Technical Report: Iron, Vitamin C, Phenolic and Phytic acid Concentration of Biofortified Potatoes as Affected by Location

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ABSTRACT

The effect of growing location on concentrations of iron, zinc, vitamin C, phenolics, phytic acid and glycoalkaloids of six iron biofortified potato clones and two potato varieties was determined. Significant variation due to location and genotype x location interaction was found for all the parameters evaluated.

The mean iron and zinc concentration was higher in localities with acidic soils and high organic matter content. However, high organic matter needs to be controlled, as it also favors glycoalkaloid production. Across the four locations, the iron biofortified potatoes showed from 23 to 54% more iron than the varieties Peruanita and Amarilla.

The mean vitamin C (a promoter of iron absorption) concentration was higher in the localities with lower total nitrogen and lower levels of Mg and K. Regarding the inhibitors of iron absorption, the mean phenolic concentration was lower in the locality with lower levels of calcium while the mean phytic acid concentration was lower in the locality with high levels of Al cation in the soil.

Across locations, the yellow fleshed biofortified potatoes have a significant amount of vitamin C and very low levels of phytic acid and phenolic compounds which suggest its strong potential to contribute to reduce anemia.

Keywords: biofortified potatoes, iron, zinc, vitamin C, phenolics, phytic acid, glycoalkaloids, location effect, anemia, highlands

1. INTRODUCTION

In the highlands of Peru, potato is a staple crop and is consumed in high amounts, with women consuming on average 800 grams of potato per day (De Haan et al. 2019). In those areas where there is little access to animal-source foods and the levels of anemia and malnutrition are high, potatoes are an important source of iron and zinc in the diet (Graham et al. 2007).

A screening of potato germplasm at the International Potato Center (CIP) found concentrations of 11 - 30 mg/kg dry weight (DW) of Fe, and 8 - 25 mg/kg DW of Zn in landraces (Burgos et al. 2007). These levels are lower than the iron and zinc concentration of legumes and cereals. However, it is thought that the bioavailability of iron in potato can be higher than that in cereals and legumes due to the presence of high levels of ascorbic acid - which facilitates iron absorption in the human body - and low levels of phytic acid, an inhibitor of iron absorption. CIP and its partners have recently shown that the bioaccessibility of iron in potato is high compared to that in other basic crops such as wheat and beans (Andre et al. 2015). Improving the iron and zinc concentration in potato can have a significant impact on reducing malnutrition and quality of life in areas where potato consumption is high, and where anemia and/or stunt growth are still pervasive. Within that context, and in order to contribute to reducing malnutrition, CIP scientists have developed biofortified potatoes with higher levels of iron than current varieties grown in the target countries. The first set of biofortified potatoes are diploid Andean type potatoes. These potatoes have similar yield and appearance to the local varieties consumed in the Peruvian highlands. They have been evaluated by participatory varietal selection in different locations in Huancavelica Peru and five clones have already been selected with strong potential to be delivered as varieties in the next two years.

It is well established that environmental and management factors can influence plant-gene expression and thereby the amount of a micronutrient accumulated in a seed or storage organ (Bouis and Welch, 2010), hence the evaluation of nutrients in contrasting environments or locations is important to guide strategies for improving iron and zinc concentration in potatoes.

The objective of this study was to evaluate the iron and zinc concentration of iron biofortified diploid potatoes grown in four locations of Huancavelica in comparison with two local varieties. The concentration of vitamin C, total phenolic acid, phytic acid and total glycoalkaloids was also evaluated. Vitamin C is considered a promoter of iron absorption, while phenolics and phytic acid are considered inhibitors of iron absorption as they chelate iron during digestion and reduce its absorption (Andre et al. 2014; Magallanes et al. 2017). In addition, glycoalkaloids were analyzed because they can be toxic for humans when present in high concentrations and can impart a bitter taste to potatoes.

2. MATERIALS AND METHODS

2.1 Plant material

Six iron biofortified potato clones from the potato breeding program at the International Potato Center (CIP) and two potato varieties were grown in four locations in the Yauli district in the department of Huancavelica in Peru: i) Tacsana (3762 m.a.s.l., 13º53’24’S 74º48’56’’W), ii)

Castillapata (3908 m.a.s.l., 12º44’12’S 74º49’23’’W), iii) Paltamachay (3945 m.a.s.l., 12º43’04’S 74º40’48’’W) and iv) Yanamachay (4130 m.a.s.l., 13º36’08’S 75º02’00’’W). The plantings were carried out in November 2016 and harvests were carried out between May and June 2017. They were cultivated and harvested using traditional farming practices. The field experiment was conducted using a randomized complete block design with three replications and ten plants by plot for each replication. The soil characteristics of each location are showed in Table 1.

2.2 Sample preparation

We followed the sampling and sample preparation procedures of Porras et al. (2014). Harvested tubers were processed as follows: raw tubers were washed thoroughly with tap water to remove any soil residue, rinsed with deionized, distilled water, and patted dry with paper towels. Tubers were peeled and then cut longitudinally from stem to bud end into four sections. Two or three slices were taken of two opposite sections of each tuber to obtain a 50 g sample, which was frozen, lyophilized, milled with 0.425 mm grid (40 mesh) to ensure particle sizes were similar, and stored in polypropylene bags before mineral and phytic acid analysis. While for phenolics and glycoalkaloids analysis the sample preparation was the same procedure, but the tubers were with peel.

2.3 Mineral analysis

All freeze dried and milled potato samples were analyzed by ICP-MS at the School of Biosciences in the University of Nottingham, UK. Briefly, the instrument was run employing three operational modes: (i) a collision-cell (Q cell) using He with kinetic energy discrimination (He-cell) to remove polyatomic interferences, (ii) standard mode (STD) in which the collision cell is evacuated, and (iii) hydrogen mode (H2-cell) in which H_2 gas is used as the cell gas. The samples were introduced from an autosampler (Cetac ASX-520) incorporating an ASXpress™ rapid uptake module through a perfluoroalkoxy (PFA) Microflow PFA-ST nebulizer (Thermo Fisher Scientific, Bremen, Germany). Internal standards were introduced to the sample stream on a separate line via the ASXpress unit and included Ge (10 µg L^{-1}), Rh (10 µg L^{-1}) and Ir (5 µg L^{-1}) in 2% trace analysis grade (Fisher Scientific, UK) HNO_3.

