

# The quinoa variety influences the nutritional and antioxidant profile rather than the geographic factors

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## ABSTRACT

Quinoa (*Chenopodium quinoa* Willd.) is attracting worldwide attention due to its nutritional and biological properties. Nowadays, this pseudocereal is cultivated worldwide in different environmental conditions. This work evaluates the nutritional profile, polyphenol content and antioxidant capacity of five quinoa cultivars (Negra Collana, Chullpi Real, Salcedo Inia, Pasankalla and Kancolla) from Spain and from the Andean region, looking for the key factor of quinoa composition. Nutritional profile was similar for the same cultivar among the locations but, protein and iron contents were higher ( $p < 0.05$ ) in Spanish seeds compared to the Andean ones. PCA and Pearson correlation coefficient reveal that the darkest quinoa cultivars, Negra and Pasankalla, had the best bioactive profile because the greater dietary fiber, polyphenol content, and antioxidant capacity ( $p < 0.05$ ), regardless of origin zone. Concluding, the genetic variability seems to have a higher influence than the geographic factors on the nutritional and antioxidant composition of quinoa.

## 1. Introduction

Quinoa (*Chenopodium quinoa* Willd.) is a dicotyledonous annual plant native to the Andean highland region of northwestern South America. It was, for centuries, a basic food of ancient Andean civilizations (Carrasco-Valencia & Serna, 2011; Pellegrini et al. 2018; Präger et al. 2018). Quinoa has been classified as a pseudocereal since it does not belong to the Gramineae family. However, it shares a similar composition to cereals regarding the starch content (Navruz-Varli & Sanlier, 2016; Jimenez et al. 2019). In 2013, the Food and Agriculture Organization of the United Nations (FAO) launched the International Year of Quinoa with the aim of promoting the production, preservation, and consumption of this crop, defined as “one of the grains of the 21st century” (Tang et al. 2015; Vilcacundo & Hernandez-Ledesma, 2017; Pellegrini et al. 2018). In fact, it has an exceptional nutritional composition that includes proteins (13–16 %) with a high biological value and an amino acid pattern close to the ideal protein balance recommended by FAO in 2011. Moreover, micronutrients such as minerals (calcium, copper, manganese, zinc, and iron) and vitamins (thiamine, riboflavin, folic acid, niacin or retinol) can be encountered among others (Nsimba et al. 2008; Lamothe et al. 2015; Vilcacundo & Hernandez-Ledesma,

2017; Jimenez et al. 2019). Furthermore, quinoa seed reveals total absence of gluten, and therefore, it is regarded as suitable for people with celiac disease (Ceyhun Sezgin, et al., 2019). Furthermore, it has been considered an alternative oilseed crop due to the quality and quantity of high levels of fatty acids (Filho et al. 2017; Miranda et al. 2012; Perreira et al. 2019). In addition, it has proved to be a functional food due to its nutritional composition and its content in bioactive components, such as flavonoids, phytosterols, carotenoids and polyphenols, with health-promoting effects. Particularly, it can prevent degenerative and inflammatory diseases, cancer, allergy, and may reduce the risk of cardiovascular problems (Hemalatha et al. 2016; Vilcacundo & Hernandez-Ledesma, 2017; Perreira et al. 2019). Because of its wide genetic variability and nutritional components, this pseudocereal has gained worldwide attention, and nowadays, it is also grown outside the Andean region, including Europe, North America, Canada, North Africa and China (Martin Gonzalez et al. 2014; Hemalatha et al. 2016; Multari et al. 2018; Pellegrini et al. 2018). The demand of quinoa products has increased in parallel with the practice of healthier lifestyles, and consequently, the price of quinoa has rapidly raised (Nsimba et al. 2008; Präger et al. 2018; Perreira et al. 2019). However, where quinoa is cultivated, there is a clear downward trend in consumption,

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which may be due to the prioritization of exports. Thus, this crop has been replaced by other cultivation with a much lower nutritional value (Pellegrini et al. 2018).

Due to the great international demand for this pseudocereal, the large producers (Bolivia and Peru) have expanded their production area, which is causing the disappearance of natural vegetation, contamination, or soil degradation. In the same line, there is considerable interest in growing quinoa in European latitudes since this crop can be a profitable source of income for European farmers, (Präger et al. 2018).

On the other hand, the nutritional and phytochemical composition of plants are influenced by different environmental factors; particularly, the great success in raising crop yields depends mainly on their production of different second metabolites, which differs according to the different cultivation areas (Miranda et al. 2012; Miranda et al., 2013; Yang et al. 2018). These molecules may strongly impact on the food quality and carry many potential beneficial effects of consumer health (Perreira et al. 2019). In fact, there are more than two hundred and fifty varieties of quinoa (Navruz-Varli & Sanlier, 2016; Präger et al. 2018).

This study was aimed at providing a detailed evaluation of the nutritional and functional value of different quinoa cultivars from different origins, Spain and Andean Region (Puno, Peru), to understand the main factors for a better nutritional profile, due to the mentioned interest of the cultivar adaption to European latitudes. Particularly, the objective of this work was to determine the nutritional and bioactive components and the antioxidant capacity of five quinoa varieties (Salcedo-INIA, Chullpi Real, Kancolla, Pasankalla and Negra Collana) in order to explore how the environmental and climatic factors and their variety type could affect the nutritional and bioactive quality of these quinoa seeds.

## 2. Materials and methods

### 2.1. Raw material

The five quinoa cultivars were Salcedo Inia, Chullpi Real, Kancolla, Pasankalla and Negra Collana. All varieties were cultivated in Spain and in the Andean region. The European location were El Pobo (1420 m a.s.l.), El Chaparrillo (628 m a.s.l.) and Villamanrique de la Condesa (34 m a.s.l.), while for the Andean region, all zones were at 3800 m a.s.l., and correspond to the Peruvian highlands in the Puno region. The cultivated areas present sandy loamy soils with good drainage. The average temperature during the year ranges between  $-4$  and  $16$  °C in Peruvian area, and in the Spanish locations,  $0$ – $24$  °C in El Chaparrillo,  $7$ – $27.6$  °C in Villamanrique de la Condesa, and  $-6$ – $28$  °C in El Pobo, being all of them mostly dry areas.

Quinoa seeds were cultivated in agricultural fields complying with mandatory regulations. They were harvested from 2014 to 2016 and sent to our laboratory inside sealed bags. Samples were kept inside desiccators at room temperature ( $18$ – $20$  °C) in darkness. The analyses described below were performed during 2016–2017, and previously, these samples were milled to flours (flour size  $< 0.1$  mm, 140 mesh) and stored in a dry atmosphere to preserve them during the analysis time. The crop origin, the altitude and the year of harvesting can be found in supplementary material (Table S1).

### 2.2. Colour

Colour of quinoa samples was measured using a tristimulus colorimeter (Minolta mod. CR-200), calibrated with a white calibration plate (CR-A43). The color measurements were expressed as  $L^*$ ,  $a^*$ ,  $b^*$  parameters.  $L^*$  indicates lightness ( $100$  = white and  $0$  = black),  $a^*$  indicates redness-greenness and  $b^*$  indicates yellowness-blueness.

