

POLYPHENOLS AND PHENOLIC ACIDS IN SWEET POTATO (*IPOMOEA BATATAS* L.) ROOTS

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ABSTRACT

Sweet potato (*Ipomoea batatas* L.) is one of the most important food crops in the world. They are rich in polyphenols, proteins, vitamins, minerals and some functional microcomponents. Polyphenols are bioactive compounds, which can protect the human body from the oxidative stress which may cause many diseases including cancer, aging and cardiovascular problems. The polyphenol content is two to three times higher than in some common vegetables. Total polyphenols (determined spectrophotometrically) and phenolic acids (i.e. caffeic acid, chlorogenic acid and isomers – using high performance liquid chromatography) contents were determined in three varieties of sweet potatoes (O'Henry – white, Beauregard-orange and 414-purple). Phenolic compounds contents were determined in raw peeled roots, jackets of raw roots and water steamed sweet potato roots. For all analysis lyophilised samples were used. Total polyphenol content ranged from 1161 (O'Henry, flesh-raw) to 13998 (414, peel-raw) mg.kg⁻¹ dry matter, caffeic acid content from the non-detected values (414, flesh-raw) to 320.7 (Beauregard, peel-raw) mg.kg⁻¹ dry matter and 3-caffeoylquinic acid content from 57.57 (O'Henry, flesh-raw) to 2392 (414, peel-raw) mg.kg⁻¹ dry matter. Statistically significant differences ($p \leq 0.05$) existed between varieties, morphological parts of the root, or raw and heat-treated sweet potato in phenolic compounds contents.

Keywords: sweet potato; polyphenols; caffeic acid; chlorogenic acid

INTRODUCTION

The sweet potato, *Ipomoea batatas* L. (Lam.), is a dicotyledonous plant belonging to *Convolvulaceae* family. Originally it was domesticated at least 5000 years ago in tropical America (Woolfe, 1992). At present sweet potato is grown mainly in China, the other major producers are Sub-Saharan Africa, Indonesia, Asia and South America. It is classified as the seventh most important food crop after rice, wheat, potatoes, maize and cassava (Pandi et al., 2016). In 2014 sweet potato world production exceeded 100 million tonnes (Esatbeyoglu et al., 2017).

Sweet potato is a crop with easy adaptability to a wide range of agro-ecological conditions (e.g. high temperature, drought, low soil fertility). It is suitable and attractive crop for agriculture with limited resources (Anbuselvi et al., 2012; Laurie et al., 2013), which leads to its increased production (Maquia et al., 2013).

The main components of sweet potato are carbohydrates representing from 80 to 90% dry weight (Pandi et al., 2016). Starch share constitutes up to 65 – 70% of dry weight (Padmaja, 2009), (amylose content ranges from 200 to 330 g per kg solids). Glucose (6.0 – 72), fructose (3.0 – 66), sucrose (21 – 77) and maltose (11 – 43 g per kg solids) are included in it as single sugars (Waramboi et

al., 2011). Non-starch polysaccharides consist of cellulose, hemicellulose and pectins. The roots of sweet potatoes are rich in minerals (P, K, Ca, Mg, Fe, Zn, Na, Cu), vitamins (B₁, B₂, B₃, B₅, B₆, H, C, E), carotenoids and conversely, have low protein content. Sulfur amino acids (Met, Cys) and Lysine are limited, (Ishida et al., 2000; Maquia et al., 2013) and simultaneously, sweet potato is a source of polyphenols. It contains phenolic compounds such as caffeoylquinic acid (CQA) derivatives, a family of esters formed from certain cinnamic acids and quinic acid, including mono-CQA (chlorogenic acid derivatives): 3-CQA (Figure 1); 4-CQA (Figure 2) and 5-CQA (Figure 3) (Clifford et al., 2003), such as three dicaffeoylquinic acids: 3,5-di CQA, 3,4-diCQA and 4,5-diCQA (isochlorogenic acid A, B, and C, respectively, Figures 4, 5, 6) (Ishiguro et al., 2007; Taira et al., 2013).