2.4 Vitamin C analysis

Tubers with peel intact were cut longitudinally into four sections. One or two slices of two opposite sections of each tuber were used to prepare each of three laboratory samples. The slices were cut and mixed and a 7.5 g laboratory sample was taken and placed in an extraction tube and analyzed in CIP’s Quality and Nutrition Laboratory (QNLAB) as described in Burgos et al., 2014. Briefly, the 7.5 g laboratory sample was extracted with an oxalic acid and acetone solution (0.4 and 20%, respectively) and homogenized in an Ultra Turrax for 1 min at 12000 rpm. The extract was filtered under vacuum through filter paper Whatman 2 and brought to 50 ml with the same extracting solution. One milliliter of the extract was reacted with 9 ml of 2,6-dichloroindophenol (1.6%) for 1 min and read at 520 nm on a spectrophotometer 160UV (Shimadzu, Japan). The vitamin C concentration was quantified through comparison with a standard curve of L-AA (Merck, Germany) and were expressed in mg/100g, FW.

Table 1. Soil characteristics of the four locations

Soil components	Tacsana	Castillapata	Paltamachay	Yanamachay
Cu available (ppm)	0.67	1.23	1.03	0.64
Zn available (ppm)	0.91	0.77	0.99	0.29
Mn available (ppm)	34.01	52.92	81.96	20.1
Fe available (ppm)	69.42	42.42	112.8	50.46
B available (ppm)	0.24	0.44	0.24	0.12
Soil type	loam sandy clay	loam	sandy	loam
Clay (%)	25.08	17.08	47.08	23.08
Silt (%)	19.71	39.78	30.78	37.86
Sand (%)	55.21	43.14	22.14	39.06
Organic matter (%)	1.55	8.21	4.93	7.38
P_2O_5 available (ppm)	8.62	12.98	28.44	38.61
K_2O available (ppm)	237.2	128.0	185.4	146.4
Total nitrogen (%)	0.09	0.48	0.29	0.43
Ca CO_3 (%)	< 0.01	< 0.01	< 0.01	< 0.01
Electrical conductivity (dS/m)	0.45	0.41	0.53	0.62
pH	5.77	6.17	5.15	4.67
Water saturation (%)	45.31	68.44	63.19	59.93
Exchangeable cations (meq/100g)				
Ca^{+2}	13.08	23.12	19.1	5.52
Mg^{+2}	3.99	0.95	2.77	0.86
K^{+}	0.60	0.31	0.47	0.35
Na^{+}	0.08	0.02	0.02	0.02
$Al^{+3} + H^{+}$	< 0.01	0.08	0.15	2.43
Effective cation exchange capacity	17.75	24.48	22.5	9.19
Interchangeable acidity (%)	< 0.06	0.31	0.68	26.47
Dissolved salts (meq/L)				
Chloride	1.72	1.82	2.66	2.29
Sulfate	0.80	0.49	0.49	0.29
Nitrate	0.09	1.32	0.95	2.14
Carbonate	< 0.02	< 0.02	< 0.02	< 0.02
Bicarbonate	2.25	0.23	0.39	0.31
Calcium	2.35	1.15	1.34	1.04
Magnesium	0.44	0.49	0.52	0.65
Sodium	1.77	1.80	2.26	2.94
Potassium	0.07	0.24	0.14	0.35
Boron (ppm)	0.09	0.08	0.09	0.03

2.5 Phenolic analysis

Total phenolics were analyzed at CIP's QNLAB according to the method reported by Waterhouse (2002). Briefly, 0.2- 1 g of the freeze dried and milled sample was weighed and extracted with 10 ml 80% methanol for 10 min, using sonication (Bransonic, CT, USA). Extraction was repeated with 10 ml of the same solvent, using in addition to sonication for 10 min, heating at 80 °C for 5 min. The methanolic extract including the phenolic compounds was filtered and adjusted to 25ml with 80% methanol. Four hundred ul of the methanolic extract was diluted in distilled water (1:20) and reacted with 500 ul of the Folin-Ciocalteu reagent (2N) for 6 min. Then 1500 ul of saturated sodium carbonate solution was added and reacted at 40 °C for 30 min. The absorbance of this solution was measured at 765 nm in a spectrophotometer UV-160A (Shimadzu Corp., Kyoto, Japan). The total phenolic concentration was calculated using a chlorogenic acid standard curve ranging from 100 to 1500 mg/mL.

2.6 Phytic acid analysis

All freeze dried and milled potato samples were analyzed using a Megazime kit at Quadram Institute Bioscience in Norwich, UK. Briefly, 1g sample was extracted with 0.66M hydrochloric acid, centrifuged and neutralized with 0.75M sodium hydroxide. For enzymatic reaction two subsamples were separated, one diluted with water and buffer to determine free phosphorous and the other diluted with water, buffer and phytase to determine total phosphorous. Samples were heated at 40°C for 10 minutes. Treatment was done twice. Further reaction was stopped in both free and total phosphorous samples by addition of trichloroacetic acid, then samples were centrifuged for 10 minutes. For determination 1 ml of free and total phosphorous samples were treated with colour reagent (5:1 mix of 10% ascorbic acid/1 M sulphuric acid and 5% ammonium molybdate). Standards of phosphorous solutions (0, 0.1, 0.5, 1.0, 1.5 μg/ml) were analysed with the samples, incubated at 40°C for 1 hour and then measured at 655nm in a spectrophotometer. Total phytic acid was calculated from the difference between total and free phosphorous for each sample after a conversion to give the phytic acid value.

2.7 Glycoalkaloid analysis

Extraction of glycoalkaloids was performed at CIP's QNLAB following the method reported in Ponnampalam & Mondy, 1983 with some modifications. Briefly, 2.5 g of freeze dried and milled sample was weighed, hydrated with 7mL of distilled water for 10 min and extracted with 60 mL of a Methanol: Chloroform (2/1) solution in an Ultra Turrax T25 D homogenizer (IKA, Staufen, Germany). The extract was concentrated using a rotary evaporator (IKA-Werke, Staufen Germany) at 65ºC and a refrigerant (Haake GH, Karlsruhe, Germany) at 16ºC. The concentrated extract was transferred to a solution of 2% acetic acid, brought to 20mL and cleaned with petroleum ether. Four ml of the aqueous extract was placed in a tube and 1.2 ml of ammonium hydroxide was added. The mixture was flocculated in a dry block (Techne, ST15 OSA, UK) at 85ºC for 10 min, refrigerated at 4ºC for 30 min and centrifuged in an Optima L-90K Ultracentrifuge (Beckman Coulter, Brea, California, USA) at 27000 rpm for 90 min. Five ml of orthophosphoric acid was added to the pellet and read at 408 nm on a spectrophotometer 160UV (Shimadzu, Japan).