### 2.3. Proximate composition and analysis of macro- and microelements

Moisture was determined by oven-drying method at  $105 \pm 1$  °C

(AOAC 945.15). Total nitrogen content was analyzed by the Kjeldahl procedure (AOAC, 945.18). The conversion factor used to transform nitrogen into protein was 6.25. Fat was measured in a Soxtec system by extraction with ethyl ether solvent (AACC n° 30-10.01). Available carbohydrates were measured using anthrone method (Southgate, 1976). Dietary fiber was analyzed through the enzymatic–gravimetric method AOAC 991.43. Ash content was determined by incineration at  $550$  °C in a microwave muffle Milestone mod. MLS-1200 Pyro (Monroe, CT, USA) following for steps ( $250$  °C/30 min,  $550$  °C/15 min,  $550$  °C/20 h, and  $100$  °C/30 min) to optimize the process and minimize the volatilization of minerals. The macro and microelements were assessed in the ash content by atomic absorption spectroscopy (AAS) in a Perkin Elmer Analyst AA200 (Shelton, CT, USA) (Mateos-Aparicio et al. 2010).

### 2.4. Functional characterization

#### 2.4.1. Dietary fiber composition

The fractions isolated from the application of the enzymatic–gravimetric AOAC method were subjected to hydrolysis with  $H_2SO_4$  12 M at  $35$  °C during 30 min followed by  $H_2SO_4$  2 M at  $100$  °C during 1 h. The released neutral sugars were transformed into alditol acetates with acetic anhydride in the presence of 1-methylimidazol. Quantification was performed in a Perkin–Elmer Autosystem chromatograph equipped with a hydrogen flame ionization detector. The column used was a SP-2330 (30 m long, 0.25 mm i.d., and 0.25  $\mu$ m film thickness) and nitrogen served as carrier gas (22 psi). Temperatures of injector and detector were  $275$  °C and oven temperature was  $235$  °C. Retention times and peak areas were registered in a PE Nelson computer mod. 1020 and  $D$ -allose (Fluka) was used as internal standard. Uronic acids content was determined in the acid hydrolysates according to the colorimetric method of 3,5-dimethylphenol, with a Pharmacia mod. LKB Ultrospec Plus Spectrophotometer, using galacturonic acid (Merck) as standard (Mateos-Aparicio et al. 2010).

#### 2.4.2. Phenolic content and antioxidant capacity

The quantification of total polyphenols and the analysis of antioxidant capacity were carried out in the whole matrix through QUENCHER methods (QUick, EASY, New, CHEAP and Reproducible) (Gökmen et al. 2009), and in the extracts obtained with constant shaking at room temperature with methanol/water (50:50 v/v) and acetone/water (70:30 v/v) following Saura-Calixto and Bravo (1998) methodology. The Folin–Ciocalteu procedure was used to quantify the total extractable polyphenols (Singleton et al., 2011). The linear regression equation for polyphenols based on gallic acid (Sigma – Aldrich) standard curves ( $0.1$ – $400$  ppm) had a  $R^2 = 0.99$  with equation  $y = 0.0028x - 0.042$ . Fast Blue method (FB) was developed as follows, quinoa extract was diluted and mixed with FB salt (aqueous solution with FB reagent SIGMA®) and NaOH (Medina, 2011). The mixture was kept at room temperature for 90 min before absorbance measurement by Perkin Elmer lambda 25 spectrophotometer, against a blank solution ( $H_2O$  with FB and NaOH). Quantification was carried out using a standard curve of gallic acid ( $R^2 = 0.99$ ; with equation  $y = 0.0019x - 0.0092$ ) obtained with concentrations ranging from  $0.1$  to  $1000$  ppm in distilled water. Results were expressed as milligrams of gallic acid per gram of sample. The ferric reducing antioxidant power (FRAP) assay was used to evaluate the reducing power of samples (Pulido et al. 2000). Increases in absorbance due to the formation of a colored TPTZ –  $Fe_2^+$  complex was monitored at  $595$  nm. A Trolox (Sigma–Aldrich) standard curve ( $y = 793x + 0.011$ ,  $R^2 = 0.98$ ) was prepared using various concentrations ( $0.1$ – $1$  mmol/L).

QUENCHER assays imply a simple polyphenols analysis procedure directly determined on the ground sample (Gökmen et al. 2009). Specifically, in the QUENCHER Fast Blue method (QUENCHER – FB), the quinoa flours were weighed in centrifuge tubes, with subsequent addition of FB reagent, followed by the addition NaOH and distilled water. After 45 min of incubation in orbital shaker, the tubes were centrifuged for 25 min and filtered by  $0.45$   $\mu$ m. The absorbance was measured at

420 nm (Palombini et al. 2016). Quantification was carried out using the prepared gallic acid standard curve for FB method ( $R^2 = 0.99$ ;  $y = 0.0019x - 0.0092$ ).

Folin-Ciocalteu QUENCHER assay (QUENCHER-FC) (Del Pino et al., 2015) consisted of weighting the milled sample in centrifuge tubes and mixed with distilled water and FC reagent. Subsequently,  $\text{Na}_2\text{CO}_3$  0.7 M solution was added, and the final volume was made up with distilled water. After 35 min of incubation in orbital shaker, the tubes were centrifuged at 6500 rpm for 25 min, filtered by  $0.45 \mu\text{m}$ , and the absorbance was measured at 750 nm. The used curve was that from F-C method ( $y = 0.0028x - 0.042$ ;  $R^2 = 0.99$ ) prepared for extracts determination.

To evaluate the reducing power in the solid sample (QUENCHER-FRAP), quinoa flours were placed in centrifuge tubes with FRAP reagent, introduced in a bath at  $37^\circ\text{C}$  for 30 min, and centrifuged (10 min at 6500 rpm) before the absorbance's measurement at 595 nm (Serpen et al. 2012). The linear regression equation was based that prepared with Trolox ( $y = 793x + 0.011$ ;  $R^2 = 0.98$ ).

## 2.5. Statistical analysis

Results were analyzed using one-way analysis of variance (ANOVA), with LSD test applied post-hoc, using SPSS Statistics 24.0 software for Mac. The data were expressed as mean  $\pm$  standard deviation (SD) and differences were considered statistically significant when  $p < 0.05$ .

In addition, principal component analysis (PCA) was performed with the nutritional components (ash, dietary fiber, protein, carbohydrates, lipid), the antioxidant data (TPC and FRAP) and the colour parameter in order to highlight the factors that could be contributed to determine whether quinoa cultivars could be grouped into different classes. TPC and FRAP data were those from the extracts because they discriminated better the studied samples. PCA was carried out using R 4.0.2.

Finally, the presence of correlation among all factors was explored by the Pearson correlation coefficients using SPSS Statistics 24.0 software for Mac.