Anthocyanins are another compounds with chemoprotective effects. Their presence is manifested by purple colouring of the flesh and jacket of the sweet potato. Cyanidin and peonidin are predominant aglycones. Authors of numerous studies refer to their antioxidant, anticarcinogenic, anti-hyperglycemic and chemoprotective properties (Esatbeyoglu et al., 2017; Nozue et al., 1998; Rumbaoa et al., 2009).

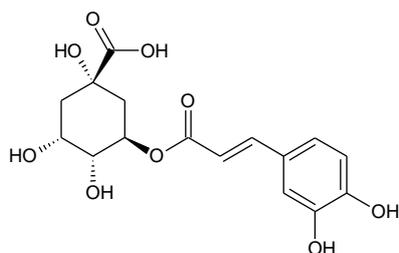


Figure 1 3-caffeoylquinic acid (3-CQA)

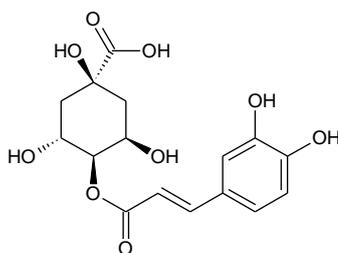


Figure 2 4-caffeoylquinic acid (4-CQA)

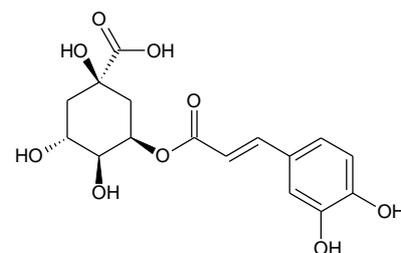


Figure 3 5-caffeoylquinic acid (5-CQA)

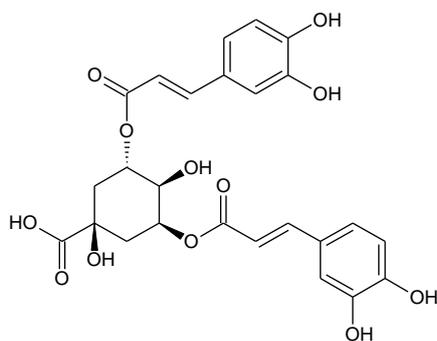


Figure 4 3,5-dicaffeoylquinic acid (3,5-diCQA)

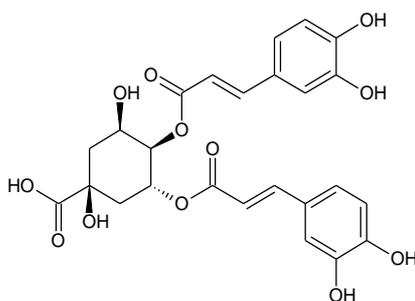


Figure 5 3,4-dicaffeoylquinic acid (3,4-diCQA)

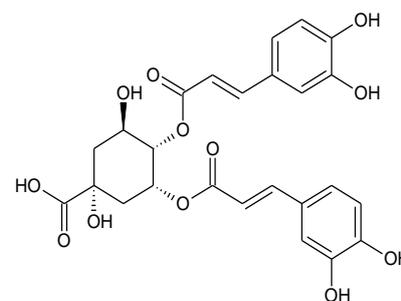


Figure 6 4,5-dicaffeoylquinic acid (4,5-diCQA)

Figures 1-6 Chemical structures of caffeoylquinic acid derivate from sweet potato (*Ipomoea batatas* L.).

