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ORIGINAL ARTICLE

Evaluation of phenolic antioxidant-linked in vitro bioactivity of Peruvian corn (*Zea mays* L.) diversity targeting for potential management of hyperglycemia and obesity

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Abstract Peruvian corn biodiversity is one of the highest in the world and may represent an important natural source of health relevant phenolic bioactive compounds whose potential needs to be investigated. This study investigated twenty-two Peruvian corn samples corresponding to five corn races (Arequipeño, Cabanita, Kculli, Granada and Coruca) in relation to their total phenolic contents (TPC), anthocyanin contents, Ultra-Performance Liquid Chromatography (UPLC) phenolic profiles and antioxidant capacity (ABTS and ORAC methods). Subsequently using both free and cell-wall bound phenolic fractions their health relevance targeting hyperglycemia (α-glucosidase and α-amylase inhibition) and obesity (lipase inhibition) potentials was evaluated using in vitro assay models. Antioxidant capacity and TPC were high in bound fractions from yellow-colored races in contrast to the purple-colored race (Kculli) which had high TPC (mainly anthocyanins) and antioxidant capacity in the free form. The major phenolic acids detected by UPLC were ferulic and *p*-coumaric acids. High α-glucosidase (32.5–76.1%, 25 mg sample dose) and moderate α-amylase inhibitory activities (13.6–29.0%, 250 mg sample dose) were found in all free fractions, but only samples from the Kculli race had lipase inhibitory activity (58.45–92.16%, 12.5 mg sample dose). Principal component analysis revealed that the variability of data was affected by the race and the α-glucosidase and lipase inhibitory activities positively correlated with anthocyanins and antioxidant capacity. Some accessions of Kculli, Granada and Cabanita races are promising for future breeding strategies focused on the development of improved corn varieties targeted for the design of functional foods relevant for hyperglycemia and obesity prevention.

Keywords *Zea mays* L. · Phenolic compounds · Antihyperglycemia · Antiobesity · Antioxidant

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Introduction

Corn (*Zea mays* L.) diversity of Peru ranks second highest in the world after Mexico and surrounding region (Serratos 2009). This is due to the result of its planned and targeted selection and cultivation since Pre-Columbian times by native farmers along with the presence of several ecological niches in the Andean and adjoining regions near Peru (Sevilla and Chura 1999). The genetic diversity of Peruvian corn is distributed in 52 native races and originally around 3931 accessions have been collected from different Peruvian regions especially from the Andean area (Sevilla and Chura 1999). However, the food and health relevant scientific knowledge of this genetic diversity is still unknown

and the original collected corn germplasm generally remains in germplasm banks without a complete integrative study of its molecular, nutritional, bioactive and agronomic potential. This is leading to the genetic erosion of certain Peruvian corn races compromising the advance of local agriculture and the possibility to identify genes that can be valuable for national and international breeding initiatives. The characterization of Peruvian corn diversity is critical for further identification of superior native varieties with potential for their use in future breeding programs and especially focused on the development of improved varieties integrating important health-relevant properties along with high-value agronomic traits relevant for food security and with resilience to climate change. The study of genetic diversity in crops can be targeted through phenotypic and molecular characterization, which then provide options for the breeders to develop new agronomically valuable varieties and hybrids (Govindaraj et al. 2015).

The rise of dietary-linked chronic diseases such as Type-2 diabetes is increasing the demand for healthier plant-based foods and natural ingredients based on local food diversity. Corn is an important source of health-relevant bioactive compounds in several regions of the world, including Andean region that needs to be investigated for further breeding and agronomic needs as it is essential for improved diets for specific locally affected regions. Previous studies mostly originating from Mexico and the United States have shown that corn kernels are important sources of bioactive constituents such as phenolics and carotenoids (Lopez-Martínez et al. 2009; Zilic et al. 2012). Further, few reports have shown that certain samples from Chile and Bolivia may be relevant sources of phenolic bioactives with antioxidant and anti-hyperglycemic potential (González-Muñoz et al. 2013; Montilla et al. 2011). Studies on the bioactive composition of Peruvian corn along with its health-relevant potential have been more focused on pigmented varieties such as purple corn, which is high in anthocyanin contents. The phenolic profile along with the antioxidant and antihypertensive effect using in vitro and in vivo models have been previously reported in Peruvian purple corn kernels and cobs (Ramos-Escudero et al. 2012). However, the potential of wider Peruvian corn diversity as a source of bioactives such as phenolic compounds and its associated health relevant functional properties needs further evaluation to support the design of food ingredients and to advance breeding strategies for developing human health targeted corn cultivars with superior phenolic bioactive profiles. Such health-relevant corn cultivars can be targeted as dietary support against chronic hyperglycemia and oxidation-linked complications commonly associated with type 2 diabetes (Bhandari et al. 2008). This knowledge would also be relevant to advance important food technology applications as those investigated in other countries such as India and Mexico (Thakur et al. 2017; Amador-Rodríguez et al. 2019).

Common therapeutic approaches for treating early stages of hyperglycemia involve the use of pharmacologically relevant inhibitors of enzymes related with the digestion of carbohydrates such as α-amylase and α-glucosidase which then prevent the postprandial increase of blood glucose after the intake of a mixed carbohydrate diet (Ademiluyi and Oboh 2013). Inhibitors of pancreatic lipase have also been targeted for obesity treatment by decreasing triglycerides digestion and absorption (De la Garza et al. 2011). Common pharmacological enzyme inhibitors have been associated with harmful side effects compared to the use of natural inhibitors from plant-based sources and therefore represent an important food-grade dietary support and complimentary strategy for targeting prevention of hyperglycemia and obesity (Filippatos et al. 2008).

Functional analysis (transcriptomics, proteomics and metabolomics) linked to genomics provide integrative phenotypic and molecular characterizations of crop genetic diversity providing information of its agricultural potential (Zivy et al. 2015). The current study is aimed at the important initial characterization of a group of land races from Peruvian corn diversity from a specific Andean area (Arequipa region) by applying metabolically focused health-relevant targeting and analysis of phenolic bioactives. Therefore in this investigation the composition of the free and dietary fiber-bound phenolic fractions and total anthocyanin contents relevant for the their antioxidant capacity and inhibitory activities against key enzymes relevant to hyperglycemia (α-amylase and α-glucosidase inhibition) and obesity (lipase inhibition) management using in vitro assay models was undertaken in 22 Peruvian originating corn samples corresponding to 5 native races. These functional variables were analyzed and compared in two sample groups: 12 corn accessions provided by a germplasm bank (originally collected from the Arequipa region) and 10 corn samples collected in situ from the same Peruvian region. Results from current initial study is important and would be integrated to information from future investigations based on the application of functional genomics for an integrative characterization of Peruvian corn diversity. This would help to target and advance promising corn races for future breeding strategies for potential diet-based functional food designs and industrial applications.

