

Breeding for nutritional quality and pest resistance: potential of a set of non-commercial tetraploid potato cultivars with purple and red flesh**R. Tierno¹ and J. I. Ruiz de Galarreta^{1/*}***Received: 27/08/2015**Accepted: 13/01/2016**Accessible on line: June 2016***Summary**

Potatoes are the most widely cultivated tubers and also a valuable source of nutrients and non-nutrient health-promoting compounds. Cultivars with pigmented skin and flesh are of especial interest due to the presence of high concentrations of carotenoids, anthocyanins, flavonoids and phenolic acids. Several investigations have focused on the characterization of native and wild potato accessions, particularly highly pigmented genotypes in order to broaden the overall genetic variation in terms of commercial and processing aptitude, pest resistance and nutritional quality. However, the incorporation of some of these traits into new breeding lines at the tetraploid level is hampered by the presence of detrimental traits as well as varying ploidy levels. Therefore, a set of ten highly-pigmented tetraploid cultivars (*Solanum tuberosum* L.) was selected and characterized on the basis of tuber morphology, pest resistance and nutritional related traits in order to select parental lines for breeding. Attractive colors and color patterns with potential for the potato snack industry have been identified. Varying degrees of resistance to *Globodera* spp. and *Phytophthora infestans*, which are considered major threats for the crop, have been found among the collection of pigmented cultivars. Nutritional quality related parameters including significantly higher levels of phenolic compounds, anthocyanins, carotenoids, vitamin C and micronutrients (Zn and Fe) were found when compared with the commercial cultivars used as checks. These findings could assist in the selection of parents (progenitors) towards the introduction of interesting traits into new potato advanced clones at the tetraploid level, without the problems derived from ploidy levels and non-commercial characteristics.

Additional Key words: characterization, minerals, vitamin C, bioactive compounds, progenitors.

Mejora para calidad nutricional y resistencia a enfermedades: potencial de un grupo de cultivares de patata tetraploides no comerciales de carne roja o morada**Resumen**

La papa es uno de los tubérculos más extensamente cultivado y constituye una fuente valiosa de nutrientes y compuestos bioactivos. Los cultivares con la piel y la carne pigmentadas son particularmente interesantes debido a la presencia de altas concentraciones de carotenoides, antocianinas, flavonoides y ácidos fenólicos. Se han realizado numerosas investigaciones enfocadas a la caracterización de introducciones nativas y silvestres de papas, especialmente centradas en el estudio de genotipos pigmentados, y con el objetivo de ensanchar la base genética en términos de aptitud comercial y de procesado, resistencia a plagas y calidad nutricional. Sin embargo, la incorporación de algunos de estos caracteres en nuevas líneas de

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mejora a nivel tetraploide se ve dificultada por la presencia de características perjudiciales así como niveles variables de ploidía. Para la realización de este trabajo, una colección de diez cultivares de patata tetraploide (*Solanum tuberosum* L.) fue seleccionada y caracterizada en base a la morfología del tubérculo, la resistencia a plagas y parámetros relacionados con la calidad nutricional, con el objetivo de seleccionar parentales de mejora. Se identificaron colores atractivos y distribuciones de colores con potencial para la industria de producción de chips. Se han hallado diferentes niveles de resistencia a *Globodera* spp. y *Phytophthora infestans*, las cuales se incluyen entre los principales patógenos del cultivo. Los parámetros relacionados con la calidad nutricional incluyeron niveles considerablemente más altos de compuestos fenólicos, antocianinas, carotenoides, vitamina C y algunos micronutrientes (Zn y Fe) comparados con los obtenidos en los cultivares comerciales usados como controles. Estos resultados pueden contribuir en la selección de progenitores para la introducción de rasgos interesantes en nuevos clones avanzados de papa a nivel tetraploide sin los inconvenientes derivados de la existencia de características no comerciales y niveles de ploidía variables.

Palabras clave adicionales: Caracterización, minerales, vitamina C, componentes bioactivos, progenitores.

