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Biochemical and pomological assessment of European pear accessions from Bosnia and Herzegovina

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Abstract

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The research on 10 old and indigenous pear cultivars was conducted during 2012 and 2013 in Bosnia and Herzegovina. The following characteristics were determined: fruit weight, fruit length and width, stalk length and width, fruit flesh firmness; soluble solids content and total dry matter content of the fruit juice; pH, titratable acidity, vitamin C, total phenolics, total flavonoids and antioxidant activity of the fruit cell juice. On the basis of the Principal Component Analysis (PCA) of pomological fruit characteristics the studied cultivars were divided into four main groups. Based on the PCA of biochemical traits the studied cultivars can be divided into three groups. Extremely high phenolic content in the cvs Mioljnjača, Žutica, Poljakinja, Karamut and Gradišćanka recommends them for their inclusion in a breeding programme. The cvs Mioljnjača and Poljakinja are also characterised by large and firm fruit and since the antioxidant capacity affects the duration of fruit storage, it is expected that these traditionally grown varieties can survive longer and keep their valuable nutritional ingredients longer.

Keywords: fruits; antioxidant activity; Principal Component Analysis

Fruit quality is an important feature in choosing starting material in the selection and breeding process (Selamovska et al. 2013). A higher content of phenolics and flavonoids which are produced as a consequence of plant response to a higher exposure to environmental stress is expected from indigenous varieties that are mainly grown without any special plant protection. There is already much evidence that these secondary metabolites have a beneficial effect on health, mainly owing to their ability to scavenge the harmful free radicals, due to which phenols and flavonoids show an increased antioxidant and antimicrobial activity (Percival 1998; Jakopič et al. 2009; Rufino et al. 2011). They also play an important physiological and morphological role in the growth and reproduction of plants and contribute to colour and sensory characteristics of fruit. The therapeutic potential of pears is directly proportional to the content of total phenols, flavonoids and tannins (Velmuru-

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gan, Bhargava 2014). In addition to these, other fruit characteristics are important for the selection of cultivars as parents in breeding. Fruit characteristics such as pH, soluble solids content in the fruit juice (SSC) and firmness are important indicators of quality of the pear fruit (Costa et al. 2002) but also decisive factors from the consumer's point of view (Li et al. 2013). In comparison to apples or plums, pears are rarely the subject of research and generally it is less known about their composition and less literature is available for comparison of different varieties grown in different conditions. The research of autochthonous pear varieties grown in Bosnia and Herzegovina, conducted for evaluation purposes using pomological and eco-physiological characteristics (Đurić et al. 2009, 2014) or using molecular markers (Gaši et al. 2013), showed that cultivars which are traditionally grown in Bosnia and Herzegovina represent an interesting genetic material. This paper is the first study of biochemical characteristics of indigenous and traditional pear cultivars from Bosnia and Herzegovina.

MATERIAL AND METHODS

The research was conducted during 2012 and 2013 on 10 old and indigenous pear cultivars (Zobnjača, Žutica, Miljevićka, Lubeničarka, Rana kolačara, Gradiščanka, Žujićeva žuta, Poljakinja, Karamut and Mioljnjača) and the results are showed as an average values of these two years. These cultivars were collected in the area of the municipalities Gradiška and Novi Grad (north-western Bosnia and Herzegovina). Older trees on garden plots (trees in *in situ* conditions) grown without special pruning, irrigation, fertilising and protection measures were selected for the study. The basic pomological and biochemical characteristics of the fruits were analysed: fruit weight, fruit length and width, stalk length and width, fruit flesh firmness; soluble solids content and total dry matter content in the fruit flesh; pH, titratable acidity, vitamin C, total phenolics, total flavonoids and antioxidant activity of the cell juice of the fruit. Thirty fruits at full maturity stage were sampled from each variety, randomly selected from all parts of the crown. A homogenised paste of the fruit flesh which was stored at –20°C until the moment of analysis was prepared for biochemical analysis in three replications for each variety.

The fruit weight was determined by weighing on the digital scale, type Kern EMB 600-2 (Kern & Sohn GmbH, Balingen, Germany), with the weighing range of 0–600 ± 0.01 g. The fruit and stalk length and width were measured by the Unior digital calliper (Unior, Zrece, Slovenia), range 0–150 mm.

The fruit firmness was determined by a Penetrometer FT 327 (Fruit Pressure Tester; Facchini, Alfonsine, Italy) with a piston diameter of 8 mm and measuring scale for reading the force of penetration in kg/cm^2.

The soluble solids content in the squeezed and filtered juice of the fruit flesh (SSC) was determined by the digital refractometer Atago (Atago Co. Ltd, Tokyo, Japan), with a working range of 0–32°Brix. The content of total dry matter was determined by the standard AOAC (1990) method, with drying at a temperature of 105°C to constant weight.