Materials and methods

Materials

Representative samples of 200–300 g (mature dried kernels) corresponding to 12 corn accessions were supplied by the Maize Research Program (germplasm bank located at Agrarian University of La Molina-UNALM, Lima-Peru). These accessions corresponded to 4 native races (Arequipeño, Kculli, Cabanita, and Coruca) from the region of Arequipa-Peru. In addition, 10 corn samples (~ 300 g, dried and mature kernels) corresponding to same above races but including the Granada race instead the Coruca were directly collected in situ in August 2015 from several Andean locations from Arequipa-Peru. The corn kernel samples were ground with a lab scale hammer mill followed by a disk mill to a powdered sample. The flours were classified with a 32 mesh sieve (500 µm) and stored at − 20 °C until next assays. Sample characteristics related to their race classification, origin and nomenclature are shown in Supplementary Table S1. Sample 8 from the germplasm group (accession Areq-084) was previously evaluated for its phenolic composition and antimicrobial activity (Ranilla et al. 2017) and was considered in the current study for the comparison reasons.

Enzymes

Baker α-glucosidase from yeast (EC 3.2.1.20), α-amylase (EC 3.2.1.1) and lipase (EC 3.1.1.3) from porcine pancreas were from Sigma Chemical Co. (St. Louis, MO).

Color characterization

The CIELAB parameters (L^*, a^*, and b^*) of all corn samples (whole kernels) were measured using a Minolta CR-400 Chroma meter (Minolta Camera Co., Japan) with an observer angle of 0° and a light source of D-65. The Hue angle (H^*) and Chroma value (C^*) parameters were also calculated according to González-Muñoz et al. (2013). Twelve measurements were replicated per sample.

Extraction of the free and bound phenolic fractions

Free and bound phenolic fractions were extracted according to the method reported by González-Muñoz et al. (2013) and modified by Ranilla et al. (2017). Free phenolic compounds were extracted using methanol/acetone/water (45:45:10, v/v/v; 0.1% HCl) whereas bound phenolic compounds were released by alkaline hydrolysis. The corn extracts were previously corrected to a pH of 7.0 only for enzymatic assays and kept at − 20 °C.

Total phenolic (TPC) and total anthocyanin (TA) contents

The TPC were assayed based on the Folin-Ciocalteu method (Singleton and Rossi 1965). The contents were expressed as mg of gallic acid equivalents (GAE) per 100 g (dry weight basis, DW). Total anthocyanin (TA) contents were determined following the method of Abdel-Aal and Hucl (1999) and the results were expressed as mg of cyanidin-3-glucoside equivalents (mg C3GE) per 100 g DW.

Analysis of phenolic profiles by UPLC

The analysis including the type of column, UHPLC system and chromatographic conditions were the same as those used by Ranilla et al. (2017). Only phenolic acids and flavonoids such as quercetin derivatives were detected and their quantification was performed based on calibration curves built with pure aglycone phenolic standards (r = 0.9990). The results were presented as mg per 100 g sample DW.

2-2's-Azino-bis(3-ethylbenzthiazoline-6-sulfonic acid) ($ABTS^{\cdot+}$) radical inhibition antioxidant capacity

The ABTS inhibition antioxidant assay was determined according to Arnao et al. (2001). The results were expressed as µmol of Trolox equivalents (TE) per 100 g DW based on a curve of Trolox as a standard (10–800 µM Trolox).

Oxygen radical absorbance antioxidant capacity (ORAC)

The ORAC assay was based on the method of Ou et al. (2001) in a 96-well microplate fluorometer (BioTek Instruments Sinergy 2, Winooski, VT, USA). The results were expressed as µmol Trolox equivalents (TE) per 100 g DW. A curve of Trolox standard was used (4–32 µM Trolox).

Inhibitory activity against α-amylase and α-glucosidase enzymes

The inhibitory activities against α-amylase and α-glucosidase were performed according to González-Muñoz et al. (2013). The results were expressed as percentage of inhibition.

Inhibitory activity against lipase enzyme

The method of Nakai et al. (2005) was used. The released 4-methylumbelliferone by the action of lipase enzyme was determined with a microplate fluorometer (BioTek Instruments Sinergy 2, Winooski, VT, USA) at 360 nm (excitation) and 460 nm (emission) wavelengths. The results were expressed as percentage of inhibition.

Statistical analysis

Assays were conducted by triplicate and results were expressed as mean ± standard deviation. A one-way analysis of variance (ANOVA) or the Kruskal–Wallis test was applied to all data ($\alpha = 0.05$). The Statgraphics Centurion XVI (StatPoint Inc., Rockville, MD, USA) was used. The multivariate principal component analysis (PCA) was performed by using the Unscrambler® X software version 10.4 (CAMO, Oslo, Norway).

Results and discussion

Color characterization

The color of corn kernels was measured using the CIELAB parameters to determine potential relationships between the grain color with the phenolic composition and related functional properties. The color parameters of corn samples provided by the germplasm bank and collected from the Arequipa region are shown in Supplementary Table S2a and S2b, respectively. In addition, photographs of all samples are shown in Supplementary Figure S1.

Both corn groups showed similar trends and the race and the type of accession/sample had a significant effect ($p < 0.05$) on color parameters. Arequipeño, Cabanita and Coruca races were characterized by high L^*, b^* and C^* values along with hue angles (H^*) close to 90° corresponding to lighter corn kernels with white to yellow pigmentations. In contrast, corn kernels from the Kculli race showed lower L^*, b^*, C^* values than samples from the other corn races, and hue angles were less than 45° indicating darker kernels with red-colored pericarps (Table S2a). For samples collected in situ (Table S2b), the Arequipeño and Cabanita races had similar color profiles as those observed in samples from the germplasm bank group. The Granada race showed intermediate L^* values with H^* values less than 45° corresponding to red-colored kernels (Figure S1). The only sample corresponding to the Kculli race (sample 4) showed the lowest L^*, b^*, and C^* values and the highest H^* value ($\sim$ 332°) among all corn samples indicating darker and purple-colored kernels. A high variability in color parameters was observed in certain samples from the Arequipeño and Cabanita races likely related to their multipigmentd nature (Figures S1). Similar variability was also found by Harakotr et al. (2015) when several waxy corn genotypes with different pigmentations were evaluated.

Total phenolic contents (TPC), total anthocyanins (TA) and antioxidant capacity by the ABTS and ORAC methods

The TPC and the antioxidant capacity results based on ABTS and ORAC methods measured in both the free and bound phenolic fractions of corn samples provided by the germplasm bank and collected from the Arequipa region are shown in Tables 1 and 2, respectively. In addition, the total anthocyanin (TA) contents were also determined in both sample groups. The race and type of accession or sample had a significant effect on the variability observed among all evaluated variables ($p < 0.05$), however certain corn races overall showed similar trends in relation to their TPC and antioxidant capacity. In case of corn samples from the germplasm bank and corresponding to the Arequipeño, Cabanita and Coruca races (with more yellow-colored pericarps), phenolic compounds were mainly found in the bound form (77–84% of TPC) and extracts from this phenolic fraction had higher antioxidant capacity than free phenolic fractions when evaluated by the ABTS and ORAC methods. The free and bound TPC varied from 23.22 to 32.27 mg GAE/100 g DW and from 106.29 to 138.63 mg GAE/100 g DW, respectively. The antioxidant capacity of the free and bound phenolic extracts evaluated by the ABTS method ranged from 34.44 to 77.39 and from 1832.10 to 2987.96 µmol Trolox equivalents/100 g DW, respectively. Higher values were obtained by the ORAC assay and varied from 654.03 to 2055.22 and from 1792.92 to 3884.37 µmol Trolox equivalents/100 g DW for the free and bound phenolic forms, respectively. Zulueta et al. (2009) pointed out that the ORAC method has a greater specificity with antioxidant compounds than the ABTS assay which was found to underestimate the antioxidant capacity especially in food samples of more complex nature.

