Natural variation of sucrose, glucose and fructose contents in Colombian genotypes of Solanum tuberosum Group Phureja at harvest

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Abstract

BACKGROUND: Potato frying quality is a complex trait influenced by sugar content in tubers. Good frying quality requires low content of reducing sugars to avoid the formation of dark pigments. Solanum tuberosum Group Phureja is a valuable genetic resource for breeding and for genetic studies. The sugar content after harvest was analyzed in a germplasm collection of Group Phureja to contribute to the understanding of the natural variation of this trait.

RESULTS: Sucrose, glucose and fructose genotypic mean values ranged from 6.39 to 29.48 g kg\(^{-1}\) tuber dry weight (DW), from 0.46 to 28.04 g kg\(^{-1}\) tuber DW and from 0.29 to 27.23 g kg\(^{-1}\) tuber DW, respectively. Glucose/fructose and sucrose/reducing sugars ratios ranged from 1.01 to 6.67 mol mol\(^{-1}\) and from 0.15 to 7.78 mol mol\(^{-1}\), respectively. Five clusters of genotypes were recognized, three of them with few genotypes and extreme phenotypic values.

CONCLUSION: Sugar content showed a wide variation, representing the available variability useful for potato breeding. The results provide a quantitative approach to analyze the frying quality trait and are consistent with frying color. The analyzed germplasm presents extreme phenotypes, which will contribute to the understanding of the genetic basis of this trait.

INTRODUCTION

Potato (Solanum tuberosum L.) is the third most important crop after wheat and rice, and therefore plays an important role worldwide for food security and nutrition. The demand for potato chips and French fries continues to grow because of changes in consumption habits. High contents of sucrose (non-reducing sugar), glucose and fructose (reducing sugars) in potato tubers represent an undesirable trait for fried processing because reducing sugars are precursors of the Maillard reaction and sucrose is the main source of reducing sugars during its enzyme-catalyzed hydrolysis. These sugars tend to accumulate during cold storage at temperatures below 10 °C. The Maillard reaction refers to a series of non-enzymatic reactions between reducing sugars and principally amino groups from amino acids during frying. This reaction leads to the production of dark pigments, the development of off-flavors and the production of toxic compounds such as acrylamide that reduce consumer acceptance and represent risks for human health. There is a wide range of sugar concentrations among potato genotypes, which is an indication that this content is under polygenic control, and therefore is highly influenced by environmental conditions.
adaptation and lack of tuber dormancy.7,8 There is a biodiversity center for this kind of potato in the southern region of Colombia.9 These potatoes are of economic importance in Andean countries, especially since they are cultivated by small farmers.2 Colombia leads the production, consumption and export of diploid Phureja genotypes known for their round tubers with yellow flesh and skin.10,11 These genotypes present an interesting possibility for widespread use because of their outstanding nutritional, organoleptic properties and processing potential.10,12,13

Group Phureja presents potential for outcrossing with tetraploid potatoes – the most extensively cultivated worldwide – and is useful in bridge crosses for the introgression of genes of Solanum wild species.14,15 Therefore, this group constitutes an important germplasm with prospective use in potato breeding programs as a source of resistance to multiple diseases such as late blight, bacterial wilt and common scab.16–18 Also, it has been shown to be valuable for the introgression of improved chip color after cold storage.19 The sequencing of the doubled-monoploid genotype DM1-3516 R44 from Group Phureja represents the potato reference genome,20 and its analysis can contribute to an understanding of the genetic basis of various agronomic traits such as frying quality with the use of the appropriate phenotypic measurements.

The phenotypic assessment of potato frying quality for linkage mapping and association mapping studies has generally been done with the use of visual scales of chip frying color.21–23 However, these studies can benefit from the additional quantification of the variability of the precursors of the Maillard reaction, considering type and content of sugars individually, on working populations and germplasm collections. This quantitative approach for the understanding of the frying quality trait will help to make more precise the identification of genotypes prone to the synthesis of dark pigments and acrylamide.24

The use of high-quality quantitative phenotypic data provides advantages to the implementation of strategies such as association mapping, where accurate quantification of the phenotypic variation in the population is crucial to find highly reliable associated variants.25,26