The pH measurement was performed by immersing the standardised pH meter electrode (Hanna pH 211; Hanna Instruments, Cluj, Romania) in the juice obtained by homogenisation of the sample, at room temperature.

Titratable acidity (TA) was determined by a modified procedure described by Mbogo et al. (2010). The homogenised sample (37.5 g) was quantitatively transferred into a measuring flask of 250 ml by rinsing with 120 ml of distilled water. Acid extraction was carried out in a water bath (Memmert WNB 7-45; Memmert GmbH, Schwabach, Germany) with occasional shaking for 30 min at a temperature of 80°C. After cooling and bringing up to the mark with distilled water, it was filtered through Whatman # 1 filter paper (GE Healthcare UK Limited, Little Chalfont, UK). An aliquot of 50 ml of the filtrate was titrated with a solution of 0.1M NaOH to pH 8.1. The results are expressed in percentage of malic acid.

Total acidity (TA) was expressed as percentage and was calculated according to the formula:

$$TA = \frac{a \times k \times v}{V_1 \times o} \times 100 \quad (\%)$$

where:

a – content of NaOH (ml) used for the titration
v – stock solution (ml)
V_1 – of the filtrate taken for titration (ml)
o – amount of sample (g)
k – acid factor for the predominant acid (0.0067 for malic acid)

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Vitamin C was determined by the standard AOAC (1990) method, using 2,6-dichlorophenolindophenol.

The fruit extract for the determination of total phenolics and total flavonoids was prepared according to the method described by Tehrani et al. (2011). The total phenol content was determined by the Folin-Ciocalteu (FC) colorimetric method (Singleton, Rosi 1965). Fruit extract (1 ml) was added to 10 ml of distilled water and 0.5 ml of FC reagent. After 5 min, 2 ml of 7.5% solution of sodium carbonate was added. After being left to stand for 2 h, the absorbance was read at 765 nm with a spectrophotometer (UV-VIS, Shimadzu 1240 mini; Shimadzu Corporation, Kyoto, Japan). The calibration curve was prepared with gallic acid (concentrations ranged from 0.25 to 0.005 mg/ml) and the results were expressed as mg gallic acid equivalent (GAE)/100 g of fresh fruit. The content of total flavonoids was determined by the colorimetric method with aluminum chloride, described by Tehrani et al. (2011). The calibration curve was prepared with catechin (concentrations ranged from 10 to 100 mg/ml) and the results were expressed as mg catechin equivalents: mg CE/100 g sample.

The antioxidant activity of the sample was determined by quenching stable free 2,2-diphenyl-1-picrylhydrazyl (DPPH) radicals. The sample preparation to determine antioxidant activity was performed according to the modified method by Tehrani et al. (2011). Fruit tissue (5 g) was mixed with methanol (15 ml), homogenised in a polypropylene tube and allowed to stand for 1 h to ensure complete extraction. Centrifugation was performed at 2,000 g for 15 min at 20°C (Centric 322 A Tehtnica; Domel, Železniki, Slovenia). The supernatant was filtered through Whatman #1 filter paper, after which the filtrate was diluted to 25 ml with methanol. Five concentrations of the sample ranging from 8 mg/ml to 56 mg/ml were prepared from the stock solution obtained in this way. Determination of antioxidant activity by quenching DPPH radicals was performed by the modified method according to Liyana-Pathirana and Shahidi (2005). In 1 ml of each different concentration of five sample 1 ml of 0.135 mM methanolic solution of DPPH was added. The mixture was vortexed and allowed to stand for 30 min in the dark, after which the absorbance was determined at 517 nm. The control was prepared in such a way that it contained methanol instead of the sample. The antiradical activity (AA) was calculated from the equation:

$$AA = \frac{A_c - A_s}{A_c} \times 100 \quad (\%)$$

where:

A_c – absorbance of control

A_s – absorbance of the sample

Based on the diagram which shows antiradical activity in relation to the different concentrations of the sample the EC_{50} value was determined. The EC_{50} value represents the effective concentration at which 50% of DPPH radicals were quenched.

Biometric analysis. The characteristics of the examined indigenous pear varieties were analysed using the analysis of variance. The pomological characteristics were analysed separately from biochemical characteristics. For those characteristics where significant statistical differences were established between varieties, further testing and grouping is determined by the Tukey's test. Total effect of the combination of different measured characteristics on the examined indigenous pear cultivars was analysed using the principal component analysis (Sneath, Sokal 1973; Williams 1976; Iezzoni, Pritts 1991, Peres et al. 2003) which assisted in the classification of the examined varieties. Statistical analyses and graphical visualisations are made in R (R Core Team 2014).