Corn samples from the Kculli race (purple kernels) showed a different pattern and exhibited higher TPC (95.33–259.75 and 140.59–206.60 mg GAE/100 g DW, free and bound fractions, respectively) and antioxidant capacity in both phenolic fractions along with total anthocyanins (TA) (92.88–310.04 mg C3GE/100 g DW) compared to the other corn races ($p < 0.05$). The data of sample 8 (Accession Areq-084) corresponding to the free and bound TPC, and TA was reported previously (Ranilla et al. 2017) and was included here for comparison and discussion purposes. Free phenolic compounds and the free

Table 1 Total phenolic contents, total anthocyanins contents (TAC), and antioxidant capacity by the ABTS and ORAC assays of corn samples provided by the germplasm bank

No.	Total phenolic contents (mg GAE/100 g DW)			TAC (mg C3GE/100 g DW)	ABTS (μmol Trolox equiv/100 g DW)		
	Free	Bound	Total		Free	Bound	Total
1	26.87 ± 2.05	138.63 ± 6.59	165.50 ± 8.27	ND	39.14 ± 1.54	2777.48 ± 128.61	2816.62 ± 127.30
2	23.22 ± 0.43	120.11 ± 10.01	143.33 ± 9.96	ND	34.44 ± 3.11	2575.71 ± 223.26	2610.15 ± 222.52
3	32.27 ± 1.65	106.29 ± 4.25	138.56 ± 4.64	ND	77.39 ± 3.28	2889.05 ± 315.65	2966.44 ± 318.89
4	26.78 ± 0.61	129.48 ± 5.96	156.27 ± 5.76	ND	46.40 ± 1.03	2905.54 ± 51.00	2951.94 ± 49.97
5	25.96 ± 0.96	131.54 ± 26.46	157.50 ± 25.71	ND	49.40 ± 4.86	2259.79 ± 240.86	2309.18 ± 245.72
6	95.33 ± 8.79	140.59 ± 13.95	235.92 ± 5.99	92.88 ± 10.37	463.08 ± 59.75	2628.35 ± 225.70	3091.43 ± 282.35
7	259.75 ± 14.58	170.34 ± 17.09	430.09 ± 31.48	251.70 ± 18.27	2547.46 ± 127.58	2922.56 ± 298.75	5470.01 ± 284.48
8	247.86 ± 33.26[a]	206.60 ± 39.42[a]	454.45 ± 62.26[a]	310.04 ± 34.16[a]	3523.06 ± 143.99	3240.65 ± 334.78	6763.70 ± 250.52
9	101.38 ± 8.26	168.72 ± 28.69	270.10 ± 25.30	76.82 ± 4.66	538.94 ± 53.27	2724.76 ± 70.31	3263.70 ± 112.73
10	24.27 ± 1.34	119.33 ± 7.74	143.60 ± 8.89	ND	47.18 ± 3.52	1832.10 ± 172.83	1879.28 ± 175.25
11	29.25 ± 1.27	126.41 ± 11.29	155.66 ± 12.47	ND	63.22 ± 1.49	2366.14 ± 233.46	2429.36 ± 232.34
12	26.92 ± 1.06	113.06 ± 7.33	139.98 ± 6.62	ND	52.70 ± 4.60	2987.96 ± 200.14	3040.66 ± 202.70
Min	23.22	106.29	138.56	76.82	34.44	1832.10	1879.28
Max	259.75	206.60	454.45	310.04	3523.06	3240.65	6763.70
Race	*	*	*	–	*	*	*
Accession	*	*	*	*	*	*	*

No.	ORAC (μmol Trolox equiv/100 g DW)		
	Free	Bound	Total
1	1127.52 ± 5.27	2051.64 ± 93.36	3179.17 ± 95.85
2	654.03 ± 36.25	1792.92 ± 86.09	2446.95 ± 91.64
3	689.20 ± 5.02	3375.61 ± 38.96	4064.80 ± 37.42
4	1474.68 ± 32.28	2650.88 ± 144.75	4125.56 ± 171.29
5	1129.54 ± 16.27	3884.37 ± 197.33	5013.92 ± 202.67
6	2496.11 ± 155.70	4821.07 ± 251.75	7317.18 ± 228.12
7	11,625.92 ± 339.34	4435.99 ± 291.48	16,061.91 ± 611.02
8	7764.58 ± 211.91	4900.03 ± 293.71	12,664.61 ± 505.56
9	4583.63 ± 116.41	4494.29 ± 41.95	9077.93 ± 136.22
10	1175.53 ± 15.22	3447.54 ± 72.99	4623.07 ± 87.80
11	2055.22 ± 30.03	2957.97 ± 237.89	5013.20 ± 233.83
12	1530.30 ± 47.01	3211.62 ± 114.51	4741.91 ± 77.40
Min	654.03	1792.92	2446.95
Max	11,625.92	4900.03	16,061.91
Race	*	*	*

Table 1 continued

No.	Accession	ORAC (μmol Trolox equiv/100 g DW)		
		Free	Bound	Total
		*	*	*

Mean ± SD; n = 3

ND not detected

*Significant at $p < 0.05$

[a]Data previously reported by Ranilla et al. (2017) and included here for comparison reasons

ORAC antioxidant capacity significantly contributed to the total phenolic content (free and bound) (38–60%) and the total antioxidant capacity (61–72%), respectively. Samples 7 and 8 (accessions Areq-035 and Areq-084, respectively) showed the highest TPC, TA, and antioxidant capacity by the two methods in both phenolic fractions among corn accessions from the germplasm bank.

Corn samples collected in situ (Table 2) showed similar trends as those observed for the germplasm bank group. Samples from the Arequipeño and Cabanita races had higher phenolic contents and antioxidant capacity in their bound fractions. Bound phenolic compounds contributed 79–86% with respect to the TPC (free and bound). Free and bound TPC varied from 21.01 to 31.86 and from 104.43 to 133.54 mg GAE/100 g DW, respectively. The antioxidant capacity of free phenolic extracts showed ranges of 39.89–69.58 and 719.81–1358.74 μmol Trolox equivalents/100 g DW when measured by the ABTS and ORAC methods, respectively; whereas the bound fraction-linked antioxidant capacity varied from 1572.82 to 2896.44 and from 2116.45 to 4948.60 μmol Trolox equivalents/100 g DW by ABTS and ORAC assays, respectively. Ranges of free and bound TPC and antioxidant capacity in corn samples from the Granada race (partially red-colored kernels) were overall similar to those found in the Arequipeño and Cabanita races but moderate amounts of anthocyanins (3.28–12.69 mg C3GE/100 g DW) were also detected. The only sample from the Kculli race (sample 4) had the highest TA contents, free and bound TPC and antioxidant capacity among all corn samples ($p < 0.05$), and values were even higher than those reported above for purple corn kernels from the germplasm bank group indicating variability within samples from the same race. Free phenolic compounds of sample 4 were the major contributors to the TPC ($\sim$ 70%) which in turn explains the contribution to the high total antioxidant capacity (67 and 74% for the ABTS and ORAC assays, respectively).