2.8 Statistical analysis

The effect of location, and of genotype by location interaction, was analyzed using mixed models and considering the genotypes and locations as fixed effects. All statistical tests were performed using SAS/STAT (version 8.2) software.

3. RESULTS AND DISCUSSION

3.1 Effect on iron and zinc concentrations

The concentration of iron and zinc of the iron biofortified clones and the varieties grown in four locations, expressed on a dry weight (DW) and fresh weight (FW) basis, is shown in Table 2 and Table 3, respectively.

The iron concentration of the biofortified clones ranged from 14.19 to 22.49 mg / kg DW in Tacsana, from 18.48 to 27.13 mg / kg DW in Castillapata, from 17.61 to 34.47 mg / kg DW in Paltamachay and from 20.06 to 33.25 mg / kg DW in Yanamachay. The iron concentration in the local variety Amarilla ranged from 11.77 mg / kg DW in Tacsana to 16.78 mg / kg DW in Yanamachay and in the variety Peruanita from 10.38 mg / kg DW in Tacsana to 15.54 mg / kg DW in Yanamachay.

The zinc concentration of the biofortified clones ranged from 7.42 to 10.98 mg / kg DW in Tacsana, from 8.02 to 11.20 mg / kg DW in Castillapata, from 8.74 to 16.33 mg / kg DW in Paltamachay and from 9.07 to 18.54 mg / kg DW in Yanamachay. The zinc concentration in the variety Amarilla ranged from 6.86 mg / kg DW in Tacsana to 10.64 mg / kg DW in Yanamachay, while concentrations in the variety Peruanita ranged from 6.49 mg / kg DW in Tacsana to 11.47 mg / kg DW in Yanamachay.

The effect of location and of the clone x location interactions on iron and zinc concentration was significant ($P < 0.001$). The mean iron concentration in Yanamachay (21.78 mg / kg DW) was higher than in Castillapata and Paltamachay (19.34 and 19.97 mg / kg DW) and higher than in Tacsana (15.31 mg / kg DW). The mean zinc concentration in Yanamachay and Paltamachay (12.48 and 11.32 mg / kg DW, respectively) was higher than in Castillapata and Tacsana (9.64 and 8.18 mg / kg DW, respectively)

It has been reported that soil composition affects the mineral concentration of crops, with acidic soils and high organic matter favoring plants' absorption of iron and zinc (Pandian et al. 2011; Alloway 2008); and sandy soils reducing iron and zinc availability to the plant (Lombardo et al. 2013).

The soil of Yanamachay, the locality with the highest mean iron concentration, has the lowest pH (4.67), while the soil of Tacsana, the locality with the lowest mean iron concentration, had the highest percentage of sand (55%) and the lowest organic matter content (1.55%).

The biofortified clones 306018.66, 306140.78 and 306417.79, as well as the variety Amarilla, presented the highest iron concentration in Yanamachay followed by Paltamachay and Castillapata. The biofortified clone 306416.68 and the variety Peruanita presented the highest iron concentration in Yanamachay and Paltamachay. The biofortified clones 306143.122 and 306140.140 presented the highest iron concentration in Yanamachay and Castillapata.

Table 2. Iron concentration (mg/kg DW and FW) in 10 potato samples of 4 locations

Clon/ Variety	Flesh color[1]	mg/kg DW[3]				mg/kg FW[3]			
		Tacsana	Castillapata	Paltamachay	Yanamachay	Tacsana	Castillapata[3]	Paltamachay	Yanamachay
CIP306018.66	Yellow	16.64 ± 0.31^{b}	18.57 ± 0.46^{ab}	19.14 ± 1.73^{a}	20.06 ± 1.97^{a}	4.79 ± 0.23^{a}	5.12 ± 0.45^{a}	4.99 ± 0.75^{a}	5.38 ± 0.54^{a}
CIP306140.140	Purple/yellow[2]	17.39 ± 0.95^{b}	23.68 ± 5.31^{a}	21.05 ± 1.24^{a}	22.52 ± 0.76^{a}	4.70 ± 0.18^{c}	7.41 ± 1.83^{a}	5.13 ± 0.27^{c}	6.26 ± 0.47^{b}
CIP306140.78	Yellow	14.64 ± 1.13^{b}	18.48 ± 1.41^{a}	19.73 ± 2.50^{a}	21.70 ± 3.13^{a}	4.23 ± 0.40^{c}	5.85 ± 0.33^{ab}	5.12 ± 0.47^{b}	6.21 ± 0.85^{a}
CIP306143.122	Yellow	14.99 ± 0.97^{c}	19.85 ± 1.52^{ab}	17.61 ± 0.92^{b}	20.69 ± 3.22^{a}	4.23 ± 0.31^{c}	5.73 ± 0.47^{ab}	5.15 ± 0.05^{b}	6.38 ± 0.78^{a}
CIP306416.68	Purple/yellow[2]	22.49 ± 3.02^{c}	27.13 ± 2.05^{b}	34.47 ± 2.20^{a}	33.25 ± 0.56^{a}	6.33 ± 0.71^{b}	7.95 ± 0.57^{a}	8.71 ± 0.51^{a}	8.30 ± 0.17^{a}
CIP306417.79	Yellow/purple[2]	14.19 ± 1.29^{c}	20.55 ± 0.74^{ab}	20.21 ± 1.04^{b}	23.67 ± 2.08^{a}	4.39 ± 0.34^{c}	6.47 ± 0.38^{ab}	5.88 ± 0.24^{b}	6.77 ± 0.24^{a}
Amarilla	Deep yellow	11.77 ± 0.35^{b}	14.01 ± 1.20^{ab}	13.45 ± 0.80^{b}	16.78 ± 2.09^{a}	3.44 ± 0.16^{b}	4.35 ± 0.46^{ab}	4.04 ± 0.24^{ab}	4.62 ± 0.37^{a}
Peruanita	Deep yellow	10.38 ± 1.30^{b}	12.43 ± 1.50^{ab}	14.09 ± 0.62^{a}	15.54 ± 1.73^{a}	3.26 ± 0.31^{c}	3.64 ± 0.26^{bc}	4.00 ± 0.11^{b}	4.86 ± 0.44^{a}
Mean		15.31 ± 1.17	19.34 ± 1.77	19.97 ± 1.38	21.78 ± 1.94	4.42 ± 0.33	5.81 ± 0.59	5.38 ± 0.33	6.10 ± 0.48

