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NUT QUALITY ASSESSMENT OF CHESTNUT CULTIVARS FROM 'IVANIK' CLONE COLLECTION

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Abstract: Chemical composition of eight native and non-native cultivars from 'Ivanik' nursery plantation – Southwestern part of Bulgaria, was investigated. High variability in chemical composition among cultivars was found out, corresponding to the high genetic variability between cultivars. Cultivar I – 5/3, which is breaded from *Castanea mollissima*, produced nuts with lowest moisture content. The highest moisture content was found for *C. crenata x sativa* cultivars. Cultivar Zlatarevo had the highest total sugar content. Marigoule and Hemus showed the lowest values in crude protein, but highest were recorded for *C. crenata* cultivars. Lower in potassium and phosphorous content were mainly cultivars, breaded from *C. mollissima* and with higher content were those, breaded from *C. crenata*.

Key words: chestnuts, chemical composition, moisture, starch, total sugar, mineral content

INTRODUCTION

European chestnut populations are widely distributed all around the Mediterranean basin and are unique communities with highly significant social, economical and environmental importance. Species from genus *Castanea* are multipurpose ones because of various useful properties and wide range of valuable products which can be obtained. One of the most valuable products is the fruits which are of great importance in pastries and in human alimentation in several European countries. Chestnuts have a quite remarkable nutritional composition that sets them apart from all other nuts and makes them an outstanding food source. In contrast to other nut species, which are characterized with large amounts of fats and proteins, in chestnut nuts mainly starch, sugar and cellulose prevail. They are extremely useful and are an indispensable source of nutrients. Therefore the area of chestnut plantations for nut production increasing continuously, and for this purpose a large number of cultivars have been created (Pavari et al., 1956; Bergonoux et al., 1978; Gomez et al., 1993; Pereira-Lorenzo et al., 1996; Solar et al., 1998).

An increasing amount of evidence shows that the consumption of fruits, particularly nuts, has become more important in human nutrition, due to the protection provided by the antioxidant compounds (Blomhoff et al., 2006). Therefore, in recent years the consumers have been showing an

increasing interest in chestnut fruits because of their nutritional qualities and potential beneficial health effects (Senter et al., 1994; Fereira-Cardoso et al., 1999; Borges et al., 2008; Borges et al., 2006).

Nevertheless the importance and multiple use *Castanea sativa* Mill. is insufficiently studied and effectively used species in Bulgaria, which was referred to the category of so-called 'negligible' genetic resources. The existing studies are related to some morphological traits, with a view for cultivation of varieties with larger fruits (Velkov, 1982; 1983; 1986) or resistant to diseases and pests (Petrov, 1982, 1990; 1992, 2005). The last few studies are based on variability of quantitative morphological and adaptive traits (Glushkova, 2006). Researches on the nutritional composition of chestnut fruits are partial, not covering the whole distribution range of the species and are not recently updated (Petrov, Ploshtakova, 1968).

This study is **aimed** to determine moisture, starch, total sugar, crude protein and mineral content of eight important for the region of Southwestern Bulgaria chestnut cultivars. This work will also provide useful information for producers and breeders about nut quality of cultivars in order to select individuals for different purposes and to produce chestnut nuts with good nutritional characteristics.

MATERIALS AND METHODS

Plant material

From the plantation in 'Ivanik' nursery-garden were chosen eight of the most important for our country native and non-native chestnut cultivars: Trajanov, Zlatarevo, Hemus, Pyreneus, I – 2/5, II – 4/2, I -5/3 and Marigoule. The trees from which the fruits were collected were 30 years old. The orchard soil type was Chromic Luvisols, the climate is fairly mild with relatively warm winters and hot, dry summer periods. The average annual temperature varied between 12.5 to 14.0 ^{o}C (mean value 13.9 ^{o}C) and is the highest for the country. Annual rainfall is about 600-700 mm, but during the summer period the precipitations are very low, only 28 mm for August.

In the 2009 ripening season three representative trees from each cultivar were selected. One kilogram of chestnuts was collected from each of the selected trees.

The size of the nuts and basic characteristics of the cultivars are reported in Table 1.

