

Characterization of carotenoids, proximal analysis, phenolic compounds, anthocyanidins and antioxidant capacity of an underutilized tuber (*Tropaeolum tuberosum*) from Bolivia

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Abstract

The tubers of *Tropaeolum tuberosum*, locally known as Isaño and native to the Andean region of South America, have been known since ancient times for their multiple uses in the Bolivian population. They are used both as food in various preparations and in traditional medicine. In this investigation, we report the study of three Isaño cultivars currently consumed in Bolivia. We determined their proximal composition, characterized carotenoids, determined antioxidant capacity, measured total phenols and total flavonoids, and quantified the major polyphenols. The results show that, apart from being a source of important nutrients such as proteins, Bolivian Isaño is a source of antioxidants and has a higher concentration of flavonoids and anthocyanidins, particularly in the purple cultivar. Additionally, we identified the presence of three carotenoids in this food for the first time: Lutein, Neoxanthin, and β -carotene. Through these types of studies, we aim to revalue this food, which is little known both in Bolivia and outside the Andean region of South America. Considering its nutritional properties, we seek to increase its consumption.

1. Introduction

Tropaeolum tuberosum, better known in the Bolivian highlands as “Isaño”, also has other vernacular names dependent on the region as isañu, mashua, mashwa, mishwa, añu among others; it is a species used and distributed in various high Andean communities of the Andes in South America [1]. In traditional medicine, it is used for the treatment of venereal, pulmonary and skin diseases, anemia, reduction of sexual appetite, wound healing, analgesic, anti-inflammatory for kidney, prostate and bladder pain [2, 3] and it also has diuretic effects [4]. These biological activities could be closely related to the presence of its secondary metabolites, such as hydroxybenzoic acids, phenolic compounds, tannins, flavonols, anthocyanins, glucosinolates, isothiocyanates, phytosterols, carotenoids, fatty acids among others [2, 5, 6].

The crop of Isaño grows from a range of 2400 to 4300 meters above sea level (m.a.s.l.) [7] and currently is considered an underutilized crop, there is a clear risk of loss as a crop due to factors such as crop change, other species with higher economic profitability, lack of public knowledge, climate change, it is poorly researched, marketed, managed and the fact that its flavor is strong (spicy-sour) making it less palatable than other tubers [8]. They are commonly confused with tubers of oca (*Oxalis tuberosa*) another crop from the Andean region, since the Isaño has similar colors ranging from gray, white, yellow, red, purple and black but easily distinguishable mainly by its conical shape [1, 7]. The strong flavor of Isaño usually improves after exposure to solar radiation where it acquires a more pleasant taste to the palate; the popular forms of Isaño consumption are as “huatia” (cooking underground) and boiled [3], but the most accepted form by the Bolivian population is frozen and it is known by the trade Aymara name “Thayacha” which is a type of Andean ice cream, marketed in winter [3, 9].

In the present investigation we report the chemical study of three Isaño cultivars (the most consumed by the population of the Bolivian altiplano). currently consumed in Bolivia, determining their proximal

composition, the characterization and identification of carotenoids, determination of antioxidant capacity, total phenols and total flavonoids, in addition to the quantification of the majority polyphenols.

2. Experimental

2.1. chemicals

The reagents used were: Folin-Ciocalteu (Sigma-Aldrich, St. Louis, USA.), gallic acid (Sigma life Science, China), sodium carbonate (Biopack, Buenos Aires, Argentina), ABTS (2,2-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid 98%) Sigma Aldrich, Canada; catechin gallate, potassium persulfate 99, 0% (Sigma-Aldrich, Japan), Trolox (6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid, 97%) Sigma Aldrich, Denmark, TPTZ (2,4,6-tripyridyl-s-triazine) Sigma Aldrich, Switzerland; ferric chloride (Biopack, Argentina), acetic acid (glacial p. a.) Merck KGaA, Darmstadt, Germany; sodium acetate (Sigma Aldrich, India), hydrochloric acid (Sigma-Aldrich, St. Louis, USA), sodium acetate (Sigma Aldrich, India), hydrochloric acid (Sigma-Aldrich, St. Louis, USA), absolute methanol from J.T. Baker (Mexico DF, Mexico), ethanol from Merck (Darmstadt, Germany), acetone from Biopack, Argentina; aluminum chloride hexahydrate (Sigma-Aldrich, Germany), sodium nitrite (Scharlab S.L., Spain), Ethyl acetate suitable for HPLC, $\geq 99.7\%$, petroleum ether p.a., sodium hydroxide (Merck KGaA, Darmstadt, Germany) β -Carotene 97% (Sigma-Aldrich, Switzerland), BHT (2,6-Di-tert-butyl-4-methylphenol) from Acros Organics, New Jersey, USA; delphinidin, cyanidin, pelargonidin obtained from Extrasynthèse (Genay, France), finally, The mili-Q ultrapure water obtained from a Millipore simplicity 185, Brazil.