The sugar content variation in tubers at the end of the growing season is mainly influenced by the biotic and abiotic stresses present during plant development, the temperature during the vegetative growth and the genotypic differences in starch degradation metabolism.4,27,28 Colombia only has two tetraploid cultivars suitable for frying purposes, and a Phureja cultivar specific for chip processing has not yet been developed.15 Núñez-López has proposed growing the potato crops for frying purposes in altitudes lower than 2600 m above sea level (m a.s.l.) to decrease the probability of a high content of reducing sugars at harvest due to cold temperatures in the course of tuber development. Nevertheless, lower altitudes generate difficulties related to diseases and pests that could affect the performance of crops. Therefore, it is relevant to contribute to potato breeding programs towards the development of specific cultivars for fried processing through the analysis of sugar content of Phureja genotypes at harvest.

Accessions from the Colombian Central Collection (CCC), which constitutes the Group Phureja Working Collection of the Breeding Program at the National University of Colombia, were characterized in a different study for their frying quality at harvest. That study identified a wide genetic basis for the trait through the assessment of chip color using a visual scale in fried tubers from plots in nine environments.19 Recently, a method using high-performance liquid chromatography (HPLC) for the analysis of sugars in Group Phureja tubers was developed and validated.29

The aim of this research was to study the natural variation of sucrose, glucose and fructose contents after harvest in Colombian Phureja genotypes using this HPLC method. This study will contribute to potato breeding programs and will generate phenotypic information with the purpose of advancing in the elucidation of the molecular basis of sugar content variation in tubers from this cultivated group.

MATERIALS AND METHODS

Plant material and experimental design

A set of 108 diploid landrace accessions from CCC collection and four commercial diploid cultivars (Criolla Colombia, Criolla Latina, Criolla Galeras and Criolla Guaneña) were analyzed for their tuber sugar content at harvest. The plants were grown through vegetative propagation in a plot from the municipality of Soacha (Cundinamarca, Colombia; altitude 2905 m a.s.l., latitude 4° 30’ 25” N and longitude 74° 11’ 28” W). To reduce the experimental error caused by soil heterogeneity from the field, tubers from each genotype were sown in pots that contained a substrate of organic moor soil with loam texture and sand in a 3:1 ratio. One tuber per pot was sown at the end of September 2013 and plants were harvested at tuber physical maturity, when plants presented foliage senescence and tubers had developed an appropriate degree of skin set.27 Plants were irrigated when required. Potato tubers from three pots of each genotype constituted three independent biological replicates in a completely randomized design.

Tuber sample preparation and HPLC analysis

Three mature and healthy raw tubers per pot without evidence of mechanical damage were used for the analysis. Tubers were washed with distilled water, sliced and prepared for freeze-drying the same day of harvest. Sucrose, glucose and fructose were extracted with 50% (v/v) aqueous methanol from 0.5 g freeze-dried ground tissue and measured by HPLC. The HPLC method was developed and validated for Group Phureja potatoes using an AMINEX 87H column, 10 mmol L−1 sulfuric acid as eluent and refraction index detector.29

Calibration curves were run every 48 samples. The lack of carryover in the long-term HPLC analysis was verified by injecting blanks every eight samples. The stability of the HPLC system during independent runs was assessed by the injection of a 40 mg L−1 mixture of standards every nine samples. The relative standard deviation (RSD) values varied from 9.43 to 11.19, from 3.03 to 5.76 and from 2.94 to 4.74, for sucrose, glucose and fructose, respectively. These RSD values (below 11.30) assured reliability of the data through long-term HPLC runs, according to levels of acceptability of RSD values proposed in the Horwitz function for analytes at concentrations ranging from 10 to 100 mg L−1.30

Statistical analyses

Mean values of each sugar content were established for each genotype and expressed as g kg−1 tuber dry weight (DW). Values of reducing sugar content were obtained from the sum of glucose and fructose, and total sugar content was calculated from the sum of the three sugars. Average and standard deviation values of reducing and total sugars were also calculated for each genotype. Mean values of glucose/fructose (mol mol−1) and sucrose/reducing sugars (mol mol−1) ratios were calculated for each genotype with the purpose of identifying differential genotypic patterns regarding the proportion of sugar content measured at harvest.