Not only tubers but also other parts of sweet potatoes have nutritionally and functionally valuable components. Young leaves can also be used for consumption (Slosar et al., 2016). Many authors refer to the leaves of the sweet potato as important sources of polyphenols, chlorogenic acid and its derivatives. Sun et al. (2014) determined polyphenols in 40 varieties from China (TPC: $2.73 \pm 0.02 - 12.46 \pm 0.62 \text{ g} \cdot 100\text{g}^{-1} \text{ DM}$). Fu et al. (2016) determined TPC in different solvents (MeOH, EtOH, acetone, water) ranged from 23.3 ± 0.9 (in water) to 43.8 ± 0.7 (in 50% acetone) $\text{mg TPC} \cdot \text{g}^{-1} \text{ DM}$. Yoshimoto et al. (2002) determined contents of caffeic acid ($2 \text{ mg} \cdot 100\text{g}^{-1}$) and caffeoyl derivatives (chlorogenic acid: 31; 3,4-diCQA: 9; 3,5-diCQA: 91; 4,5-diCQA: 49; 3,4,5-triCQA: $4 \text{ mg} \cdot 100\text{g}^{-1}$) of lyophilized powder. Xu et al. (2010) determined TPC in the leaves of 116 varieties ranged from 8.943 to 27.333 $\text{mg CHA} \cdot \text{g}^{-1} \text{ DM}$.

The aim of this study was to determine and evaluate total polyphenol and phenolic acids contents (i.e. caffeic acid and its esters with quinic acid) depending on the variety, morphological part of root morphology (flesh vs. jacket), and thermal treatment.

MATERIAL AND METHODOLOGY

Three varieties of potatoes were used for the analyses as follows: O'Henry (white), Beauregard (orange) and 414 (purple), which were grown in the cadastral area of Šoporňa (N: 48.243421; E: 17.813596) in the Slovak Republic. About 2 kg of plant material was taken from two sampling sites for each variety.

The roots were peeled after washing and average samples were prepared for each variety from the jackets, or fleshes.

About 150 g of sweet potato were cut up, mixed and lyophilised and all jackets were mixed and lyophilised. About 30 g of the homogenized sample was used for the determination of dry matter.

Another portion of prepared average samples of flesh was used for the steam cooking as follows: about 200 g of sweet potatoes were cut into slices the thickness of which was 3 mm and cooked 20 minutes in steam at a temperature of $98 \pm 2 \text{ }^\circ\text{C}$. The samples were lyophilised and mixed after cooling.

Chemicals

Authentic standards of chlorogenic acid (purity $\geq 95.0\%$) and *trans*-caffeic acid (purity $\geq 95.0\%$), acetonitrile (gradient HPLC grade), phosphoric acid (ACS grade), 80% EtOH and Folin-Ciocalteu agents were purchased from Sigma-Aldrich (Sigma Aldrich Chemie GmbH, Steiheim, Germany); gallic acid (p.a.) was provided by Merck (Germany); double deionized water (ddH_2O) was treated ($0.054 \mu\text{S} \cdot \text{cm}^{-1}$) in a Simplicity 185 purification system (Millipore, UK).

Preparation of extracts

The lyophilized samples (1 g) were after homogenization in a mortar extracted with 20 mL of 80% EtOH at laboratory temperature for 8 h by horizontal shaker (Unimax 2010; Heidolph Instrument GmbH, Germany). Extract was filtered through Munktell No 390 paper (Munktell & Filtrac, Germany) and stored in closed 20 mL vial tubes. Prior to injection the standard solutions and extracts were filtered through syringe filter Q-Max (0.22 mm, 25 mm; Frisenette ApS, Knebel, Denmark).

Determination of total polyphenols content (TPC)

TPC was determined spectrophotometrically (Spectrophotometer UV-VIS 1601, Shimadzu) in ethanol extract using Folin-Ciocalteu agents. Measurement of absorbance (against blank) was at wavelength $\lambda = 765$ nm and total polyphenols content was expressed as mg gallic acid eqv. (Lachman et al., 2006).