Introduction

Potatoes are a valuable source of carbohydrates, proteins, minerals and vitamins (Ferne and Willmitzer, 2001). Highly-pigmented potato cultivars have also been reported to show high levels of health promoting phytochemicals, even higher than those of certain berries and fruits (Tierno *et al.*, 2015). Moreover, worldwide annual potato consumption per capita reaches to more than 30 kg, exceeding 80 kg in several countries of America and Northern and Eastern Europe (Helgi-Library, 2015). Thus, potatoes and particularly deep colored genotypes could be considered as a very important source of minerals, vitamins and bioactive compounds for the human diet.

For this reason, during the last decades several researches have focused both on the characterization of native and wild potato genotypes as a source of nutritional quality related traits and on the design of suitable breeding strategies (Spooner *et al.*, 2005; Burgos *et al.*, 2007; Lindhout *et al.* 2011). However, the utilization of native and wild germplasm in potato breeding programs at the tetraploid level is hampered by the presence of non-commercial characteristics, ploidy levels

and lack of adaptation to temperate zones (Ruiz de Galarreta *et al.* 2015).

In this context, a collection of ten highly-pigmented tetraploid *Solanum tuberosum* L. cultivars was selected and characterized in terms of tuber morphology, pathogens resistance and nutritional related traits with the aim of selecting parental lines at the tetraploid level without problems derived from the utilization of non-adapted native or wild potato cultivars with different ploidy levels and detrimental characteristics. fertilization with Fe y Zn, in order to make it possible for this nutrients to be absorbed in higher proportion by the tubers and so for to be available in the needed quantities for human nutrition, particularly in areas where there is deficiency of this micronutrients (Devaux, 2013).

Previous studies, in agronomic biofortification, were performed by Val Verde *et al.* (2013) in Ecuador. They applied Fe and Zn to the soil and to the foliage in five cultivar of potatoes (some native others bred), their results showed that the concentrations of Fe and Zn in the tubers were high, though not of significance. Besides, in Perú, Burgos *et al.* (2007) characterised 37 native cultivars of potato for its content of Fe y Zn and

found out that some of the cultivars reached the deliverance of 29 y 26 % of the recommended ingest (6 mg.day⁻¹ and 4.1 mg.day⁻¹) of Fe y Zn respectively. These results, for children 1 to 3 years old, with an average consumption of potato of 200 g.day⁻¹.

This present research had two objectives: a) determinations of the differentiated response of two native potato cultivars to different levels of fertilization of Fe and Zn in soil in three different towns in Bolivia and b) determine the content of Fe and Zn in the tubers for each level of fertilization applied in each of the two native potato cultivars.

Materials and Methods

Ten highly-pigmented cultivars were selected from potato accessions of the Potato Germplasm Collection, NEIKER (Table 1) and grown during the year 2013 in a precise field trial in Arkaute (Alava) in the north of Spain (500 masl) with a humid climate and annual rainfall of about 800 L m⁻². The soil with a clay loam texture was previously subjected to conventional wheat cropping. Plants were grown from mid-May to mid-October 2013 afre pre-sowing fertilization with 800 kg ha⁻¹ (N-P-K 4-8-16). Watering was performed using an automatic spray irrigation system. After harvesting potatoes were stored at 4°C in a darkened cold room for 1 month.

Table 1. Collection of ten highly-pigmented *Solanum tuberosum* L. cultivars.

Cultivar	Origin
Bleu de La Manche	France
Blue Congo	Sweden
British Columbia Blue	Canada
CIP-7002/34	Peru
Highland Burgundy	
Red	France
Morada	Spain
Purple Peruvian	Peru
Rouge de Flandes	Belgium
Valfi	Sweden
Vitelotte	France

Morphological characterization of tubers was performed considering six descriptors from UPOV (1984). The evaluation of resistance of the set of colored potato cultivars to *Globodera pallida* (Stone) and *G. rostochiensis* (Woll.) was performed in triplicate with greenhouse bioassays following the methodology described by Ruiz de Galarreta *et al.* 1998 and Alor *et al.* 2015. Cultivars were classified according to Rousselle-Burgeois and Mugniery (1998) (< 5 cysts were classified as resistant, 5-20 cysts as partially resistant and > 20 cysts as susceptible cultivars).