## 3. Results and discussion

### 3.1. Color of quinoa seeds and flours

The seed samples of South America were brighter as compared to their Spanish counterparts (Fig. 1), specifically the Salcedo and Kancolla cultivars, which had the highest value of  $L^*$  parameter (73.97 and 74.29, respectively). These cultivars had also a high level of  $a^*$  and  $b^*$  factors, suggesting that the color tends to yellow-reddish. The Negra variety showed the lowest values ( $p < 0.05$ ) of all color parameters, indicating a dark color. In the case of flours, all cultivars showed a decrease ( $p < 0.05$ ) in  $a^*$  and  $b^*$  parameters, except for  $a^*$  parameter in Andean Pasankalla and Andean Negra. However, an increased  $L^*$  parameter (brightness) for all flours was observed. These results indicated that the color pigments were mainly located in the pericarp and, once the pseudocereal was ground, these molecules were mixed with the endosperm's whiter particles. The absence of a predominant color in quinoa flour may be due to a color mixture (Abderrahim et al. 2015). Regardless of the origin zone, Negra and Pasankalla varieties were the darkest cultivars. The analysis of  $L^*$ ,  $a^*$ , and  $b^*$  parameters (low  $L^*$ , positive  $a^*$  and negative  $b^*$  values) indicates a very small shift on a dark-reddish shades. Results are in accordance with Escribano et al. (2017), that found a high luminosity parameter in white samples (72.04–75.06) while the black varieties had very low values for  $a^*$  and  $b^*$  with  $L^*$  ranged between 41.57 and 45.26.

### 3.2. Proximate composition and analysis of macro- and microelements

The nutritional compositions of the different cultivars of quinoa are presented in Table 1. Results are in accordance with Pellegrini et al.

(2018), which determined a moisture content ranging between 5.2 and 8.6 %. Besides, other studies have shown higher values of moisture content (9.3–14 %) (Carrasco-Valencia & Serna et al. 2011; Hemalatha et al. 2016; Perreira et al. 2019).

The main constituent of quinoa seeds was the available carbohydrates (CHO), ranged up from 55 to 77 %, and followed by protein (13–19 %) (Carrasco-Valencia et al. 2010; Carrasco-Valencia & Serna et al. 2011; Miranda et al. 2012; Nowak et al. 2016; Vilcacundo & Hernandez-Ledesma, 2017; Perreira et al. 2019). No significant differences in the CHO were observed as comparing the same varieties from different origin, except for Andean Kancolla, which featured a higher level than the Spanish one (74.9 g/100 vs 59.6 g/100 g;  $p < 0.05$ ). The Negra variety showed the lowest content for both regions. This aspect may be due to the similar environmental conditions (climate, soil, biological factors) of both fields. The cultivated areas are soils with good drainage, being all of them dry areas, and generally, with good soil quality, high nitrogen bioavailability and similar organic matter content regardless the geographical area considered. On the other hand, Farajzadeh and co-workers (2020) did not find differences of moisture and carbohydrates content among Giza1, Red Carina and Sajama quinoa cultivars, which grown in the same area (Iran).

The protein content was found similar for all varieties. Data are in accordance with Miranda et al. (2012) and Präger et al. (2018) studies, which did not find differences in protein amounts of different quinoa cultivars (Regalona, Villarrica, Ancovinto, Cansosa, Faro, Cáhuil and Puno, Titicata, Jessie, Zeno). Regarding the origin, a trend was observed suggesting a higher content in the Spanish seeds ( $p < 0.05$ ). However, Gonzalez et al. 2012 found differences in the protein content of ten quinoa cultivars grown in two different agroecological regions (the Andean highland and the Argentinean northwest).

The fat amount was similar for all different cultivars coming from the same zones, and also concerning the two regions. Results are in accordance with Vega-Galvez et al. (2010), Martin Gonzalez et al. (2014) and Wang and Zhu (2016) the fat content in quinoa was ranged 5 to 8 %.

Pasankalla and Negra varieties had the highest ( $p < 0.05$ ) dietary fiber content of all samples, with values ranging from 11 to 16 %. These values are slightly higher than those provided by Navruz-Varli & Sanlier, (2016) and Vilcacundo & Hernandez-Ledesma (2017) (7–12 %). However, Pellegrini et al. (2018) reported an amount of 13–18.6 g/100 g. According to Lamothe (2015) and Pellegrini et al. (2018), the total dietary fiber in all the analyzed samples, was mainly composed by insoluble fraction (about 70 %). Curiously, dietary fiber seems to be influenced by the cultivation area more than variety appearing the Spanish quinoa richer in this fraction as compared to Andean ones, except for Pasankalla. On the other hand, Andean quinoas had the lowest IDF/SDF relationship, indicating a more balanced dietary fiber in comparison to the Spanish ones.

The ash content of quinoa ( $2.6 \pm 0.2$  g/100 g) is similar to that obtained by Carrasco-Valencia et al. (2010), Vilcacundo & Hernandez-Ledesma (2017), Jimenez et al. (2019), Perreira et al. 2019). The cultivation zone seems to be important because the Spanish Negra and Chullpi varieties had the higher ash content as compared to the Andean ones (Table 1); this can be noted as well in macro- and microelements amounts (Fig. 2). According to literature, the main minerals were potassium, sodium and magnesium. These minerals can be encountered in their bioavailable forms and in an adequate quantity for a balanced human diet (Navruz-Varli & Sanlier, 2016; Filho et al. 2017; Vilcacundo & Hernandez-Ledesma, 2017). Potassium was the major component in all varieties, particularly in the Negra ( $193 \pm 13$  mg/100 g), Kancolla ( $193 \pm 6$  mg/100 g) and Chullpi cultivars. The Spanish Negra and Chullpi showed a high level of magnesium ( $31 \pm 1$  mg/100 g and  $24 \pm 1$  mg/100 g, respectively), while Andean Kancolla had the lowest sodium content ( $37 \pm 8$  mg/100 g). Nevertheless, other authors have reported different values for macroelements ranging 192–502 mg/100 g for magnesium and 530–1200 mg/100 g for potassium in quinoa seeds (Filho et al. 2017; Vega-Galvez et al. 2010; Wang et al. 2020).