RESULTS AND DISCUSSION

Pomological characteristics

A statistically significant difference was observed for all examined pomological characteristics (Table 1). The fruit weight, as the most prominent pomological characteristics of the studied indigenous cultivars exhibited a statistically significant difference (F = 32.58, P < 0,001) and according to the Tukey's test the cultivars were grouped into seven groups with transitions. The highest average fruit weight was recorded in cv. Mioljnjača (109.40 g) and the lowest in cv. Zobnjača (31.10 g). Max. fruit firmness was observed in relation to cv. Mioljnjača (6.30 kg/cm^2) and minimum was recorded in cv. Žujićeva Žuta (2.07 kg/cm^2). According to the fruit firmness, the examined indigenous cultivars were grouped into four groups with transitions.

$S_{\bar{x}}$

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Table 1. Mean values ($\bar{x}$) and standard errors ($\pm S_{\bar{x}}$) of the pomological and biochemical characteristics of the studied autochthonous pear cultivars, with *F*-values and significance scores

Cultivar	Weight (g)		Firmness (kg/cm^2)		SS (°Brix)		Fruit height (mm)		Fruit width (mm)		Stalk length (mm)		Stalk width (mm)	
	$\bar{x}$	$S_{\bar{x}}$	$\bar{x}$	$S_{\bar{x}}$	$\bar{x}$	$S_{\bar{x}}$	$\bar{x}$	$S_{\bar{x}}$	$\bar{x}$	$S_{\bar{x}}$	$\bar{x}$	$S_{\bar{x}}$	$\bar{x}$	$S_{\bar{x}}$
Gradišćanka	56.43^{cd}	3.61	5.10^{cd}	0.28	13.51^{a}	0.23	57.41^{b}	1.07	48.37^{c}	0.63	28.56^{bc}	1.74	3.89^{a}	0.25
Karamut	39.85^{ab}	2.82	3.72^{abc}	0.93	18.90^{de}	0.50	36.85^{a}	0.82	40.51^{ab}	1.36	22.36^{ab}	1.20	3.98^{ab}	0.14
Lubeničarka	63.10^{cd}	3.21	4.78^{cd}	0.23	14.59^{ab}	0.34	55.44^{b}	1.26	49.87^{cd}	0.59	33.99^{cd}	2.22	4.07^{ab}	0.25
Miljevićka	98.19^{fg}	7.91	4.28^{bc}	0.22	15.07^{ab}	0.48	67.01^{c}	2.96	54.00^{def}	1.75	46.75^{e}	2.01	5.07^{bc}	0.29
Mioljnjača	109.04^{g}	9.22	6.30^{d}	0.21	18.68^{de}	0.40	67.50^{c}	4.93	58.23^{f}	1.50	30.45^{cd}	1.91	4.15^{ab}	0.30
Poljakinja	84.99^{ef}	5.20	6.24^{d}	0.57	17.34^{cd}	0.47	57.53^{b}	0.82	55.20^{ef}	1.12	19.21^{a}	0.95	5.40^{c}	0.26
R. Kolačara	73.49^{de}	2.66	3.76^{abc}	0.36	13.80^{a}	0.59	50.59^{b}	0.91	53.06^{cde}	0.66	30.64^{cd}	0.96	5.45^{c}	0.25
Zobnjača	31.10^{a}	1.85	2.80^{ab}	0.32	19.72^{e}	0.57	51.36^{b}	1.09	38.24^{a}	0.93	33.16^{cd}	1.50	3.81^{a}	0.16
Ž. Žuta	46.75^{abc}	1.89	2.07^{a}	0.10	14.18^{a}	0.21	58.35^{bc}	1.26	43.12^{b}	0.66	36.43^{cd}	0.92	3.79^{a}	0.26
Žutica	36.05^{ab}	1.64	2.36^{a}	0.30	16.28^{bc}	0.35	41.31^{a}	0.93	42.05^{ab}	0.74	28.19^{bc}	1.75	3.71^{a}	0.28
F	32.58**		12.68**		28.68**		23.20**		42.02**		23.02**		7.72**	
	A-oxi. (mg/ml)		pH		Dry matter (%)		TA (%)		Vitamin C (mg/100g)		Phenols (mgGAE/100g)		Flavonoids (mgCE/100g)	
Gradisćanka	32.26	22.26	4.24^{ab}	0.06	17.17	0.06	0.18^{a}	0.01	1.61	0.47	693.85	257.24	84.64	21.71
Karamut	9.87	7.79	3.85^{a}	0.06	25.45	1.12	0.44^{b}	0.03	1.31	0.23	570.75	9.55	82.07	9.89
Lubeničarka	30.39	9.98	3.99^{ab}	0.04	18.64	0.33	0.28^{ab}	0.01	1.19	0.27	416.65	49.63	77.76	14.44
Miljevićka	43.16	4.60	4.37^{bc}	0.06	18.16	0.61	0.17^{a}	0.02	0.90	0.17	307.06	17.34	43.73	4.63
Mioljnjača	17.18	12.33	4.07^{ab}	0.02	22.31	1.15	0.34^{ab}	0.01	0.77	0.06	652.95	8.08	120.20	29.62
Poljakinja	24.90	3.32	3.97^{ab}	0.04	24.34	1.13	0.34^{ab}	0.01	1.37	0.28	717.08	157.32	53.31	0.24
R. Kolačara	35.26	15.49	4.19^{ab}	0.06	19.25	1.49	0.28^{ab}	0.03	1.35	0.33	463.80	78.96	85.45	21.24
Zobnjača	38.33	5.04	4.85^{v}	0.07	23.91	0.85	0.16^{a}	0.01	0.81	0.08	413.97	30.42	44.74	2.71
Ž. Žuta	43.72	9.62	4.34^{abc}	0.06	18.24	0.15	0.15^{a}	0.01	1.27	0.25	473.74	117.08	62.99	4.20
Žutica	23.74	3.47	4.08^{ab}	0.05	20.97	0.26	0.32^{ab}	0.02	0.99	0.11	715.80	17.54	91.04	14.11
F	1.01		10.01**		2.27		5.64**		0.23		0.40		0.47	