Processing samples

Samples were stored unshelled at 2 ^{o}C until analyses were performed for a maximum of 3 to 4 days. About 15 to 20 fruits were randomly selected and the shells and pellicle were manually removed. The raw shelled samples were

Table 1
Basic characteristics of cultivars

Клон	Произход от	Тип плод	Тип	Fruit weight, g
Trajanov	*Castanea crenata*	depressa	longistaminea	12.87
Hemus	*Castanea sativa*	depressa	longistaminea	15.85
Marigoule	*C. crenata x sativa*	depressa	longistaminea	16.00
Pyreneus	*C. crenata x sativa*	typica	longistaminea	16.00
Zlatarevo	*Castanea crenata*	depressa	longistaminea	11.95
I – 2/5	*Castanea crenata*	depressa	longistaminea	12.42
II – 4/2	*Castanea crenata*	depressa	longistaminea	9.94
I – 5/3	*Castanea molissima*	depressa	longistaminea	10.10

broken into small pieces and was dried in an oven at 105 °C until constant weight was reached (for at least 24 h), to determine moisture content. Dried samples were ground with a coffee grinder (Siemens, Germany) for further use in various analyses.

Proximate analysis

The total nitrogen content was determined using Kjeldahl method (AOAC, 1997). The percentage of nitrogen was transformed into protein content by multiplying the total nitrogen content by conversion factor of 5.30, which is specific for chestnut fruits (McCarthy, Meredith, 1988). The quantitative analyses of total carbohydrates content, starch content and soluble sugar content were assessed according to the method of Pochinok (1976). The studied parameters were determined by spectrophotometric measurement at 620 nm (Spectrophotometer UV/VIS, Perkin Elmer, Lambda 2).

Mineral analysis

The content of macro- and microelements K, Ca, Mg, Cu, Fe, Mn, Na and Zn was determined by atomic absorption (AAS, Perkin Elmer 310 A) and P was estimated using a spectrophotometer (UV/VIS, Perkin Elmer, Lambda 2).

Statistical analysis

All analyses were performed in triplicate and each replicate was quantified in duplicate. Data were presented as means ± standard deviation. The coefficient of variation was expressed as relative standard deviation (%). An analysis of variance (ANOVA) was applied to estimate the effect of cultivar. F-values were used to determine the significance of the changes of variables. Mean separations were made using Tukey's test. A Pearson correlation analysis was also performed in order to reveal possible relationships between studied parameters.

RESULTS AND DISCUSSION

Proximate analysis

Chestnuts have very high moisture content, in most cases over 50%, which is essential for storage supply to the fresh market. High moisture presents also a mould problem during the storage and delivery. Results obtained for proximate composition of cultivars are shown in Table 2. The average moisture content was 42.95%, lower than the mean values reported by McCarthy, Meredith (1988) and Breisch (1995) for European chestnuts, the range of which is between 49 and 60%, but very close to the mean values, referred to Desmaison, Adrian (1986), who found out the moisture values for fresh chestnuts between 41 and 59% for different French and Italian cultivars.

Analysis of variance showed that cultivar had significant effect on average moisture content of nuts $P < 0.001$ (Table 2). Cultivar I – 5/3, which is breaded from *C. mollissima*, produced nuts with lowest moisture content – only 31.87%. This value was lower than American and Chinese chestnuts, with 44% moisture content (McCarthy, Meredith, 1988). The cultivars, breaded from *C. sativa* and *C. crenata* showed intermediate moisture values. The highest moisture content was found out for *C. crenata x sativa* cultivars, which were greater than 50%. The higher moisture content showed cv. Marigoule – 55.40%.

The coefficients of variation of moisture content were quite low and ranged from 4.24 to 9.18 % (not shown), but similar results were also found by Neri et al. (2010), who reported for coefficient of variation from 2.05 to 11.0%. Different results were found by De La Montaña Miguelez et al. (2004), who pointed out for very high coefficient of variation (up to 20%), even within the same cultivar. According to Bounous, Torello Marinoni (2005) this behaviour is due to the chestnut epicarp, which is porous and not lignified and causes chestnuts to dry out rapidly in comparison to other nuts.

