRODUCTION

In Bosnia and Herzegovina, sweet chestnut stand covers approximately 7,000 ha of land at altitudes from 150 to 700 m. Most of these stands are located between 300 and 600 m above sea level (Wraber, 1958). The prevailing opinion about the origin of the sweet chestnut in Bosnia and Herzegovina is that chestnut forests in this area are part of a unique, indigenous area extending from Slovenia through Croatia, Bosnia and Herzegovina (Sučić, 1953; Wraber, 1958; Zelić, 1998). The largest and most important site of sweet chestnuts in Bosnia and Herzegovina is in the northwestern part of the country, Bosanska Krajina.

Studies on chestnuts in Bosnia and Herzegovina are very limited in their scope. Mujagić-Pašić and Ballian (2012, 2013a, 2013b), Ballian et al. (2012, 2013), and Daničić et al. (2018, 2019) studied morphological traits. Mujić et al. (2010), Skender (2010), and Al-ibabić (2018, 2019) studied pomological properties. Mujić et al. (2006) and Daničić et al. (2008) studied the chemical composition of the fruit. Treštić et al. (2009) studied diseases of sweet chestnut. Daničić (2019) and Daničić et al. (2019) studied the genetic diversity of Bosanska Krajina sweet chestnut populations. Fussi et al. (2016) investigated sweet chestnut populations in Central and Southeast Europe, including Bosnia and Herzegovina.

It is well known that the phenotypic and genotypic variability of the starting material is the first stepping point for breeding because only in highly variable material can we find desired properties-in this case, high-quality fruits. The morphological and phenological properties of sweet chestnuts are often used for studying variability (Neophytou et al., 2007). Therefore, the knowledge gained from

testing the variability of sweet chestnut fruit in the Bosanska Krajina can be a starting point for further research on the fruit quality in these species in natural stands, as well as forming a basis for selecting genetically suitable material for the conservation of natural chestnut populations in Bosnia and Herzegovina.

This study aims to determine the quality of sweet chestnut subpopulations based on selected morphological properties of fruits to inform future selection and breeding of this valuable tree species.

MATERIALS AND METHODS

Material for analysis was collected within a large population of sweet chestnuts in Bosanska Krajina; divided into 3 locations and 16 subpopulations (Figure 1, Table 1) under different ecological conditions.

Within the subpopulations, 146 chestnut trees were selected: solitary adults and individuals at the edges of forests. Fruits were collected in the autumn of 2009. Thirty fruits were collected from each selected tree by random selection.

The analyzed variables on the chestnut fruit were shape (Figure 2), embryo (Figure 3), and the penetration of seed coat into embryo (Figure 4). An international descriptor for the sweet chestnut was used for the assessment of these fruit characteristics (UPOV, 1989).



Figure 1. Researched subpopulations of sweet chestnut on the area of Bosanska Krajina

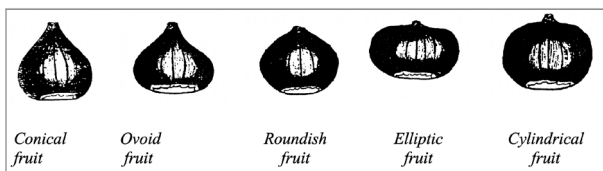


Figure 2. Fruit shape (UPOV, 1989)

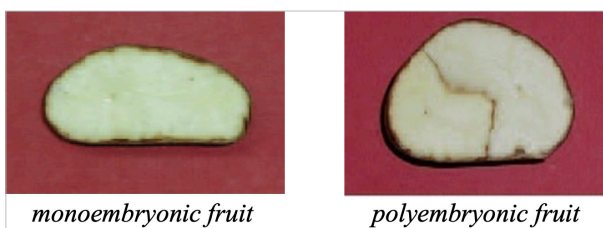


Figure 3. Fruit embryo (UPOV, 1989)