[1] Primary color
[2] Secondary color
[3] Mean values ± standard deviation (n = 3).
Different letters indicate significant differences between sites for each clone or variety ($\alpha = 0.05$)

Table 3. Zinc concentration (mg/kg DW and FW) in 10 potato samples of 4 locations

Clon/ Variety	mg/kg DW[1]				mg/kg FW[1]			
	Tacsana	Castillapata	Paltamachay	Yanamachay	Tacsana	Castillapata	Paltamachay	Yanamachay
CIP306018.66	7.42 ± 0.54^{a}	8.02 ± 0.52^{a}	8.74 ± 0.83^{a}	9.07 ± 2.21^{a}	2.14 ± 0.23^{a}	2.21 ± 0.24^{a}	2.43 ± 0.14^{a}	2.43 ± 0.55^{a}
CIP306140.140	8.36 ± 1.29^{b}	10.93 ± 1.90^{a}	12.09 ± 1.84^{a}	10.96 ± 0.28^{a}	2.26 ± 0.30^{b}	3.18 ± 0.44^{a}	2.95 ± 0.44^{a}	3.05 ± 0.21^{a}
CIP306140.78	7.95 ± 0.94^{b}	11.20 ± 1.08^{a}	11.96 ± 1.07^{a}	13.28 ± 3.20^{a}	2.30 ± 0.32^{b}	3.54 ± 0.15^{a}	3.11 ± 0.18^{a}	3.80 ± 0.90^{a}
CIP306143.122	8.20 ± 0.26^{b}	8.88 ± 1.89^{ab}	9.57 ± 1.21^{ab}	11.60 ± 3.19^{a}	2.31 ± 0.13^{c}	2.57 ± 0.60^{bc}	2.79 ± 0.22^{b}	3.57 ± 0.88^{a}
CIP306416.68	10.98 ± 1.17^{b}	9.90 ± 1.47^{b}	16.33 ± 1.27^{a}	18.54 ± 2.10^{a}	3.10 ± 0.35^{b}	3.02 ± 0.39^{b}	4.13 ± 0.27^{a}	4.63 ± 0.54^{a}
CIP306417.79	9.18 ± 0.59^{c}	11.02 ± 1.52^{b}	12.33 ± 1.36^{ab}	14.29 ± 2.02^{a}	2.84 ± 0.16^{b}	3.46 ± 0.38^{a}	3.59 ± 0.34^{a}	4.10 ± 0.59^{a}
Amarilla	6.86 ± 0.18^{b}	7.08 ± 0.58^{b}	10.13 ± 1.02^{a}	10.64 ± 1.23^{a}	2.01 ± 0.09^{c}	2.20 ± 0.22^{bc}	3.04 ± 0.29^{a}	2.94 ± 0.32^{ab}
Peruanita	6.49 ± 0.07^{b}	10.09 ± 0.18^{a}	9.43 ± 0.55^{a}	11.47 ± 1.57^{a}	2.04 ± 0.05^{c}	2.97 ± 0.14^{ab}	2.67 ± 0.03^{b}	3.58 ± 0.24^{a}
Mean	8.18 ± 0.63	9.64 ± 1.14	11.32 ± 1.15	12.48 ± 1.98	2.37 ± 0.20	2.89 ± 0.32	3.09 ± 0.24	3.51 ± 0.53

[1] Mean values ± standard deviation (n = 3).
Different letters indicate significant differences between sites for each clone or variety ($\alpha = 0.05$)

The biofortified clones 306140.140, 306140.78 and the variety Peruanita showed the highest zinc concentration in Yanamachay, Paltamachay and Castillapata. The biofortified clone 306416.68 and the variety Amarilla presented the highest zinc values in Yanamachay and Paltamachay and the biofortified clones 306143.122 and 306417.79 presented the highest values in Yanamachay.
The highest iron and zinc concentrations in Yanamachay and Paltamachay can be explained by the fact that Yanamachay and Paltamachay (4.67 pH and 5.15 pH, respectively) presented a more acidic soil than Castillapata and Tacsana (6.17 pH and 5.77 pH, respectively). The high iron concentration in Castillapata is difficult to explain as the pH values were very high, but it seems that the loamy soil type and the high organic matter content (8.21%) favored iron absorption. The low iron and zinc concentration in Tacsana can be explained by the fact the soil of Tacsana presented the lowest percentage of organic matter (1.55 vs 8.21, 4.93 and 7.38% in Castillapata, Paltamachay and Yanamachay, respectively) and a high proportion of sand, which favors the oxidation of iron and zinc to insoluble polymers which reduces mineral availability to the plant (Lombardo et al. 2013).

3.2 Effect on vitamin C concentration

The vitamin C concentration of the biofortified clones and varieties grown in four locations, expressed on a DW and FW basis, is shown in Table 4.
The concentrations of vitamin C in the biofortified clones and varieties ranged from 41.03 to 67.93 mg / 100g DW in Tacsana, from 34.09 to 51.06 mg / 100g DW in Castillapata, from 46.53 to 58.04 mg / 100g DW in Paltamachay and from 38.86 to 58.61 mg / 100g DW in Yanamachay.
The effect of the location and of the clone x location interactions on vitamin C concentrations was significant ($P < 0.001$). Similar results have been reported by Burgos et al. 2009; Skrabule et al. 2013; Hamouz et al. 2018. Some authors suggest that the concentration of vitamin C in potato tubers may be related to soil type, temperature and N fertilization (Hamouz et al. 2007), while other studies suggest that plants can increase their vitamin C content as a consequence of stress conditions as part of defense responses (Skrabule et al. 2013; Locato et al. 2013).
The mean vitamin C concentration in Paltamachay (50.57 mg / 100g DW), Tacsana (50.94 mg / 100g DW) and Yanamachay (47.67 mg / 100g DW) was higher than in Castillapata (41.97 mg / 100g DW). A negative relation between nitrogen nutrition and vitamin C content in potatoes has also been observed by Skrabule et al. 2013 and Lin et al. 2004. The soil of Tacsana, Paltamachay and Yanamachay, the localities with the higher mean vitamin C concentration, had a total nitrogen percentage (0.09%, 0.29% and 0.43%, respectively) lower than in Castillapata (0.48%), the locality with the lower mean vitamin C concentration. In addition, Hamouz et al. 2007 found that high levels of Mg and K favor the production of vitamin C in potato. The Mg and K concentration of the soil of Tacsana and Paltamachay (3.99 and 2.77 for Mg, respectively and 0.60 and 0.47 for K, respectively) was higher than in Castillapata (0.95 for Mg and 0.31 for K). Furthermore, Hamouz et al. 2009 reported that the vitamin C concentration of potatoes grown in sandy, loamy brown soil was higher than in potatoes grown in loamy soil. The soil of Tacsana and Paltamachay was of a sandy loamy type while the soil of Castillapata was loamy type.