**indicates statistically significant difference at $P < 0.01$ (ANOVA); letters indicate significant differences and grouping of the cultivars according to Tukey's multiple range tests with 95% significance; SS – soluble solids content; A-oxi. – antioxidant activity; TA – total acidity

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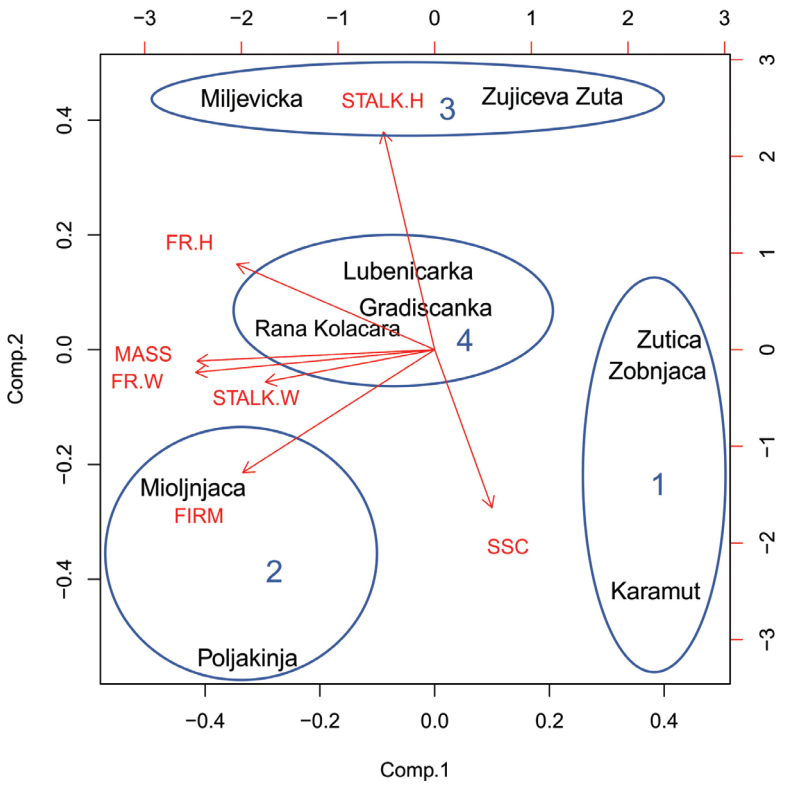

Fig. 1. Principal Component Analysis for the pomological characteristics of the studied autochthonous pear cultivars (black letters), and grouping of the cultivars in accordance with their morphological characteristics (red letters with eigen-vectors)

MASS – fruit weight; FIRM – fruit flesh firmness; SSC – soluble solids content; FR.H – fruit height; FR.W – fruit width; STALK.H – stalk height; STALK.W – stalk width

The highest content of SSC was noted in relation to cv. Karamut (18.90°Brix), while the smallest was recorded in cv. Gradišćanka (13.51°Brix). Four different groups of cultivars with transitions were observed according to the content of SSC. The highest average fruit height was recorded in cv. Mioljnjača (67.50 mm), while cv. Karamut was the smallest and all cultivars were grouped into three groups. The greatest fruit width was observed in cv. Mioljnjača (58.23 mm), while the smallest was noted in relation to cv. Zobnjača (38.34 mm), and the cultivars were grouped into five groups with transitions. Cv. Miljevićka had the highest stalk length (46.25 mm), while the shortest stalk was noted in cv. Poljakinja (19.21 mm). Based on this trait, the studied indigenous varieties were grouped into four groups with transitions. The highest stalk width was registered in cv. Rana Kolačara (5.45 mm), the smallest in cv. Žutica (3.71 mm) and three groups of cultivars with transitions were formed.