2.2 Samples

The samples (Fig. 1) were obtained in June 2022 from Apuvillque community (Municipality of Huarina, Omasuyos province), La Paz department, Bolivia with coordinates 16°08'02.9"S 68°38'32.8"W. Each Isaño sample has different traditional names according to its shape and color where sample **1** is known as "Santo jonk'ori", sample **2** as "Achakani Isaño" and sample **3** as "Ch'iyaara Isaño".

2.2 Proximate analysis

All fresh samples were analyzed five times for moisture percentage using a moisture analyzer (Radwag, MAC 110/WH, Poland). For pH the method recommended by AOAC 943.02 [10] was using a pH meter (pH-mV-Temp 305225, ISOLAB, Germany), Total solids were analyzed with a hand refractometer (RF.5532, type 0–32% ATC, The Netherlands) described in [11]. For ash determination a microprocessed muffle furnace (QUIMIS Q318M21 Diadema-SP-, Brazil) was used using the AOAC method 923.03 [10]. For total proteins the AOAC method 960.52A [10], was adapted to the characteristics of the equipment Compact digestion system (RAYPA MBCM-40, R. ESPINAR, S.L. Spain) and Micro Kjeldahl (RAYPA Distillation unit DNP, R. ESPINAR, S.L. Spain) and finally for the total percentage of fats the soxhlet liquid solid extraction method was used. The total carbohydrate content was determined by 100 - (moisture + protein + fat + ash) % difference.

2.3 Extraction

Approximately 100 mg of sample were weighed to which 1 mL of methanol 90% was added. The samples were sonicated at 0°C for 15 min. They were then centrifuged at 12000 rpm for 2 min. The extraction was used for the determination of antioxidants, phenols and total flavonoids, the supernatant was stored at 4° C before analysis.

2.4 Determination for ABTS

Through the mixture of ABTS (2,2'-azinobis(3-ethylbenzothiazoline-6-sulfonic acid)) 7 mM (colorless) with potassium persulfate 140 mM (24 h of rest, minimally prior to the analysis), the oxidized product ABTS⁺ (green color) is obtained, this radical cation in solution was diluted in acetic acid-sodium acetate buffer (pH = 5) until obtaining an absorbance of approximately 0.700 ± 0.02 at 734 nm. Where 100 μ L of sample or standard was mixed with 1 mL of ABTS⁺ solution with 6 min rest for stabilization, then the measurement at 734 nm was performed in a multichannel reader (Biotek uQuant, USA). The results were fitted to a Trolox standard curve (20–200 μ mol/L) [12].

2.5 Determination for FRAP

FRAP (Ferric reducing antioxidant power) solution is formed through the ferric complex from the mixture of acetic acid-sodium acetate buffer solution (pH = 3.6) with 10 mM TPTZ and 20 mM iron chloride. 30 μ L of sample, 120 μ L of distilled water and 900 μ L of FRAP solution were used. The absorbance was measured at 593 nm. The final absorbance of the samples was adjusted with a Trolox standard curve (100–1000 μ mol/L) [12].

2.6 Total phenol content (TPH)

50 μ L of sample were used with 1 mL of Folin-Ciocalteu reagent solution diluted to 10% and 500 μ L of 7.5% sodium carbonate. They were shaken and incubated for 30 min at 45°C to stabilize the reduction reaction and the formation of the blue complex. Subsequently, they were cooled to room temperature to measure their absorbance at 765 nm. The readings of the samples were adjusted with a standard curve of gallic acid (230–1170 μ mol/L) [12].

2.7 Total flavonoids content (TF)

The method described in [12] slightly modified was used where 150 μ L of sample, 45 μ L of 10% sodium nitrite and 45 μ L of 20% aluminum chloride hexahydrate were used and left to stand for 10 min for the formation of the pink colored flavonoid-aluminum complex, then 300 μ L of 1 M sodium hydroxide and 600 μ L of water were added. The absorbance readings were taken at 510 nm, the results fit a standard curve for catechin (69–689 μ mol/L).

2.8 Total carotenoids content (CT)

For the extraction we slightly modified the method described by [13]. Where from 0.5 g of sample we added 5 mL of ethanol-acetone mixture (1:1 v/v) containing 200 mg/L of BHT, the mixture was shaken in

a vortex for 30 s, and centrifuged at 12000 rpm for 5 min at 4 °C, the procedure was repeated until the disappearance of color in the extraction solvent. The supernatants were read at an absorbance of 450 nm; the concentration of total carotenoids was calculated from a standard curve of β -carotene in a range of 5 to 50 $\mu\text{g/mL}$.