Table 4. Vitamin C concentration (mg/100g DW and FW) in 10 potato samples of 4 locations

Clon/ Variety	mg / 100g DW[1]				mg / 100g FW[1]			
	Tacsana	**Castillapata**	**Paltamachay**	**Yanamachay**	**Tacsana**	**Castillapata**	**Paltamachay**	**Yanamachay**
CIP306018.66	55.19 ± 2.86^{a}	36.98 ± 3.22^{b}	50.50 ± 2.19^{a}	42.82 ± 2.41^{b}	15.07 ± 1.44^{a}	9.98 ± 1.28^{b}	13.90 ± 0.88^{a}	11.33 ± 0.97^{b}
CIP306140.140	49.57 ± 5.83^{b}	51.06 ± 3.90^{b}	58.04 ± 3.29^{a}	58.61 ± 3.47^{a}	13.63 ± 1.09^{b}	15.40 ± 1.89^{ab}	14.20 ± 0.32^{ab}	15.45 ± 1.24^{a}
CIP306140.78	45.65 ± 0.80^{a}	45.00 ± 1.34^{a}	47.32 ± 6.01^{a}	50.71 ± 2.78^{a}	13.18 ± 0.45^{ab}	14.34 ± 0.74^{a}	12.04 ± 0.65^{b}	13.80 ± 0.98^{a}
CIP306143.122	50.44 ± 7.35^{a}	40.28 ± 5.07^{b}	48.21 ± 3.45^{a}	48.73 ± 1.97^{a}	13.92 ± 0.55^{a}	11.62 ± 1.27^{b}	14.73 ± 0.98^{a}	15.42 ± 0.62^{a}
CIP306416.68	41.03 ± 0.80^{b}	44.39 ± 7.57^{b}	51.03 ± 1.81^{a}	39.72 ± 0.99^{b}	11.19 ± 0.15^{b}	13.23 ± 2.44^{a}	13.41 ± 0.47^{a}	9.36 ± 0.30^{c}
CIP306417.79	46.00 ± 1.76^{a}	34.09 ± 4.15^{b}	47.21 ± 1.08^{a}	38.86 ± 4.11^{b}	13.73 ± 0.70^{a}	10.12 ± 1.02^{b}	13.38 ± 0.74^{a}	10.14 ± 1.22^{b}
Amarilla	67.93 ± 7.70^{a}	38.53 ± 4.43^{d}	55.69 ± 2.64^{b}	47.98 ± 3.40^{c}	20.83 ± 1.63^{a}	12.11 ± 1.67^{c}	15.99 ± 0.90^{b}	13.27 ± 1.09^{c}
Peruanita	51.69 ± 3.45^{ab}	45.43 ± 0.48^{b}	46.53 ± 4.21^{b}	53.96 ± 3.40^{a}	16.26 ± 0.15^{a}	13.66 ± 0.54^{b}	13.51 ± 0.44^{b}	16.41 ± 1.73^{a}
Mean	50.94 ± 3.82	41.97 ± 3.77	50.57 ± 3.09	47.67 ± 2.82	14.73 ± 0.77	12.56 ± 1.36	13.90 ± 0.67	13.15 ± 1.02

[1] Mean values ± standard deviation (n = 3).

Different letters indicate significant differences between sites for each clone or variety ($\alpha = 0.05$)

3.3 Effect on phenolic concentration

The concentration of phenolics in the iron biofortified potato clones and the varieties grown in four locations of Huancavelica, Peru, expressed on a DW and FW basis, is shown in Table 5.

The phenolic concentration ranged from 204.40 to 1031.49 mg / 100g DW in Tacsana, from 210.07 to 899.56 mg / 100g DW in Castillapata, from 231.74 to 998.15 mg / 100g DW in Paltamachay and from 220.60 to 758.11 mg / 100g DW in Yanamachay, with the purple fleshed biofortified clone 306416.68 showing the highest phenolic concentration in the four locations and the yellow fleshed clone 306140.78 showing the lowest phenolic concentration.

The effect of the location and of the clone x location interactions on phenolic concentrations was significant ($P < 0.001$). The mean phenolic concentration in Castillapata and Paltamachay (449.77 and 457.24 mg / 100g DW) was higher than in Yanamachay (383.03 mg / 100 g DW).

Some studies have reported that calcium contributes to increased caffeic and chlorogenic acid in potatoes. Furthermore, calcium soil amendment also improved the concentration of polyphenol oxidase (PPO) and peroxidase (POD) enzymes, which are involved in the metabolism of phenolics (Ngadze et al. 2014). In our study, Yanamachay, the location with the lowest calcium concentration (5.52 meq / 100g of calcium), also has a lower mean value of phenolics (383.03 mg / 100g DW) than Tacsana, Castillapata and Paltamachay (13.08, 23.12 and 19.10 meq of calcium soil / 100 g respectively) with phenolics levels of 425.54, 449.77 and 457.24 mg/100g DW; respectively.

Previous studies concluded that the higher altitude locations, the lower temperature during the overall vegetative period and maturation, the higher annual precipitation and the lowest soil fertility produced an increase of the total phenolic content in potato tubers (Lachman et al. 2008, Hamouz et al. 2007). In this study, the potatoes grown in Yanamachay, the location situated at the highest level (4130 m.a.s.l.) showed the lowest phenolic concentration. However, the differences in altitude were not so contrastable as the other three localities were also at a very high altitude (3672, 3908 and 3945 m.a.s.l for Tacsana, Castillapata and Paltamachay, respectively).

3.4 Effect on phytic acid concentration

The phytic acid concentration of the biofortified clones and varieties grown in four locations, expressed on a DW and FW basis, is shown in Table 6.