Determination of phenolic acids

Chlorogenic acids (CA) and *trans*-caffeic acid were determined by Lukšič et al. (2016), using an Agilent 1260 Infinity high performance liquid chromatograph (Agilent Technologies, Waldbronn, Germany) with quaternary solvent manager coupled with degasser (G1311B), sample manager (G1329B), column manager (G1316A) and DAD detector (G1315C). All HPLC analyses were performed on a Purosphere reverse phase C18 column (4 mm x 250 mm x 5 mm) (Merck, KGaA, Darmstadt, Germany).

The detection wavelengths were conducted at 327 nm (chlorogenic acids) and 325 nm (*trans*-caffeic acid). The data were collected and processed using Agilent OpenLab ChemStation software for LC 3D Systems. Limit of detection for chlorogenic acids and *trans*-caffeic acid were 0.98 and 1.09 $\mu\text{g}\cdot\text{mL}^{-1}$, respectively. Limit of quantification for chlorogenic acids and *trans*-caffeic acid were 3.27 and 3.63 $\mu\text{g}\cdot\text{mL}^{-1}$, respectively.

Statistical analysis

Results were statistically evaluated by Analysis of Variance (ANOVA – Multiple Range Tests, Method: 95.0 percent LSD) using statistical software STATGRAPHICS (Centurion XVII, USA) and a regression and correlation analysis (Microsoft Excel) was used.

RESULTS AND DISCUSSION

Content of mineral and trace elements

Mineral and trace elements content was determined in the lyophilised samples of raw sweet potatoes (in the peeled roots and jackets) and in the peeled roots cooked in steam. The results shown in Table 1 are comparable to the values determined by Suárez et al. (2016) in 30 varieties of sweet potatoes from Tenerife Island and La Palma Island. The other two varieties from Japan show lower mineral contents in roots of sweet potatoes (Ishida et al., 2000) compared to our varieties. Waramboi et al. (2011) published comparable contents of K and Mg, lower contents of Ca and P, and higher content of Na in the variety Beauregard from Papua (New Guinea). The

differences in mineral contents in sweets potato may be due to their different content in the soil.

Total polyphenol content (TPC)

TPC was determined by the Folin-Ciocalteu spectrophotometrically. The content of polyphenols, which was the highest in the purple cultivar 414, was more than 8.4 times higher than in the O'Henry variety. The average levels of TPC ranged from 1161 to 9800 $\text{mg}\cdot\text{kg}^{-1}$ DM, which was similar to the findings reported by Rumbaoa et al. (2009) (192.7 – 1159.08 mg GAE.100 g^{-1} dry sample). The results shown by Padda, Picha (2008) indicate that sweet potato genotypes differ greatly in total phenolic content. The TPC in all of the 14 genotypes ranged from 1.4 to 4.7 $\text{mg}\cdot\text{g}^{-1}$ DW. Polyphenol content determined by Teow et al. (2007) was 9.646 in the white sweet potato, ranged from 440.8 to 742.9 in orange cultivars and from 1523.9 to 2955.2 mg CA equiv. kg^{-1} DM in purple cultivars. Shin et al. (2009) compared the orange and yellow varieties of sweet potatoes. Total phenolic compounds of freeze-dried samples of orange sweet potato (Tainong 66) were higher than those of yellow sweet potatoes (Tainong 57) (10.9 and 6.38 mg catechin equiv/g DM respectively).

There are statistically significant differences in TPC between varieties Niele, but also between flesh and peel in a single variety. The most significant difference is evident in the Beauregard variety: $\text{TPC}_{\text{peel}}/\text{TPC}_{\text{flesh}} = 4.27$. The differences in TPC are statistically significant in all varieties ($p \leq 0.05$), which corresponds with the results published by Steed and Truong (2008), showing that the TPC in the jackets of purple-fleshed sweet potatoes was more than 3.5 times higher than in their flesh.