Chestnut fruits generally contained high rates of carbohydrates. Total carbohydrate quantities found out in this study changed between 65.49 and 88.43 g/100 g d. m., depending on cultivar, with a mean value of 76.56 g/100 g d. m. (Table 2). These data are within the range reported in American chestnuts – 86.26 g/100g d. m. (Mc Carthy, Meredith, 1988), in Chinese chestnuts – 87.50 g/100 g d. m. (Mc Carthy, Meredith, 1988) and in European chestnuts – 71.68 g/100 g d. m. (Künsch et al., 1999; Bounous et al., 2000).

The predominant component of dry matter is starch. The analysis of variance showed significant effect of cultivar on starch content $P<0.001$. Starch quantities ranged from 36.09 to 56.32 g/100 g d. m. with regard to the cultivar (Table 2). Higher starch content showed cultivars I – 2/5,

Table 2
Proximate composition of chestnut cultivars

Cultivar	Moisture (%)	Total Carbohydrates g 100 g^{-1}d.m.	Starch g 100 g^{-1}d.m.	Soluble Sugar g 100 g^{-1}d.m.	Crude Protein g 100 g^{-1}d.m.
Trajanov	36.83±1.55 b	80.78±2.04 c	55.64±1.54 d	21.17±0.48 bc	4.42±0.10 cd
Hemus	41.48±1.49 c	79.69±1.78 c	54.98±1.03 cd	21.85±1.08 bc	3.68±0.42 b
Pyreneus	50.43±2.11 e	78.15±1.25 c	48.24±3.13 b	21.78±1.43 bc	3.76±0.76 b
I – 2/5	43.03±1.09 cd	80.84±1.98 c	56.32±1.08 d	20.74±1.65 bc	4.88±0.16 d
II – 4/2	40.48±1.26 bc	68.56±0.64 ab	36.09±0.47 a	24.21±1.32 c	4.85±0.32 d
Marigoule	55.40±1.17 f	70.57±2.17 b	49.97±1.47 bc	16.54±1.62 a	2.92±0.19 a
I – 5/3	31.87±1.28 a	65.49±2.03 a	40.85±2.95 a	20.08±1.74 ab	3.88±0.66 bc
Zlatarevo	44.15±1.87 d	88.43±1.45 d	55.43±2.06 d	30.02±1.24 d	4.52±0.16 d
F-value	54.842 ***	58.448 ***	46.698 ***	23.779 ***	13.602 ***

Note: Values are average of three individual samples per tree each analysed in triplicate ±SD.
For each columns different letters denote statistically significant differences ($P<0.05$) by ANOVA and Tukey's multiple range test.
Statistical significance *P<0,05, **P<0,01 and ***P<0,001

Trajanov and Zlatarevo, all of them breaded from *C. crenata*. The lowest quantities were found for II – 4/2 and I – 5/3, the first one, breaded from *C. crenata* and the second one – from *C. mollissima*. The values found by most researchers were slightly higher than these ones, generally ranging from 49.60 to 69.70 g/100 g d. m. in different species (Pinavaia et al., 1993; Fereira-Cardoso et al., 1993; Bounous et al., 2000; Ertürk et al., 2006). However, some researchers found the value much lower – 29.80 g/100 g d. m. (Üstun et al., 1999) or much higher – 80.00 g/100 g d. m. (Demiate et al., 2001) than these. Our findings are in accordance with those of Borges et al. (2008) for chestnut cultivars in Portugal and for most of the Spanish cultivars, studied by Pereira-Lorenzo et al. (2006). As we know the quantity of starch in chestnut fruits is very important for their use in food industry, and the higher starch content should be taken into account in selection of cultivars for flour production or for animal food.

The mean soluble sugar content changed between 16.54 and 30.02 g/100 g d. m. (Table 2). Cultivar Zlatarevo had higher content of soluble sugar – 30.02 g/100 g d. m., which is higher than the average values reported by most of the researchers. The data for the rest of the cultivars are within the range 14.01-20.60 g/100 g d. m., reported in some Italian chestnuts (Pinavaia et al., 1993; Bounous et al. 2000), and in Spanish chestnut varieties (Pereira-

Lorenzo et al., 2006). However these values are higher than the average value reported by De La Montaña Miguelez et al. (2004) for chestnuts from the region of Galicia. Lowest quantity of soluble sugar was observed in cv. Marigoule and cv. I – 5/3, which have to be taken into account in selecting cultivars for fresh market.