The phytic acid concentration ranged from 164.58 to 484.18 mg / 100g DW in Tacsana, from 158.83 to 317.99 mg / 100g DW in Castillapata, from 120.30 to 353.49 mg / 100g DW in Paltamachay and from 123.48 to 281.68 mg / 100g DW in Yanamachay. These values are similar to the phytate values reported by the FAO global food composition database for raw potatoes (289.26 to 367.53 mg / 100g DW) (FAO, 2018). Previous studies reported similar values, Phillippy et al. 2003 found 273.62 mg / 100g DW, while Glahn et al. 2017 reported 330.00 mg/100g DW of phytic acid content for potatoes.

Table 5. Phenolic compounds concentration (mg/100g DW and FW) in 10 potato samples of 4 locations

Clon/ Variety	mg / 100g DW[1]				mg / 100g FW[1]			
	Tacsana	Castillapata	Paltamachay	Yanamachay	Tacsana	Castillapata	Paltamachay	Yanamachay
CIP306018.66	302.66 ± 27.32^{ab}	368.74 ± 69.53^{a}	264.29 ± 32.12^{b}	276.67 ± 45.35^{ab}	82.63 ± 9.55^{a}	98.66 ± 13.34^{a}	72.56 ± 7.00^{a}	72.95 ± 10.33^{a}
CIP306140.140	441.12 ± 24.46^{c}	498.20 ± 48.42^{bc}	622.79 ± 28.79^{a}	534.23 ± 36.68^{b}	121.88 ± 11.56^{b}	149.97 ± 16.33^{a}	152.52 ± 6.06^{a}	140.95 ± 15.26^{ab}
CIP306140.78	204.40 ± 36.98^{a}	210.07 ± 23.89^{a}	231.74 ± 13.84^{a}	220.60 ± 24.51^{a}	58.96 ± 10.34^{a}	66.88 ± 7.53^{a}	59.30 ± 5.08^{a}	60.07 ± 7.24^{a}
CIP306143.122	449.07 ± 2.44^{a}	281.97 ± 48.44^{b}	241.88 ± 15.47^{b}	270.80 ± 8.49^{b}	125.40 ± 14.88^{a}	81.06 ± 9.84^{b}	73.89 ± 4.45^{b}	85.68 ± 3.26^{b}
CIP306416.68	1031.49 ± 98.01^{a}	899.56 ± 72.26^{b}	998.15 ± 97.46^{ab}	758.11 ± 80.03^{c}	281.17 ± 24.27^{a}	267.80 ± 22.36^{a}	262.22 ± 25.71^{a}	178.75 ± 20.19^{b}
CIP306417.79	474.05 ± 38.26^{b}	676.86 ± 147.30^{a}	748.75 ± 112.67^{a}	381.06 ± 42.42^{c}	141.52 ± 13.36^{b}	201.22 ± 43.43^{a}	211.13 ± 22.13^{a}	99.45 ± 12.46^{c}
Amarilla	225.91 ± 34.74^{b}	334.83 ± 51.84^{a}	298.62 ± 7.46^{ab}	282.98 ± 47.61^{ab}	69.17 ± 7.55^{b}	105.25 ± 18.53^{a}	85.80 ± 4.59^{ab}	78.49 ± 15.30^{ab}
Peruanita	275.62 ± 23.72^{ab}	327.92 ± 10.07^{ab}	251.68 ± 40.43^{b}	339.76 ± 15.48^{a}	86.81 ± 7.94^{ab}	98.60 ± 5.76^{ab}	73.09 ± 10.60^{b}	103.05 ± 1.13^{a}
Mean	425.54 ± 35.74	449.77 ± 58.97	457.24 ± 43.53	383.03 ± 37.57	120.94 ± 12.43	133.68 ± 17.14	123.81 ± 10.70	102.42 ± 10.65

[1] Mean values ± standard deviation (n = 3).
Different letters indicate significant differences between sites for each clone or variety ($\alpha = 0.05$)

Table 6. Phytic acid concentration (mg/100g DW and FW) in 10 potato samples of 4 locations

Clon/Variety	mg / 100 g DW[1]				mg / 100 g FW[1]			
	Tacsana	Castillapata	Paltamachay	Yanamachay	Tacsana	Castillapata	Paltamachay	Yanamachay
CIP306018.66	215.26 ± 28.01^{ab}	262.80 ± 24.04^{a}	167.30 ± 12.72^{bc}	149.48 ± 12.24^{c}	58.50 ± 6.87^{ab}	72.25 ± 5.51^{a}	46.59 ± 2.88^{bc}	39.97 ± 1.34^{c}
CIP306140.140	235.46 ± 35.56^{a}	291.18 ± 68.99^{a}	271.89 ± 60.74^{a}	138.53 ± 8.14^{b}	63.63 ± 8.71^{a}	84.64 ± 18.32^{a}	66.24 ± 14.61^{a}	38.43 ± 0.62^{b}
CIP306140.78	250.29 ± 32.18^{ab}	317.99 ± 75.00^{a}	203.53 ± 44.79^{bc}	167.26 ± 38.00^{c}	72.41 ± 10.45^{b}	99.84 ± 17.62^{a}	52.93 ± 11.10^{c}	47.82 ± 10.40^{c}
CIP306143.122	256.83 ± 28.79^{a}	244.97 ± 70.98^{a}	134.17 ± 11.43^{b}	172.39 ± 49.81^{b}	72.35 ± 6.06^{a}	70.96 ± 22.40^{ab}	39.24 ± 2.11^{c}	53.09 ± 13.77^{b}
CIP306416.68	484.18 ± 62.13^{a}	274.89 ± 60.94^{c}	353.49 ± 44.40^{b}	281.68 ± 23.01^{c}	136.09 ± 12.44^{a}	84.71 ± 22.93^{bc}	89.21 ± 9.50^{b}	70.41 ± 7.46^{c}
CIP306417.79	362.66 ± 43.23^{a}	257.72 ± 58.32^{b}	244.01 ± 9.46^{b}	228.66 ± 37.99^{b}	112.40 ± 14.49^{a}	81.39 ± 20.70^{b}	71.08 ± 4.06^{b}	65.21 ± 7.79^{b}
Amarilla	231.60 ± 20.74^{a}	158.83 ± 16.82^{ab}	120.40 ± 22.92^{b}	123.48 ± 2.58^{b}	67.63 ± 4.84^{a}	49.19 ± 4.38^{ab}	36.14 ± 7.06^{b}	34.13 ± 2.00^{b}
Peruanita	164.58 ± 16.53^{b}	257.56 ± 33.36^{a}	120.30 ± 5.21^{b}	141.27 ± 6.52^{b}	51.64 ± 4.24^{b}	75.57 ± 7.45^{a}	34.15 ± 2.23^{c}	44.22 ± 1.05^{bc}
Mean	275.11 ± 33.40	258.24 ± 51.06	201.89 ± 26.46	175.34 ± 22.29	79.33 ± 8.51	77.32 ± 14.91	54.45 ± 6.69	49.16 ± 5.55