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A multi-environmental chip darkening index of CCC accessions estimated by Ñúñez-López through a Bayesian approach was included as an additional variable in the correlation analysis. This index reflects the chip darkening percentage of genotypes from the tuber analysis in nine environments. The index was calculated using a scale proposed by the author with equivalences between the International Potato Center (CIP) chip frying color scale and a chip darkening percentage. This proposed scale shows the chip darkening percentage range that corresponds to each one of the five color degrees of the CIP scale (supporting information, supplementary Table S2). Thus the first degree, with the lightest color, represents a darkening percentage of 0, while the fifth degree, with the darkest color, represents a darkening percentage range that corresponds to each one of the five color degrees of the CIP scale (supporting information, supplementary Table S2). Thus the first degree, with the lightest color, represents a darkening percentage of 0, while the fifth degree, with the darkest color, represents a darkening percentage range that corresponds to each one of the five color degrees of the CIP scale (supporting information, supplementary Table S2). Thus the first degree, with the lightest color, represents a darkening percentage of 0, while the fifth degree, with the darkest color, represents a darkening percentage range that corresponds to each one of the five color degrees of the CIP scale (supporting information, supplementary Table S2). Thus the first degree, with the lightest color, represents a darkening percentage of 0, while the fifth degree, with the darkest color, represents a darkening percentage range that corresponds to each one of the five color degrees of the CIP scale (supporting information, supplementary Table S2).

### RESULTS AND DISCUSSION

#### Sugar content

The sugar content analysis of Phureja genotypes showed an extensive range of variation among the three sugars (Table 1; supplementary Table S3), supporting the high variability for this trait in Group Phureja. The most abundant sugar found in tubers was sucrose, followed by glucose and fructose. Different from our results, a study of the sugar content at harvest of ten landraces from Group Tuberosum and Group Andigena reported ranges with lower variation of sucrose, glucose and fructose contents of 1.82–7.96, 0.13–2.39 and 0.10–1.08 g kg⁻¹ tuber DW, respectively. The sucrose content of commercial cultivars is close to the average value of CCC genotypes (Table 1). Potato genotypes with low sucrose levels at harvest have a greater ability to control sucrose accumulation as soon as tuber development is completed. These genotypes present a high potential for processing because they have a reduced pool of sucrose that by hydrolysis can produce glucose and fructose during the transport and storage of harvested tubers. Criolla Guaneña is the commercial cultivar with the highest sucrose content, and therefore it is not a good candidate for frying purposes.

Commercial cultivars show values closer to the minimum values of reducing sugars observed in CCC, with the exception of Criolla Colombia (Table 1). A previous analysis of commercial cultivars sown in a plot at 3400 m a.s.l. showed an increased concentration of reducing sugars at harvest (from 3.6 to 4.0 g kg⁻¹ tuber DW; Duarte-Delgado et al. 2015). This result supports the high environmental influence on the trait and reveals that commercial cultivars present a similar reducing sugar content at high altitudes with lower temperatures. The higher content of reducing sugars found in Criolla Colombia among the commercial cultivars in the current study at a lower altitude (2905 m a.s.l.) supports the view that this cultivar is not suitable for frying purposes. Criolla Colombia is a clonal selection from CCC accessions, differing from the other cultivars that were selected from breeding populations.

The five Phureja accessions with the lowest sucrose content at harvest are CCC 80, CCC 61, CCC 2, CCC 20 and CCC 4 (from 6.39 to 8.65 g kg⁻¹ tuber DW), while the five accessions with the lowest reducing sugar content are CCC 27, CCC 4, CCC 91, CCC 35 and CCC 41 (from 0.87 to 0.98 g kg⁻¹ tuber DW). It is noteworthy that CCC 27 and CCC 35 are the accessions with the lowest multi-environmental estimate of chip darkening index of 3.33% and 4.45%, respectively (supplementary Table S2). Genotype CCC 4 is interesting because it accumulates low amounts of both sucrose and reducing sugars. The nine aforementioned accessions are interesting subjects for further analysis as it is important to study genotypic variants that present not only low reducing sugar content at harvest but also a diminished potential for the generation of reducing sugars by having low concentrations of sugars.

