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# Comparative Analysis Of Total Polyphenol Content, Total Flavonoid Content And Antioxidant Potential Of Four Colored Flesh Potato Genotypes And Some Berries Fruits

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**ABSTRACT**
Antioxidants contained in vegetables and fruits are important for preventing the oxidative damages produced by free oxidant radicals, having a positive impact on the human health. Potato tubers with colored flesh contain antioxidants as do some of the berries fruits. This study evaluated the total polyphenol content (TPC), total flavonoids content (TFC) and antioxidant capacity (% of DPPH inhibition) of four potatoes genotypes with colored flesh tubers in comparison with cranberries, blueberries, blackberries and raspberries. The range for total polyphenol content (TPC) and total flavonoids content (TFC) in the food material tested was: (5464.3±87.643 - 10049.67±326.44 μg GAE/g) and (46.067±9.259 - 510.4±2.113μg QE/g) respectively. The potential antioxidant activity of the purple potato tubers backed was closed to that obtained for raspberries. DPPH scavenging activity in all samples (berries and backed purple potato tubers) was positively correlated with TPC and TFC. The advantages of the colored flesh potato tubers consumption are: the special bioavailability of this plant, economic considerations (lower price compared to other rich sources of polyphenols like the berries) and the presence of valuable antioxidants (increasing the nutritional value of this food).
**Keywords -** antioxidant capacity, berries, flavonoids, polyphenols, purple potato

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## I. INTRODUCTION

Antioxidant activity describes the ability of redox molecules in foods and biological systems to scavenge free radicals, considering the additive and synergistic effects of all antioxidants rather than the effects of single compound [1, 2]. Therefore, in terms of total phenols (TPC), total flavonoids (TFC), dietary antioxidants are associated with Total Antioxidant Capacity (TAC) [2]. However, the TAC consumed by an individual depends on the type and amount of food intake. Therefore, the estimation TPC and TFC and their respective TAC provides valuable information on potential health benefits [3, 4]. Moreover, the marketing of so called superfoods is commonly based on their antioxidant potential. In fact, a superior antioxidant activity with health benefits has been claimed for a number of antioxidants foods on in vitro antioxidant assays. There is a great variability in the phenolic compounds and the antioxidant activity in the commercial fruits [5-7]. Cranberries, blackberries, black blueberries, raspberries are considered to be superfood because of their antioxidant activity. They are some of the richest sources of antioxidant phytonutrients having higher level of anthocyanins, pro-anthocyanins, phenolic acid and flavonoids than many other fruits and vegetables. Because of their diverse range of these phytonutrients, cranberries blackberries, black blueberries, raspberries have beneficial effects on human health including antidiabetic, anti-neurodegenerative, anti-inflammatory and anti-obesity effects [2, 3, 8, 9].

On the other hand, potato is one of the major food crops and breeding new potato cultivars whit high level of antioxidant compounds is considered a realistic approach to increasing dietary antioxidant intake [10, 11]. Potato tubers with red and purple flesh are considered to have high phenolic contents [11, 12, 13, 14, 15]. Common phenolic acids in potatoes are chlorogenic acid, caffeic acid, cinnamic acid, p-cumaric acid, ferulic acid and sinapic acid [16]. The predominant phenolic acid in potatoes is the chlorogenic acid, constitutes about 80% of the total phenolic compounds [16]. The frequently flavonoid compounds in potato tubers are catechins and epicatechins. In addition to common phenolic compounds, purple fleshed potato tubers contains acylated glucosidases of pelargonidin, acylated glucosides of malvidin, petunidin, peonidin and

delphinidine [17]. There were identified and the flavonols: catechin, rutin and kaempferol-3-o-rutinoside. About 30 μg per 100 g FW of flavonoids (predominant catechin and epicatechin) are present in the flesh of white-fleshed potatoes with roughly twice the amount present in red purple-fleshed potatoes [4]. Purple potato with flesh strongly pigmented represents a rich source of anthocyanin pigments. Whole unpeeled with complete pigmentation in the flesh may have up to 40 mg per 100 g FW of total anthocyanin [17]. Even though many species produce polyphenols and anthocyanins, availability and the cost of certain materials vegetal origin limited commercial exploitation of these natural sources of anthocyanin pigments only for a few species [18].