[1] Mean values ± standard deviation (n = 3). Different letters indicate significant differences between sites for each clone or variety ($\alpha = 0.05$)

The effect of the location and of the clone x location interactions on phytic acid concentrations was significant ($P < 0.001$). The mean phytic acid concentration in Tacsana and Castillapata (275.11 and 258.24 mg / 100g DW, respectively) was higher than in Paltamachay and Yanamachay (201.89 and 175.34 mg / 100g DW respectively). Mazetti et al. 2014 found higher phosphorus (P) concentrations in potato tubers at increased rates of P_2O_5 in soil. Since phytic acid is a P reserve in plants, it was expected that the phytic acid concentration of Yanamachay, the location with the highest P2O5 available (38.61 ppm) had the highest phytic acid values; and that the phytic acid concentration of Tacsana and Castillapata, the locations with the lowest P_2O_5 available (8.62 and 12.98 ppm respectively) had the lowest phytic acid concentration. However, opposite results were found. Since soils with elevated aluminum (Al) cations, reduce the P uptake (FAO 2008), the lowest phytic acid concentration found in Yanamachay can be explained by the fact that its soil has higher level of $Al^{+3} + H^{+}$ cations (2.43 meq/100g) compared to Tacsana, Castillapata and Paltamachay (<0.01, 0.08 and 0.15 meq/100g, respectively). In addition, as it has been reported that sandy soils increase the P uptake by the potato plant (Lopes et al. 2018), hence, the highest phytic acid mean concentration in Tacsana could be attributed to a high percentage of sand in its soil (55.21%)

3.5 Effect on glycoalkaloid concentration

The concentration of total glycoalkaloids in the biofortified clones and varieties grown in four locations, expressed in DW and FW, is shown in Table 7. The total glycoalkaloid concentration in the biofortified clones ranged from 3.36 to 23.27 mg / 100g DW in Tacsana, from 4.53 to 30.09 mg / 100g DW in Castillapata, from 4.18 to 42.20 mg / 100g DW in Paltamachay and from 5.94 to 42.92 mg / 100g DW in Yanamachay. The total glycoalkaloid concentration in the variety Amarilla ranged from 5.29 mg / 100g DW in Tacsana to 28.50 mg / 100g DW in Yanamachay, and in the variety Peruanita from 11.50 mg / 100g DW in Tacsana to 29.94 mg / 100g DW in Yanamachay.

The effect of the location and of the clone x location interactions on total glycoalkaloid concentrations was significant. The mean total glycoalkaloid concentration in Paltamachay (21.09 mg / 100g DW) and Yanamachay (24.32 mg / 100g DW) was significantly higher than in Castillapata (16.18 mg / 100g DW) and higher than in Tacsana (9.52 mg / 100g DW).

These results can be explained by the fact that a higher nitrogen concentration in the soil favors a greater synthesis and accumulation of nitrogen compounds such as glycoalkaloids in the tuber. The soil of Tacsana has a very low total nitrogen content (0.09%) compared to Castillapata, Paltamachay and Yanamachay (0.48, 0.29 and 0.43%, respectively). Zolnowski 2010, reported that glycoalkaloid content was positively correlated with the total nitrogen content in the soil. In addition, Skrabule et al. 2013 suggested that a higher percentage of organic matter produced greater availability of nitrogen. The soils of Castillapata, Paltamachay and Yanamachay (8.21, 4.93 and 7.38%, respectively) have a higher organic matter percentage than the soil of Tacsana (1.55%). In addition, Castillapata and Yanamachay presented loamy soil which has been reported as favoring glycoalkaloid tuber concentration (Haase, 2010).

Table 7. Glycoalkaloids concentration (mg/100g DW and FW) in 10 potato samples of 4 locations

Clon/ Variety	mg/100g DW[1]				mg/100g FW[1]			
	Tacsana	**Castillapata**	**Paltamachay**	**Yanamachay**	**Tacsana**	**Castillapata**	**Paltamachay**	**Yanamachay**
CIP306018.66	10.50 ± 1.00^{c}	30.09 ± 5.23^{b}	42.20 ± 8.24^{a}	42.92 ± 4.67^{a}	2.87 ± 0.35^{c}	8.06 ± 1.00^{b}	11.57 ± 2.00^{a}	11.32 ± 0.86^{a}
CIP306140.140	4.86 ± 1.03^{b}	5.79 ± 2.83^{b}	12.73 ± 2.31^{a}	12.79 ± 2.52^{a}	1.35 ± 0.31^{c}	1.77 ± 0.96^{bc}	3.12 ± 0.52^{ab}	3.38 ± 0.77^{a}
CIP306140.78	6.02 ± 1.74^{b}	6.57 ± 2.94^{b}	12.10 ± 2.70^{ab}	15.44 ± 0.38^{a}	1.73 ± 0.48^{b}	2.11 ± 1.01^{b}	3.07 ± 0.51^{ab}	4.20 ± 0.02^{a}
CIP306143.122	3.36 ± 0.86^{a}	4.53 ± 0.63^{a}	4.18 ± 0.81^{a}	5.94 ± 2.08^{a}	0.93 ± 0.17^{a}	1.32 ± 0.25^{a}	1.28 ± 0.25^{a}	1.88 ± 0.68^{a}
CIP306416.68	23.27 ± 2.53^{b}	13.87 ± 0.44^{c}	29.14 ± 2.44^{ab}	30.48 ± 5.57^{a}	6.35 ± 0.78^{a}	4.13 ± 0.27^{b}	7.65 ± 0.61^{a}	7.18 ± 1.29^{a}
CIP306417.79	11.34 ± 1.34^{c}	28.46 ± 4.46^{b}	39.39 ± 10.51^{a}	28.53 ± 2.73^{b}	3.39 ± 0.45^{c}	8.48 ± 1.51^{b}	11.08 ± 2.49^{a}	7.45 ± 0.82^{b}
Amarilla	5.29 ± 1.95^{c}	25.70 ± 3.07^{a}	16.03 ± 1.10^{b}	28.50 ± 6.71^{a}	1.65 ± 0.71^{c}	8.07 ± 1.07^{a}	4.61 ± 0.45^{b}	7.81 ± 1.33^{a}
Peruanita	11.50 ± 4.36^{b}	14.41 ± 0.55^{b}	12.98 ± 2.26^{b}	29.94 ± 3.27^{a}	3.65 ± 1.50^{b}	4.33 ± 0.09^{b}	3.75 ± 0.41^{b}	9.11 ± 1.23^{a}
Mean	9.52 ± 1.85	16.18 ± 2.52	21.09 ± 3.80	24.32 ± 3.49	2.74 ± 0.59	4.78 ± 0.77	5.77 ± 0.90	6.54 ± 0.88