2.9 Quantification by HPLC-DAD

Polyphenols were analyzed by HPLC; for hydrolysis of glycosides, extracts were subjected to reflux (90°C) where 300 μL of sample extract were mixed with 300 μL 3.0 M HCl and 300 μL methanol for 75 min. Phenolic compounds were separated using an Agilent liquid chromatography system (1100 Series, USA), consisting of a vacuum degasser (G1322A), a solvent supply module (Quat Pump-G1311A), a column oven (ColCom-G1316A). The reverse phase column used was a 5 μm Agilent Eclipse Plus C18 (150 x 4.6 mm) protected by a 10 mm pre-column. The injection volume was 20 μL with a flow rate of 0.8 mL/min. The mobile phase was a binary solvent system consisting of A (1% acetic acid in water) and B (methanol) using a gradient program of 0 min:40% B; 5 min:65% B; 10 min: 90% B; 15 min: 40% until 17 min. The UV-VIS absorbance of the eluate was recorded with a diode array detector (G1315B) (Agilent, Santa Clara, CA) at 280, 360 and 530 nm. Compounds were identified by comparing them with the standards of each compound identified using the retention time, the profile of the absorbance spectrum and also by analyzing the samples by comparing them with the pure standards.

2.10 Characterization of carotenoids by HPLC-MS

Prior the analysis, the Isaño tuber was cleaned, dried and ground, it was treated with petroleum ether, the ethereal dry extract was subjected to column chromatography on silica gel, eluting initially with petroleum ether and later with mixtures of different concentrations of petroleum ether. petroleum and ethyl acetate of increasing polarity gradient, the fraction eluted with petroleum ether-ethyl acetate (5:1 v/v) was the one that showed the presence of reddish colored substances.

HPLC-MS analysis was carried out on a Purospher® RP-18 column (5 μm) in a Waters 2695 separation module, with a mobile phase composed of 0.1% formic acid in water as solvent A and methanol as solvent B. Flow rate: 1 mL min⁻¹. The analyte was separated by isocratic elution using A/B = 35/65 (v/v) in 15 min. The compound was detected with a Waters Quattro ESI mass spectrometer operated in positive mode, model 4micro (Milford Massachusetts, USA).

2.11 Statistic analysis

All data are reported as the average of five analyses and its standard deviation. To determine significant differences in the data, they were subjected to analysis of variance (ANOVA). Differences between means were compared using the Tukey test at 5% with IBM SPSS Statistics 22.0, USA. Principal component analysis (PCA) and Pearson's correlation were carried out using as performed using the statistical language R version 4.2.3 (R Foundation for Statistical Computing, Vienna, Austria).

3. Results

3.1 Proximate analysis

According to Table 1, sample 2 has the highest moisture content and sample 3 has the lowest, but there are no significant differences. The soluble solids in the initial material are between 8.4–8.9 and when soaked for seven days, they increase a third part of their initial state.

Table 1
Comparison of the proximal analysis in fresh *T. tuberosum* cultivars.

Sample			
Parameter	1	2	3
Moisture (%)	81,5 ± 2,7	83,0 ± 0,5	80,3 ± 1,4
Soluble solids (°Brix)	8,9 ± 0,3	8,4 ± 0,6	8,7 ± 0,8
Soluble solids (°Brix)*	11,9 ± 0,4	11,6 ± 0,5	12,2 ± 0,6
pH	6,2 ± 0,5	6,3 ± 0,3	6,0 ± 0,1
Ash (%)	0,77 ± 0,05	0,78 ± 0,07	0,89 ± 0,06
Protein (%)	2,1 ± 0,7	2,3 ± 0,6	1,5 ± 0,7
Lipid material (%)	0,30 ± 0,05	0,25 ± 0,02	0,21 ± 0,03
Carbohydrate (%)	15,3	13,7	17,1
Energy (kCal/100g)	72,3	66,3	76,3
* Samples sunned 5h/day for seven days			

It was found that the pH values maintained a similar value among the three varieties, in addition to the percentage of ashes, which had values from 0.77–0.89% in the different varieties, with no significant differences. The protein percentage is between 1.5–2.3% and finally the fatty material in all samples is below 0.3%. While the percentage of carbohydrate was obtained from the difference of 100%, which is between 13.7–17.1%.

3.2 Carotenoids

Although previous studies show the quantification of carotenoids, these do not report which carotenoids are present in *T. tuberosum*, therefore, this study will be the first to attempt to characterize carotenoids in Isaño samples by HPLC-MS. The major carotenoids are described in Table 2 and, such compounds are supported by literature [14]. For the first time, the characterization was performed by comparing the UV-VIS max value, molecular mass and fragments where: Lutein shows a molecular weight of 569 (M + H) with daughter fragments of 551 (M + H-H₂O), 430 (M + H-139).

Table 2
Carotenoids found in *T. tuberosum*.