In particular, as in the case of the fruits tested in this paper, potato antioxidants have been shown to have favorable impacts on several measures of cardio-metabolic health, including lowering blood pressure, improving lipid profiles and decreasing markers of inflammation [3, 8, 9, 19]. 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 [19].

The main objective of this research work was to measure the TPC and TFC in purple potato tubers (four genotypes very appreciated by the consumers and producers), cranberries, black blueberries, blackberries, raspberries fruits and assess their potential antioxidant capacity (the percentage of DPPH inhibition). This study provides information on content of important phytochemicals in several potato cultivars with improved nutritional quality, the results being compared whit the levels of these compounds in some berries fruits.

## II. MATERIAL AND METHODS

### 2.1. Chemical and materials

The following chemicals (analytical grade) were used:

- Gallic acid (prod. no. 2699.1), quercetin hydrate (prod. no. 7138.1), aluminum chloride (prod. no. CN86.1) all obtained from Roth (Germany)
- Sodium hydroxide (prod. no. S5882), sodium nitrite (prod. no. 237213), Trolox (6-hydroxy-2, 3, 7, 8-tetramethylcroman-2 carboxylic acid, prod. no. 23881-2) and DPPH (2, 2-diphenyl-1-picrylhydrazyl, prod. no. D9132), Folin-Ciocâlteu Reagent (Folin-Denis' Reagent, prod. no. 47742), all were obtained from Sigma Aldrich Corporation (USA).

### 2.2. Biological material

The following potato genotypes with strong pigmentation in the flesh were studied:

-'Blue Congo', 'Blue de la Manche', 'Patraque Auvergne'

-'Albastru Violet Galanesti '('Blue Purple of Galanesti')

Seed tubers were planted in May in Brasov (coordinates lat. 45.6744234, long. 25.539622) in 2017. Mature tubers were harvested 152 days after planting in Brasov. After harvest, marketable tubers (medium size and free of damage and defects) were selected, washed, stored at 4ºC until the sample preparation.


**Fig. 1.** Potato with purple flesh (samples).

The potato tubers (Fig. 1) were pierced four time on which side using a fork and baked at 200ºC for 1 hour in an oven (method adapted by Kalita and Jayanty (2014) [2].

The berries fruits: cranberries (*Vaccinium myrtillus* L.), black blueberries (*Ribes Nigrum* L.), blackberries (*Rubus fruticosus* L.) and raspberries (*Rubus idaeus* L.) were purchased from commercial market.

All samples were freeze dried with a 0.12 mBa vacuum at -50ºC (ScanVac CoolSafe 55-9 Pro Freeze Dryer, Denmark), ground to a fine powder (using a coffee grinder) and stored in plastic zipper bags at -20ºC until analysis.

### 2.3. Extraction

Phenolic, flavonoids and antioxidant compounds were extracted from the material obtained and stored as described above. One gram of freeze dried food material was weight into a centrifuge tube (50 ml) and 50ml of aqueous 96% ethanol was added. The mixture was homogenized for 5 min. (Vortex), centrifuged at 10000rpm for 10 min. and filtered through Whatman (Number 400) filter paper.

### 2.4. Total Phenolic Content (TPC) Analysis

The total phenolic content was determined spectrophotometrically by Folin Ciocâlteu method [21] with several modifications [2]. 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 Ciocâlteu 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 (TecanSun Rise, software Magellan). Gallic acid was used as standard and total phenolic content was expressed as μg GAE (Gallic acid equivalents) per gram of dry weight (DW) materials.

**2.5. Total flavonoids content analysis**

It is based on the nitration of any aromatic ring bearing a catechol group (two contiguous hydroxyls in the aromatic ring) with its three or four positions unsubstituted or not sterically blocked, followed by the formation of an aluminum complex which turns to red in basic medium [2].