Table 1. List and labels of researched subpopulations

Location	Subpopulation label	North latitude	East longitude	Altitude (m)
Vrnograč (Velika Kladuša)	V1	45°10'11 "	15°56'07 "	268.80
	V2	45°09'56 "	15°58'13 "	262.10
	V3	45°09'54 "	15°58'12 "	252.10
	V4	45°10'06 "	15°58'13 "	258.60
	V5	45°10'02 "	15°58'19 "	285.20
Zaradostovo (Bužim)	B6	45°04'44 "	16°03'42 "	282.60
	B7	45°04'44 "	16°03'53 "	271.10
	B8	45°04'40 "	16°03'43 "	319.60
	B9	45°04'35 "	16°03'38 "	323.30
Gornja Koprivna (Cazin)	C10	44°58'47 "	15°57'35 "	503.00
	C11	44°58'44 "	15°57'40 "	490.10
	C12	44°58'49 "	15°57'39 "	484.10
	C13	44°58'59 "	15°57'45 "	498.80
	C14	44°59'08 "	15°57'46 "	486.40
	C15	44°58'52 "	15°57'41 "	498.60
	C16	44°58'47 "	15°57'35 "	492.00

Statistical Analysis

Data were statistically processed using IBM Statistical Package for the Social Sciences version 26.0 (IBM SPSS Corp.; Armonk, NY, USA). The authors tested the normality of data distribution and analyzed the variability of chestnut fruit within and among locations, and within and among subpopulations, through frequency analysis and nonparametric Kruskal–Wallis testing (ANOVA).

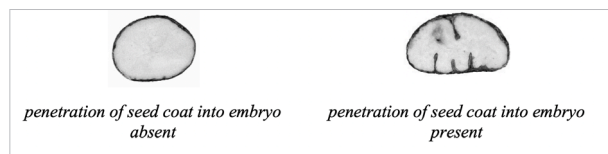


Figure 4. Penetration of seed coat into embryo (UPOV, 1989)

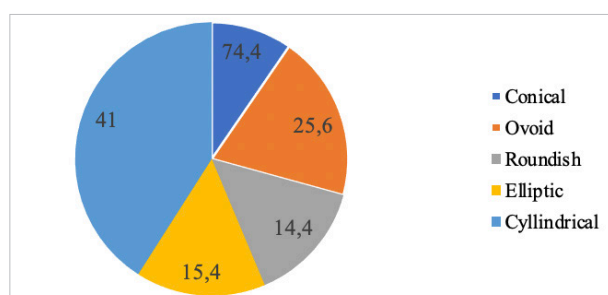


Figure 5. Frequencies of fruit shapes (in %) for all localities/subpopulations

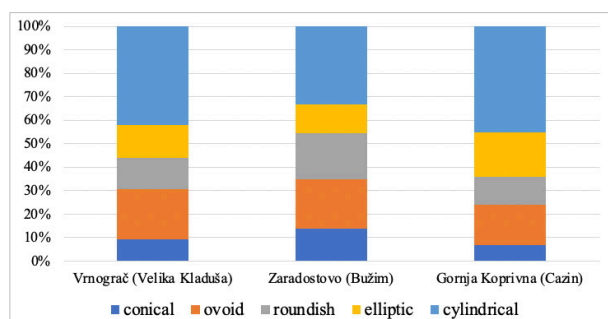


Figure 6. Frequencies of different fruit shapes (in %) per localities

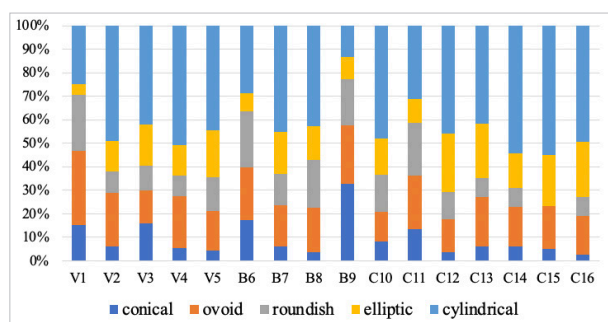


Figure 7. Frequencies of different fruit shapes (in %) per subpopulations

RESULTS AND DISCUSSION

Fruit Shape

The results of the fruit shape frequency analysis for all locations and subpopulations together are shown in Figure 5. The most common fruit shape was cylindrical, 41.0%, whereas the rarest was conical, 9.6%. The results of fruit shape frequencies per location are shown in Figure 6.