Resistance to *Phytophthora infestans* (Mont.) de Bary was evaluated according to Vleeshouwers *et al.* (1999) by estimating the area under disease progress curve (AUDPC), as described by (Bonierbale *et al.* (2008). A total of eight tubers of each cultivar were selected, peeled and freeze-dried. Analyses were performed in triplicate. Sample preparation and chemical analysis of total soluble phenolics (TSP), total monomeric anthocyanins (TMA), total carotenoids (TC), vitamin C (VC) and minerals (Zn and Fe) for nutritional characterization of

tubers were performed according to Tierno *et al.* (2015). The commercial cultivars Panda and Monalisa were selected as controls for nutritional quality related parameters because they are commonly

Results and Discussion

Morphological characterization of tubers showed different colors and color-patterns among the collection of colored potatoes, including interesting combinations consisting in purple or red potato hearts surrounded by light or yellow fleshed halos, mosaic patterned cultivars and also totally anthocyanin-colored tubers (Table

cultivated potato varieties with different commercial characteristics and varying processing quality parameters. Statistical analyses were performed using R (R Development Core Team, 2008). 2). The incorporation of these interesting traits into new potato advanced clones could satisfy the increasing demand of special and exotic food products, such as colored chips and snacks, with advantages in terms of organoleptic and nutritional quality (Ruiz de Galarreta *et al.* 2015). Most cultivars produced oval or long shaped tubers with medium or deep anthocyanin base-colored eyes.

Table 2. Morphological characterization of the collection of tubers.

Cultivars	Skin/Flesh type ¹	Tuber shape ²	Eye depth ³	Color of base of eye ⁴
Bleu de La Manche	P/PW	SOV	M-D	B
Blue Congo	P/PW	OV-L	M-D	B
British Columbia Blue	P/PW	OV	M-D	B
CIP-7002/34	R/R Y	R	D	R
Highland Burgundy				
Red	P/R Y	L	S	R
Morada	P/Y P	OV	S	B
Purple Peruvian	P/P	VL	D	B
Rouge de Flandes	P/R Y	L	S	R
Valfi	P/W P	OV	M-D	B
Vitelotte	P/P	VL	D	B

¹ Key to skin and tuber flesh types: R = Red, P = Purple, Y = Yellow and W = White. ² Key to tuber shape: R = Round, SOV = Short Oval, OV = Oval, L = Large and VL = Very large. ³ Key to eye depth: VS = Very shallow, S = Shallow, M = Medium and D = Deep. ⁴ Key to color of base of eye: L = Long and VL = Very long.

The evaluation of resistance against *Globodera* spp. and *Phytophthora infestans*, which are considered major threats affecting potato production worldwide (Rich, 2013), revealed interesting results (Table 3). A total of four accessions (cvs. Blue Congo, Morada, Rouge de Flandes and Vitelotte) were identified as partially resistant to *G. pallida*

(Pa 2/3). Three of them also showed total (cv. Blue Congo) or partial resistance (cvs. Morada and Vitelotte) to *G. rostochiensis* (Ro 1/4). On the other hand, the cultivar Valfi was classified as moderately resistant to *P. infestans* (13_A2_68). The levels of resistance found among the set of colored cultivars could be utilized to introduce pest resistance related traits in the context of a potato breeding program.

Table 3. Resistances found among the collection of cultivars.

Cultivars	<i>Globodera pallida</i> ¹	<i>Globodera rostochiensis</i> ²	<i>Phytophthora infestans</i> ³
Bleu de La Manche	S	S	S
Blue Congo	PR	R	S
British Columbia Blue	S	S	S
CIP-7002/34	S	S	S
Highland Burgundy			
Red	S	S	S
Morada	PR	PR	S
Purple Peruvian	S	S	S
Rouge de Flandes	PR	S	S
Valfi	S	S	PR
Vitelotte	PR	PR	S