Crude protein varied significantly among cultivars ($P<0.001$). The quantities of crude protein ranged from 2.92 to 4.88 g/100 g d. m. (Table 2). Between the studied cultivars, Marigoule and Hemus showed the lowest values – 2.92 and 3.68 g/100 g d. m. respectively. The highest values were recorded for *C. crenata* cultivars – I – 2/5 and II – 4/2, greater than 4.85 g/100 g d. m. The average value of 4.12 g/100 g d. m. is much higher than reported by Borges et al. (2008), Bellini et al. (2005), and close to those observed in Chinese chestnuts – being between 2.12 and 7.49 g/100 g d. m. (McCarthy, Meredith, 1988). Pereira-Lorenzo et al. (2005) founded similar protein values, but did not find significant variations among 47 Spanish cultivars. Similar to these results were reported by Sacchetti, Pinavaia (2005) for some Italian ecotypes. Our data were in agreement with those, reported by Petrov, Ploshtakova (1968) for some selected chestnut trees in the region of Berkovitsa, Western part of Balkan Mt.

Mineral content

Regarding nutritional quality, chestnuts have a valuable mineral composition. They are a source of K, Mg, P, Fe, Ca, Mn and Cu, which play an important role in many physiological processes in human organisms. The contents of nine minerals were determined in the studied cultivars from 'Ivanik' nursery plantation (Table 3). All of the minerals varied significantly among cultivars, with exception of Zn. The main macrominerals are K, P, Ca and Mg, but potassium and phosphorous are among the most representative minerals in chestnut and are those, which characterized with greater variability within different cultivars. K content ranged between 790.33 in cv. I – 5/3 and 1034.67 g/100 g d. m. in cv. Zlatarevo, with mean value 920.21 g/100 g d.m. P varied significantly in the range from 135.00 in cv. Trajanov to 188.33 g/100 g d. m. in cv. Pyreneus. The mean value for P content was 155.58 g/100 g d. m. Lower in potassium and phosphorous content were mainly cultivars, breaded from *C. mollissima* and with higher content were those, breaded from *C. crenata*. Mg content ranged between 41.28 for cv. Pyreneus and 102.44 g/100 g d. m. for cv. Hemus, with 85.34 g/100 g d. m. on average. Calcium has lower concentration among all other macronutrients. Ca content changed between 40.15 in cv. Hemus and 86.22 g/100 g d. m. in cv. I – 5/3, with mean value – 66.06 g/100 g d. m. The average content for main macronutrients found out in our study were slightly higher, but comparable with those, reported by Fereira-Cardoso et al. (1999); Borges et al. (2008) for European chestnuts and by McCarthy, Meredith (1988) for