1 Mean values + standard deviation (n = 3).

Different letters indicate significant differences between sites for each clone or variety ($\alpha = 0.05$)

Experiments with human taste panels revealed that potato varieties with glycoalkaloid levels exceeding 14 mg / 100 g FW tasted bitter (Friedman 2006). According to FDA/WHO recommendations, glycoalkaloid content in potato tubers should not exceed 20 mg / 100 g FW, as this level is dangerous to human health (Ruprish et al., 2009). In this study, the total glycoalkaloid concentration of all biofortified clones and local varieties in the four localities was below 12 mg / 100 g FW. However, from a breeder's perspective it is recommended that concentrations of glycoalkaloids be as low as possible, in order to avoid the possibility that any exposure of the tubers to abiotic or biotic stress may result in an increase of levels to above acceptable limits.

3.6 Potential of iron biofortified potato to reduce iron deficiency

On a fresh weight basis, the iron concentration of biofortified potatoes ranged from 0.42 to 0.87 mg / 100 g, which is 23 to 54% more iron than the varieties Peruanita and Amarilla (0.32 to 0.48 mg / 100 g). The iron values in the biofortified potatoes are lower than the iron concentration of beans (4.45 to 7.62 mg / 100g; Hass et al., 2016), pearl millet (2.59 to 8.49 mg / 100g; Tako et al.,2015) and wheat (3.33 mg / 100g; Araujo et al., 2008). However, biofortified potatoes have a significant amount of vitamin C concentration, a promoter of iron absorption, ranging from 9.36 to 15.45 mg / 100 g FW. Cereals and legumes have a non-detectable vitamin C content and have a high concentration of phytates which inhibit iron absorption. The phytic acid concentration in the biofortified potatoes and control varieties analyzed in this study ranged from 34.15 to 136.09 mg / 100 g FW. These values are significantly lower than the phytic acid concentration reported for wheat (801.35 – 942.24 mg/100g FW; Erdal et al. 2002), beans (1082.95 mg/100g FW; Glahn et al. 2017) and pearl millet (618.15 - 980.33 mg/100g FW; Krishnan and Meera 2017).

Phenolics are also inhibitors of iron absorption. The biofortified potatoes contain a total phenolic compound concentration ranging from 58.96 to 281.17 mg / 100 g FW. The yellow fleshed biofortified clones, as well as the variety controls, have a lower phenolic concentration (below 130 mg / 100 g, FW) than beans (around 300 mg / 100 g FW, Oliveira et al, 2018; Hanis et al., 2017) and pearl millet (around 250 mg / 100 g FW, Kumar and Kaul., 2017). However, the purple fleshed biofortified clone 306416.68, that presents the highest iron concentration, has a phenolic concentration reaching 281 mg / 100 g FW, which is similar to that of beans and pearl millet. Hence, it is very important to determine if the type of phenolic contained in potato inhibits iron absorption, and if so to what extent.

Considering 500 g of potato consumption per day for women, as is the case in the highlands of Huancavelica (De Haan et al., 2019) and considering the iron estimated average requirement (EAR) assuming 10% of potato iron bioavailability (15 mg per day, IOM 2001), the biofortified potatoes contribute 14 to 29% of EAR for women of fertile age. However, potato iron bioavailability should be determined in humans. Progress in this regard is limited to the finding that potato Fe has high in vitro bioaccessibility, with 63 to 79 % of potato Fe released from the food matrix during in vitro gastro-intestinal digestion, and therefore available at the intestinal level (Andre et al., 2015). Potato Fe bioaccessibility compares very favorably with various cereals and legumes. For example, pearl millet which is considered a success among biofortified crops, has an

Fe bioaccessibility varying from 10 to 24%, while fava bean, soybean, and rice has an Fe bioaccessibility ranging from 6 to 32%. Nevertheless, to assess the full potential of the Fe potato biofortification program, human studies are required to gain insight on how much of the iron from biofortified potatoes is absorbed by the human body.

4. CONCLUSIONS

Iron, zinc, vitamin C, phenolic, phytic acid and glycoalkaloid concentrations are affected by the location and by the interaction between the clone and the location.

The mean iron and zinc concentration was higher in Yanamachay and Paltamachay, localities with acidic soils and high organic matter content; and was lower in Tacsana, a locality with a low percentage of organic matter and high proportion of sand.

The mean vitamin C concentration was higher in Tacsana and Paltamachay, localities with lower levels of nitrogen and higher levels of Mg and K in the soil.

The mean phenolic and phytic acid concentration was lower in Yanamachay. Low levels of calcium and high levels of Al^{+3} + H^{+} cations in the soil of Yanamachay could be the reason of low phenolic and phytic acid accumulation in Yanamachay, respectively.

The mean glycoalkaloid concentration was higher in Yanamachay, locality with high level of nitrogen and high organic matter content in the soil.

Acidic soils with high organic matter content can favor the production of iron in biofortified potatoes. High organic matter needs to be controlled, as it also favors glycoalkaloid production. High levels of magnesium and potassium and lower levels of nitrogen could favor production of vitamin C, a promotor of iron absorption, while low levels of calcium and high levels of Al cation in the soil can reduce the production of the inhibitors of iron absorption (phenolic compounds and phytic acid respectively).

Across locations, the yellow fleshed biofortified potatoes have a significant amount of vitamin C and very low levels of phytic acid and phenolic compounds which suggest its great potential to contribute to reduce anemia.

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