Considering all evaluated corn samples, TPC (free and bound) quantified in the current research (133.14–548.87 mg GAE/100 g DW) were higher than ranges found by González-Muñoz et al. (2013) (132.2–262.5 mg GAE/100 g DW, samples from Chile), Zhang et al. (2017) (190–285.2 mg GAE/100 g DW, samples from China) and Trehan et al. (2018) (102.3–206.4 mg GAE/100 g DW, samples from India) whereas comparable ranges were reported by De la Parra et al. (2007) (243.8–320.1 mg GAE/100 g DW), Lopez-Martínez et al. (2009) (170–671 mg GAE/100 g DW), Montilla et al. (2011) (311.0–611.7 mg GAE/100 g DW) in corn samples with different pigmentations from the United States, Mexico and Bolivia, respectively. In addition, total anthocyanin contents found in current study (76.82–510.51 mg C3GE/100 g DW) were higher when

Table 2 Total phenolic contents, total anthocyanins contents (TAC), and antioxidant capacity by the ABTS and ORAC assays of corn samples collected from the Arequipa region

No.	Total phenolic contents (mg GAE/100 g DW)			TAC (mg C3GE/100 g DW)	ABTS (μmol Trolox equiv/100 g DW)		
	Free	Bound	Total		Free	Bound	Total
1	25.75 ± 1.45	128.88 ± 16.29	154.63 ± 16.28	ND	69.58 ± 6.09	2364.47 ± 141.39	2434.05 ± 138.28
2	31.39 ± 4.03	134.71 ± 9.97	166.10 ± 13.99	ND	46.96 ± 2.26	2425.80 ± 277.26	2472.76 ± 277.92
3	28.71 ± 1.77	104.43 ± 36.76	133.14 ± 35.16	ND	39.89 ± 2.72	1572.82 ± 172.31	1612.72 ± 174.91
4	378.49 ± 2.92	170.38 ± 8.22	548.87 ± 6.76	510.51 ± 25.21	6370.00 ± 380.36	3086.37 ± 243.79	9456.37 ± 139.07
5	21.01 ± 1.54	133.54 ± 4.91	154.55 ± 3.37	ND	57.14 ± 4.67	2742.55 ± 96.30	2799.69 ± 100.96
6	23.89 ± 1.74	109.94 ± 5.04	133.82 ± 3.56	ND	45.67 ± 1.98	2896.44 ± 171.56	2942.11 ± 170.69
7	31.86 ± 3.28	116.28 ± 9.71	148.14 ± 9.69	ND	61.63 ± 1.47	2204.20 ± 203.52	2265.84 ± 203.51
8	30.57 ± 2.05	127.79 ± 9.09	158.36 ± 10.67	ND	59.44 ± 3.05	2519.83 ± 209.16	2579.28 ± 212.07
9	33.78 ± 2.29	127.22 ± 2.57	161.01 ± 4.83	12.69 ± 0.23	94.54 ± 9.80	3063.88 ± 262.85	3158.42 ± 259.75
10	33.35 ± 0.62	137.04 ± 2.05	170.39 ± 1.56	3.28 ± 0.17	86.51 ± 4.33	2599.34 ± 204.74	2685.85 ± 201.23
Min	21.01	104.43	133.14	3.28	39.89	1572.82	1612.72
Max	378.49	170.38	548.87	510.51	6370.00	3086.37	9456.37
Race	*	*	*	–	*	*	*
Sample	*	NS	*	*	*	*	*

No.	ORAC (μmol Trolox equiv/100 g DW)		
	Free	Bound	Total
1	1063.16 ± 28.89	2909.65 ± 251.30	3972.81 ± 279.80
2	1116.34 ± 7.35	2116.45 ± 185.20	3232.79 ± 185.84
3	1066.96 ± 32.40	2445.45 ± 142.12	3512.41 ± 173.84
4	18,271.64 ± 296.72	6465.75 ± 513.31	24,737.38 ± 530.82
5	719.81 ± 29.87	3597.09 ± 226.57	4316.90 ± 243.84
6	986.65 ± 63.97	4948.60 ± 330.20	5935.25 ± 387.50
7	1358.74 ± 98.62	4013.51 ± 191.65	5372.25 ± 255.17
8	1069.88 ± 20.00	4803.36 ± 44.47	5873.23 ± 62.95
9	1153.45 ± 2.83	3524.69 ± 164.10	4678.14 ± 165.87
10	1065.42 ± 74.68	3619.25 ± 7.37	4684.67 ± 78.68
Min	719.81	2116.45	3232.79
Max	18,271.64	6465.75	24,737.38
Race	*	*	*
Sample	*	*	*

Mean ± SD; n = 3

ND not detected, *NS* not significant

*Significant at $p < 0.05$

compared to those found by Montilla et al. (2011), Zilic et al. (2012) and Harakotr et al. (2015) (1.97–71.68, 1.5–69.6 and 0.25–106.3 mg C3GE/100 g DW, respectively) for purple pigmented and red-blue corn samples from different origins. These differences might be related to several factors such as the corn origin, the genotype, the different agro-climatic conditions for the growth and the specific analytical procedures for phenolic analysis.

In case of results from the current study, the observed variability seems to be linked to the race effect likely associated with genetic factors. However, differences on growing conditions such as the altitude of growth and the geographical origin might be also involved (Supplementary Table S1). Further investigations with more promising corn accessions should be undertaken for better understanding of the potential relationships among above factors and their influence on phenolic biosynthesis.

UPLC-PDA phenolic profiles

The contents of specific phenolic compounds were determined by UPLC-PDA in samples provided by the germplasm bank (Table 3) and collected from the Arequipa region (Table 4). In addition, representative chromatograms from each evaluated corn race from both corn groups are shown in Supplementary Figures S2a and S2b.

Excluding anthocyanins which were not quantified by UPLC, phenolic compounds detected in corn samples were mainly found linked to the dietary fiber fraction and the free and bound phenolic extracts had different compositions. Variability among results was overall influenced by the race and the type of accession/sample ($p < 0.05$), but in some cases the effect was not statistically significant.

Considering both sample groups (germplasm bank and collected in situ), major phenolic compounds found in the free fraction were hydroxycinnamic acids such as *p*-coumaric acid derivatives (2.55–6.96 mg/100 g DW), followed by caffeic acid derivatives (0.70–2.18 mg/100 g DW) and ferulic acid derivatives (0.21–1.89 mg/100 g DW). Also, low amounts of *p*-coumaric (0.55–0.56 mg/100 g DW; sample 1 and 3, germplasm bank group) and ferulic acids (0.40–0.48 mg/100 g DW, samples 5 and 3 from the collected in situ and germplasm group, respectively) were detected in some corn extracts. Guo and Beta (2013) have reported low levels of ferulic and *p*-coumaric acids linked to the soluble dietary fiber structure from yellow corn kernel. The solvents used for the extraction of the free phenolic fraction in current study might have partially extracted those phenolic acids.