Table 3
Mineral content of chestnut cultivars

Клон	P g 100 g^{-1} d.m.	K g 100 g^{-1}d.m.	Ca g 100 g^{-1}d.m.	Mg g 100 g^{-1}d.m.	Fe g 100 g^{-1}d.m.	Na g 100 g^{-1}d.m.	Mn g 100 g^{-1}d.m.	Zn g 100 g^{-1}d.m.	Cu g 100 g^{-1}d.m.	Na:K ratio	Ca:P ratio
Trajanov	135,00±6,56 b	956,67±32,23 cd	41,14±3,95 a	88,84±9,63 c	2,04±0,35 ab	3,39±0,48 ab	3,67±0,72 bc	6,88±3,24 a	0,59±0,04 a	0,004 a	0,306 ab
Hemus	150,33±2,08 bc	849,33±48,35 ab	40,15±7,66 a	102,44±7,22 c	1,73±0,19 a	2,86±0,26 a	3,84±0,44 c	3,41±1,38 a	0,45±0,06 a	0,003 a	0,267 ab
Pyreneus	188,33±5,51 e	950,33±45,08 cd	83,54±8,22 b	41,28±5,98 a	2,70±0,26 ab	3,02±1,09 ab	3,37±0,04 b	6,34±1,17 a	2,42±0,49 c	0,003 a	0,443 ab
I – 2/5	163,00±7,55 cd	893,67±7,51 bc	86,08±27,49 b	94,76±14,31 c	2,08±0,30 ab	3,72±0,43 ab	4,55±0,11 d	2,08±0,43 a	1,65±0,05 b	0,004 ab	0,524 bc
II – 4/2	173,67±4,51 de	942,00±26,06 bcd	74,39±6,56 ab	96,23±11,28 c	3,34±1,26 b	4,29±0,69 ab	3,93±0,22 c	10,36±7,75 a	2,28±0,54 bc	0,005 ab	0,428 ab
Marigoule	180,33±7,09 e	872,67±18,56 abc	43,50±1,18 a	66,37±1,42 b	1,91±0,19 ab	2,91±0,07 a	2,38±0,06 a	3,41±0,31 a	0,64±0,08 a	0,003 a	0,241 a
I – 5/3	114,33±8,74 a	790,33±25,38 a	86,22±23,37 b	96,44±10,08 c	2,71±0,44 ab	4,49±0,49 b	5,17±0,07 e	5,07±1,09 a	2,37±0,17 c	0,006 b	0,759 c
Zlatarevo	139,67±6,11 b	1034,67±62,18 d	73,48±5,71 ab	96,36±6,52 c	2,81±0,79 ab	3,89±0,34 ab	3,93±0,08 c	5,89±1,64 a	1,61±0,20 b	0,004 a	0,526 bc
F-value	48,078 ***	12,172 ***	6,857 **	15,912 ***	2,889 *	3,776 *	56,930 ***	2,062	27,334 ***	5,168 **	8,472 ***

Note: Values are average of three individual samples per tree each analysed in triplicate ±SD.
For each columns different letters denote statistically significant differences (P<0.05) by ANOVA and Tukey's multiple range test.
Statistical significance *P<0.05, **P<0.01 and ***P<0.001

Chinese cultivars. These results were close to the results, obtained by Ertürk et al. (2006) for chestnuts in Turkey.

From the microelements the most important role has Manganese (Table 3), which ranged from 2.38 in cv. Marigoule to 5.17 g/100 g d. m. in cv. I – 5/3, with mean value 3.86 g/100 g d. m. Fe content varied between 1.73 in cv. Hemus and 3.36 g/100 g d. m. in cv. II – 4/2, with 2.42 g/100 g d. m. on average. Copper content changed in the range from 0.45 in cv. Hemus to 2.42 g/100 g d. m. in Pyreneus. The average value for Zn content was 5.43 g/100 g d. m. with variation from 2.08 in cv. I – 2/5 to 10.36 g/100 g d. m. in cv. II – 4/2. These data are slightly different from the values reported in other studies. The Zn content was higher than referenced in literature by McCarthy, Meredith (1988); Fereira-Cardoso (1999); Borges et al. (2008), but close to those reported by Ertürk et al. (2006). Fe and Mn values were in good agreement with the data from Fereira Cardoso et al. (1993); Künsch et al. (1999); Bounous (1999); Ertürk et al. (2006), but were lower than those obtained by Borges et al. (2008). The data about Cu content were comparable with these ones obtained by Ertürk et al. (2006) for Turkish chestnuts and by Borges et al. (2008) for chestnut cultivars in Portugal.

For good nutrition characteristics of fruits the ratios of the minerals Na:K and Ca:P are very important too. According to Stamler (1994) low Na:K ratio makes chestnuts preferable in many diets. In our study the mean values for Na:K ratio ranged between 0.003 to 0.006, which defined all the studied cultivars as good for diets. The mean Ca:P ratio was 0.437 and reveals a high concentration of P compared to Ca. According to Iqbal et al. (2006) this ratio should not be less than 1.0. Otherwise a mineral supplementation is needed in order to correct this imbalance. In our study the highest Ca:P ratio had the cultivars I – 5/3, Zlatarevo and I –2/5 (over 0.52). Nevertheless, these cultivars also needed a mineral supplementation, according to Iqbal et al. (2006).