Aliquots of 150μl of extracts were also transferred to 1.5ml tubes. Volumes of 600μl distillated water and 45μl of a 7.5% solution sodium nitrite were added to each tube, mixed by inversion and left to react for 5 min. A volume of 45μl aluminum chloride 10% solution was pipetted onto the tubes, mixed by inversion and allowed to react for 1min. Finally, 300μl of a 1N sodium hydroxide solution and 360 μl distillated water were added and the tubes vortex mixed. The absorbance of every solution was measured at 510nm against the blank using a spectrophotometer DR2800 (Hach Lange, USA).

Values of the absorbance samples were interpolated into a minimum squares regression equation (a 5-point calibration curve with an $R^2$ value of 0.998), which was calculated with the absorbance and the corresponding concentration of each quercetin standard. Quercetin hydrate in ethanol was used as the standard. Final results were calculated taking into account sample weight, extraction volumes and dilution factors applied and were expressed as μg quercetin equivalents (QE) per gram of dry weight (DW) of sample.

**2.6. Antioxidant activity analysis (DPPH assay)**

Volumes 20μl of the extracts were added to 20μl of distillated water in a 96 –well bottom microplate. 200μl of 120mg/l DPPH radical solution (using ethanol as a solvent) was then added and mixed thoroughly. The absorbance was measured using a plate reader (TecanSun Rise, software Magellan) at 515nm after keeping the plates in the dark for 30 min. A control with 20 μl of ethanol (no extract) was also included in each plate. The DPPH radical scavenging activity was calculated with the following formula:

DPPH radical scavenging activity (%) =

= [(A control – A sample)/ A control] x 100 (%)

where A is the absorbance at 515 nm.

**2.7. Statistical interpretation**

The experiments were carried out in triplicates and statistical analysis was performed by one way analysis of variance (ANOVA) at $p< 0.05$ significance level using. Also, Duncan's multiple range test was used. A linear regression analysis was performed to evaluate the correlation coefficient.

## III. RESULTS AND DISCUSSION

Phenolic and flavonoids are an important phytonutrients group of various vegetable and fruits because they have health benefits. Many reports are available regarding the profiling of these phytochemicals in cranberries, blueberries, blackberries and raspberries [6, 12]. The antioxidant capacity is caused by all individual antioxidants and thus, in this paper there were measured only TPC and TFC in several berries fruits and purple potato tubers backed (provided from four genotypes).

**3.1. Total polyphenol content (TPC)**

The results for total polyphenol content (determined using Folin Ciocâlteu method) are presented in Fig. 2A. Significant differences ($p<0.05$) were observed in total polyphenol content among the samples tested. The cranberries and blueberries samples had the highest levels of these compounds (10049.67±326.44μg GAE/g, respectively 10012.00±100.02 μg GAE/g). The TPC was in the following order: cranberries > blueberries > blackberries > raspberries > potato cv. 'Patraque Auvergne' > potato cv. 'Blue Purple of Galanesti' > potato cv. 'Blue de la Manche' > potato cv. 'Blue Congo'. There is a short range of total polyphenol content values (5464.3 - 10049.67 μg GAE/g) from potato tubers to cranberries. This range could indicate a comparable antioxidant capacity in potato tubers and the berries samples tested. Previous reports indicated that some purple flesh potato tubers had a TPC comparable to phenolic rich vegetables and fruits (berries, pomegranate and grapes) [2, 11]. As potato are consumed after cooking, we tested the total phenolic content after processing using the most popular cooking method (baking) and observed a 54% (cv. 'Blue Purple of Galanesti'), 52% (cv. 'Blue Congo' and 'Blue de la Manche') and 49% (cv. 'Patraque Auvergne') reduction of TPC after baking in these purple potato tubers (unpublished data). Close similar values of the reduction of TPC in potato after processing using different cooking methods (boiling, baking and microwaving) have been reported [2, 11, 13].

**3.2. Total flavonoid content (TFC)**

Flavonoids are natural phenolic having a large spectrum of biochemical and biological activities, including antioxidant, radical-scavenging properties. The results regarding TFC of the berries and colored potatoes tested are presented in Fig. 2B.

There were observed significant differences ($p<0.05$) between the samples. Cranberries had the highest value of (510.4±2.113µg QE/g DW), whereas purple potato cv. 'Blue de la Manche' had the lowest value of this indicator (46.067±9.259 µg QE/g DW).