The results of variability among and within subpopulations are shown in Figure 7. Conical fruits were found to be the most common in subpopulation B9 (32.9%). All other subpopulations predominantly had cylindrical fruits.

The study findings correspond with those of Skender (2010), who studied fruit shape in three Bosnian populations (Pećigrad, Konjic, and Bratunac). Results showed the highest occurrence of cylindrical shaped fruits (47%), followed by round (36%), and elliptical (17%). Ovoid and conical shapes were not found.

Testing of the normality of the distribution showed non-normal distribution by location as well as by subpopulation. Consequently, nonparametric Kruskal–Wallis tests for fruit shape per location and subpopulation were performed. The test results showed statistically significant variability among subpopulations (Sig.<0.05) (Table 2).

Fruit Embryony

Fruit embryony was determined based on the descriptor scheme for chestnuts (UPOV, 1989, Characteristic 28), according to which fruits can be monoembryonic or polyembryonic. Figure 8 shows the frequencies of fruit embryony for all locations, i.e., all subpopulations combined.

Figure 9 shows the frequencies of fruit embryony by location, and Figure 10 by subpopulation.

Our results correspond to those of Skender (2010), where monoembryonic fruits also dominated in all three studied populations in Bosnia and Herzegovina, with rare cases of polyembryonic fruits.

Testing the normality of the distribution showed a non-normal distribution by location as well as by subpopulation. Consequently, nonparametric Kruskal–Wallis tests for fruit embryony per location and subpopulation were performed. The test results showed statistically significant variability among subpopulations (Sig. <0.05), but not locations (Sig. >0.05) at the level of 0.05 significance (Table 3).

Table 2. Kruskal–Wallis test for fruit shape by location and subpopulation

	Grouping variable: location	Grouping variable subpopulation
Kruskal–Wallis H	60.504	287.100
df	2	15
Asymp. Sig.	0.000	0.000

Penetration of seed coat into embryo

Degree of penetration of seed coat into embryo was determined according to the descriptor for chestnuts (UPOV, 1989, Characteristic 30). Figure 11 shows the absence of penetration

Table 3. Kruskal–Wallis test for fruit embryony

	Grouping variable: location	Grouping variable: subpopulation
Kruskal–Wallis H	2.502	33.343
df	2	15
Asymp. Sig.	0.286	0.004