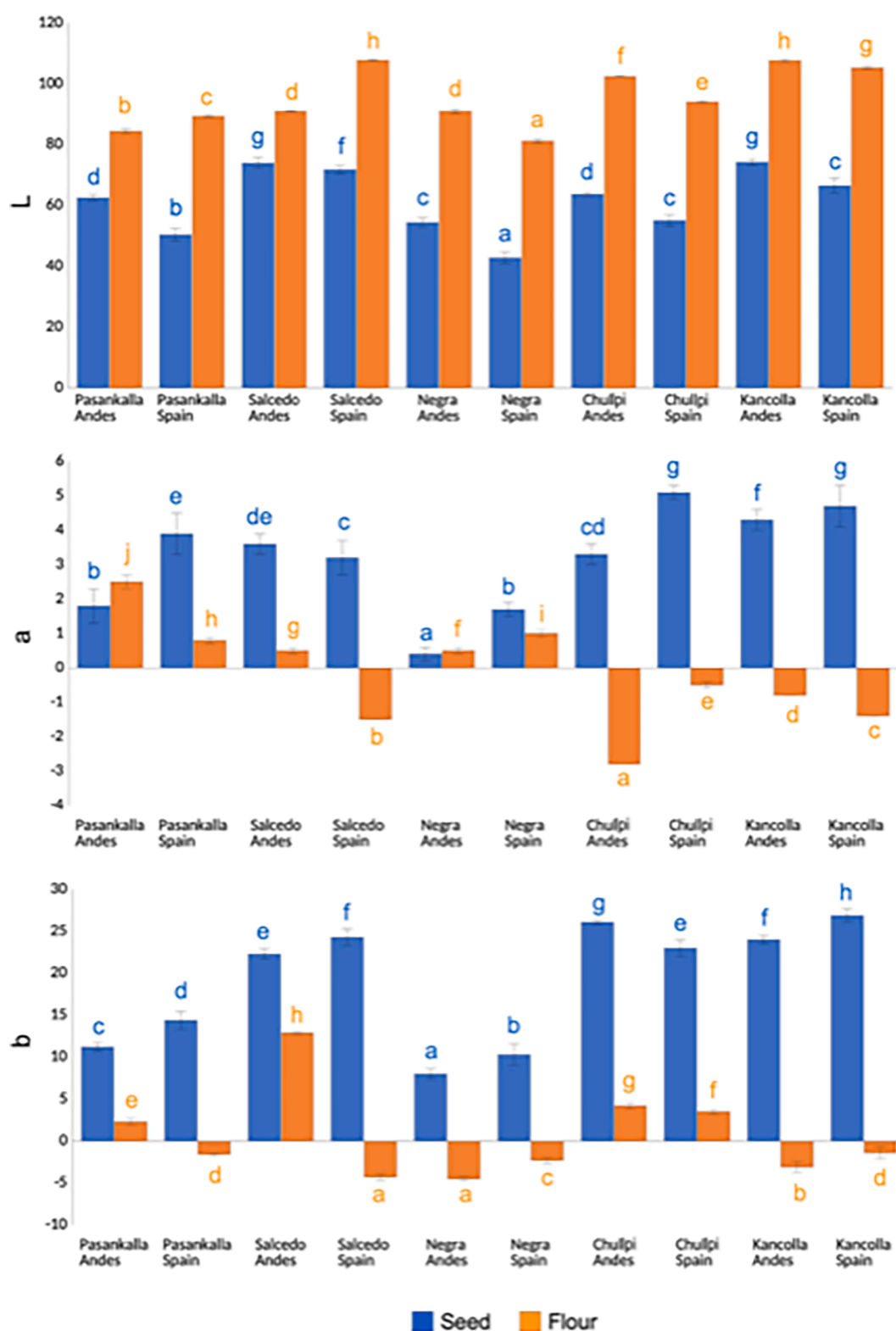


Fig. 1. Color parameters of quinoa cultivars (seeds appear in blue and the flours from seeds in orange). Values are Mean  $\pm$  SD. Different superscript letters are significantly different  $p < 0.05$ . (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

Regarding microelements, iron (7.24 mg/100 g) and zinc (5.75 mg/100 g) were the most remarkable in the Spanish varieties that showed the highest iron content. These contents are higher than in rice (1 mg/100 g), wheat (4 mg/100 g) and maize (2.17 mg/100 g) (FAO, 2011; Martín Gonzalez et al. 2014; Outi, 2014). In the case of zinc, there is no relation between the zinc content and the quinoa origin or the type of

variety. The Spanish Kancolla and Andean Pasankalla were the varieties with the highest levels of zinc. Thus, 6–20 g/day of quinoa might cover the recommended zinc's intake (8–14 mg/day) (FAO, 2011). Copper is similar in all varieties with a range of 0.7–2.2 mg/100 g, being the lowest content in the Andean Kancolla and Chulpi, and the highest in Andean Salcedo. However, manganese is greater ( $p < 0.05$ ) in the



**Table 1**

Nutritional profile of the different varieties of quinoa (g/100 g dry matter).

SAMPLES	MOISTURE	LIPID	PROTEIN	CHO	Dietary fiber			ASH
					IDF	SDF	TDF	
Negra Andes	8.3 ± 0.3 <sup>d</sup>	7.7 ± 0.0 <sup>c</sup>	14.6 ± 0.6 <sup>ab</sup>	57.2 ± 4.7 <sup>a</sup>	9.0 ± 0.4 <sup>cd</sup>	4.1 ± 0.9 <sup>d</sup>	12.6 ± 0.9 <sup>e</sup>	2.8 ± 0.4 <sup>c</sup>
Negra Spain	6.8 ± 0.1 <sup>c</sup>	5.8 ± 0.1 <sup>ab</sup>	15.9 ± 0.3 <sup>abcd</sup>	54.3 ± 3.9 <sup>a</sup>	12.4 ± 0.5 <sup>e</sup>	3.4 ± 0.5 <sup>cd</sup>	15.9 ± 1.0 <sup>f</sup>	4.2 ± 0.1 <sup>d</sup>
Chullpi Andes	6.3 ± 0.1 <sup>ab</sup>	6.0 ± 0.3 <sup>ab</sup>	15.5 ± 4.3 <sup>abcd</sup>	78.4 ± 7.0 <sup>c</sup>	6.1 ± 0.6 <sup>b</sup>	1.5 ± 0.4 <sup>ab</sup>	7.7 ± 0.8 <sup>a</sup>	2.5 ± 0.4 <sup>bc</sup>
Chullpi Spain	6.1 ± 0.3 <sup>a</sup>	4.9 ± 0.1 <sup>a</sup>	16.8 ± 0.9 <sup>abcd</sup>	69.5 ± 9.4 <sup>bc</sup>	8.7 ± 0.8 <sup>c</sup>	1.3 ± 0.4 <sup>a</sup>	10.2 ± 0.5 <sup>cd</sup>	3.9 ± 0.1 <sup>d</sup>
Salcedo Andes	6.8 ± 0.1 <sup>bc</sup>	6.9 ± 0.1 <sup>bc</sup>	14.0 ± 0.3 <sup>ab</sup>	71.6 ± 6.8 <sup>c</sup>	4.8 ± 0.2 <sup>a</sup>	3.8 ± 0.5 <sup>d</sup>	8.6 ± 0.7 <sup>ab</sup>	1.8 ± 0.5 <sup>ab</sup>
Salcedo Spain	7.9 ± 0.2 <sup>d</sup>	5.9 ± 0.1 <sup>ab</sup>	18.8 ± 0.9 <sup>d</sup>	69.3 ± 1.7 <sup>bc</sup>	6.5 ± 0.3 <sup>b</sup>	2.5 ± 0.8 <sup>bc</sup>	9.0 ± 1.1 <sup>abc</sup>	1.6 ± 0.0 <sup>a</sup>
Pasankalla Andes	6.1 ± 0.3 <sup>a</sup>	5.7 ± 1.7 <sup>ab</sup>	15.3 ± 0.3 <sup>abc</sup>	75.9 ± 2.3 <sup>c</sup>	10.0 ± 1.6 <sup>d</sup>	3.9 ± 1.4 <sup>d</sup>	13.9 ± 0.6 <sup>e</sup>	2.3 ± 0.4 <sup>abc</sup>
Pasankalla Spain	6.1 ± 0.3 <sup>a</sup>	6.0 ± 1.2 <sup>ab</sup>	15.9 ± 2.9 <sup>abcd</sup>	77.5 ± 10.9 <sup>c</sup>	9.4 ± 0.8 <sup>cd</sup>	1.7 ± 0.8 <sup>ab</sup>	11.1 ± 1.3 <sup>d</sup>	2.9 ± 0.1 <sup>c</sup>
Kancolla Andes	7.1 ± 0.3 <sup>c</sup>	5.0 ± 0.2 <sup>a</sup>	13.1 ± 0.3 <sup>a</sup>	74.9 ± 2.8 <sup>c</sup>	5.3 ± 0.5 <sup>ab</sup>	3.3 ± 0.9 <sup>cd</sup>	8.7 ± 0.5 <sup>ab</sup>	1.7 ± 0.1 <sup>a</sup>
Kancolla Spain	7.0 ± 0.0 <sup>c</sup>	7.0 ± 0.8 <sup>bc</sup>	17.9 ± 2.5 <sup>cd</sup>	59.6 ± 1.2 <sup>ab</sup>	6.2 ± 0.4 <sup>b</sup>	3.7 ± 0.4 <sup>cd</sup>	9.9 ± 0.2 <sup>bcd</sup>	2.3 ± 0.5 <sup>abc</sup>