Flavonols such as quercetin derivatives were only detected in corn samples from the Kculli race (purple kernels) (0.27–0.58 mg/100 g DW; samples 7 and 4 from the germplasm and collected in situ group, respectively) along with higher caffeic acid derivatives contents (1.61–1.78 and 2.18 mg/100 g DW; samples 7–8 and 4 from the germplasm bank and collected in situ groups, respectively) than those quantified in the other corn races. Paucar-Menacho et al. (2017) and Giordano et al. (2017) also detected caffeic acid derivatives in the free phenolic fraction of Peruvian and Italian pigmented corns but at lower levels (0.55 and 0.27–1.16 mg/100 g DW, respectively) than those found in the current study. Flavonol derivatives such as quercetin, kaempferol and isorhamnetin derivatives have been previously detected in purple corn (Paucar-Menacho et al. 2017). Further, caffeic and ferulic acids along with rutin, quercetin and kaempferol have been also reported in Peruvian corn kernels by Ramos-Escudero et al. (2012). Both the above authors used improved Peruvian purple corn varieties (PVM-581 and INIA-601) from different Peruvian regions (Lima and Cajamarca) which could explain differences with respect to the results from the current study.

Chromatograms of bound phenolic fractions were similar among all corn races and only hydroxycinnamic acid derivatives were detected (Supplementary Figures S2a and S2b). Major bound phenolic compound was ferulic acid (72.33–156.30 mg/100 g DW) followed by *p*-coumaric acid (19.01–35.51 mg/100 g DW) and a ferulic acid derivative (11.71–23.07 mg/100 g DW). This latter compound may be a derivative of ferulic acid dehydrodimer or dehydrotrimer as it was also found in the bound fraction of Bolivian purple corn (Montilla et al. 2011). The bound ferulic acid represented approximately 73% with respect to the TPC (free and bound). Other studies have also shown that ferulic acid is the major bound phenolic acid in corn grains (Giordano et al. 2017; Thakur et al. 2017). Ranges of total bound phenolic acids measured by UPLC (104.22–208.57 mg/100 g DW) were similar to values found by the Folin-Ciocalteu method in the same fraction (104.43–206.60 mg GAE/100 g DW) confirming that bound phenolic compounds of evaluated Peruvian corn samples are mainly phenolic acids specially hydroxycinnamic acids.

The total ferulic acid contents (free and bound) reported here (72.33–156.30 mg/100 g DW) were in the range of values reported by previous studies (Lopez-Martínez et al. 2009; Montilla et al. 2011; González-Muñoz et al. 2013) when various pigmented corn samples and from different origins were evaluated. Moreover, the *p*-coumaric acid contents found in current study (19.45–35.51 mg/100 g DW) were comparable to those reported by González-Muñoz et al. (2013) and Giordano et al. (2017) in corn samples from different origins. Samples 7 and 8 (Kculli race, purple kernel) showed the highest total ferulic and *p*-coumaric acid contents among corn accessions provided by the germplasm bank whereas sample 10 (Granada race, partially red-colored kernel) had the highest levels of ferulic acid in the collected in situ corn group. In comparison sample 4 (Kculli race, purple kernel) had the highest amount of *p*-coumaric acid in this group. Giordano et al. (2017) also reported that contents of ferulic and *p*-coumaric acids were the highest in Italian red-colored corn kernels compared to yellow or white samples.

In vitro α-amylase, α-glucosidase and lipase inhibitory activities

The potential to inhibit enzymes relevant for targeting hyperglycemia (α-amylase and α-glucosidase) and obesity (lipase) management were evaluated in the free and bound phenolic fractions of corn samples provided by the germplasm bank (Table 5) and collected from the Arequipa region (Table 6). The race and the accession/sample factors

Table 3 Phenolic compounds detected by UPLC-PDA in corn samples provided by the germplasm bank (mg/100 g DW)

No.	*p*-Coumaric acid			*p*-Coumaric acid derivat[b]	Ferulic acid			Ferulic acid derivatives[c]		
	Free	Bound	Total	Free	Free	Bound	Total	Free	Bound	Total
1	0.55 ± 0.08	21.36 ± 0.18	21.91 ± 0.21	3.62 ± 0.52	ND	143.16 ± 2.68	143.16 ± 2.68	1.66 ± 0.11	19.66 ± 5.01	21.32 ± 5.12
2	ND	19.83 ± 0.46	19.83 ± 0.46	4.86 ± 0.27	ND	119.89 ± 7.71	119.89 ± 7.71	0.23 ± 0.02	18.60 ± 3.81	18.83 ± 3.80
3	0.56 ± 0.01	19.01 ± 0.97	19.58 ± 0.97	5.24 ± 0.30	0.48 ± 0.05	103.58 ± 13.20	104.06 ± 13.24	0.60 ± 0.09	18.19 ± 6.70	18.79 ± 6.72
4	ND	19.45 ± 0.97	19.45 ± 0.97	3.86 ± 0.30	ND	124.88 ± 13.82	124.88 ± 13.82	0.51 ± 0.06	19.21 ± 3.13	19.72 ± 3.19
5	ND	19.88 ± 0.92	19.88 ± 0.92	4.50 ± 0.21	ND	124.68 ± 21.67	124.68 ± 21.67	0.32 ± 0.03	19.26 ± 6.95	19.58 ± 6.97
6	ND	25.15 ± 1.68	25.15 ± 1.68	3.53 ± 0.46	ND	132.02 ± 11.69	132.02 ± 11.69	0.88 ± 0.23	15.97 ± 2.64	16.85 ± 2.45
7	ND	29.51 ± 1.40	29.51 ± 1.40	3.84 ± 0.15	ND	151.41 ± 9.94	151.41 ± 9.94	ND	16.71 ± 3.53	16.71 ± 3.53
8	ND	33.49 ± 1.44[a]	33.49 ± 1.44[a]	3.86 ± 0.80[a]	ND	156.30 ± 14.41[a]	156.30 ± 14.41[a]	1.44 ± 0.35[a]	18.78 ± 7.16[a]	20.21 ± 7.12[a]
9	ND	23.39 ± 0.26	23.39 ± 0.26	3.37 ± 0.62	ND	153.78 ± 4.18	153.78 ± 4.18	0.81 ± 0.28	17.33 ± 3.47	18.14 ± 3.37
10	ND	20.16 ± 1.38	20.16 ± 1.38	3.71 ± 0.27	ND	106.93 ± 20.30	106.93 ± 20.30	0.21 ± 0.01	18.92 ± 3.99	19.13 ± 3.99
11	ND	22.49 ± 0.76	22.49 ± 0.76	4.93 ± 0.22	ND	128.98 ± 21.32	128.98 ± 21.32	0.55 ± 0.09	20.17 ± 2.10	20.72 ± 2.01
12	ND	22.08 ± 1.04	22.08 ± 1.04	3.51 ± 0.16	ND	110.72 ± 12.81	110.72 ± 12.81	1.89 ± 0.20	17.02 ± 3.29	18.91 ± 3.13
Min		19.01	19.45	3.37		103.58	104.06	0.21	15.97	16.71
Max		33.49	33.49	5.24		156.30	156.30	1.89	20.17	21.32
Race		*	*	*	–	*	*	*	NS	NS
Accession	NS	*	*	*	–	*	*	*	NS	NS