¹ Pathotype: Pa 2/3; ² Pathotype: Ro 1/4; ³ Isolate: 13_A2_68

S = Susceptible; PR = Partially Resistant and R = Resistant

Statistical analysis revealed significance differences between cultivars for all the studied nutritional parameters ($P \leq 0.05$) (Table 4). Total soluble phenolics (TSP) concentrations, ranging from 0.189 ± 0.035 (cv. Monalisa) to 2.149 ± 0.048 g GAE kg⁻¹ fw (cv. Highland Burgundy Red), were higher in purple and red fleshed cultivars (Figure 1A). The highest TSP levels (cvs. Highland Burgundy Red and Purple Peruvian) were more than five times higher than those of the commercial cultivars used as testers. Higher levels of phenolic acids and other bioactive compounds have been previously reported in purple and red fleshed cultivars (Brown, 2005). Total monomeric anthocyanins (TMA) levels found in this study ranged from 0 to 1.311 ± 0.096 g CGE kg⁻¹ fw (Figure 1B). As shown in Figure 1C, total carotenoid levels (TC) were higher in partially yellow

fleshed accessions (cv. CIP-7002/34 and Morada) and ranged from 0.009 ± 0.002 (cv. Bleu de La Manche) to 0.097 ± 0.018 g LE kg⁻¹ fw (cv. CIP-7002/34). On the other hand, concentration of vitamin C (VC) ranged from 0.040 ± 0.007 (cv. CIP-7002/34) to 0.107 ± 0.020 g kg⁻¹ fw (cv. Blue Congo) (Figure 1D). When comparing with Panda and Monalisa, accessions with significantly higher levels of TC (cvs. CIP-7002/34 and Morada) and VC (cvs. Blue Congo, Morada and Purple Peruvian) have been identified within the collection. Despite the fact that potato is considered a valuable source of vitamin C, data extracted from the present study revealed significantly low concentrations of this vitamin among the collection of pigmented cultivars. In this sense, several studies have shown that vitamin C levels are highly dependent on many environmental factors (Dale *et al.* 2003).

Table 4. Comparison of means of the characters: total carotenoids expressed as g lutein equivalents per kg fresh weight (TC), vitamin C (VC) expressed as g vitamin C per kg fresh weight, total soluble phenolics (TSP) expressed as g gallic acid equivalents per kg fresh weight, total monomeric anthocyanins expressed as g cyanin-3-O glucoside equivalents per kg fresh weight (TMA), zinc expressed as g of Zn per kg fresh weight (Zn) and iron expressed as g Fe per kg fresh weight (Fe) evaluated in ten potato cultivars.

Cultivar	TSP		TMA		TC		VC		Zn		Fe	
Highland Burgundy Red	2.15	a	1.311	A	0.036	c	0.058	def	0.021	abc	0.038	c
Purple Peruvian	2.14	a	0.601	C	0.019	de	0.093	ab	0.020	bcd	0.032	f
Vitelotte	1.68	b	0.455	D	0.035	c	0.067	cd	0.020	bcd	0.051	a
Blue Congo	1.18	c	0.298	E	0.010	e	0.107	a	0.022	abc	0.023	h
British Columbia Blue	1.13	c	0.331	E	0.016	de	0.058	def	0.023	A	0.030	g
Rouge de Flandes	1.09	c	1.011	B	0.035	c	0.068	cd	0.017	F	0.034	de
Bleu de La Manche	0.92	d	0.334	E	0.009	e	0.083	bc	0.018	Ef	0.033	ef
CIP-7002/34	0.66	e	0.068	G	0.097	a	0.04	f	0.020	cde	0.043	b
Valfi	0.52	f	0.274	E	0.020	d	0.059	de	0.022	Ab	0.039	c
Morada	0.51	f	0.193	F	0.059	b	0.106	a	0.020	cde	0.035	d
Panda	0.21	g	0	G	0.021	d	0.047	ef	0.015	G	0.020	i
Monalisa	0.19	g	0	G	0.035	c	0.056	def	0.018	def	0.019	i
<i>LSD (0.05)</i>	0.11		0.071		0.010		0.020		0.002		0.002	

Means followed by different letters are significantly different at $P \leq 0.05$.