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**Table 1.** Average content of sugars and average sugar ratios from accessions of CCC (108 genotypes) and four commercial cultivars from *Solanum tuberosum* Group Phureja with their respective standard deviations

| Genotype       | Sucroseᵇ | Glucoseᵇ | Fructoseᵇ | Reducing sugarsᵇ | Total sugarsᵇ | Glucose/fructoseᶜ | Sucrose/reducing sugarsᶜ |
|----------------|----------|----------|-----------|------------------|---------------|-----------------|------------------------|
| CCC            | 14.32 ± 4.70 | 2.68 ± 4.27 | 1.77 ± 3.76 | 4.45 ± 7.98 | 18.77 ± 10.25 | 1.79 ± 0.71 | 2.96 ± 1.50 |
| Criolla Guaneñaᵈ | 14.87 ± 3.49 | 0.80 ± 0.14 | 0.41 ± 0.10 | 1.21 ± 0.22 | 16.07 ± 2.24 | 1.95 ± 0.36 | 6.50 ± 1.18 |
| Criolla Latinaᵈ | 10.44 ± 1.35 | 0.69 ± 0.14 | 0.37 ± 0.07 | 1.05 ± 0.20 | 11.49 ± 2.53 | 1.88 ± 0.14 | 5.39 ± 1.33 |
| Criolla Galerasᵈ | 11.15 ± 0.32 | 0.79 ± 0.11 | 0.38 ± 0.14 | 1.17 ± 0.25 | 12.32 ± 2.01 | 2.07 ± 0.58 | 5.16 ± 1.06 |
| Criolla Colombiaᵈ | 12.00 ± 1.26 | 1.67 ± 0.14 | 1.31 ± 0.33 | 2.99 ± 0.47 | 14.99 ± 1.07 | 1.28 ± 0.22 | 2.16 ± 0.56 |
| Minimum        | 6.39      | 0.46      | 0.29      | 0.87              | 7.53          | 1.01            | 0.15               |
| Maximum        | 29.42     | 28.04     | 27.23     | 55.25            | 74.05         | 6.67            | 7.78               |

ᵃ CCC denotes the accessions from the Colombian Central Collection that constitute the Working Collection from the Potato Breeding Program at the National University of Colombia.
ᵇ Average sugar content is expressed in g kg⁻¹ dry weight tuber.
ᶜ Average ratios are given in mol mol⁻¹.
ᵈ Commercial cultivars.
of sucrose. Therefore, it is important to study the stability of such germplasm under different environmental conditions and to determine whether they present specific allelic variants contributing to the reduced content of sugars in tubers at harvest.

A reducing sugar content of 1–5 g kg\(^{-1}\) tuber DW is reported as appropriate for acceptable flavor,27–29 browning and acrylamide content in roasted and baked potato products.30 In contrast, there are chip processing industries that have established a limit value of reducing sugar content of 0.35 g kg\(^{-1}\) tuber DW.31 Considering the value reported by the chip industry, the accessions with the lowest reducing sugar content and the commercial cultivars analyzed in this study (Table 1; supplementary Table S3) present fructose and glucose contents in the current environment (2905 m a.s.l.) that are not suitable for chip processing of tubers. This result supports the hypothesis that altitudes higher than 2600 m a.s.l. increment reducing sugar content after harvest in Phureja genotypes to undesirable levels for chip processing,32 although they might be adequate for other kinds of preparation. Therefore, for a better comprehension of the trait it is necessary to perform an evaluation of chip color and sugar content in tubers harvested in the same experimental field to compare both assessments and establish a limit value of reducing sugar content that predicts an adequate frying color in Group Phureja.

Sugar ratios

Glucose/fructose ratios in general terms presented values greater than 1.00, indicating that fructose tends to be present in lesser amounts than glucose in Phureja tubers at harvest (Table 1). Glucose concentration can be found up to six times higher than fructose, as in the case of CCC 2 (supplementary Table S3). Accessions CCC 42 and CCC 108 were the exception, as glucose and fructose contents were nearly equimolar (supplementary Table S3).

The variability of glucose and fructose contents in cold-stored tubers at 2 °C has also been studied across wild Solanum species, showing either balanced amounts of glucose and fructose or higher contents of fructose than glucose.33–34 The balanced amount of both reducing sugars is consistent with sucrose cleaving by invertase in cold-stored tubers.35 Analyzing the sugar content of 22 genotypes from a botanical seed accession of Group Phureja, McCann et al.34 found a mean glucose/fructose ratio of 0.52 after cold storage. These results suggest the influence of other sucrose-cleaving enzymes. Sucrose synthase is related to cold stress response and converts sucrose into UDP-glucose and fructose, thus causing higher concentrations of fructose in cold-stored tubers,34 which might explain the different ratios found by these authors in Group Phureja.