Nº	t _R (min)	Compound	Molecular formula	UV MAX (nm)	[M + H] ⁺	Fragment
1	10.54	Lutein	C ₄₀ H ₅₆ O ₂	421; 443; 470	569	551; 430
2	11.77	Neoxanthin	C ₄₀ H ₅₆ O ₄	416; 442; 463	601	583; 565; 547; 509
3	14.67	β-carotene	C ₄₀ H ₅₆	329; 423; 446; 475	537	413; 400; 269

Neoxanthin with a molecular weight of 601 plus fragments of 583(M + H-H₂O), 565(M + H-2H₂O), 547 (M + H-3H₂O), 509 (M + H-H₂O-92) and β-carotene with a molecular weight [M + H] of 537 and daughter fragments 413, 400, 269. According to their structures it can be seen that the presence of xanthophyll type carotenoids (except β-carotene), characteristic for having oxygen in their structures, was identified.

3.3 Antioxidant capacity and polyphenols

Table 3 shows the results of the antioxidant capacity reported in trolox equivalent (TE), the results indicate that by the ABTS method it is between 2.9–11.6 µmol/g and by the FRAP method it is between 4.5–30.0 µmol/g of fresh matter (FM). In the case of total phenols, they indicate that they are between 2.0-11.7 µmol/g of fresh sample expressed in gallic acid equivalents (GAE), finally the analysis of total flavonoids expressed in catechin equivalents (CE) shows a range of 1.8–2.4 µmol/g of fresh sample, it can be seen that between samples 1 and 2 there is no significant difference, compared to 3 which evidently has greater antioxidant capacity in addition to higher concentration of phenolic compounds and flavonoids. Finally, the analysis of total carotenoids provides a range between 57.3.-144.0 µg β-carotene/g, it was observed that samples 1 and 2 have a higher concentration of total carotenoids, however, a lower concentration was obtained in sample 3, which reveals an inversely proportional relationship with its antioxidant capacity.

Table 3
Comparison of antioxidant activity (ABTS, FRAP), total phenols (TPH), total flavonoids (TF), total carotenoids (CT) in fresh matter.

Sample			
Determination	1	2	3
ABTS ($\mu\text{mol TE/g}$)	$2,9 \pm 0,4^a$	$1,8 \pm 0,2^a$	$13,2 \pm 2,9^b$
FRAP ($\mu\text{mol TE/g}$)	$7,1 \pm 0,6^a$	$4,5 \pm 0,5^b$	$30,0 \pm 1,0^c$
TPH ($\mu\text{mol GAE/g}$)	$2,6 \pm 0,2^a$	$2,0 \pm 0,3^a$	$11,7 \pm 1,6^b$
TF ($\mu\text{mol CE/g}$)	$1,8 \pm 0,1^a$	$1,8 \pm 0,2^a$	$2,4 \pm 0,3^b$
CT ($\mu\text{g } \beta\text{-caroteno/g}$)	$115,9 \pm 15,9^a$	$144,0 \pm 10,4^a$	$57,3 \pm 4,2^b$

Different superscript in row indicates significant difference $p < 0.05$, by ANOVA analysis and Tukey test.

Table 4 shows the main polyphenols found in the Isaño samples, where they present 31.95–40.11 $\mu\text{g/g}$ of gallic acid, with no significant difference between samples. 7.0–13.3 $\mu\text{g/g}$ of epicatechin where sample 2 presented lower concentration; 0.7–4.8 $\mu\text{g/g}$ of catechin gallate where sample 2 presented higher concentration. In addition, after the hydrolysis of the anthocyanins, it was possible to quantify the anthocyanidins delphinidin, cyanidin and pelargonidin (Fig. 2), where clearly sample 3 presents higher concentration of these polyphenols, which would be related to its high antioxidant capacity.

Table 4
Polyphenols measured by HPLC-DAD.

Sample [$\mu\text{g/g}$ (FM)]			
Compound	1	2	3
Gallic acid	$31,9 \pm 6,9^a$	$40,1 \pm 2,0^a$	$33,8 \pm 2,4^a$
Epicatechin	$13,3 \pm 5,4^{a,b}$	$7,0 \pm 1,5^b$	$13,3 \pm 0,8^a$
Catechin gallate	$1,1 \pm 0,3^a$	$4,8 \pm 1,3^b$	$0,7 \pm 0,4^a$
Delphinidin	$0,017 \pm 0,001^a$	$0,008 \pm 0,002^a$	$0,577 \pm 0,145^b$
Cyanidin	$0,0049 \pm 0,0002^a$	ND	$0,030 \pm 0,007^b$
Pelargonidin	$0,006 \pm 0,001^a$	$0,0008 \pm 0,0003^a$	$0,022 \pm 0,007^b$
Different superscript in row indicates significant difference $p < 0.05$, by ANOVA analysis and Tukey test. ND: not detected			

4. Discussions

4.1 Proximate analysis

The results of proximal analysis are shown in Table 1: The moisture content, in the samples of the present study showed values between 80.3 to 83.0%, which is in a similar range reported in previous studies [2, 15, 16]. The range of soluble solids in the samples (without sunlight) of this study are between 8.4 to 8.9 °Brix which are comparable to literature (6.1–8.5 °Brix) [15]; but this value can increase significantly when samples are exposed to solar radiation as can be seen in Table 1. These changes in sugar content was also observed in Oca (*Oxalis tuberosa*) previously [9].