No.	Caffeic acid derivat[d]	Total phenolic acids			Quercetin derivat[e]
	Free	Free	Bound	Total	Free
1	0.85 ± 0.02	6.69 ± 0.69	184.18 ± 7.51	190.86 ± 8.04	ND
2	0.84 ± 0.06	5.94 ± 0.24	158.32 ± 8.01	164.25 ± 8.24	ND
3	ND	6.89 ± 0.41	140.78 ± 9.38	147.67 ± 9.56	ND
4	0.97 ± 0.04	5.34 ± 0.28	163.54 ± 15.39	168.89 ± 15.30	ND
5	ND	4.82 ± 0.19	163.81 ± 29.14	168.63 ± 29.24	ND
6	ND	4.42 ± 0.69	173.15 ± 10.76	177.57 ± 11.40	ND
7	1.61 ± 0.15	5.44 ± 0.30	197.63 ± 13.99	203.08 ± 14.14	0.27 ± 0.03
8	1.78 ± 0.20[a]	7.08 ± 1.34[a]	208.57 ± 17.86[a]	215.65 ± 18.91[a]	ND
9	ND	4.17 ± 0.89	194.50 ± 3.06	198.68 ± 2.28	ND
10	0.89 ± 0.12	4.81 ± 0.38	146.02 ± 20.86	150.82 ± 21.24	ND
11	0.70 ± 0.04	6.19 ± 0.31	171.63 ± 20.54	177.82 ± 20.48	ND
12	0.75 ± 0.08	6.15 ± 0.44	149.82 ± 10.94	155.97 ± 11.38	ND
Min	0.70	4.17	140.78	147.67	
Max	1.78	7.08	208.57	215.65	
Race	NS	NS	*	*	–

Table 3 continued

No.	Caffeic acid derivat[d]	Total phenolic acids			Quercetin derivat[e]
	Free	Free	Bound	Total	Free
Accession	*	*	*	*	–

Mean ± SD; n = 3

ND not detected, *NS* not significant

*Significant at $p < 0.05$

[a]Data previously reported by Ranilla et al. (2017) and included here for comparison reasons

[b]Quantified as *p*-coumaric acid

[c]Quantified as ferulic acid

[d]Quantified as caffeic acid

[e]Quantified as quercetin aglycon

had a significant effect on the variability of results ($p < 0.05$). Considering both corn sample groups, the evaluated in vitro health-targeted functionality was mostly detected in the free phenolic fractions and the inhibitory activity followed a dose-dependent trend. Some level of α-glucosidase inhibitory activity was found in bound phenolic extracts, but values were overall less than 10% (data not shown). Only free phenolic extracts of samples from the Kculli race (darker purple kernels) significantly inhibited the obesity-relevant pancreatic lipase enzyme. In addition, corn extracts showed higher α-glucosidase inhibitory activity even at lower sample doses (5–25 mg) compared to the α-amylase inhibitory activity (evaluated at 50–250 mg). This is particularly important since high inhibition of pancreatic α-amylase could conduct to abnormal bacterial fermentation of undigested starch in the gut leading to undesired abdominal side effects (Bischoff 1994). Therefore, high α-glucosidase inhibition with mild α-amylase inhibitory activity may have potential for lowering glycemic response and its modulation; however current results should be further validated with future animal/clinical studies and reflecting common dietary corn consumption (cooked or with other technological processes) and cultural adaptations.

The α-glucosidase inhibitory activities in the samples provided by the germplasm bank ranged from 19.57 to 54.22% (at 12.5 mg) and from 32.50 to 70.05% (at 25 mg) whereas the α-amylase inhibitory activity varied from 13.67 to 29.07% at the highest evaluated sample dose (250 mg). Overall, samples from the Kculli race (purple grains) had higher α-glucosidase and α-amylase inhibitory activities than the other corn races. Further, the ability to inhibit the lipase enzyme was found only in extracts from the Kculli race at all tested doses (2.5–12.5 mg) and values ranged from 42.68 to 79.02% and from 58.45 to 85.44% at 6.25 and 12.5 mg, respectively. Samples 7 and 8 (corresponding to accessions Areq-035 and Areq-084, respectively) had the highest inhibitory activity against α-glucosidase (at all sample doses), α-amylase (at 250 mg) and lipase (at all doses). These accessions also showed the highest TA contents, free and total TPC and antioxidant capacity as stated above.

In case of samples collected in situ, similar α-amylase (14–26.75% at 250 mg) and α-glucosidase (47.61–76.10% at 25 mg) inhibitory activities to those of the germplasm bank were also found and sample 4 from the Kculli race showed the highest inhibition ($p < 0.05$) against all enzymes. The lipase inhibitory activity of sample 4 was even higher (88.21 and 92.16% at 6.25 and 12.5 mg of sample dose, respectively) than values detected in samples 7 and 8 from the germplasm bank group (79.20% and 85.44% for samples 7 and 8; dose 12.5 mg, respectively). This corn sample also had the highest TA contents, free

Table 4 Phenolic compounds detected by UPLC-PDA in corn samples collected from the Arequipa region (mg/100 g DW)

No.	*p*-Coumaric acid	*p*-Coumaric acid derivat[a]	Ferulic acid			Ferulic acid derivatives[b]		
	Bound	Free	Free	Bound	Total	Free	Bound	Total
1	21.50 ± 2.40	5.08 ± 0.32	ND	136.30 ± ± 22.60	136.30 ± 22.60	0.70 ± 0.12	18.64 ± 4.83	19.34 ± 4.81
2	22.33 ± 2.32	6.96 ± 0.76	ND	142.51 ± 30.14	142.51 ± 30.14	1.00 ± 0.33	23.07 ± 0.46	24.07 ± 0.23
3	20.18 ± 0.56	4.82 ± 0.54	ND	72.33 ± 7.83	72.33 ± 7.83	0.28 ± 0.03	11.71±1.76	11.99 ± 1.79
4	35.51 ± 3.10	5.97 ± 0.32	ND	140.92 ± 20.57	140.92 ± 20.57	ND	14.66 ± 3.20	14.66 ± 3.20
5	20.70 ± 1.38	2.55 ± 0.44	0.40 ± 0.01	138.04 ± 3.82	138.44 ± 3.83	0.44 ± 0.12	21.26 ± 2.73	21.70 ± 2.65
6	19.55 ± 0.42	4.35 ± 0.62	ND	119.27 ± 11.09	119.27 ± 11.09	0.41 ± 0.10	19.52 ± 1.95	19.92 ± 1.87
7	20.76 ± 1.02	5.30 ± 0.41	ND	119.60 ± 12.08	119.60 ± 12.08	0.91 ± 0.20	20.05 ± 3.01	20.97 ± 2.86
8	21.32 ± 1.04	5.49 ± 0.61	ND	138.73 ± 17.26	138.73 ± 17.26	0.74 ± 0.25	17.06 ± 2.08	17.80 ± 1.89
9	19.52 ± 0.55	5.60 ± 0.58	ND	135.71 ± 8.13	135.71 ± 8.13	1.53 ± 0.38	21.10 ± 1.77	22.63 ± 1.40
10	20.75 ± 0.56	4.52 ± 0.37	ND	153.69 ± 11.81	153.69 ± 11.81	1.06 ± 0.05	21.27 ± 3.20	22.33 ± 3.17
Min	19.52	2.55		72.33	72.33	0.28	11.71	11.99
Max	35.51	6.96		153.69	153.69	1.53	23.07	24.07
Race	*	NS	–	NS	NS	*	NS	NS
Sample	*	*	–	*	*	*	*	*