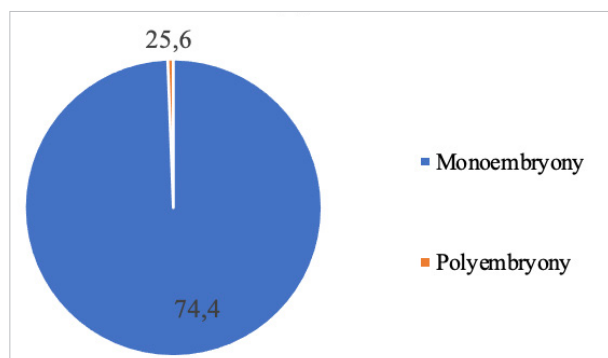


Figure 8. Frequency of fruit embryony (in %) for all researched subpopulations

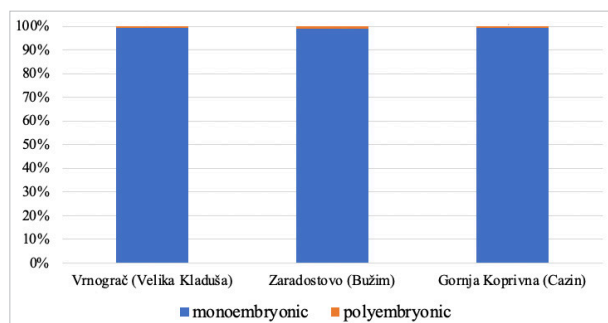


Figure 9. Frequencies of fruit embryony (in %) per localities

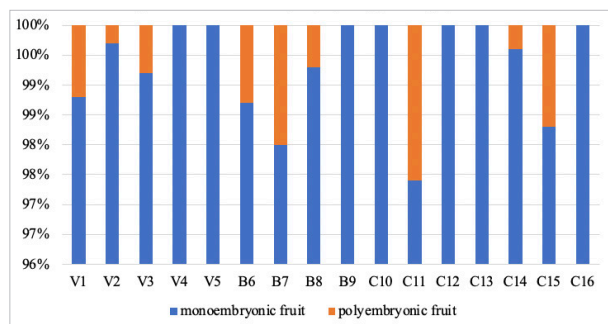


Figure 10. Frequencies of fruit embryony (in %) per subpopulations

of seed coat into embryo in most analyzed fruits from all locations and subpopulations. These results correspond to those obtained by Skender (2010).

Figure 12 shows the frequencies of degree of penetration of seed coat into embryo by location and Figure 13 by subpopulation.

By location, the highest frequency of penetration of seed coat into embryo was recorded at Vrnograč (Velika Kladuša). Regarding subpopulations, C14 in Gornja Koprivna, Cazin had the highest frequency of penetration of seed coat into embryo (Figure 13).

Testing of the normality of the distribution showed a non-normal distribution by location as well as by subpopulation. Consequently, nonparametric Kruskal–Wallis tests for penetration of seed coat into embryo per location and subpopulation were performed. The test results showed statistically significant variability among locations as well as among subpopulations (Sig. <0.05) at the level of 0.05 significance (Table 4).

In this study, the variability of sweet chestnut (*Castanea sativa* Mill.) in Bosanska Krajina was investigated based on three morphological characteristics of the fruit.

Several existing studies utilized different morphological analyses to assess the variability of sweet chestnut populations and confirmed that fruit, leaf, and cupule parameters may be

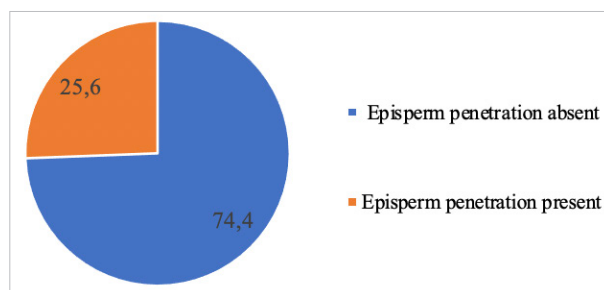


Figure 11. Frequency of penetration of seed coat into embryo (in %) for all localities/subpopulations

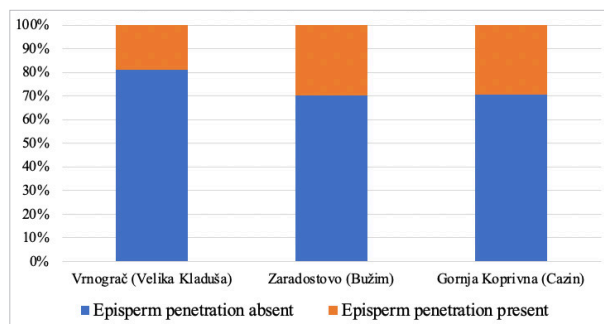


Figure 12. Frequencies of penetration of seed coat into embryo (in %) per localities

Table 4. Kruskal–Wallis for penetration of seed coat into embryo per localities and subpopulations

	Grouping variable: location	Grouping variable: subpopulation
Kruskal–Wallis H	5.243	173.801
df	2	15
Asymp. Sig.	0.000	0.000

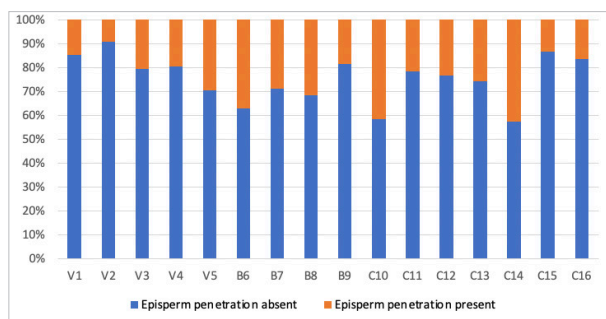


Figure 13. Frequencies of penetration of seed coat into embryo per subpopulations in percents

suitable variables to detect the level of phenotypic variability among natural populations.