The pH values were around 6, which are in agreement with previous studies [15], but it was observed that there are also cultivars that have more acid pH values (3.9–4.2) [16], this pH variation could be related to the post-harvest time showing that the cultivars of the present study were fresh. The values of ash determined in Isaño ranged from 0,77 to 0,89 and they are in accordance with the literature [2, 15], The ash content is related to the minerals present in the samples particularly phosphorus, calcium, magnesium, manganese, zinc, iron, sodium and potassium [2].

The protein values in Isaño were determined in several studies and in the present study a wide variability that could be linked to the type of study accession, sampling site, sampling method used among other parameters but the range is between 1.1 to 2.7% in FM [2, 15], while the values measured in this study is between 1.5 to 2.3%, showing higher protein content the cultivar 2. The fatty material in the Isaño is scarce as it is reported values less than or equal to 1% [2], which was replicated in this study when values below 0.3% were found. The content of carbohydrates measured in fresh Isaño range from 13,78 – 17,1 while in previous studies, a range of 9,8 to 12.8 were reported [2, 15].

4.2 Carotenoids

Carotenoids are the most extensive group of natural pigments, characterized by being indispensable cellular components in living beings [17], responsible for the attractive colorations in various foods [18]. These compounds are classified into two groups according to their structural characteristics: carotenes consisting of carbon and hydrogen (β -carotene, α -carotene and lycopene), and xanthophylls, consisting of carbon, hydrogen and additionally oxygen (lutein, zeaxanthin, astaxanthin and fucoxanthin) [19]. The importance of carotenoid intake in food is due to the fact that no member of the animal kingdom, including humans, is able to biosynthesize these compounds and they are closely associated with good health, due to the properties attributed to them such as the effect against oxidative stress [20], as well as preventing the onset of certain types of cancer and macular degradation, protecting against sunburn reactions and improving immune function [18].

The coloration of the different varieties of *T. tuberosum* ranging from yellow to purple and black, these were presumed to contain natural pigments such as anthocyanins and carotenoids. The presence of carotenoids in Isaño was confirmed by several spectrophotometric studies revealing carotenoid

concentration between 1 to 25 $\mu\text{g g}^{-1}$ [21], 2.6 to 96.7 $\mu\text{g g}^{-1}$ [22], 12.8 to 85.8 $\mu\text{g g}^{-1}$ [23] and 70 to 132 $\mu\text{g g}^{-1}$ [24] expressed in β -carotene equivalents. In this study it was found that the cultivars studied present similar concentrations since they are between 57.3 to 144.0 $\mu\text{g } \beta\text{-carotene/g}$. It should be taken into account that the concentration of carotenoids tends to vary according to the stage of maturity, variety and post-harvest storage, and it was also found that the darker the variety, the lower the concentration of carotenoids it contains [24]. Moreover, this study reports for the first time the presence of lutein, neoxanthin and β -carotene characterized by HPLC-MS/MS through the comparison of molecular weight, daughter fragments and UV-VIS spectra. As it was mentioned below, it is the first time that this molecules are identified in *Tropaeolum tuberosum* but not in the Tropaeolaceae family since they were identified previously in *Tropaeolum majus* [25].

4.3 Antioxidant capacity and polyphenols

Some of the compounds previously found in Isaño were caffeic acid, chlorogenic acid, gallic acid, hydroxycinnamic acid and hydroxybenzoic acid; the flavonoids rutin, quercetin, anthocyanins derived from delphinidin, cyanidin, pelargonidin, as well as other uncommon compounds such as glucosinolates, alkamides, phytosterols, among others [2, 26], which could contribute to the antioxidant activity in Isaño.

Previously, several studies of antioxidant capacity were reported [2, 16, 22] that could not be compared with the present study because the differences in sample preparation or because they are expressed in other units, as shown in Table 5. The results obtained in the present research are comparable with other Andean tubers such as Oca (*Oxalis tuberosa*), Ullucu (*Ullucus tuberosus*), Arracacha (*Arracacia xanthorrhiza*) and colored potatoes (*Solanum tuberosum*), while the purple Isaño (sample 3) has a higher antioxidant capacity than all the tubers in Table 5, except for the concentration of total phenols and flavonoids, which is lower than that of the colored potatoes. This can be explained due to the presence of anthocyanins.

From literature a previous study presented the quantification of polyphenols in Isaño from different cultivars. The study shows the presence of caffeic acid, rutin, chlorogenic acid and quercetin [27]. In the present study (Table 4), it was quantified gallic acid, epicatechin and catechin gallate also present in other tubers [28, 29]. Isaño. After hydrolysis of the extract, the main anthocyanidins malvidin, cyanidin and pelargonidin were quantified for the first time in Isaño. these anthocyanidins are also present in various fruits, berries, tubers, vegetables, wines and flowers, and they are responsible for the red to blue colour [30, 31, 32].