CHO: total carbohydrates; IDF: insoluble dietary fiber; SDF: soluble dietary fiber; TDF: total dietary fiber.

Data in the same column with different superscript letters are significantly different  $p < 0.05$ .

Andean varieties but Chullpi variety. In general, quinoa appears to be a good source of minerals, with a higher content than traditional flour (Ando et al. 2002; Miranda et al. 2012; Navruz-Varli & Sanlier, 2016; Wang et al. 2020). Despite some minerals, microelements, seem to be influenced by the quinoa variety and the cultivation area, the latter being a key factor of mineral content. According to Vega-Galvez et al. (2010) some mineral elements appear to be largely influenced by the variety while others were more sensitive to the environment's conditions, soil type and/or different applied fertilizers. Therefore, the different mineral composition identified among the samples may be due to the mineral composition of soils.

### 3.3. Functional components

#### 3.3.1. Dietary fiber composition

The monomeric composition of dietary fiber was analyzed (Table 2). For all samples, the insoluble dietary fiber (IDF) was more than 65 % of the TDF. The most important monosaccharides of IDF were glucose, arabinose and uronic acids. Results are in accordance to Zhu (2020) who studied the dietary fiber composition of pseudocereals, and, in quinoa, he determined the presence of the same sugars in IDF. The composition indicated the presence of cellulose, homogalacturonans and arabinans, as the most important polysaccharides in the insoluble fraction. The association of pectic material with cellulose indicated the high ramification degree of that material, which implies its insolubility (Mateos-Aparicio et al., 2010). Lamothe et al. (2015) proposed a similar polysaccharide composition for IDF. However, they founded that the pectic polysaccharides are the dominant polymers. The present study identified as cellulose, although the glucose and uronic acids contents were very similar, especially in Kancolla and Salcedo, independently of origin, and the Andean Negra.

Negra cultivars in both, Spanish and Andean area, showed the highest content of the mentioned monomers, while the lowest glucose and uronic acids values were found in Salcedo and Kancolla cultivar. However, arabinose element, was quantified in minor amounts in Andean Chullpi and Spanish Pasankalla. Regarding SDF, Negra cultivars had the greatest amount, mainly due to glucose, uronic acids and arabinose. Andean Pasankalla had the highest ( $p < 0.05$ ) level of uronic acids, while all the other cultivars did not show differences based on their cultivation area. According to Lamothe (2015), xylose appears in low amount and, galacturonic acid and arabinose are the main monomers in quinoa SDF as we observed. For Negra cultivars, glucose was the most important monomer, and had a remarkable xylose content as compared to the others. Therefore, this composition suggest that SDF is mainly composed of pectic polysaccharides, namely, homogalacturonan and arabinan, except Negra that also has xyloglucans. On the other hand, mannose and galactose were found in slight contents or not detected depending on variety, so galactomannan may be in a small quantity or does not appear.

#### 3.3.2. Polyphenols and antioxidant capacity

The quinoa varieties were assayed for total extractable polyphenol content (TPC) using the Fast Blue (FB) and Folin (F-C) methods and antioxidant capacity using the FRAP assay. These methods were carried out directly in the flours (QUENCHER), and also, in the polyphenolic extracts (Table 3).

In all these methods, the QUENCHER values were higher ( $p < 0.05$ ) than those from the polyphenol extracts as it was expected. Results could be explained because some phenolics are covalently bound to the cell wall structural components, e.g., cellulose, hemicellulose, lignin (Mullari et al. 2018), and thus, they cannot be totally extracted by the used solvents. Consequently, the antioxidant capacity measured by based-extracts traditional procedures might be underestimated. For this reason, a direct procedure (QUENCHER), which does not require a previous extraction could be more accurately estimating the antioxidant capacity of the samples.

Considering the quinoa varieties and their location, the polyphenols content did not change among the two locations for all the samples in the QUENCHER F-C assay (Table 3). However, the extracts showed differences among location and cultivar, being the highest values for Kancolla, Negra and Pasankalla. These results were in accordance with Abderrahim et al. (2015) and Balakrishnan and Schneider (2020), who showed TPC values comprised between 1.23 and 3.41 mg GAE/g sample. Regarding QUENCHER FB assay, TPC did not change for Chullpi and Kancolla considering the two cultivation zones. However, notable differences in TPC from extracts were detected in all samples. Negra and Pasankalla samples, independently from the growing site and the determinations (QUENCHER or extracts), showed the highest TPC. These results are in agreement with those presented by Tang et al. (2015) and Hemalatha et al. (2016), who detected a higher TPC in dark quinoa seeds. Thus, the influence of variety seems to be more important than origin zone regarding polyphenol content presented in quinoa. There is a great variability in TPC among type of methodology, F-C and FB, and the performance, QUENCHER and extracts. TPC was higher when determined with FB than with F-C in all the extracts. Regarding the QUENCHER method, greater TPC values were reported as compared to extracts. However, TPC from F-C method was not higher for all the samples. Indeed, Negra samples and Andean Pasankalla presented major TPC determined with FB method. Therefore, it seems that the usual interferences of non-phenolic compounds with the F-C reagent are not so important in the studied quinoa samples than in other vegetable samples, in which the capacity of the diazonium group from Fast-Blue salt to specifically couple with phenolic hydroxyl groups can give an accurate TPC determination (Medina, 2011).