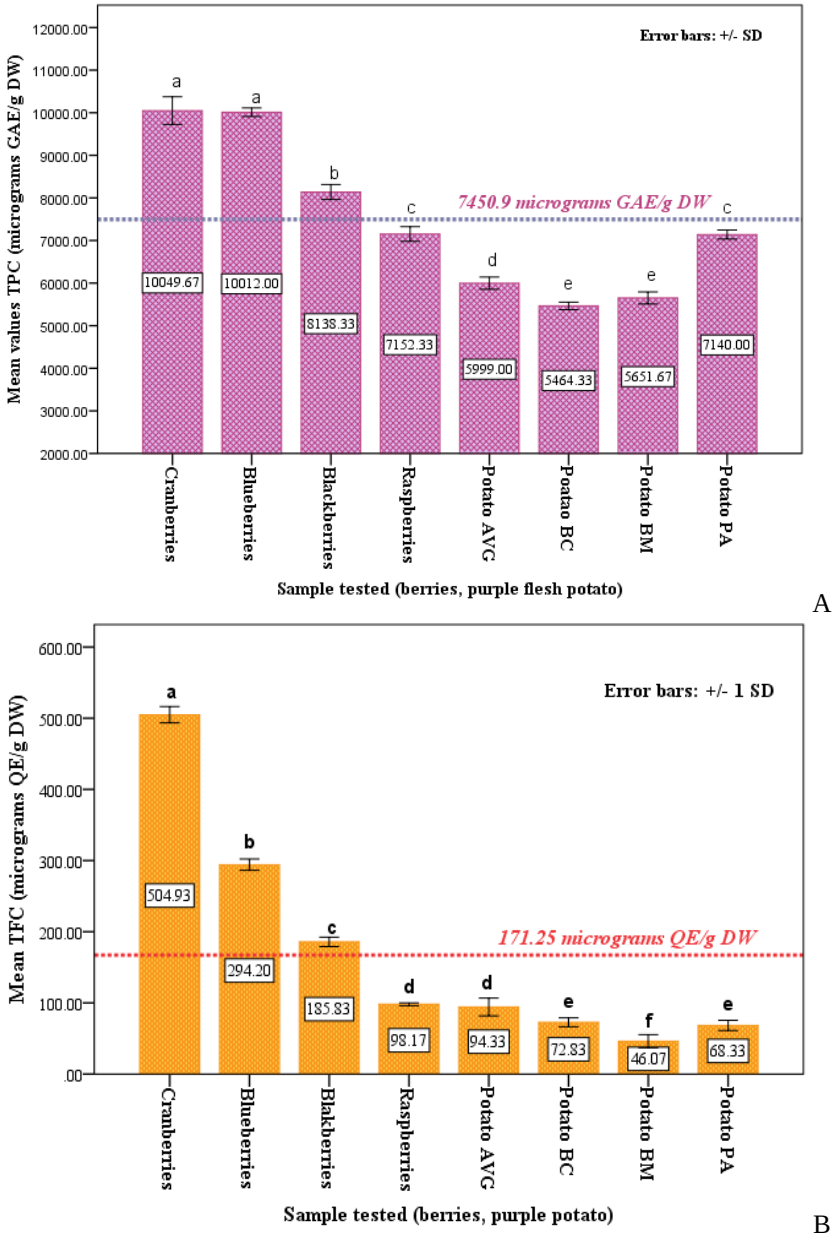


**Fig. 2.** Total polyphenol content (A), total flavonoid content (B) in several berries (cranberries, blueberries, blackberries, raspberries) and different purple potato tubers backed (AVG = Albastru Violet de Galanesti – cv. 'Blue Purple of Galanesti'; BC = cv. 'Blue Congo'; BM = cv. 'Blue de la Manche'; PA = cv. 'Patraque Auvergne'). The line represents the mean values of indicators tested. Values not followed by the same letter are significantly different (P=0.05) according to Duncan's test. Abbreviations: TPC=Total Polyphenol Content; TFC=Total Flavonoid Content; GAE= Gallic Acid Equivalent; QE= Quercetin Equivalent; DW=Dry Weight; SD=standard deviation; cv.=cultivar.