Table 5
Comparison of antioxidant capacity among andean tubers.

Sample	ABTS ($\mu\text{mol TE/g}$)	FRAP ($\mu\text{mol TE/g}$)	TPH ($\mu\text{mol GAE/g}$)	TF ($\mu\text{mol CE/g}$)	Reference
Isaño	1,8–13,2	4,5–30,0	2,0–11,7	1,8–2,4	This study
	1,1–45,7	-	4,6–22,0	0,1–2,7	[26, 27]
Oca	1,8–14,7	3,8–7,6	2,8–6,4	0,4–1,9	[32–34]
Olluco	2,6	2,7	1,7–6,4	0,4–2,0	[33, 34]
Arracacha	2,0	2,4	-	-	[33]
Colored potatoes	0,2–7,0	1,0–10,0	8,0–17,0	2,0–19,0	[31, 35]

The methods presented a high correlation between FRAP and ABTS with the content of phenols and total flavonoids, it was also found a positive correlation of the ABTS, FRAP, TPH and TF with malvidin, cyanidin and pelargonidin, while all of these methods showed negative correlations with the concentration of total carotenoids. These results could indicate that there is a trend related to color where darker (purple) cultivars have higher antioxidant capacity and anthocyanins but low carotenoid concentrations and conversely for lighter (yellow) cultivars. All the correlations are presented in Fig. 3.

While the PCA reveals that the three samples are classified into different clusters according to the parameters analyzed, as shown in the Fig. 4. PC1 and PC2 together explained more than 95% of the total variance. It could also be confirmed that sample 3 has the highest antioxidant capacity due to its high content of anthocyanidins, while sample 1 and 2 showed a high content of carotenoids and proteins.

5. Conclusions

This study reports for the first time the presence of the carotenoids lutein, neoxanthin and β -carotene in samples of Isaño from Bolivia, determined by HPLC-MS/MS through the comparison of molecular weight, daughter fragments and UV-VIS spectra. In addition, the antioxidant capacity and quantification of polyphenols were study, showing higher values in comparison with other tubers in particular sample 3 (purple cultivar).

Proximal analysis was carried out among the varieties, and it was possible to find high protein and carbohydrates content in Isaños. It was also seen that the results of the antioxidant capacity have statistically significant correlation with the anthocyanins content and an inverse correlation with the carotenoids content; these results can indicate that the intensity of purple color in the tubers will have higher antioxidant capacity but lower concentration of carotenoids.

The locally consumed varieties in Bolivia appear to be a source of important active compounds for the nutrition of the Andean population. These findings contribute, from both a chemical and nutritional

perspective, to the revaluation of this food, which is significant for local populations as well as the general public. Through these kinds of studies, it is intended to revalue this food, little known both in Bolivia and outside the South American region, and considering its nutritional properties, increase its consumption.

Declarations

Author contributions

GC conceptualized, designed the research and writing the original draft. DMF prepared and analysed the samples, designed the methodology. MB analysed the samples, and interpreted the data. MP sourced the funding, supervised the study, and reviewed and edited the final manuscript draft.

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Consent for publication

Not applicable.

Data availability

All data generated or analysed during this study are included in this published article

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Competing interests

No potential conflicts of interest were reported by the authors.

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Figures



Figure 1

Cultivars of *T. tuberosum* tubers studied.

