TOTAL CONTENT OF SOME ANTIOXIDANTS IN TEN ROMANIAN POTATO GENOTYPES

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Abstract

Being a valuable source of important nutrients (starch, fibres, amino-acids, vitamins, minerals) potato tubers represent a complex matrix in the human diet, having potential health benefits. This vegetable has a significant contribution to the dietary intake of some antioxidants such vitamin C, carotenoids and phenolic compounds. The present study evaluated the total content of phenolic compounds (TPC) and of carotenoids (TCC) in skin and flesh of ten Romanian potato genotypes grown in Brasov, over two years. Significant differences of all parameters tested were observed across genotypes. Lower levels of TPC and TCC were found in the flesh than in the skin of the tubers. Blue genotype 'Blue Purple of Galanesti' had the highest values for all parameters excepting the carotenoids in flesh and skin which were higher in the variety 'Sevastia'. This study contents information regarding the content of valuable micronutrients of several Romanian potato cultivars with good nutritional quality.

Key words: antioxidants, potato, total carotenoids content, total phenolic content.

INTRODUCTION

Many studies on potato composition revealed that there is much more in potatoes than starch. Indeed, the potato tubers contain important phytochemicals (as phenolic compounds, carotenoids, vitamin C) that may prevent the development of diabetes, cardiovascular diseases and certain types of cancers (Ezekiel et al., 2013).

Some of the reasons that led the choice of this subject (study polyphenol and carotenoids content of potato tubers) were: the special of this bioavailability plant, economic considerations (lower price compared to other sources of antioxidants), the content of anthocyanin and the presence of valuable polyphenols (chlorogenic acid, quercetin and kaempferol glycosylated with rutinose significant amounts of catechin, usually acylated glycosides of rutinose and glucose) (Andre et al., 2006; Ezekiel et al., 2013).

Regarding the carotenoids, violaxanthin, antheraxanthin, lutein and zeaxanthin are found in potatoes (Morris et al., 2004).

The variation of carotenoids content among potatoes species and among the varieties in the

same species is wide, with total carotenoids content up to 60 times higher in *S. phureja* and *S. tuberosum* and up to a 20-fold difference within the same species (Morris et al., 2004; Burgos et al., 2009; Lu et al., 2001; Andre et al., 2006).

In particular, potato antioxidants have been shown to have favourable impacts on several measures of cardio-metabolic health, including lowering blood pressure, improving lipid profiles and decreasing markers of inflammation (Valcarcel et al., 2015; Wang et al., 1999).

Tubers with increased level of polyphenols or carotenoids could have an impact on human health, because these compounds posses antioxidant, antiglycemic, antiviral and antiinflammatory activities (Duthie et al., 2000; Wang et al., 1999). This impact could be strong especially for people where potato is the most important food crop and therefore would be of interest to consumers and producers (Valcarcel et al., 2015).

The main objective of this work was to evaluate the amount of total polyphenols and carotenoids content of ten Romanian potato cultivars grown in Brasov over two years with different climatic conditions (Table 1).

MATERIALS AND METHODS

Biological material. The following potato genotypes were studied:

- BV 1791/1, BV 1871/4 Romanian breeding lines from NIRDPSB Brasov;
- 'Christian', 'Roclas', 'Sevastia', 'Marvis', 'Castrum', 'Brasovia', 'Cosiana' (new Romanian varieties);
- 'Albastru Violet Galanesti' ('Blue Purple of Galanesti') (genotype with strong pigmentation in the flesh).

Seed tubers were planted in May in Brasov (coordinates lat. 45.6744234, long. 25.539622) in 2016 and 2015, with three replicates. Similar fertilizer chemical inputs were applied in both years. The climatic conditions of the experimental years are presented in table 1. Mature tubers were harvested 160 days after planting in Brasov in 2016 and 148 days in Brasov in 2015. After harvest, marketable tubers (medium size and free of damage and defects) were selected, washed, stored at 4°C until the sample preparation.

Month	Year	Mean temperature (° C)	Rainfall (mm)	
Mari	2015	13.2	82.6	
wiay	2016	12.4	100.4	
June	2015	16.3	107.7	
	2016	19.0	121.2	
July	2015	17.9	95.9	
	2016	19.7	28.8	
August	2015	17.3	78.5	
	2016	18.4	85.8	
September	2015	13.5	54.7	
	2016	15.0	38.0	
October	2015	8.2	42.7	
	2016	6.9	96.0	
Average / Sum	2015	14.4	462.1	
	2016	15.2	470.2	

Table 1. Climatic conditions in the experimental years

Sample preparation

Composite samples (4 to 10 tubers from each cultivar, depending of their size) were

prepared by pooling tubers with a potato peeler. The tuber flesh was quartered from stem to bud and one of the quarters sliced. The tissues were freeze-dried (ScanVac CoolSafe 55-9 Pro Freeze Dryer, Denmark), ground to a fine powder (using a coffee grinder) and stored to -20°C until analysis.