Phenolic acids content

Trans-caffeic acid (CfA) and 3-CQA were identified by HPLC method (Figures 7 – 9). Other phenolic acids (5-CQA, 4-CQA and dicaffeoylquinic acids) are defined as a sum of the CQA isomers. CfA and 3-CQA were significantly higher ($p \leq 0.05$) in the jackets of all the three varieties of sweet potatoes. Purple variety 414 is an exception with the statistically significant difference between the content of 3-CQA in the jacket and flesh (CfA was not detected in the jackets of this variety) (Table 2). The content of total phenolic acids (CfA, 3-CQA, sum CQA-isomers) determined in raw sweet potatoes ranged from 169.5 (O'Henry – flesh) to 7952.5 (414 – flesh) $\text{mg}\cdot\text{kg}^{-1}$ DM. Esatbeyoglu et al. (2016) determined

Table 1 Content of mineral and trace elements in sweet potato ($\text{mg}\cdot\text{kg}^{-1}$ DM).

		K	Na	Ca	Mg	P	Cu	Zn	Mn
O'Henry	flesh (raw)	13353	598.2	3364	751.6	2159	9.051	9.503	7.605
	peel (raw)	21807	706.9	20561	1104	2572	13.35	13.61	29.80
	flesh steaming)	10241	534.7	3231	646.9	2173	8.904	9.401	8.151
Beauregard	flesh (raw)	15342	370.5	4241	554.5	1686	7.452	8.853	9.057
	peel (raw)	21294	568.2	24885	1022	2391	14.30	13.32	20.751
	flesh steaming)	10521	355.0	5198	581.4	1796	7.110	7.851	6.956
414	flesh (raw)	7418	932.4	6445	950.4	2626	7.904	9.202	9.804
	peel (raw)	12421	2260	26649	2030	3118	12.25	12.12	32.35
	flesh steaming)	6385	957.2	7713	960.8	2561	7.252	8.754	9.302

1070 ± 8.7 mg.kg⁻¹ DM chlorogenic acid and 430.4 ± 2.3 mg.kg⁻¹ DM isomers 3,4-, 3,5- and 4,5-diCQA in Chinese purple sweet potato. Jung et al. (2011) determined six phenolic acids (3-CQA, 5-CQA, 4-CQA, 3,5-diCQA, 4,5-diCQA and 3,4-di CQA) in eight Korean

sweet potatoes. The sums of acids ranged from 19.77 mg.100 g⁻¹ FM in the middle part of potato roots to 300.3 mg.100 g⁻¹ FM in the stem end of potato roots. In two varieties of sweet potatoes, Koganesengan and Beniazuma (Japan) determined Ishuida et al. (2000)

Table 2 Total polyhenols content (TPC, mg gallic acid equiv.k⁻¹g DM), phenolic acids (mg chlorogenic acid equiv.kg⁻¹ DM) contents of three cultivar sweet potatoes roots.

		TPC	Caffeic acid	3-CQA	Sum CQA-isomers
O'Henry	flesh (raw)	1161±81.01 ^{a;A}	1.276±0.005 ^{a;A,B}	57.57±8.333 ^{a;A}	110.6 ^{a;A}
	peel (raw)	4263±123.16 ^b	317.3±23.41 ^b	584.1±96.69 ^{b,c}	1734 ^b
	flesh steaming)	1543±72.45 ^B	ND ^A	108.7±8.312 ^A	126.0 ^A
Beauregard	flesh (raw)	1186±60.41 ^{a;A}	12.41±3.281 ^a	193.3±28.59 ^{a,b;A}	387.0 ^{a;A}
	peel (raw)	5062±75.06 ^c	320.7±6.328 ^{b;D}	715.8±92.72 ^c	2,790 ^c
	flesh steaming)	2904±67.64 ^C	6.297±1.336 ^{B,C}	615.7±61.97 ^B	1423 ^B
414	flesh (raw)	9800±145.0 ^{d;E}	ND ^{a;A}	2163±280.5 ^{d;C}	5790 ^{e;D}
	peel (raw)	13998±386.8 ^e	272.3±142.7 ^b	2392642.9 ^d	4350 ^d
	flesh steaming)	7644±561.9 ^D	6.895±0.146 ^C	2282±304.9 ^C	3424 ^C

Note: ^{a,b,c,d,e} – statistically significant differences between content of caffeic acid (3-CQA and sum CQA isomers. resp.) in raw flesh (raw peel) of sweet potatoes from different cultivar (Multiple Range Tests; Method: 95.0 percent LSD).