On the basis of the Principal Component Analysis on the pomological characteristics of the fruit (Fig. 1) the studied indigenous cultivars were grouped into four main groups. The first two groups are characterised by an increased amount of soluble solids content in the fruit. The key characteristics of the third and fourth group are small soluble solids content and long stalk. The first group is distinguished by small fruit size, especially low fruit weight and other dimensions of the fruit as well as small fruit firmness. This group is also characterised by an increased amount of soluble solids content. It includes cvs Žutica, Zobnjača and Karamut. The second group is also characterised by a higher amount of soluble solids content, but also a higher fruit firmness, larger dimensions and higher fruit weight. This group includes cvs Mioljnjača and Poljakinja. The third group is characterised by a greater stalk length and smaller soluble solids content. Cvs Miljevićka and Žujićeva Žuta belong to this group. The fourth group is characterised by a mix of characteristics. Compared to other varieties, these are pears of medium firmness, medium fruit size and soluble solids content. This group includes cvs Lubeničarka, Rana Kolačara and Gradišćanka.

Biochemical characteristics

The weakest antioxidant activity was exhibited by cv. Žujićeva žuta (43.72 mg/ml) and the strongest antioxidant activity was noted in cv. Karamut (8.87 mg/ml). The highest pH value was noted in Zobnjača (4.85), and the lowest in relation to Kara-

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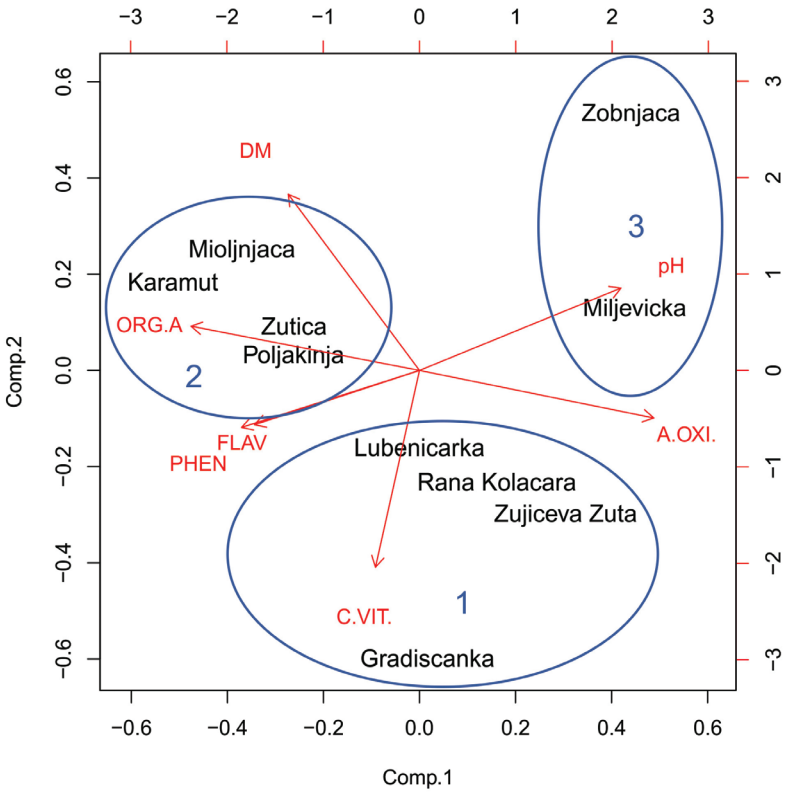

Fig. 2. Principal Component Analysis for the biochemical characteristics of the studied autochthonous pear cultivars (black letters), and grouping of the cultivars in accordance with their biochemical characteristics (red letters with eigen-vectors)

A.OXI – antioxidant activity; pH – acidity; DM – dry matter; ORG.A. – organic acid; C.VIT. – vitamin C; FEN. – phenols; FLAV. – flavonoids

mut (3.85), while cultivars were grouped into three groups (Table 1). The highest amount of total dry matter was displayed in cv. Karamut (25.45%) and the lowest in cv. Gradišćanka (17.17%). Cv. Karamut had the highest content of organic acids (0.44%), while cv. Žujićeva Žuta had the lowest (0.15%). The studied cultivars were grouped into two groups. In terms of the content of vitamin C, cv. Gradišćanka had the highest amount (1.61 mg/100 g) and the smallest amount of vitamin C was recorded in cv. Mioljnjača (0.77 mg/100 g). Considering the content of phenols, the highest amount was measured in the cv. Poljakinja (717.08 mg GAE/100 g of fresh fruit) while the smallest amount of phenols was observed in cv. Miljevićka (307.06 mg GAE/100 g of fresh fruit). Of all studied indigenous varieties, cv. Mioljnjača had the highest flavonoid content (120.20 mg CE/100 g of fresh fruit) and the smallest flavonoid content was observed in cv. Miljevićka (43.73 mg CE/100 g of fresh fruit).