Extraction. The extraction was carried out following the method described by Valcarcel et al. (2015). So, 0.2 g of freeze-dried potato skin or 0.6 g of flesh were weight into a 50 ml centrifuge tubes and 5 ml of ethanol solution 80% (v/v) in pure water were added. The tubes were shaken 5 min at room temperature and centrifuged 15 min at 10 000 rot/min. A part of supernatant was transferred to 1.5 ml tubes and stored at -20°C until analysis.

Total Phenolic Content (TPC) Analysis. The TPC was determined spectrophotometrically by Folin Ciocalteu method (Singleton et al., 1999) with several modifications (Digantha, Sastry, 2014). 20 µl of skin extracts and 50 µl of flesh extracts were mixed with 50µl, respectively 100µl pure water in a 96 well flat bottom assay plate (NUNC, Denmark). 50ml Folin Ciocalteu reagent were added and mixed for 1 min. After 5 min., 80 µl of a 20% solution (w/v) of Na_2CO_3 were added and mixed with a pipette: the microplates were shaken for 5 min. in the plate reader. After that, the plates were incubated at room temperature in the dark, agitating at 150 rpm on a MicroPlate Shaker (Biosan PST-60HL-4, Latvia) for 90 min. The absorbance of the samples was determined at 725 nm (Tecan SunRise, software Magellan). Gallic acid was used as standard and total phenolic content was expressed as milligrams GAE (Gallic acid equivalents) per gram of dry weight (DW) materials.

Total carotenoids content (TCC) analysis. Total carotenoids content was determined according to Burgos et al. (2009) without alkaline hydrolysis. Extraction of TCC from 0.5 g of powdered skin or 2 g of powdered flesh was sequentially carried out in triplicate with acetone, shaking in 50 ml tubes at 10 000 rot/min for 15 minutes. The supernatants were combined and 5 ml of petroleum ether and 20 ml of ultra-pure water added. The tubes were shaken vigorously by hand and centrifuged at 10 000 rot/min for 1 minute. The top organic phase was removed and washed with pure water, separating both phases as described above. The top organic phase was again removed and the absorbance of an aliquot was measured at 450 nm against petroleum ether using a UV VIS spectrophotometer Spectronic Genesys 5 (Milton Roy).

Statistical interpretation

Analysis of variance (ANOVA) and Duncan's multiple range test were used.

RESULTS AND DISCUSSIONS

Total polyphenol content

The experiments conducted in Brasov in 2015 and 2016 show that cv. 'Blue Purple of Galanesti' had the highest mean TPC value in skin and flesh tissue, in the years 2015 and 2016. The samples with the lowest TPC values were from the breeding line BV 1791/1 in both years for both tissues flesh and skin, respectively (Table 2).

The levels of TPC ranged from 0.59 to 3.38 and 2.68 to 10.79 mg GAE g⁻¹ DW in the flesh and skin, respectively, with flesh and skin contents showing a significant difference between the cultivars (Table 2). The skin of the potatoes studied contained on average 4.62 and 4.12 times more phenolic compounds (TPC) than the flesh, in 2015 respectively in 2016 (Table 2). TPC in both tissue (skin and flesh) was positively correlated with the colour of the tissue, with a Pearson coefficient 0.813 (p<0.01) and 0.596 (p<0.01) for skin and flesh respectively.

The results obtained are comparable with those reported by other researchers, with values ranging from 1.56 to 12.9 and 0.54 to 3.59 mg GAE g^{-1} DW in the skin, respectively in the flesh (Valcarcel et al., 2015), from 1.4 to 2.4 mg GAE g^{-1} DW in the flesh (Cornacchia et al., 2011), from 0.92 to 12.37 mg GAE g^{-1} DW for whole tubers (Andre et al., 2006; Xu et al., 2009) from 1.00 to 4.3 mg GAE g^{-1} DW in the skin (Kahkonen et al., 1999; Makris et al., 2007).

Despite the fact that some vegetable sources have higher TPC than potatoes, in many countries the potatoes are consumed in higher quantities and so, potatoes make an important contribution to the phenolic compounds daily intake. A recent study in USA estimated that potatoes were the third highest contributor to the daily intake of phenolic compounds, after oranges and apples, with a daily intake consumption of 171 g day⁻¹ (Chun et al., 2005). These properties of potatoes could be greater if the cultivars with high TPC level become popular for the people. Unfortunately, the cv. 'Blue Purple of Galanesti' (reported in this study with the higher TPC level in the flesh and skin) is not accepted with pleasure by the consumers because the tubers are small, elongated and with deep eyes. Maybe in the future, the potato breeders correct these quality parameters by developing new cultivars with functional food characteristics.