Regarding QUENCHER-FRAP, the Spanish samples presented greater values for FRAP in QUENCHER and extracts, except for Andean Kancolla and Pasankalla, which have higher antioxidant capacity than the Spanish ones. Andean Pasankalla and Kancolla, and Spanish Negra were the varieties with the highest FRAP. These results were in accordance

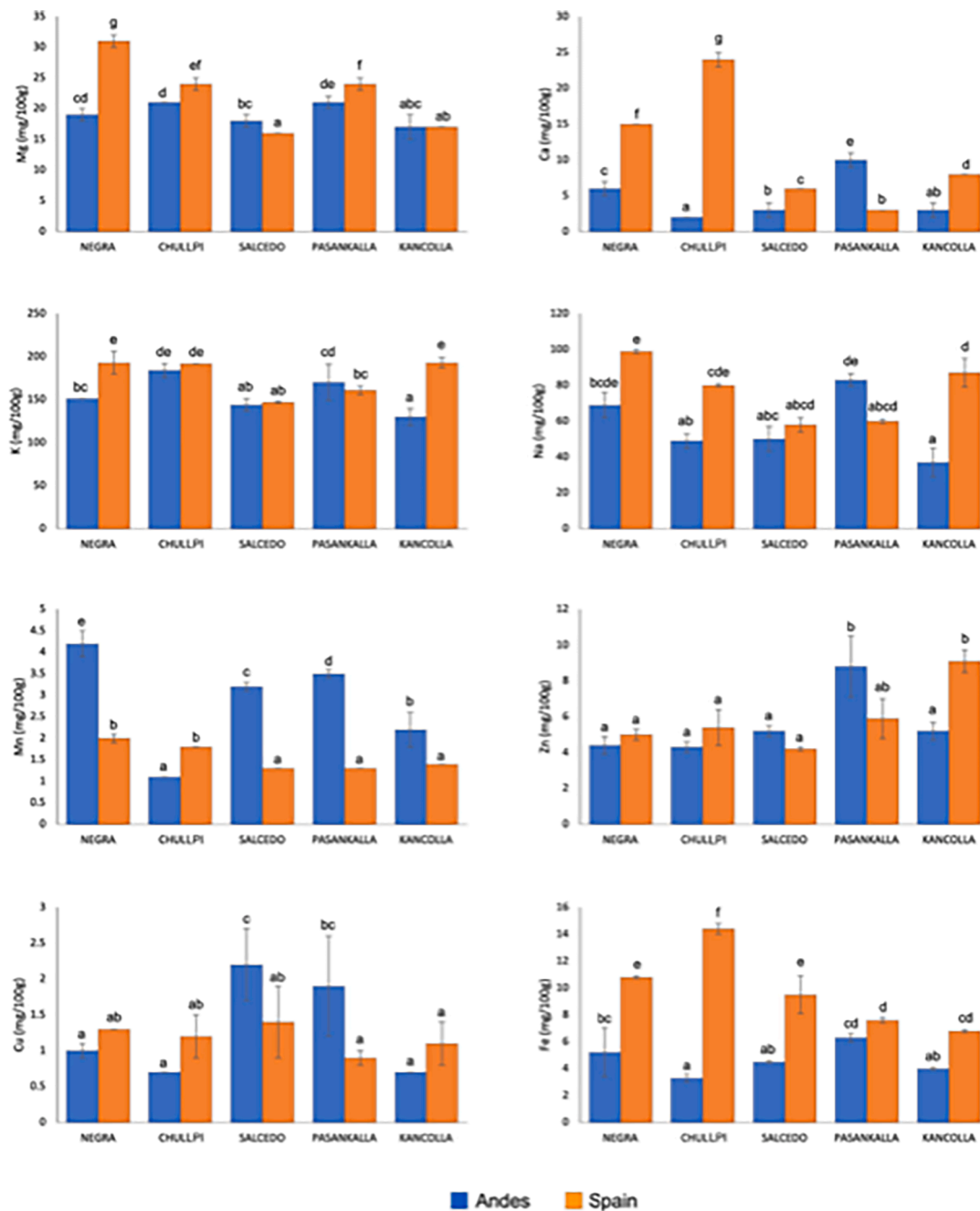


Fig. 2. Minerals of quinoa (mg/100 g). Figure reported the macroelements sodium (Na), potassium (K), calcium (Ca) and magnesium (Mg) and the microelements manganese (Mn), zinc (Zn), copper (Cu) and iron (Fe). Values are Mean  $\pm$  SD. Different superscript letters are significantly different  $p < 0.05$ .

with those showed by Pellegrini et al. (2018), who found a higher antioxidant capacity in darker quinoa seeds. There seems to be no correlation between polyphenols and antioxidant capacity in Chullpi and Kancolla cultivars. Therefore, the antioxidant capacity measured as FRAP might be related to the type of polyphenols and the different content of them in the samples. Furthermore, they may be less sensible to FRAP colorimetric reaction as well (Hemalatha et al. 2016).

### 3.4. Principal component analysis

Principal component analysis (PCA) was applied to observe any possible cluster within analyzed quinoa samples; it was performed including colour parameters, polyphenol content, antioxidant capacity, and nutritional components (Fig. 3). The first two principal components

could explain 65.3 % of total variance (PC1 = 43.54 % and PC2 = 21.76 %, respectively). PCA graph (Fig. 3) revealed that the quinoa cultivars with high antioxidant capacity, polyphenols content and dietary fiber were located to the right in the score plot, whereas seed samples with low bioactive compounds were situated at the left in the diagram. Furthermore, darker crops seem rightward positioned in the score plot while seed with higher  $L^*$  value were located on the west side.

Two quinoa varieties, Negra and Pasankalla, showed a longer distance from other cultivars. Both had the highest values of FRAP, TPC, and dietary fiber, due to the insoluble fraction, and resulted the darkest samples as well. The Kancolla, Salcedo and Chullpi cultivars appear in the same region of the biplot relatively close and separated from the other two due to their high  $L^*$  value and low content of functional compounds.

**Table 2**

Monomeric composition of insoluble (IDF) and soluble dietary fiber (SDF) of the different quinoa varieties (g/100 g dry matter).