On the basis of the Principal Component Analysis (PCA) in relation to the biochemical traits (Fig. 2) a more clear grouping and separation of certain studied varieties can be seen in accordance with the action of the examined factors. The studied cultivars can be divided into three groups. The first group are cultivars with an increased content of vitamin C. These are cvs Gradišćanka, Lubeničarka, Rana Kolačara and Žujićeva Žuta. The second group is distinguished by an increased amount of dry matter, flavonoids, phenols and organic acids as well as increased antioxidant activity. These are the cvs Mioljnjača, Karamut, Poljakinja and Žutica. The third group is characterised by an increased acidity and decreased antioxidant activity and decreased amount of vitamin C, phenols and flavonoids. The third group includes the cvs Zobnjača and Miljevićka.

The content of secondary metabolites depends on the degree of ripeness, climatic factors and other environmental impacts, species, variety and applied technological measures (Milošević et al. 2012). The available literature data originating from a pear growing region in Turkey (Ozturk et al. 2009) show that the amount of titratable acidity, the content of ascorbic acid and the pH are 0.21 to 0.56%; 0.96–4.90 g/l and 3.84–4.52, respectively, which corresponds to our results (0.15–0.44%; 0.77–1.61 mg/100 g and 3.85–4.85, respectively). Variations are probably the result of different climatic conditions but also cultivars. The difference among cultivars in the content of phenols and flavonoids is significant, which, as expected, has resulted in a significant difference in the antioxidant

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activity. Cvs Poljakinja, Gradišćanka and Mioljnjača have a very high content of phenolic compounds (717.08, 693.85 and 652.95 mg GAE/100 g of fresh fruit, respectively), particularly in comparison with literature data for pear varieties from Turkey, Chile and Pakistan (Galvic-Sanches et al. 2003; Ozturk et al. 2009; Manzoor et al. 2013), where the total phenol content does not exceed 500 mg GAE/1 kg, 2,000 mg/kg, 620 mg GAE/100 g, respectively. This certainly supports the fact that indigenous varieties produce a greater amount of compounds that protect them against free radicals and pests by adapting to environmental conditions and growing without technological protection measures (Veberič et al. 2005). When we add the fact that the pears generally have lower antioxidant activity when compared to other fruits (Prior, Cao 2000; Karadeniz et al. 2005), the high phenolic content in some cultivars is even more significant. The cv. Mioljnjača has the highest content of flavonoids (120 mg CE/100 g of fresh fruit), which, along with its very high phenolic content, provides excellent antioxidant activity (expressed as EC, i.e. effective concentration at which 50% of the radicals are quenched) in 17.18 mg of fresh fruit/ml.

However, compared with Pakistani pears (Manzoor et al. 2013) which have over 200 mg CE/100 g in the pulp and over 500.00 mg CE/100 g in the skin, this is not such a high content of flavonoids, which means that the antioxidant capacity of these varieties rests largely on the high content of phenols. Besides cv. Mioljnjača, cultivars that are best in terms of the content of total phenols and flavonoids and antioxidant activity are cvs Žutica, Poljakinja, Karamut and Gradišćanka. Although ascorbic acid also has antioxidant activity because it protects membranes from lipid peroxidation (Shewfelt, Del Rosario 2000), given the very low concentration in all studied varieties, it is assumed that there is no significant impact on the total antioxidant activity, which is in line with the conclusions of the study on Chilean pear varieties (Galvic-Sanches et al. 2003). In comparison with an overview of pear cultivars from Turkey, where the soluble solids content ranged from 7–15°Brix (Bostan 2009), it can be said that the content of SSC in cultivars from B&H is significant (13.51–18.90°Brix), especially because this is a very important characteristics of fruit quality for consumers. The content of total soluble solids in pear cultivars from Macedonia varied in a similar range between 12.8 and 18.3, neglecting slight variations that result from different varieties and climatic conditions (Selamovska et al. 2014) and 14–18.9°Brix (Selamovska et al. 2013). Karamut is the cultivar in which the highest content of SSC was registered and that is an important characteristics of this cultivar, which was previously considered as the most valued cultivar in Bosnia and Herzegovina. On the other hand, cv. Karamut had the highest content of total organic acids (0.44%), which is certainly interesting, while by reviewing data for the assortment of Macedonia, values of this parameter did not exceed 0.39%.

The results of this study allow for the assumption of diversity of indigenous pear cultivars in Bosnia and Herzegovina. This diversity is reflected primarily in differences between the studied indigenous varieties which showed different results in the measured pomological and biochemical characteristics. However, besides diversity, grouping of varieties by individual characteristics and groups of characteristics was performed. Such grouping is the result of similarities in certain characteristics and groups of characteristics that are most likely the result of similar selection pressure on individual traits with the aim to obtain the best possible pear fruits in conditions in which they are predominantly grown. This situation is ideal for further work on pear breeding in order to create new varieties with improved pomological and biochemical characteristics.