No.	Caffeic acid derivat[c]	Total phenolic acids			Quercetin derivat[d]
	Free	Free	Bound	Total	Free
1	0.84 ± 0.04	6.62 ± 0.35	176.43 ± 23.19	183.06 ± 23.17	ND
2	0.89 ± 0.20	8.86 ± 1.22	187.91 ± 32.53	196.77 ± 32.78	ND
3	ND	5.10 ± 0.56	104.22 ± 7.20	109.32 ± 6.65	ND
4	2.18 ± 0.21	8.15 ± 0.53	191.08 ± 21.48	199.23 ± 21.09	0.58 ± 0.19
5	0.81 ± 0.17	4.20 ± 0.64	180.01 ± 3.95	184.20 ± 3.50	ND
6	0.80 ± 0.06	5.55 ± 0.74	158.34 ± 13.39	163.89 ± 12.66	ND
7	1.08 ± 0.10	7.30 ± 0.67	160.41 ± 12.42	167.71 ± 12.61	ND
8	1.32 ± 0.18	7.56 ± 1.00	177.10 ± 17.32	184.66 ± 18.10	ND
9	1.30 ± 0.12	8.43 ± 1.05	176.33 ± 7.31	184.76 ± 8.23	ND
10	1.22 ± 0.11	6.79 ± 0.52	195.71 ± 10.22	202.50 ± 10.36	ND
Min	0.80	4.20	104.22	109.32	
Max	2.18	8.86	195.71	202.50	
Race	*	NS	NS	NS	–
Sample	*	*	*	*	–

Mean ± SD; n = 3

NS not significant, *ND* not detected

*Significant at $p < 0.05$

[a]Quantified as *p*-coumaric acid

[b]Quantified as ferulic acid

[c]Quantified as caffeic acid

[d]Quantified as quercetin aglycon

and total TPC and antioxidant capacity. In addition, sample 9 (partially red-colored kernel) which showed the highest TA contents among corn samples from the Granada race, had the second highest α-glucosidase inhibitory activity at all sample doses.

Pearson coefficients revealed a direct correlation between the α-glucosidase inhibitory activity and the TA contents (r = 0.63–0.69, $p < 0.05$, for both corn groups) along with the free antioxidant capacity (r = 0.64–0.72 and 0.63–0.73, $p < 0.05$; for ABTS and ORAC results, respectively in both corn groups). Further, a strong

correlation was observed between the TA contents and the lipase inhibitory activity (r = 0.90–0.96, $p < 0.05$; for the germplasm bank group). This suggests that the inhibition of α-glucosidase and pancreatic lipase seems to be mainly associated with the anthocyanin content especially in those corn samples from the Kculli and Granada races. However, a potential synergistic action of anthocyanins with the other detected soluble phenolic compounds may also explain the results. Yao et al. (2013) found that cyanidin-3-glucoside and peonidin-3-glucoside from black rice showed a significant in vitro inhibition against pancreatic lipase in a competitive manner. This might explain the high lipase inhibitory activity found in purple corn samples in current study. Although the inhibition of lipase is only one of many mechanisms whereby phenolic compounds such as anthocyanins may influence obesity (Hsu and Yen 2007), the potential of certain samples from the Peruvian corn diversity to inhibit this digestive enzyme is reported in current study for the first time and has relevance for further studies towards targeting in food applications.

Anthocyanins may also partially contribute to the α-amylase inhibitory activity in samples from the Kculli race although other undetected soluble phenolic or nonphenolic compounds also solubilized under current extraction method may also be involved especially in yellow-colored corn races and further research is needed to elucidate this.

González-Muñoz et al. (2013) determined similar ranges of α-glucosidase inhibitory activity (10.8–72.5%, at 25 mg sample dose) in Chilean corn races as those reported here, but no correlation with the TPC and the antioxidant capacity was found. Lee et al. (2010) had a similar observation when ethanolic extracts of colored Mexican corns were analyzed. However, Yao et al. (2010) found a significant correlation between the total anthocyanin contents and the DPPH antioxidant capacity with the α-glucosidase inhibitory activities of several colored grain cereals from China including purple corn as also was observed in the current study.

Principal component analysis (PCA)

This multivariate technique was performed to better analyze obtained results and to reveal the underlying relationships among evaluated variables (González-Muñoz et al. 2013). The PCA model for corn samples provided by the germplasm bank retained two principal components (PC) which explained 61% of the total variability of the dataset. The loading plots and score of the first two PC are shown in the Supplementary Figure S3. Corn accessions from the Kculli race showed a different pattern with respect to the other corn races and formed a separate group (left of score plot), but some intra racial variability was also observed (samples 7 and 8 versus samples 6 and 9). No defined groups were observed among samples from the Arequipeño, Cabanita and Coruca races. The PC 1 (51% of explained variance) revealed a direct relationship among the free and bound TPC, anthocyanin contents, bound and total ferulic acid, bound and total *p*-coumaric acid, bound and total phenolic acids contents with the free and total antioxidant capacity (ABTS and ORAC methods), bound ORAC values, and the α-glucosidase and lipase inhibitory activities (at all evaluated doses). These variables were significantly higher in samples 7 and 8. In addition, an inverse relationship between above variables and the color parameters (H^*, L^*, b^* and C^*) was observed. On the contrary, the PCA of data from several colored Chilean corn accessions determined that variability was more related to the accession type whereas no relationship between the in vitro functionality (DPPH and ABTS antioxidant capacity and α-glucosidase inhibitory activity) and the TPC or any phenolic compound was found (González-Muñoz et al. 2013). This indicates important differences related to the origin of corn germplasm.

In case of samples collected from the Arequipa region (score and loading plot in Supplementary Figure S4a), the PCA model retained five PC that explained 89.9% of the total variability. The PC 1 (55% of explained variance) allowed to separate sample 4 (Kculli race) (left of score plot) from the other races. This sample had the highest H^* value, free and total TPC, anthocyanins, bound *p*-coumaric acid, free and bound antioxidant capacity (ABTS and ORAC results) and α-amylase (at 50 and 125 mg), α-glucosidase and lipase inhibitory activities (at all sample doses). Such variables were highly correlated to each other and were inversely proportional to L^*, C^* and b^* color parameters. When data from sample 4 was excluded (Supplementary Figure S4b), the PCA model retained 5 PC that explained 85.6% of the total variability. According to the first PC (36% of explained variance), samples were clearly separated based on the race type. The Granada race (samples 9 and 10) showed the highest bound ferulic acid and total phenolic acid contents, anthocyanins, free ABTS antioxidant capacity and α-glucosidase inhibitory activity. These variables were inversely correlated to the color parameters L^*, H^*, b^* and C^*. Finally, samples from the Cabanita race (5–8) were characterized by their high bound and total ORAC antioxidant capacity and high α-amylase inhibitory activity (50 and 125 mg sample dose).