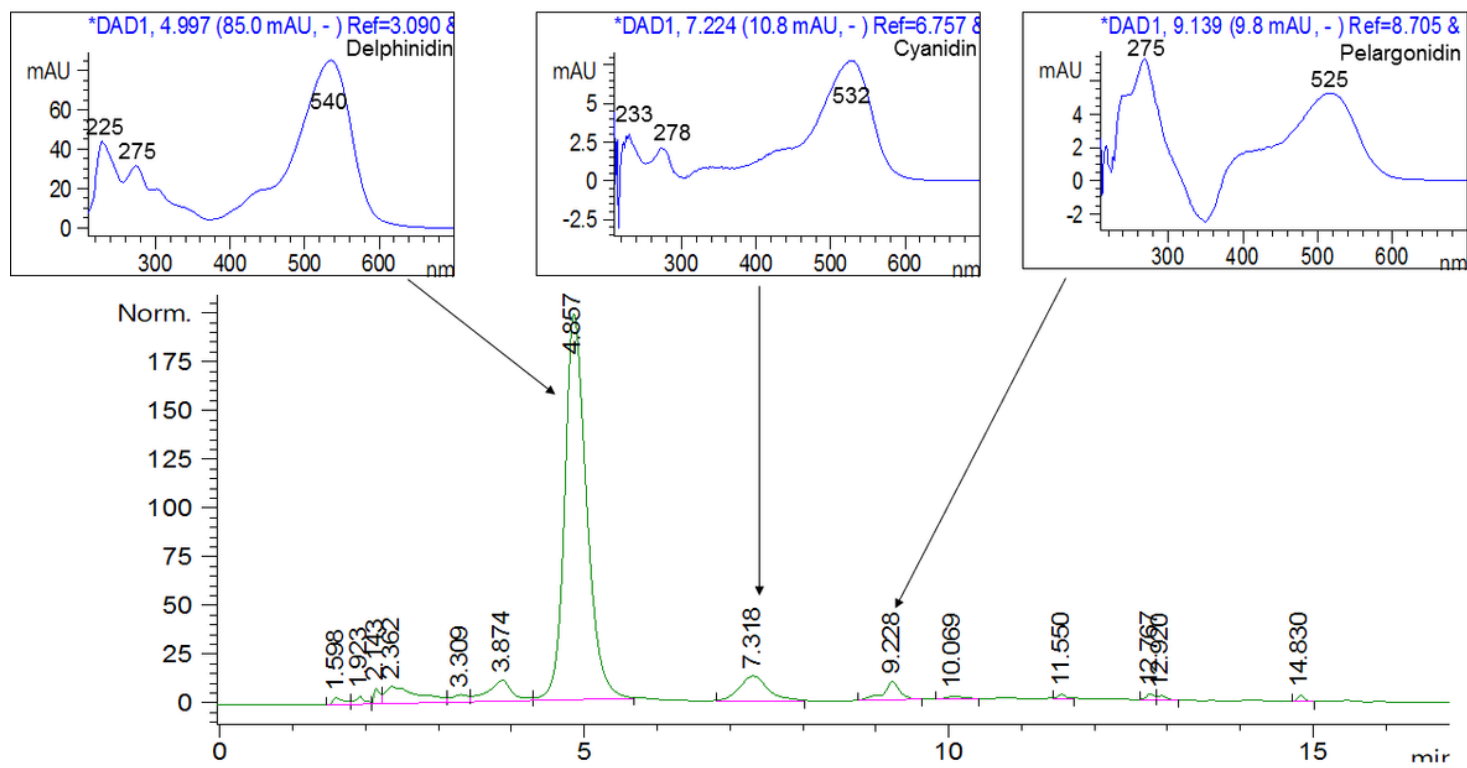


Figure 2

Chromatogram of the separation of anthocyanidins in sample 3, at 530 nm and their respective UV-Vis.