Thus, in order to work on increasing fruit size, the cv. Mioljnjača certainly needs to be considered, which, in addition to superior fruit size, also showed a high firmness as well as a very high content of dry matter. In addition cv. Mioljnjača has a high content of phenols and flavonoids, excellent antioxidant activity, but lower acidity and low vitamin C content. Considering the agronomic side of fruit traits and biochemical characteristics, cv. Mioljnjača proved to be superior to other studied indigenous pear cultivars. Concerning the content of vitamin C, cv. Gradišćanka showed the best results, with medium fruit size in comparison to the other studied indigenous varieties. Similar conclusions can be drawn for groups of cultivars in relation to specific groups of characteristics. If the breeding interest is focused on fruits of larger size and higher firmness, then the cvs Mioljnjača and Poljakinja will be selected. These cultivars also belong to the same group according to the analysis of biochemical characteristics, i.e. increased amount

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of dry matter, phenols, flavonoids, higher antioxidant capacity and organic acids.

Besides, all the above mentioned varieties are grown in specific conditions with the specific aim of growing adjusted to traditional fruit production and unfavourable climatic conditions. For example, the cultivar Karamut has adapted and survived primarily due to the production of traditional high-quality brandy of special characteristics which is named after the pear cultivar from which it is made (Đurić et al. 2009). Indigenous cultivars, as they are unchanged, represent an important resource for production, particularly in regions where climatic conditions do not allow practicing intensive fruit production. This has special significance in light of the current climate changes, where old indigenous varieties with better adaptability and natural resistance to harsh and changing climatic conditions may have a more dominant role in specific growing conditions.

In this respect, it must be borne in mind that the analysed varieties were grown in absolutely natural conditions, without any cultural measures. Values of some of the studied traits were lower and some were higher than in standard varieties (Sanches et al. 2003; Kevers et al. 2011). Although the studied varieties showed lower levels of vitamin C compared to standard varieties (Sanches et al. 2003; Kevers et al. 2011) or even other grown fruits (Venkatachalam et al. 2014), this can be explained, to a certain degree, by large difference in the content of vitamin C in the skin and flesh of the fruit (Sanches et al. 2003). On the other hand phenol content was higher than in standard varieties. Growing results would be significantly better if agrotechnical measures are improved. Therefore, it is necessary to continue the research of each cultivar individually in order to apply the appropriate agrotechnical measures for achieving optimal results.

Exposure to stress due to a lack of protection from pests and diseases in indigenous varieties of pears may result in a high content of secondary metabolites; their fruits can thus play an important role in reducing the risk of diseases caused by the action of free radicals. Extremely high phenolic content in the cvs Mioljnjača, Žutica, Poljakinja, Karamut and Gradišćanka is definitely a recommendation for their inclusion in the crossing programme. Cultivars Mioljnjača and Poljakinja are also characterised by large and firm fruit and since antioxidant capacity affects the duration of fruit storage, it is expected that these traditionally grown varieties can survive longer and keep longer their valuable nutritional ingredients. Accordingly, there is a need to further evaluate indigenous pear varieties selection of high-quality cultivars in relation to the examined characteristics, as well as their dissemination and conservation.

References

AOAC (1990): Official Methods of Analysis of the Association of Official Analytical Chemists. 15th Ed. Arlington.

Costa G., Noferini M., Andreotti C. (2002): Non-destructive determination of internal quality in intact pears by near infrared spectroscopy. Acta Horticulturae (ISHS), 596: 821–825.

Boston Z.S. (2009): Pomological traits of local apple and pear cultivars and types grown in trabzon province (Eastern Black Sea Region of Turkey). Acta Horticulturae (ISHS), 825: 293–298.

R Core Team (2014): A language and environment for statistical computing. Foundation for Statistical Computing. Vienna, Austria.

Đurić G., Tomić L., Mićić N., Cvetković M., Radoš Lj., Pašalić B. (2009): Fruit genetic resources in Republika Srpska. Acta Agriculturae Serbica XIV, 28: 31–40.

Đurić G., Mićić N., Salkić B. (2014): Evaluation of pear (*Pyrus communis* L.) germplasm collected in Bosnia and Herzegovina using some pomological and ecophysiological characteristics. Acta Horticulturae (ISHS), 1032: 105–115.

Galvic-Sanches A.C., Gil-Izquierdo A., Gil M.I. (2003): Comparative study of six pear cultivars in terms of their phenolic and vitamin C contents and antioxidant capacity. Journal of the Science of Food and Agriculture, 83: 995–1003.

Gaši F., Kurtović M., Kalamujić B., Pojskić N., Grahić J., Kaiser C., Meland M. (2013): Assessment of European pear (*Pyrus communis* L.) genetic resources in Bosnia and Herzegovina using microsatellite markers. Scientia Horticulturae, 157: 74–83.

Iezzoni A.F., Pritts M.P. (1991): Applications of principal component analysis to horticultural research. HortScience, 26: 334–338.

Jakopič J., Štampar F., Veberič R. (2009): The influence of exposure to light on the phenolic content of "Fuji" apple. Scientia Horticulturae, 123: 234–239.