Monomer	Fraction	Negra Andes	Negra Spain	Chullpi Andes	Chullpi Spain	Salcedo Andes	Salcedo Spain	Pasankalla Andes	Pasankalla Spain	Kancolla Andes	Kancolla Spain
Arabinose	IDF	1.7 ± 0.12 <sup>de</sup>	1.9 ± 0.2 <sup>e</sup>	1.1 ± 0.3 <sup>ab</sup>	1.6 ± 0.1 <sup>cde</sup>	1.3 ± 0.1 <sup>bc</sup>	1.9 ± 0.4 <sup>e</sup>	1.6 ± 0.1 <sup>cd</sup>	1.0 ± 0.1 <sup>a</sup>	1.4 ± 0.0 <sup>bc</sup>	1.7 ± 0.1 <sup>de</sup>
	SDF	0.5 ± 0.0 <sup>e</sup>	0.4 ± 0.1 <sup>d</sup>	0.3 ± 0.1 <sup>bc</sup>	0.2 ± 0.0 <sup>bc</sup>	0.2 ± 0.0 <sup>cd</sup>	0.2 ± 0.0 <sup>ab</sup>	0.2 ± 0.0 <sup>abc</sup>	0.1 ± 0.0 <sup>a</sup>	0.3 ± 0.0 <sup>bc</sup>	0.3 ± 0.0 <sup>bc</sup>
Xylose	IDF	0.2 ± 0.0 <sup>ab</sup>	0.5 ± 0.0 <sup>ef</sup>	0.1 ± 0.0 <sup>a</sup>	0.4 ± 0.0 <sup>de</sup>	0.2 ± 0.0 <sup>ab</sup>	0.2 ± 0.0 <sup>bc</sup>	0.4 ± 0.0 <sup>de</sup>	0.3 ± 0.1 <sup>cd</sup>	0.2 ± 0.1 <sup>ab</sup>	0.6 ± 0.3 <sup>f</sup>
	SDF	0.5 ± 0.0 <sup>g</sup>	0.1 ± 0.0 <sup>bc</sup>	0.2 ± 0.1 <sup>e</sup>	0.2 ± 0.0 <sup>d</sup>	0.1 ± 0.0 <sup>f</sup>	nd	0.1 ± 0.0 <sup>c</sup>	nd	nd	nd
Mannose	IDF	0.1 ± 0.0 <sup>a</sup>	0.4 ± 0.1 <sup>c</sup>	0.8 ± 0.0 <sup>a</sup>	0.7 ± 0.0 <sup>b</sup>	nd	0.1 ± 0.0 <sup>a</sup>	0.3 ± 0.3 <sup>b</sup>	nd	0.3 ± 0.0 <sup>b</sup>	0.7 ± 0.5 <sup>b</sup>
	SDF	1.0 ± 0.0 <sup>c</sup>	0.2 ± 0.0 <sup>a</sup>	0.3 ± 0.0 <sup>b</sup>	nd	0.4 ± 0.0 <sup>b</sup>	nd	0.2 ± 0.0 <sup>a</sup>	nd	nd	nd
Galactose	IDF	0.3 ± 0.0 <sup>de</sup>	0.4 ± 0.0 <sup>h</sup>	0.1 ± 0.0 <sup>ab</sup>	0.3 ± 0.0 <sup>fg</sup>	0.3 ± 0.0 <sup>ef</sup>	0.2 ± 0.2 <sup>bc</sup>	0.3 ± 0.2 <sup>g</sup>	0.1 ± 0.0 <sup>a</sup>	0.2 ± 0.0 <sup>cd</sup>	0.3 ± 0.1 <sup>g</sup>
	SDF	0.2 ± 0.0 <sup>f</sup>	nd	0.1 ± 0.0 <sup>cd</sup>	nd	0.1 ± 0.0 <sup>e</sup>	0.1 ± 0.0 <sup>e</sup>	0.1 ± 0.0 <sup>de</sup>	nd	nd	0.1 ± 0.0 <sup>cd</sup>
Glucose	IDF	2.2 ± 0.2 <sup>c</sup>	3.4 ± 0.3 <sup>e</sup>	2.5 ± 0.1 <sup>c</sup>	2.4 ± 0.4 <sup>c</sup>	1.2 ± 0.0 <sup>a</sup>	1.2 ± 0.1 <sup>a</sup>	3.0 ± 0.2 <sup>d</sup>	1.7 ± 0.1 <sup>b</sup>	1.1 ± 0.1 <sup>a</sup>	1.4 ± 0.3 <sup>a</sup>
	SDF	1.6 ± 0.4 <sup>cd</sup>	2.2 ± 0.3 <sup>e</sup>	nd	0.1 ± 0.1 <sup>a</sup>	0.6 ± 0.1 <sup>bc</sup>	0.4 ± 0.0 <sup>ab</sup>	0.3 ± 0.0 <sup>a</sup>	0.2 ± 0.0 <sup>a</sup>	0.1 ± 0.0 <sup>a</sup>	0.4 ± 0.1 <sup>de</sup>
Uronic acids	IDF	2.1 ± 0.1 <sup>f</sup>	2.5 ± 0.0 <sup>g</sup>	1.1 ± 0.1 <sup>b</sup>	1.5 ± 0.0 <sup>d</sup>	1.4 ± 0.0 <sup>c</sup>	1.0 ± 0.0 <sup>a</sup>	1.8 ± 0.0 <sup>e</sup>	1.4 ± 0.0 <sup>c</sup>	1.0 ± 0.0 <sup>ab</sup>	1.4 ± 0.0 <sup>c</sup>
	SDF	0.4 ± 0.0 <sup>abc</sup>	0.5 ± 0.0 <sup>ab</sup>	0.5 ± 0.4 <sup>a</sup>	0.5 ± 0.1 <sup>abc</sup>	0.9 ± 0.0 <sup>d</sup>	0.7 ± 0.0 <sup>cd</sup>	1.3 ± 0.0 <sup>e</sup>	0.7 ± 0.0 <sup>bcd</sup>	0.6 ± 0.1 <sup>abcd</sup>	0.9 ± 0.0 <sup>d</sup>

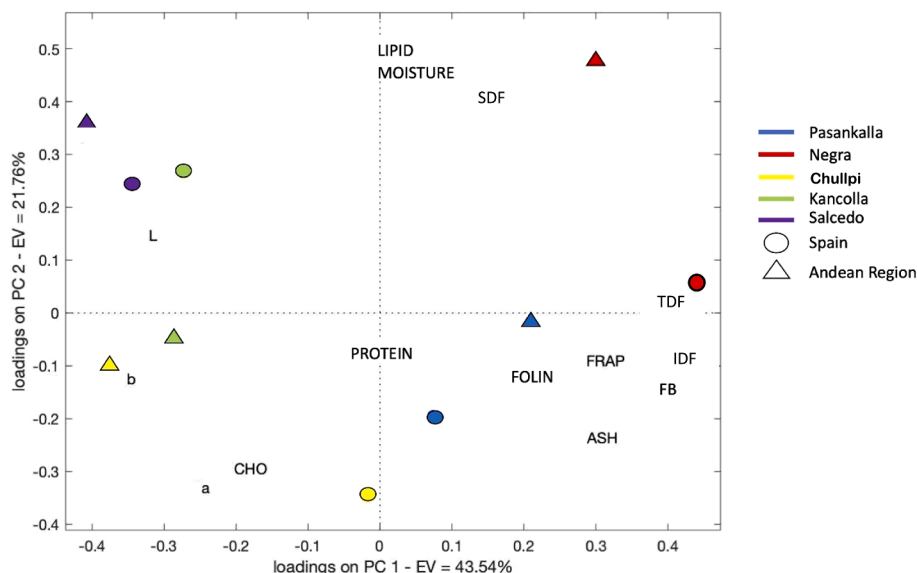
Rhamnose and fucose were not detected in any sample.