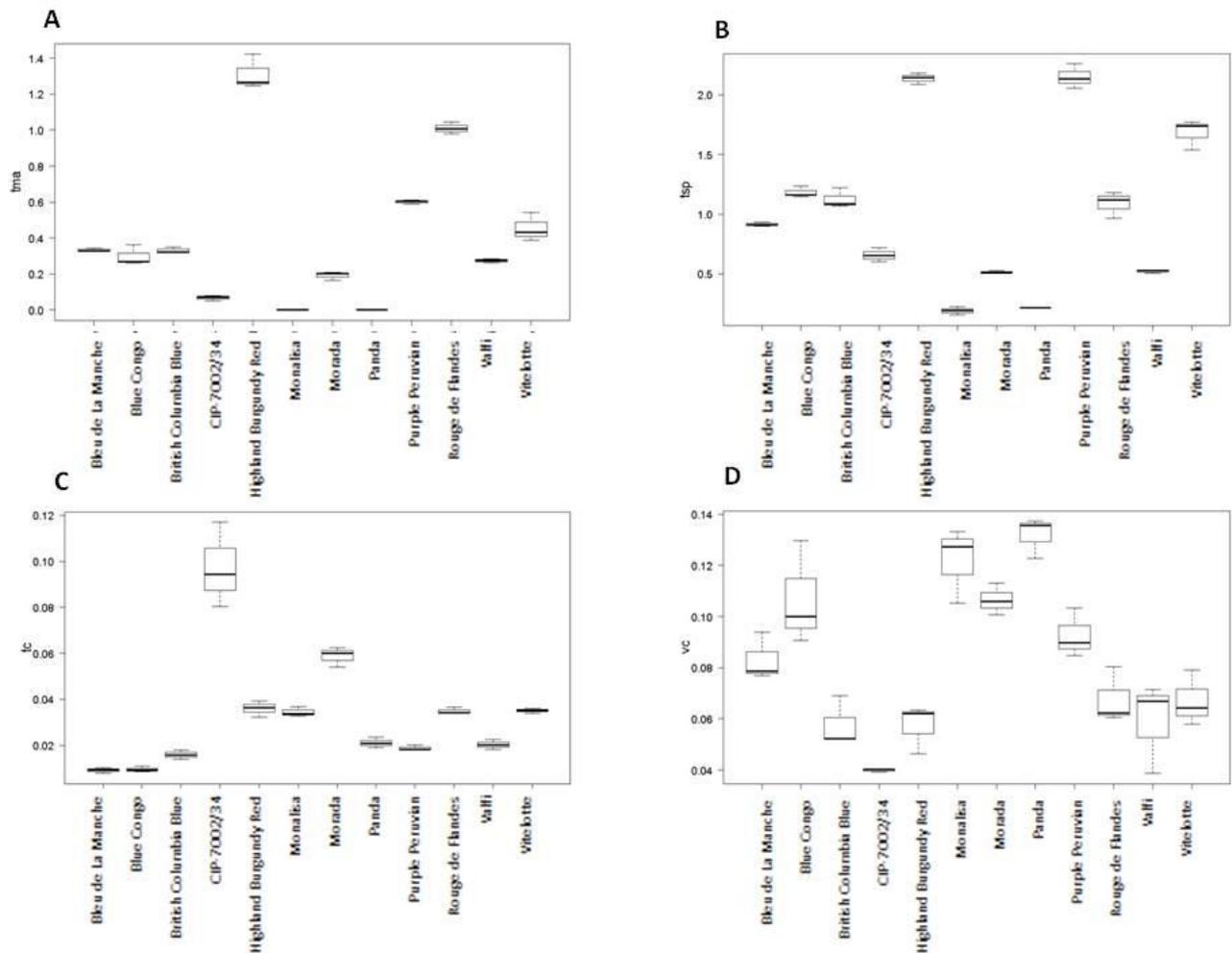


Figure 1. Boxplots indicating the distribution of the following phytochemical composition related parameters: A) total soluble phenolics (TSP), B) total monomeric anthocyanins (TMA), C) total carotenoids (TC) and D) vitamin C (VC) in peeled tubers of the collection of ten highly-pigmented cultivars. Means and standard deviations of: TSP are expressed as g gallic acid equivalents per kg fresh weight; TMA are expressed as g cyanidin 3-O-glucoside equivalents per kg fresh weight, TC are expressed as g lutein equivalents per kg fresh weight and VC are expressed as g vitamin C per kg fresh weight (n=3).