Properties that were analyzed included leaves (Aravanopoulos et al., 2005; Serdar and Demirsoy, 2006; Serdar and Kurt, 2011; Zarafshar et al., 2010), fruit (Aravanopoulos et al., 2001; Bolvansky and Uzic, 2005; Ertan, 2007), and molecular structure (Koyuncu et al., 2008; Pandit et al., 2009; Ramos-Camber et al., 2005; Solar et al., 2005).

As stated by Poljak et al. (2016), the ideal characteristics of sweet chestnut include large, easily peeled fruit; a low percentage of penetration of seed coat into embryo; and monoembryony. The results of the present study showed that all researched subpopulations have a good fruit shape (mostly cylindrical) and a very low percentage of polyembryony and penetration of seed coat into embryo. These traits make them desirable for future selection and breeding.

Daničić et al. (2019) investigated the genetic diversity of Bosnian–Herzegovinian sweet chestnut populations using simple sequence repeat (SSR) markers. The results showed that the Bosnian–Herzegovinian sweet chestnut is a rich source of genetic diversity and is suitable for future cultivation. In the study of genetic diversity of sweet chestnut populations in the northwestern part of Bosnia and Herzegovina, Daničić (2019) found negative values of fixation index, which indicates good stability of the studied populations and the absence of inbreeding. Fussi et al. (2016) studied the genetic makeup of sweet chestnut populations from Central and Southeast Europe, which included populations from Bosnia and Herzegovina. All populations showed a high degree of genetic diversity. Given that all subpopulations in this study showed desirable morphological properties of the

fruit and the fact that they have a good genetic structure (Fussi et al., 2016), selection and breeding efforts should include trees that show other desirable traits as well.

The results of Kruskal–Wallis nonparametric tests showed statistically significant differences among subpopulations for all investigated traits. However, Kruskal–Wallis nonparametric tests among locations showed statistically significant differences in fruit shape and penetration of seed coat into embryo but not in fruit embryony.

This study can be a starting point for further research on sweet chestnuts in Bosnia and Herzegovina. The results of these studies can serve as a good basis for the selection of seed stands, breeding, and conservation of genetic diversity.

CONCLUSION

In this research, the qualitative traits of sweet chestnut fruit within natural populations in Bosanska Krajina were analyzed. Based on the conducted research, the following conclusions can be made:

A total of 41% of all tested fruits in all subpopulations had a cylindrical shape, which can be considered the most desirable shape given the accessibility of the edible part of the fruit.

Only subpopulation B9 from Zaradostovo Bužim predominantly had conically shaped fruits (32.9%).

Overall, 99.4% of the examined fruits in all subpopulations were monoembryonic, which is desirable for fruit quality.

In 74.4% of the examined fruits, episperm penetration of seed coat into embryo was absent, which is preferred for the quality of the fruit.

Results of the Kruskal–Wallis test showed statistically significant differences in fruit shape frequencies among locations as well as among subpopulations; statistically significant differences in fruit embryony frequencies among locations but not among subpopulations; and statistically significant differences in penetration of seed coat into embryo frequencies among locations as well as among subpopulations.

This form of research of variability can be the starting point for further research on the quality of sweet chestnuts in Bosnia and Herzegovina, and the results can serve as a good basis for the selection of seed stands, breeding, and conservation of genetic diversity. The obtained results should be confirmed at the molecular/genetic level.

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or Processing – A.T., M.M.H.; Analysis and/or Interpretation – A.T., M.M.H., D.B., A.K.; Literature Search – D.B.; Writing Manuscript – A.T., M.M.H., A.K.; Critical Review – D.B.

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