Conclusion

Phenolic compounds in corn races with yellow-colored corn kernels (Arequipeño, Cabanita, Coruca) were mostly found in the cell wall-bound fraction showing higher antioxidant capacity than free forms. In contrast, samples

Table 5 α-Amylase, α-glucosidase and lipase inhibitory activities (%) at different sample doses of corn samples provided by the germplasm bank

No.	α-Amylase Inhibitory Activity (%)			α-Glucosidase Inhibitory Activity (%)			Lipase Inhibitory Activity (%)		
	Free			Free			Free		
	50 mg	125 mg	250 mg	5 mg	12.5 mg	25 mg	2.5 mg	6.25 mg	12.5 mg
1	ND	10.78 ± 1.27	21.76 ± 1.39	15.89 ± 2.27	26.83 ± 1.19	39.68 ± 2.28	ND	ND	ND
2	ND	8.39 ± 1.12	22.17 ± 1.95	12.16 ± 2.28	24.32 ± 1.82	35.27 ± 5.38	ND	ND	ND
3	2.26 ± 0.28	6.43 ± 0.69	18.45 ± 0.60	24.66 ± 2.40	39.98 ± 1.71	57.08 ± 2.53	ND	ND	ND
4	1.18 ± 0.19	10.25 ± 0.37	20.13 ± 1.97	19.18 ± 1.88	31.34 ± 2.00	45.43 ± 3.37	ND	ND	ND
5	3.05 ± 0.32	8.83 ± 1.19	13.67 ± 1.93	18.91 ± 2.06	37.65 ± 4.38	55.73 ± 3.59	ND	ND	ND
6	ND	9.02 ± 0.72	19.72 ± 1.35	11.73 ± 1.66	19.60 ± 0.22	35.76 ± 1.27	25.37 ± 2.63	42.68 ± 3.55	58.45 ± 4.18
7	ND	9.92 ± 0.72	29.07 ± 2.06	34.84 ± 2.39	54.22 ± 4.11	70.05 ± 0.55	53.33 ± 1.94	69.84 ± 2.22	79.20 ± 1.62
8	3.97 ± 0.56	12.63 ± 1.06	22.19 ± 1.94	30.61 ± 2.25	50.07 ± 0.80	68.07 ± 0.82	64.82 ± 4.21	79.02 ± 2.07	85.44 ± 0.78
9	1.38 ± 0.31	12.99 ± 0.60	28.37 ± 0.83	17.59 ± 3.09	32.32 ± 3.71	53.67 ± 3.10	32.88 ± 0.44	50.07 ± 2.42	66.50 ± 2.67
10	ND	11.37 ± 0.71	22.50 ± 2.22	18.64 ± 2.92	30.44 ± 2.55	47.90 ± 1.96	ND	ND	ND
11	2.61 ± 0.50	11.37 ± 1.03	20.28 ± 3.03	23.04 ± 2.93	40.95 ± 3.73	58.03 ± 2.56	ND	ND	ND
12	2.81 ± 0.45	10.27 ± 0.89	23.09 ± 3.29	10.27 ± 0.33	19.57 ± 3.01	32.50 ± 3.42	ND	ND	ND
Min	1.18	6.43	13.67	10.27	19.57	32.50	25.37	42.68	58.45
Max	3.97	12.99	29.07	34.84	54.22	70.05	64.82	79.02	85.44
Race	NS	*	*	*	*	*	–	–	–
Accession	*	*	*	*	*	*	*	*	*

Mean ± SD; n = 3

ND not detected, *NS* not significant

*Significant at $p < 0.05$

Table 6 α-Amylase, α-glucosidase and lipase inhibitory activities (%) at different sample doses of corn samples collected from the Arequipa region

No.	α-Amylase Inhibitory Activity (%)			α-Glucosidase Inhibitory Activity (%)			Lipase Inhibitory Activity (%)		
	Free			Free			Free		
	50 mg	125 mg	250 mg	5 mg	12.5 mg	25 mg	2.5 mg	6.25 mg	12.5 mg
1	ND	ND	21.38 ± 1.53	15.08 ± 4.12	26.06 ± 3.24	53.39 ± 4.27	ND	ND	ND
2	ND	ND	25.77 ± 2.52	24.43 ± 5.20	37.38 ± 6.62	61.93 ± 3.01	ND	ND	ND
3	ND	ND	16.16 ± 0.65	15.51 ± 4.24	30.54 ± 5.14	47.61 ± 5.98	ND	ND	ND
4	9.72 ± 0.93	17.46 ± 1.71	26.75 ± 1.91	37.40 ± 7.78	60.25 ± 6.04	76.10 ± 2.77	77.86 ± 3.72	88.21 ± 2.44	92.16 ± 1.93
5	2.13 ± 0.28	9.05 ± 0.68	14.59 ± 1.43	25.83 ± 1.51	40.81 ± 1.88	63.62 ± 1.94	ND	ND	ND
6	3.51 ± 0.49	10.17 ± 0.99	19.07 ± 1.77	22.45 ± 0.93	40.53 ± 0.30	57.60 ± 1.51	ND	ND	ND
7	2.61 ± 0.39	8.95 ± 0.86	16.31 ± 1.62	22.01 ± 4.57	40.07 ± 7.12	57.20 ± 6.25	ND	ND	ND
8	4.02 ± 0.66	9.32 ± 0.78	17.27 ± 1.67	20.69 ± 1.77	34.92 ± 3.36	50.00 ± 0.99	ND	ND	ND
9	ND	7.31 ± 0.68	15.29 ± 1.50	29.22 ± 1.51	49.96 ± 1.34	68.07 ± 1.80	ND	ND	ND
10	1.94 ± 0.27	6.31 ± 0.51	14.68 ± 0.91	23.92 ± 2.90	43.04 ± 2.31	61.48 ± 3.71	ND	ND	ND
Min	1.94	6.31	14.59	15.08	26.06	47.61			
Max	9.72	17.46	26.75	37.40	60.25	76.10			
Race	*	*	*	*	*	*	–	–	–
Sample	*	*	*	*	*	*	–	–	–

Mean ± SD; n = 3

ND not detected, *NS* not significant

*Significant at $p < 0.05$

from the Kculli race (purple kernels) had higher free TPC (mainly anthocyanins) and antioxidant capacity than the bound fraction. Major phenolic acids detected by UPLC in free fractions were derivatives of *p*-coumaric, ferulic and caffeic acids, whereas ferulic, *p*-coumaric acid and a ferulic acid derivative were found in all bound fractions. All samples (free fraction) showed high inhibition of α-glucosidase with moderate α-amylase inhibitory activity whereas high lipase inhibitory activities were only detected in samples from the purple Kculli race. The PCA revealed that the variability of data was significantly affected by the corn race and confirmed that the in vitro functionality linked to the α-glucosidase and lipase inhibitory activities was high in purple-colored kernels and significantly correlated to the anthocyanin contents and the antioxidant capacity. This study provides the initial phenolic-linked biochemical evidence and confirms the potential of Peruvian corn diversity as a source of health relevant bioactives with important in vitro enzyme model-based health-targeted functionality relevant for hyperglycemia and obesity management and with potential for future breeding studies.

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