Total carotenoids content

The experiments conducted in Brasov show that cv 'Sevastia' had the highest mean TCC value in both skin and flesh. Genotypes with the lowest quantified values were BV 1791/1 in 2015 and 2016 for both tissues, flesh and skin respectively (Table 3).

The levels of TCC ranged from negligible quantities to 8.92 and 4.38 mg kg⁻¹ DW in the skin and flesh, respectively, with flesh and skin contents showing a significant difference for both years (Table 3). On average, the skin of the potatoes analysed contained between two and three times more TCC than in the flesh (2.24 times in 2015, 3.24 times in 2016).

These data are in agreement with other studies: it has been reported (Breithaupt, Bamedi, 2002) that the total quantity of the four main carotenoids analysed in eight commercial potato varieties was between 0.38 and 1.75 mg kg⁻¹, which would be equivalent to 1.90-8.75mg kg⁻¹ DW assuming 80% of water in the fresh samples. Other authors (Morris et al., 2004, Lu et al., 2001) found that for varieties 'Pentland Javelin' (white flesh) TCC was 1.60 mg kg⁻¹ DW and for variety 'Yukon Gold' (yellow flesh) and 'Superior' (white flesh) were 1.11 and 0.64 mg kg⁻¹ FW, respectively (equivalent to approximately 5.55 and 3.20 mg kg⁻¹ DW). Excepting the cultivars 'Sevastia', 'Christian' and 'Roclas', the majority of the varieties included in this work had relatively low content of TCC in the flesh, with values below 0.24 and 1.14 mg kg⁻¹ DW (Table 3). TCC in both tissue (skin and flesh) was very weak positively correlated with the colour of the tissue, with a Pearson coefficient 0.357 (p<0.01) and 0.196 (p<0.01) for skin and flesh, respectively.

Tubers grown in Brasov in 2016 had an average higher TCC and TPC content than those grown in Brasov in 2015. As reported and other researchers, potato peels are a great source of phenolic compounds because almost 50% of phenolic are located in the peel and adjoining tissues (Albishi et al., 2013; Al-

Weshahy et al., 2009). The results presented in this study revealed higher TPC and TCC in skin compared with the flesh tissue. There is a lot of information in the literature about TPC in potato varieties grown in different controlled conditions. In this study, there were analysed several new Romanian varieties and some cultivars with strong colour in the flesh and skin tissue.

Table 2. Total polyphenol content (ing GAE/g D w) of potato samples (Brasov, 2015-2010	Table 2. Total polyphenol	content (mg GAE/g DV	<i>I</i>) of potato samples	(Brasov, 2015-2016
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Cultivars/ genotypes	Flesh/ Skin colour	Year 2015		Year 2016	
		Flesh	Skin	Flesh	Skin
BV 1791/1	W/LY	0.59±0.012 (g)*	2.68±0.354 (h)	0.82±0.207 (f)	2.72±0.247 (i)
BV 1871/4	C/R	0.78±0.136 ef)	7.96±0.914 (b)	0.88±0.118 (ef)	8.32±0.604 (b)
AVG	B/B	3.03±0.208 (a)	9.39±1.262 (a)	3.38±0.400 (a)	10.79±1.114 (a)
Brasovia	WY/Y	0.92±0.057 (d)	3.35±0.130± (f)	1.04±0.158 (e)	3.90±0.307 (gh)
Castrum	LY/Y	1.15±0.113 (c)	4.01±0.264(e)	$1.43 \pm 0.246(c)$	4.55±0.164 (j)
Christian	Y/R	1.25±0.088(c)	6.54±0.614(cd)	$1.53 \pm 0.341(c)$	6.93±0.108 (d)
Cosiana	WY/R	0.73±0.027 (ef)	7.16±0.828 (bc)	1.03±0.249 (e)	7.68±0.420 (c)
Marvis	WY/Y	0.75±0.017 (ef)	4.51±0.317 (de)	0.91±0.315 (ef)	4.66± 0.350(f)
Roclas	Y/Y	0.88±0.230 (<i>de</i>)	$3.15 \pm 0.167(g)$	1.27±0.218 (d)	$3.56 \pm 0.807(g)$
Sevastia	DY/Y	1.66±0.150 (b)	$4.99 \pm 0.063(d)$	1.95±0.142 (b)	5.57±0.226 (e)
Mean		1.164	5.383	1.424	5.868

*Means with different letters are significantly different at p<0.05 in each column. Values reported for the two Romanian trials are the mean of three filed replicates. Abbreviations: GAE-Gallic Acid Equivalents; DW-dry weight; BV 1791/1, BV 1871/4 breeding lines; AVG-Albastru Violet de Galanesti (Blue Purple of Galanesti); W-white; WYwhite yellow; C-cream; LY-light yellow; Y-yellow; DY-dark yellow; R-red; B-blue; C-Cream.