Micronutrient concentrations ranged from 0.015 ± 0.001 to 0.023 ± 0.003 for Zn 0.021 ± 0.005 to 0.051 ± 0.002 for Fe (Figure 2A and 2B). Moreover, most highly pigmented cultivars showed significantly higher Zn (cvs. Blue Congo, British Columbia Blue, Valfi and Highland Burgundy Red) and Fe (Vitelotte, CIP-7002/34, Highland Burgundy Red, Morada and Rouge de Flandes) concentrations. An average edible portion of Vitelotte contains

more than 20% of the recommended daily intake of Fe and also about 10% of the dietary reference intake of Zn (Institute of Medicine: Food and Nutrition Board, 2001). Results obtained in this study, regarding tuber morphology, pest resistance and nutritional quality of a small set of highly-pigmented *Solanum tuberosum* L. cultivars revealed the existence, among them, of interesting traits: such as resistances to *Globodera*

spp. and *Phytophthora infestans*; as well as significantly higher concentrations of phenolic compounds, carotenoids, vitamin C, zinc and iron. These findings could be useful to assist in parental line selection for

breeding with the aim of obtaining health-promoting advanced clones with lower incidence of pests and diseases.

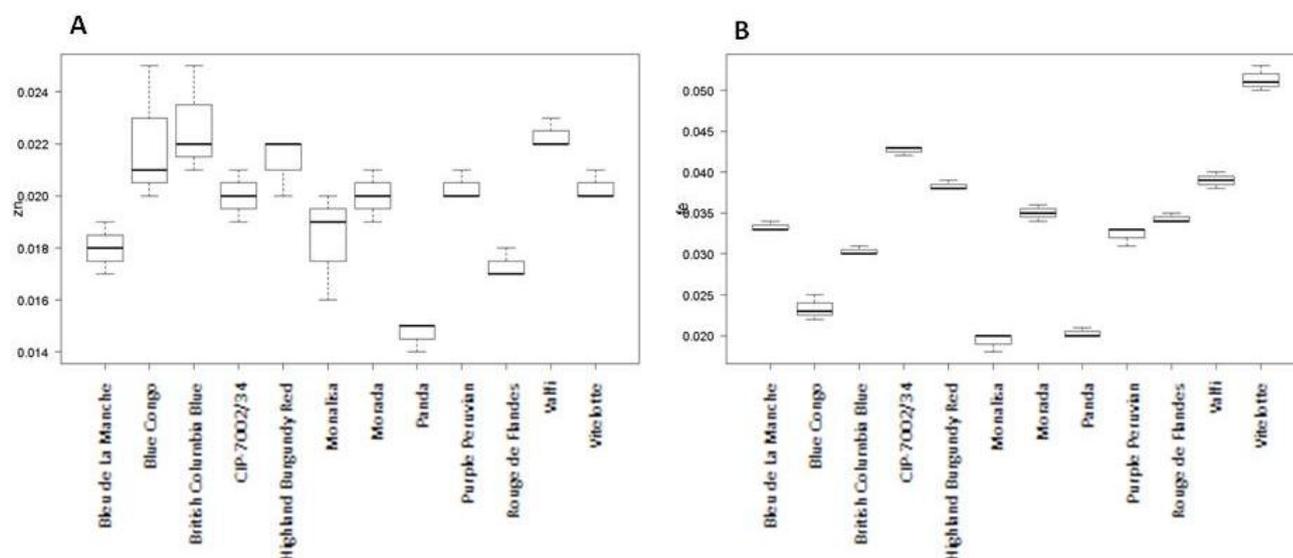


Figure 2. Boxplots indicating the distribution of the following mineral composition related parameters: A) zinc content (Zn) and B) iron content (Fe). Means and standard deviations of Zn and Fe are expressed as g per kg fresh weight (n=3).

Conflict of interests

The authors declare there is no conflict of interest in the publication of the results of this research.

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