^{A,B,C,D} – statistically significant differences between content of caffeic acid (3-CQA and sum CQA-isomers. resp.) in raw flesh (steaming flash) of sweet potatoes from different cultivar (Multiple Range Tests; Method: 95.0 percent LSD).

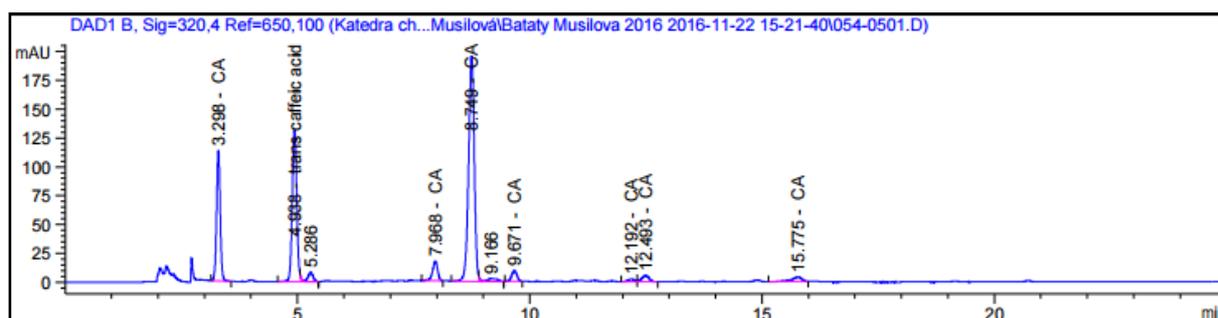


Figure 7 Chromatogram of trans-caffeic acid and chlorogenic acid isomers in peel of sweet potato (cv. O'Hara).

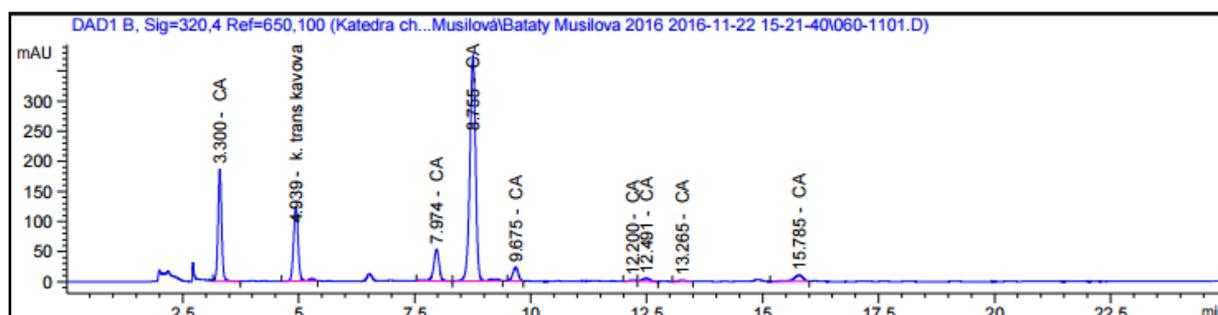


Figure 8 Chromatogram of trans-caffeic acid and chlorogenic acid isomers in peel of sweet potato (Beauregard – peel).