Cultivars/ genotypes	Flesh/	Year 2015		Year 2016	
	colour	Flesh	Skin	Flesh	Skin
BV 1791/1	W/LY	0.24±0.048 (h)*	1.24±0.424 <i>(f)</i>	0.32±0.033 (g)	1.53±0.025 (g)
BV 1871/4	C/R	1.18±0.634 (d)	2.08±0.632 (d)	1.42±0.392 (de)	2.98±0.392 (e)
AVG	B/B	1,14±0.325 (d)	2,43±0.286 (c)	1,31±0.273 (de)	nd
Brasovia	WY/Y	0,38±0.105 (g)	1,57±0.122 (e)	0,38±0.105 (g)	1,84±0.048(ef)
Castrum	LY/Y	0,82±0.208 (e)	1,46±0.162 (e)	1,30±0.049 (de)	2,68±0.035(ef)
Christian	Y/R	1,45±0.942 (c)	2,44±0.622 (c)	2,22±0.825 (c)	4,55±0.692 (c)
Cosiana	WY/R	0,69± 0.092(ef)	1,32±0.102 (ef)	1,68±0.412 (d)	3,53±0.528 (d)
Marvis	WY/Y	0,47±0.026 (g)	1,43±0.094 (e)	0,79±0.033 (f)	1,54±0.077 (f)
Roclas	Y/Y	3,22±0.725 (b)	5,10±0.944 (b)	3,42±0.046 (b)	8,04±0.831 (b)
Sevastia	DY/Y	4.84±0.492 (a)	7.83±0.863 (a)	4.78±0.082 (a)	8.92±0.723 (a)
Mean		1.443	4.685	1.762	3.957

Table 3. Total carotenoids content (mg /kg DW) of potato samples (Brasov, 2015-2016)

*Means with different letters are significantly different at p<0.05 in each column. Values reported for the two Romanian trials are the mean of three filed replicates. Abbreviations: GAE-Gallic Acid Equivalents; DW-dry weight; BV 1791/1, BV 1871/4 breeding lines; AVG-Albastru Violet de Galanesti (Blue Purple of Galanesti); W-white; WY-white yellow; C-cream; LY-light yellow; Y-yellow; DY-dark yellow; R-red; B-blue; C-Cream; nd-non determined.



Figure 1. Mean total polyphenols content (TPC) and total carotenoids content (TCC) according to the colour of the skin (A) and flesh (B) colour of the tubers. Bars represents mean values of samples collected in 2015 and 2016 with different skin and flesh colour. Error bars represent standard deviations. The letters different upper the bars indicate significant difference at p<0.05. Abbreviations: TPC-total polyphenols content; TCC-total carotenoids content; DW-dry weight; SD-standard deviation

Results regarding the influence of skin and flesh colour on TPC and TCC are presented in Figure 1. The tubers with blue skin had the highest TPC values. These values were significantly different from other colours, except red-skinned varieties (Figure 1). In the flesh tissue, blue potatoes had the highest TPC values, significant different from all the other colour variants. Blue and red colours are due to the presence of anthocyanins (Andre et al., 2008) and the higher TPC values obtained for this kind of tubers colour can be attributed to these pigments with high antioxidant potential. Strong correlations between TPC and antioxidant activity were reported in previous studies (Andre et al., 2006; Hu et al., 2012; Reyes et al., 2005; Valcarcel et al., 2015) which specified that both phenolic and flavonoid compounds are the main contributors to the antioxidant potential. Climate data for the growing season in Brasov (Table 1) show that average temperatures in 2016 were slightly higher than in 2015. This difference was accentuated in June, July and August and was accompanied also by increased rainfall. TCC and TPC content seems to be higher in early developing tubers (Morris et al., 2004) so these climatic conditions during the summer months could contributed to difference observed. Reddivari et al. (2007) reported that tubers planted in the location with higher average

temperature and increased rainfall contained higher levels of carotenoids.

CONCLUSIONS

Ten Romanian potato genotypes with different colour of the skin and flesh tissue (grown in Brasov two years) were analysed for estimation their total polyphenols and carotenoids content. The results obtained were in the range of values reported in the literature.

Significant differences between the different cultivars tested were observed for TCC and TPC. Higher contents of antioxidants (polyphenols, carotenoids) were found in intense coloured fleshed tubers than in white counterparts. which should allow visual selection of varieties with enhanced levels of these compounds. So, the genotype 'Blue Purple of Galanesti' with blue skin and flesh had higher values of total polyphenols content and the cultivar 'Sevastia' (with a dark yellow colour of the flesh) had the highest values of total carotenoids content.

Potato consumption has a great importance in population's food and this study offer preliminary information to researchers and producers on the level of some phytochemicals such carotenoids and polyphenols, antioxidants with functional properties.

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