The range of glucose/fructose ratio found in Group Phureja at harvest suggests differential activities and affinities of fructokinases and hexokinases, which are enzymes responsible for fructose and glucose irreversible phosphorylation before their use in metabolic processes.27,36–38 It is more frequent to find glucose at greater concentrations in potato cells during growth. High concentrations of fructokinase have been found in potato extracts that have the ability to cycle fructose back into the hexose-phosphate pool.27,38 A greater content of glucose than of fructose is also common in other types of potato at harvest.32,39 and Colombian Phureja genotypes also present this pattern.

The range observed for the sucrose/reducing sugars ratio (Table 1) indicated the presence of genotypes with values lower than 1.00, representing individuals with greater content of reducing sugars than that of sucrose at harvest. Besides, there were genotypes with nearly eight times more sucrose than reducing sugars, as was the case for CCC 41 (Table 1; supplementary Table S3), which is caused by the low reducing sugar content present in this accession. We did not perform experiments in cold storage; nevertheless, the variability in this ratio has been correlated with vacuolar invertase activity in cold-stored tubers;35 hence there might be a differential enzymatic activity in Phureja accessions evident at harvest because of the cold temperatures present during tuber development. This is notable in the genotypes with ratios lower than 1.00, which might present a potential increased vacuolar invertase activity that causes greater content of reducing sugars at harvest time.

The previous results show that it is necessary to carry out experiments to test the differences in enzymatic activity, protein structure, expression levels and nucleotide sequences of fructokinases, hexokinases and vacuolar invertases in Colombian accessions of Group Phureja to understand their effect in the variation observed in sugar ratios of tubers at harvest.

Correlation among variables

The correlation coefficients that were found among the eight variables considered in this study are shown in Table 2. A strong and significant correlation was found between glucose and fructose contents. Sucrose was significantly correlated with reducing sugar content, revealing that the disaccharide content has a moderate relationship with the reducing sugar content in Phureja genotypes at harvest. Sucrose/reducing sugar ratio presented a strong negative correlation with reducing sugar content and a non-significant correlation with sucrose content. These results indicate that this ratio variability is more related to reducing sugar variation.

Chip darkening index presented moderate significant correlations with glucose, reducing sugars and total sugar content with coefficients greater than 0.4. The correlation results show that, despite the specific interactions of the genotypes and the environment, there is a positive relationship between sugar content and frying color in Colombian accessions of Group Phureja. Consequently, it is proposed that the total sugars measured in the current environment moderately influence the variation of the multi-environmental estimate of chip darkening index.

Furthermore, studies with chip color and sugar content data from the same experimental field do not report strong correlations between reducing sugar content and frying color,34,40 mainly because reducing sugar content alone does not explain frying color trait. Non-enzymatic browning can also be influenced by the contents of amino acids, ascorbic acid and phenolic acids; therefore, when low reducing sugar content is present in the tubers, these other compounds might be important in explaining frying color.40 Hence it is necessary to measure amino acid, ascorbic acid and phenolic acids content to study their individual role in non-enzymatic browning in order to obtain a comprehensive understanding of chip color development in Group Phureja.

Principal component analysis

The previous results reveal the extensive variation present in Phureja genotypes regarding sugar content and sugar ratios. This variation is depicted in the PCA biplot (Fig. 1), where the two principal components explain 77.0% of the total inertia, thus displaying a high amount of the total variability. The magnitude calculated for the vectors of the variables in the biplot (supplementary Table S1) supports that all of them are well represented in the factorial space, with the exception of glucose/fructose ratio, which shows a value of 0.57. This magnitude is close to 0.60, which is the limit
value that indicates an appropriate representation of the variables in the factorial space.