The mean values of TFC obtained for the material tested were in the following order: cranberries > blueberries > blackberries > raspberries > potato cv. 'Blue Purple of Galanesti' > potato cv. 'Blue Congo' > purple potato cv. 'Patraque Auvergne' > potato cv. 'Blue de la Manche'. Various papers reported that potato tubers with purple flesh have significant level of flavonoids [2, 11, 13, 14]. Excepting the cranberries, the results indicate a short range of TFC from purple potato tubers cv. 'Blue Purple of Galanesti' to blueberries (94.3-294.2µg QE/g DW), these values could

explain the comparable antioxidant potential of potato tubers with that of the berries samples tested.

### 3.3. Antioxidant activity (DPPH radical-scavenging activity)

The reduction capability of DPPH radical was determined by decrease in absorbance at 515 nm induced by antioxidants (included in the samples studied). Fig 3 presents the DPPH scavenging activity of the ethanol extracts of berries and potato tubers. As shown in this figure, the blueberries had the highest level of % DPPH inhibition, followed by cranberries, blackberries, purple potato AVG (cv. 'Blue Purple of Galanesti'), 'Patraque Auvergne', 'Blue Congo' and 'Blue de la Manche' (with significantly different (P=0.05). As in the case of TFC and TPC content, the raspberries samples had close values of DPPH scavenging activity with some purple potato cultivars

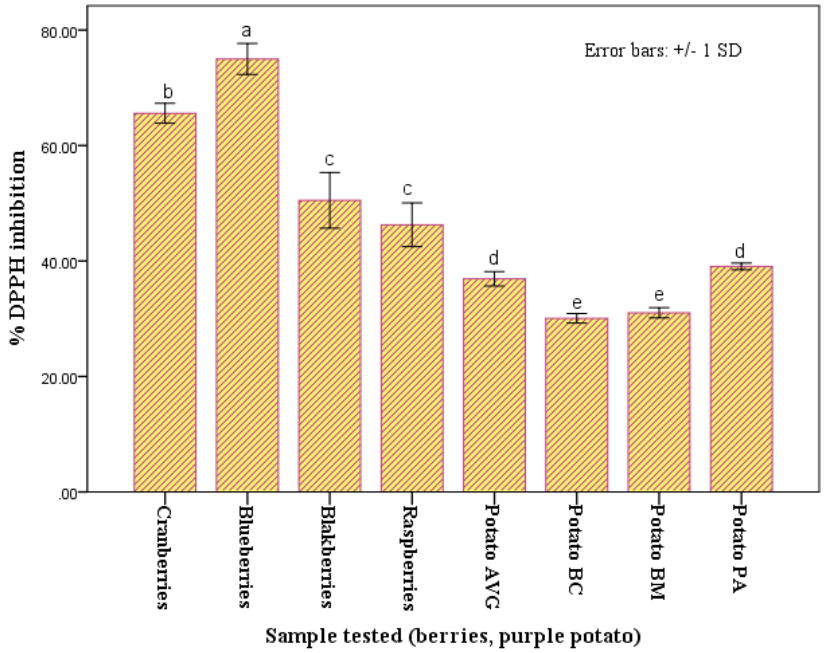


**Fig. 3.** Antioxidant activity of several berries (cranberries, blueberries, blackberries, raspberries) and different purple potato tubers backed (AVG = Albastru Violet de Galanesti – cv. 'Blue Purple of Galanesti'; BC = cv. 'Blue Congo'; BM = cv. 'Blue de la Manche'; PA = cv. 'Patraque Auvergne') by DPPH assay. Values not followed by the same letter are significantly different (P=0.05) according to Duncan's test. Abbreviations: DPPH=2, 2-diphenyl-1-picrylhydrazyl; SD=standard deviation; cv.=cultivar.