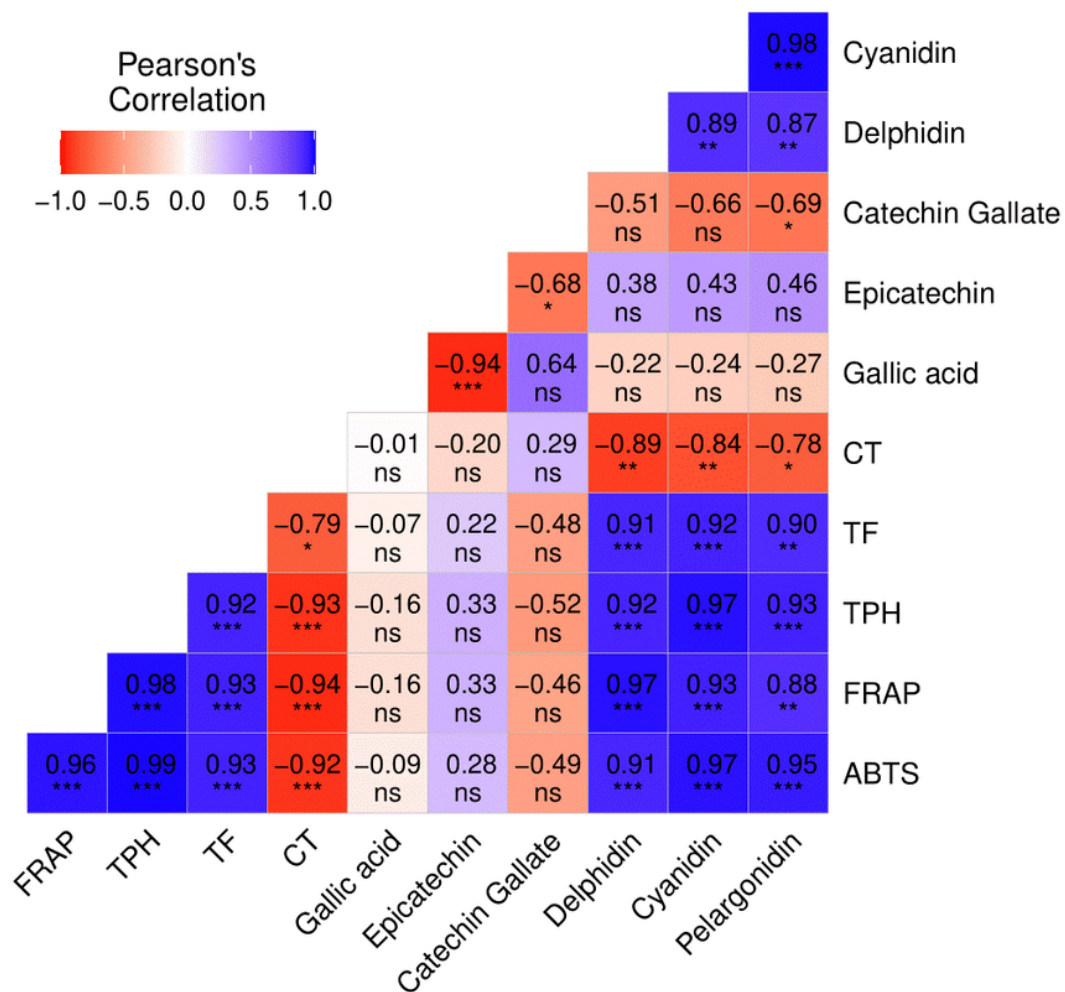


Figure 3

Statistic correlations among different measurements.

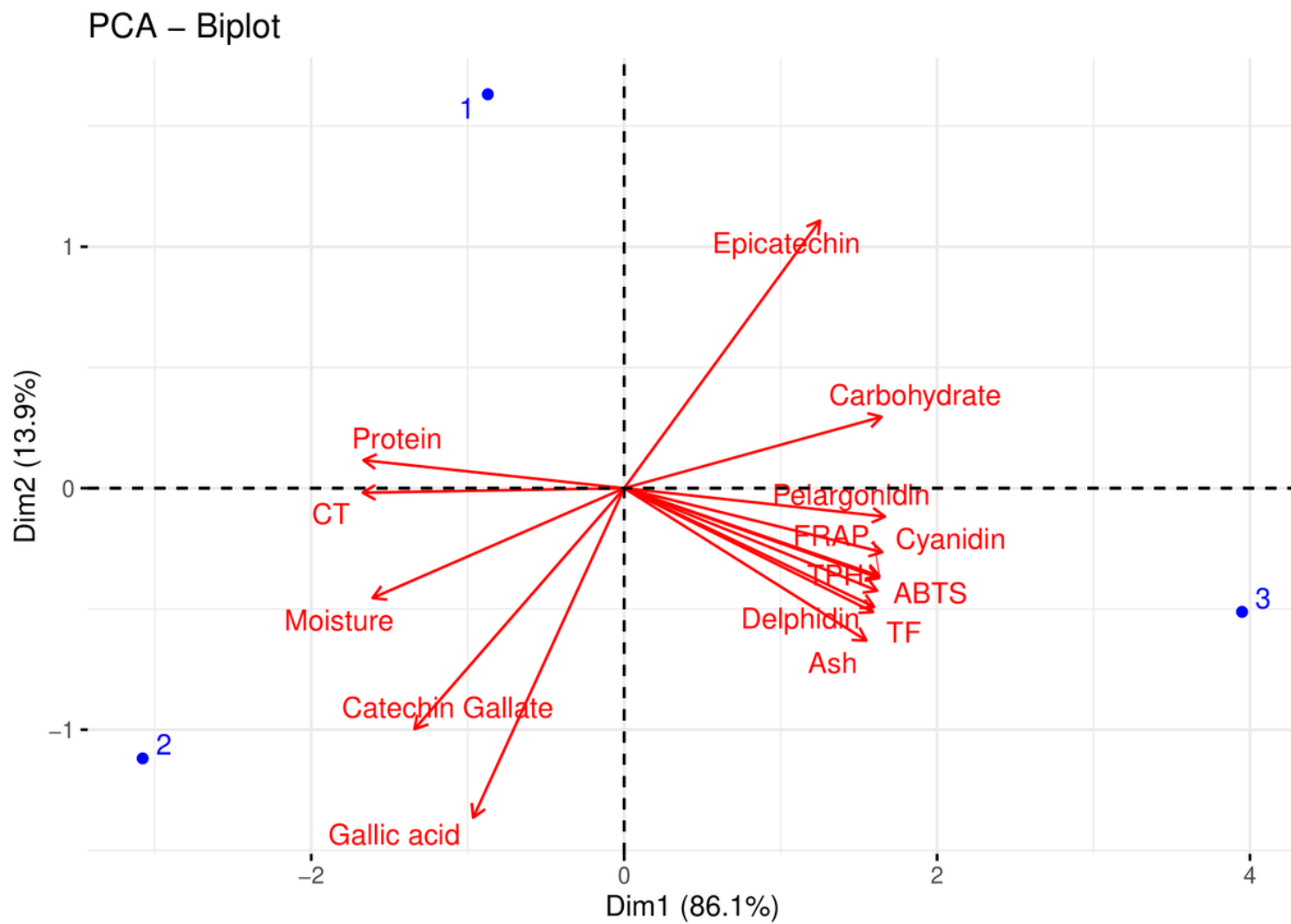


Figure 4

PCA biplot showing the samples and study variables.