Four of the studied eight cultivars showed coefficients of variation, higher than 20% (not shown). These were chestnuts from cultivar Pyreneus – for crude protein and I – 5/3 for Ca content. The higher coefficient of variation, greater than 30%, was observed for Ca content in cv. I – 2/5. The cultivars with coefficients of variation, lower than 12% are characterized with relative intravarietal homogeneity considering nutritional parameters.

The correlation matrix (Table 4) shows some relationships between studied parameters. High positive correlation was found for P content and moisture content ($r = 0.828$, $P<0.01$) and for starch content and carbohydrates ($r = 0.834$, $P<0.01$). K content was positively correlated with carbohydrates content ($r = 0.683$, $P<0.05$) and total sugar content ($r = 0.725$, $P<0.05$). The level of Mg correlate negatively with moisture content ($r = – 0.693$, $P<0.05$) and potassium content ($r = – 0.660$, $P<0.05$). According to Pereira-Lorenzo

Table 4
Correlations between chemical parameters of chestnut cultivars

	Mois-ture	Total Carbo-hydrates	Starch	Soluble Sugar	Crude Protein	P	K	Ca	Mg
Moisture	1	0.161	0.222	-0.195	-0.501	0.828**	0.265	-0.179	-0.693*
Carbohydrates			0.834**	0.585	0.313	-0.033	0.683*	-0.125	0.065
Starch				0.086	-0.023	-0.085	0.377	-0.390	0.044
Soluble Sugar					0.589	-0.208	0.725*	0.279	0.330
Crude Protein						-0.193	0.450	0.449	0.497
P							0.237	0.003	-0.660*
K								0.037	-0.154
Ca									-0.097
Mg									

Statistical significance: * P<0,05; ** P<0,01; *** P<0,001

et al. (2006) the correlation between mineral content and environmental peculiarities was low and not significant and means that differences observed among Spanish cultivars were due to genetic differences.

CONCLUSION

The variability found in chemical composition among studied cultivars may reflect genetic structure and environmental conditions. These results could be very useful in selecting cultivars for different purposes. For instance, cultivars, from *Castanea mollissima* are not suitable for fresh market conservation, because they produced nuts with lowest moisture content. Most suitable are *C. crenata x sativa* cultivars, with higher moisture content. It also recommended that cultivar Zlatarevo have to be involved in breeding programs for fresh market production, because of higher quantities of total sugar, much higher than in many Italian, Spanish and Turkish cultivars. Due to their high starch content for industry purposes are suitable cultivars, breaded from *Castanea crenata*. Regarding mineral composition, chestnuts are a good source of K, Mg, Fe, Cu and Mn. Higher in potassium and phosphorous content were cultivars, breaded from *C. crenata*.

The mean values for Na:K ratio found out in our study showed that all of the studied chestnut cultivars are characterized with good nutrition parameters, which defined them as good source for many diets. The mean Ca:P ratios reveal a high concentration of P compared to Ca and means

that chestnut trees in 'Ivanik' nursery plantation are needed a mineral supplementation.

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КАЧЕСТВО НА ПЛОДОВЕТЕ НА КЕСТЕНОВИ КУЛТИВАРИ ОТ КЛОНОВА КОЛЕКЦИЯ „ИВАНИК“

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(Резюме)

Изследван е химичният състав на плодовете на осем от най-често използваните при създаването на култури за плодопроизводство кестенови култивари в клоновата колекция в разсадник „Иваник“. Установена е висока вариабилност в химичния състав на култиварите, съответстваща на висока генетична вариабилност между отделните култивари. Култивар I – 5/3, който е селекциониран от *Castanea mollissima*, дава плодове с най-ниско водно съдържание, а най-високо съдържание на вода в плодовете е установено за култиварите *C. crenata x sativa*. Плодовете на култивар Zlatarevo имат най-високо съдържание на общи захари. При култиварите Marigoule и Hemus са установени най-ниски стойности за съдържанието на общ белтък, а най-високи са отчетени при култиварите, селекционирани от *C. crenata*. С най-ниско съдържание на калий и фосфор се характеризират култиварите, селекционирани от *C. mollissima*, а най-ниски стойности са отчетени при селекционираните от *C. crenata* култивари.

Ключови думи: кестени, химичен състав, водно съдържание, скорбяла, общи захари, минерален състав

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