The TPC values are correlated with the antioxidant activity. It is possible that some differences between polyphenol content and antioxidant capacity because the complex samples (vegetable and fruits extracts) could content some polyphenol non antioxidants. However, in this research the data show that the DPPH values were strongly correlated with the TPC ($r^2$ = 0.919). A good correlation was found and between TFC and DPPH scavenging activity ($r^2$ = 0.688) (Fig. 4 A&B). DPPH scavenging activity in all tissue (berries and backed potato tubers) was positively correlated with TPC and TFC, with a Pearson coefficient 0.959 ($p<0.01$) and 0.827 ($p<0.01$) for TPC and TFC respectively.

There are great differences between the price of polyphenol rich berries and potato tubers. So, for beneficial and economic alternatives for improving health in poor communities, where the access to meat or the high cost of fruits such as the berries (like cranberries, blueberries, blackberries, raspberries) cause them to be limited, cheaper purple flesh potatoes could be considered a good choice [2, 14].

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 [14].

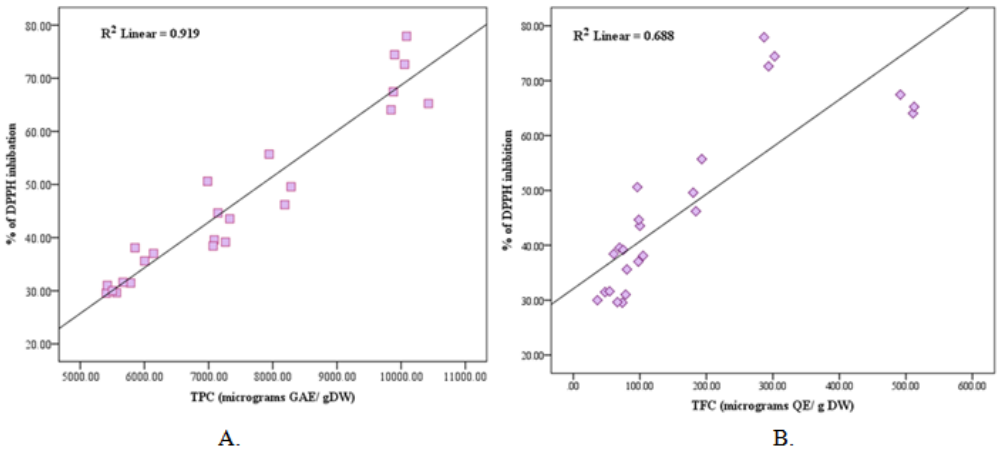


**Fig. 4.** Relationship of total polyphenolic content (A), total flavonoid content (B) with DPPH assay. Abbreviations: DPPH=2, 2-diphenyl-1-picrylhydrazyl; TPC=Total Polyphenol Content; TFC=Total Flavonoid Content; GAE= Gallic Acid Equivalent; QE= quercetin Equivalent; DW=Dry Weigh; SD=standard deviation.

A 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$^{-1}$ [22]. These properties of potatoes could be greater if the cultivars with high TPC level become popular for the people. Unfortunately, the Romanian genotype 'Blue Purple of Galanesti', (reported in this study with high TPC and TFC level) 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 [14].

## IV. CONCLUSION

Potato consumption has a great importance in population's food and this study offer information to researchers and producers on the level of polyphenols and flavonoids (antioxidants with functional potential), the content of these phytochemicals being compared with the one from cranberries, black blueberries, blackberries and raspberries.

As the polyphenols and flavonoids from the berries and other fruits, those from the purple potato tubers have been shown to have favorable impacts on several measures of cardio-metabolic health, including lowering blood pressure, improving lipid profiles and decreasing markers of inflammation. For the total polyphenol content, total flavonoids content and DPPH scavenging activity, the results obtained in this study were in the range of values reported in the literature.

Significant differences between the samples of the food tested were observed for all the indicators studied. Higher contents of polyphenols and flavonoids were found in cranberries, black blueberries and blackberries and close value for raspberries and several intense colored fleshed potato tubers. Among the potato studied, the genotype 'Blue Purple of Galanesti' and the cv. 'Patraque Auvergne' (all with blue skin and purple flesh) had higher values of total polyphenols content and percentage of DPPH inhibition, these value were closed to that obtained for raspberries samples. Therefore, colored potatoes contribute to the daily intake of antioxidants and their consumption thereby may have positive effects on the human health.

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