Data in the same row with different superscript letters are significantly different for  $p < 0.05$ . nd = not detected.

**Table 3**

Total polyphenols content (TPC) and ferric reducing antioxidant power (FRAP).

Samples	Folin (mg gallic acid /g)		Fast Blue (mg gallic acid/g)		FRAP (mg Trolox/g)	
	Quencher	Extract	Quencher	Extract	Quencher	Extract (*10 <sup>-2</sup> )
Negra Andes	119.6 ± 4.7 <sup>bc</sup>	2.3 ± 0.0 <sup>c</sup>	164.5 ± 27.2 <sup>d</sup>	26.5 ± 0.7 <sup>f</sup>	0.9 ± 0.1 <sup>c</sup>	1.65 ± 0.1 <sup>b</sup>
Negra Spain	121.9 ± 2.8 <sup>bcd</sup>	2.7 ± 0.1 <sup>e</sup>	127.6 ± 10.6 <sup>c</sup>	43.5 ± 0.2 <sup>i</sup>	1.1 ± 0.0 <sup>d</sup>	2.24 ± 0.5 <sup>c</sup>
Chullpi Andes	128.8 ± 17.4 <sup>cd</sup>	1.8 ± 0.0 <sup>a</sup>	119.9 ± 12.3 <sup>c</sup>	11.1 ± 1.2 <sup>a</sup>	0.6 ± 0.0 <sup>a</sup>	0.52 ± 0.06 <sup>a</sup>
Chullpi Spain	137.3 ± 16.6 <sup>d</sup>	2.5 ± 0.1 <sup>d</sup>	114.9 ± 3.4 <sup>c</sup>	26.3 ± 2.0 <sup>f</sup>	1.1 ± 0.0 <sup>d</sup>	1.47 ± 0.3 <sup>b</sup>
Salcedo Andes	104.3 ± 4.9 <sup>ab</sup>	2.0 ± 0.1 <sup>b</sup>	43.9 ± 2.8 <sup>a</sup>	13.6 ± 0.8 <sup>b</sup>	0.5 ± 0.0 <sup>a</sup>	0.46 ± 0.04 <sup>a</sup>
Salcedo Spain	95.4 ± 2.1 <sup>a</sup>	2.2 ± 0.1 <sup>c</sup>	66.1 ± 6.3 <sup>b</sup>	15.4 ± 0.3 <sup>c</sup>	0.6 ± 0.0 <sup>a</sup>	0.65 ± 0.3 <sup>a</sup>
Pasankalla Andes	115.7 ± 9.2 <sup>abc</sup>	2.8 ± 0.1 <sup>e</sup>	178.5 ± 13.3 <sup>d</sup>	37.9 ± 1.2 <sup>h</sup>	1.6 ± 0.0 <sup>f</sup>	3.06 ± 0.18 <sup>d</sup>
Pasankalla Spain	119.3 ± 0.9 <sup>bcd</sup>	2.3 ± 0.0 <sup>c</sup>	108.3 ± 4.7 <sup>c</sup>	33.8 ± 0.2 <sup>g</sup>	0.9 ± 0.0 <sup>c</sup>	1.47 ± 0.03 <sup>b</sup>
Kancolla Andes	122.4 ± 0.3 <sup>bcd</sup>	3.3 ± 0.0 <sup>f</sup>	57.6 ± 8.1 <sup>ab</sup>	18.9 ± 1.2 <sup>d</sup>	1.3 ± 0.0 <sup>e</sup>	2.49 ± 0.5 <sup>c</sup>
Kancolla Spain	118.6 ± 1.7 <sup>bcd</sup>	2.2 ± 0.1 <sup>c</sup>	54.4 ± 1.5 <sup>ab</sup>	22.8 ± 0.4 <sup>e</sup>	0.7 ± 0.0 <sup>b</sup>	1.45 ± 0.1 <sup>b</sup>

Data in the same column with different superscript letters are significantly different for  $p < 0.05$ .**Fig. 3.** Principal component analysis (PCA) biplot of data from antioxidant capacity, total amounts of phenolics, proximal composition and color parameters of five quinoa varieties. Note: CHO (carbohydrates); IDF (insoluble dietary fiber); SDF (soluble dietary fiber); TDF (total dietary fiber); FB (fast blue); FRAP (ferric reducing antioxidant power). (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

The PCA ordination was confirmed by Pearson correlation results shown as [supplementary material \(Table S2\)](#). Pearson coefficients showed a strong positive correlation between the phenolic compounds and the antioxidant capacity ( $p < 0.01$ ), as well as a negative correlation between phenolic compounds, antioxidant capacity and  $L^*$  values ( $p < 0.01$ ); these results are in accordance with [Abderrahim et al. \(2015\)](#) and [Hemalatha et al. \(2016\)](#). [Liu et al. \(2020\)](#) and [Escribano et al. \(2017\)](#) found that darker quinoa seeds had high TPC values.

Results are supported by the correlation analysis ( $p < 0.05$ ; [Table S2](#)) suggesting that an improvement in phenolic compounds and functional properties is accompanied by an increase in pigments content; probably attributed to betalains and/or betacyanin. ([Abderrahim et al. 2015](#); [Tang et al. 2015](#); [Wang and Zhu, 2016](#)).

Finally, a high positive association was found between total phenolics and total and insoluble dietary fiber ( $p < 0.01$ ). This result can express the presence of polyphenols compound which are covalently linked with same sugar of fiber and that an improvement in phenolic compounds was linked with a high fiber content.

#### 4. Conclusion

This work evaluated the nutritional and antioxidant properties of five quinoa varieties cultivated in the Andean region and Spain. It emerged that the major differences so nutritional composition as bioactive compounds in quinoa seeds seems to be attributable to the

genetic diversity instead to geographic factors. The effects of genetic diversity seem to have a higher impact on the nutritional composition of the seeds grown in the same geographical location; while the protein and iron content appear influenced by cultivation area. In fact, the value of these parameters was found higher in Spanish cultivars. Regarding Negra and Pasankalla cultivars, the darkest quinoa samples, had a better functional profile (high content of polyphenols, antioxidant capacity and amount of dietary fiber). Thereby, PCA distribution and Pearson correlation coefficient reveal that the most pigmented seeds appear as the best bioactive quinoa. More studies are required with the aim of analyzing others quinoa cultivars ground in different location to explore the complex interactions between genotypic properties and environmental factors.

#### CRedit authorship contribution statement

**D. Pedrali:** Formal analysis, Investigation, Methodology, Writing – original draft. **L. Giupponi:** Formal analysis, Data curation. **R. De la Peña-Armada:** Formal analysis, Writing – review & editing. **M.J. Villanueva-Suárez:** Conceptualization, Methodology, Validation, Investigation. **I. Mateos-Aparicio:** Conceptualization, Validation, Writing – review & editing, Supervision.



## Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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## Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.foodchem.2022.133531>.

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