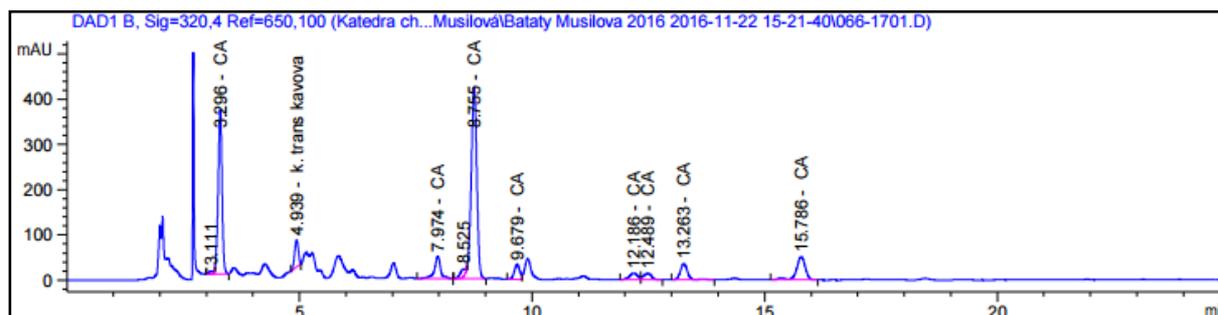


Figure 9 Chromatogram of trans-caffeic acid and chlorogenic acid isomers in peel of sweet potato (414 – peel).

21.2 and 18.8 mg chlorogenic acid in 100 g DM.

Statistically significant differences were found out in the content of polyphenols, caffeic acid and the sums of CQA-isomers between the raw sweet potato and those boiled in water. With the exception of cv. Beauregard (CfA) and 414 (sum CQA-isomers) phenolic compounds contents are higher in the steamed potatoes compared to the raw ones. TPC in O'Henry variety was 1.33 times and in Beauregard 2.45-times higher. TPC was 1.2 times lower in the steamed sweet potatoes compared to the raw potatoes in purple variety 414. **Bellail et al. (2012)** compared the effect of different processing methods (raw – boiled – baked – microwaved – deep fried) on total phenolics in four cultivars of sweet potato. For each cultivar of sweet potato, the TPC of the processed samples were higher than that of raw sample, and the result indicates that all home processing methods resulted in a significant increase ($p \leq 0.05$) in phenolic content of the flesh tissues. The increasing rate was in the following order: deep-frying >baking >boiling >microwaving. Boiling and microwaving showed the highest total phenolics with Beauregard cultivar (2.8 and 2.6 times, respectively), as compared to the raw samples.

The influence of steaming reduced the CFA content in the varieties O'Henry and Beauregard and increased it in the purple variety 414. 3-CQA content was increased in all the three varieties.

Rautenbach et al. (2010) observed an increase in the chlorogenic acid content in all the varieties of sweet potato after heat treatment. The increase was between 21.1% and 79.1%. **Bellail et al. (2012)** presents a significant increase ($p \leq 0.05$) in phenolic acids in the processed sweet potatoes. The caffeic acid and 3,4-diCQA content was more than 7 times higher in the cooked sweet potatoes cv. Beauregard in comparison with their content in the raw roots.

The increase in the efficiency of extraction of phenolic compounds can be explained by the damage to cellular structures caused by the peeling, or by the heat treatment of the plant material (**Bellail et al., 2012; Huang et al., 2006**).

CONCLUSION

The sweet potato is a crop which is relatively undemanding in respect of the plant growing conditions. It is the source of many nutritional and bioactive substances. Its cultivation is widespread mainly in African and Asian countries and often is concentrated in the poorest growing areas and among farmers with limited-resources. Sweet potatoes are grown by the small growers in the Slovak Republic. They reach our consumers in particular as imported goods. It would be appropriate to increase consumer interest in this kind of crops consumed less frequently, because it has a high content of mineral substances, vitamins and antioxidants as well as dietary fibre, carotenoids and anthocyanins.

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