Karadeniz F., Burdurlu H.S., Koca N., Soyer Y. (2005): Antioxidant activity of selected fruits and vegetables grown in Turkey. Turkish Journal of Agriculture & Forestry, 29: 297–303.

Kevers C., Pincemail J., Tabart J., Defraigne J.O., Dommes J. (2011): Influence of cultivar, harvest time, storage conditions and peeling on the antioxidant capacity and phenolic

doi: 10.17221/53/2015-HORTSCI

and ascorbic acid contents of apples and pears. Journal of Agricultural and Food Chemistry, 59: 6165–6171.

Li J., Huang W., Zhao C., Zhang B. (2013): A comparative study for the quantitative determination of soluble solids content, pH and firmness of pears by Vis/NIR spectroscopy. Journal of Food Engineering, 116: 324–332.

Liyana-Pathirana C.M., Shahidi F. (2005): Antioxidant activity of commercial soft and hard wheat (*Triticum aestivum* L.) as affected by gastric pH conditions. Journal of Agricultural and Food Chemistry, 53: 2433–2440.

Manzoor M., Anwar F., Bhatti I.A., Amer J. (2013): Variation of phenolics and antioxidant activity between peel and pulp parts of pear (*Pyrus communis* L.) fruit. Pakistan Journal of Botany, 45: 1521–1525.

Mbogo G.P., Mubofu E.B., Othoman C.C. (2010): Post harvest changes in physico-chemical properties and levels of some inorganic elements in off vine ripened orange (*Citrus sinensis*) fruits cv (Navel and Valencia) of Tanzania. African Journal of Biotechnology, 9: 1809–1815.

Milošević T., Milošević N., Glišić I., Mladenović J. (2012): Fruit quality attributes of blackberry grown under limited environmental conditions. Plant, Soil and Environment, 58: 322–327.

Ozturk I., Ercisli S., Kalkan F., Bunyamin D. (2009): Some chemical and physico-mechanical properties of pear cultivars. African Journal of Biotechnology, 8: 687–693.

Percival I.C. (1998): Quantum transfer functions, weak nonlocality and relativity. Physics Letters A, 244: 495–501.

Peres L., Jackson D., Somers K. (2003): Giving meaningful interpretation to ordination axes: Assessing loading significance in principal component analysis. Ecology, 84: 2347–2363.

Prior R.L., Cao G. (2000): Antioxidant phytochemicals in fruits and vegetables: diet and health implications. HortScience, 35: 588–592.

Rufino M.S.M., Alves R.E., Fernandes F.A.N., Brito E.S. (2011): Free radical scavenging behaviour of ten exotic tropical fruits extracts. Food Research International, 44: 2072–20175.

Sanchez A.G., Gil-Izquierdo A., Gil M. (2003): Comparative study of six pear cultivars in terms of their phenolic and vitamin C contents and antioxidant capacity. Journal of the Science of Food and Agriculture, 83: 995–1003.

Selamovska A., Milevska M.E., Najdenovska O. (2013): Genetic resources of traditional pear varieties in the region of Skopje. Section of Natural, Mathematical and Biotechnical Sciences, 34: 93–100.

Selamovska A., Milevska M.E., Najdenovska O., Dimovska D. (2014): Traditional pear varieties in the west region of Republic of Macedonia. Acta Agriculturae Serbica, 19: 47–60.

Shewfelt R.L., Del Rosario B.A. (2000): The role of lipid peroxidation in storage disorders of fresh fruits and vegetables. HortScience, 35: 575–579.

Singleton V.L., Rosi J.A. (1965): Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. American Journal of Enology and Viticulture, 16: 144–158.

Sneath P.H.A., Sokal R.R. (1973): Numerical taxonomy. The principles and practice of numerical classification. San Francisco, W.H. Freeman & Co.

Tehrani M., Chandran S., Sharif Hossain A.B.M., Nasrulhaq-Boyce A. (2011): Postharvest physico-chemical and mechanical changes in jumbu air (*Syzygium aqueum* Alston) fruits. Australian Journal of Crop Science, 5: 32–38.

Veberič R., Trobec M., Herbinger K., Hofer M., Grill D., Štampar F. (2005): Phenolic compounds in some apple (*Malus domestica* Borkh) cultivars of organic and integrated production. Journal of the Science of Food and Agriculture, 85: 1687–1694.

Velmurugan C., Bhargava A. (2014): Total phenolic, flavonoids and tannin content of various extracts from *Pyrus communis* fruit. International Journal of Pharmacy and Analytical Research, 3: 384–390.

Venkatachalam K., Rangasamy R., Krishnan V. (2014): Total antioxidant activity and radical scavenging capacity of selected fruits and vegetables from South India. International Food Research Journal, 21: 1039–1043.

Williams W.T. (1976): Pattern analysis in Agricultural Science. New York, Elsevier Scientific.

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