The disposition of the vectors in the biplot is representative of the correlation coefficients found among the variables (Table 2). The proximity of the vectors representing glucose, fructose and reducing sugar contents supports the high positive correlation found among them. The opposite direction of the sucrose/reducing sugars vector reflects the negative correlation with reducing sugar and total sugar contents. Finally, the independent tendency that shows the orientation of the vector of glucose/fructose ratio reveals the non-significant or weak correlation between this ratio and the other variables. Therefore, the PCA biplot (Fig. 1) is an appropriate tool to demonstrate the relationship among all the analyzed variables and to show their variability.

Cluster analysis

The factor-based cluster analysis shows five groups in the factorial space (Fig. 1). The resulting dendrogram is presented in supplementary Fig. S1, with a red line showing the level of partition that displays the five groups with biological relevance. These clusters distributed in the factorial space are conditioned by the direction of the vectors of the variables. Thus the correspondence of the group position with the vector direction supports the greater weight of that variable in the definition of that particular cluster. Supplementary Table S3 also includes the group that corresponds to each genotype.

The biplot and the dendrogram show that Groups 3 and 4 are closely related clusters mainly conditioned by the vector of sucrose/reducing sugars ratio (Fig. 1; supplementary Fig. S1). Group 3 (49.1% of genotypes) is the most frequent and defines genotypes with lower sucrose/reducing sugar ratios than Group 4; hence these genotypes tend to present greater content of reducing sugars. Group 4 (27.7% of genotypes) comprises accessions with high values of sucrose/reducing sugar ratios, which includes the Phureja genotypes with the lowest content of reducing sugars, including commercial cultivars with the exception of Criolla Colombia. The cluster analysis indicates that, in fact, the most common feature of Phureja genotypes at harvest in the current environment assessed is a high concentration of sucrose and a low concentration of reducing sugars (represented in Groups 3 and 4), which is also reflected in the PCA biplot by the dense cumulus of genotypes towards the minimum values of this variable in the factorial space.

Group 2 (17.0% of genotypes) is mainly conditioned by the sucrose vector and covers accessions with high content of the disaccharide and intermediate values of reducing sugars. Group 1 (3.6% of genotypes) is influenced by the vector of glucose/fructose ratio and represents the accessions with the highest values. This cluster includes only four genotypes with glucose/fructose ratios higher than 3.50 (supplementary Table S3), revealing that this elevated content of glucose at harvest is rare in the natural variation of Phureja accessions. Therefore, these accessions might use mechanisms that favor fructose phosphorylation; this needs to be proven, as discussed above in the sugar ratios analysis.

Group 5 (2.7% of genotypes) includes the lowest percentage of accessions. This cluster is mainly conditioned by the vectors of glucose, reducing sugars and fructose, and presents the genotypes with the greatest values of reducing sugars. Accessions CCC 108 and CCC 120, which show the highest reducing sugar content, agree with high and undesirable estimates of chip darkening index of 79.46% and 68.26%, respectively (supplementary Table S2). The rare Phureja genotypes with extreme phenotypic values are of great interest for further studies because they are often enriched with uncommon genetic variants that can contribute to explain complex trait variability and present a large effect on the trait.41,42

CONCLUSIONS

Sugar content analysis in Colombian genotypes of Group Phureja revealed an extensive variability and is consistent with previous data of frying color from the collection because of the moderate significant correlations found between both results. These moderate correlations are supported by the correspondence of

### Table 2. Spearman’s correlation coefficients among average sugar contents, average sugar ratios and multi-environmental chip darkening indexes studied in accessions of CCC (108 genotypes) and four commercial cultivars from Solanum tuberosum Group Phureja

| Sucrose<sup>b</sup> | Glucose<sup>b</sup> | Fructose<sup>b</sup> | Reducing sugars<sup>b</sup> | Total sugars<sup>b</sup> | Glucose/fructose<sup>c</sup> | Sucrose/reducing sugars<sup>c</sup> | Chip darkening index<sup>d</sup> |
|---------------------|---------------------|---------------------|--------------------------|--------------------------|--------------------------|-----------------------------------|-------------------------------|
|                     | 0.48***             | 0.51***             | 0.49***                  | 0.94***                  | −0.10 ns                 | −0.01 ns                         | 0.34**                        |
| Glucose<sup>b</sup> |                     |                     |                          |                          |                         |                                   |                               |
|                     | 0.48***             | 0.90***             | 0.99***                  | 0.72***                  | −0.05 ns                 | −0.01 ns                         | 0.43**                         |
| Fructose<sup>b</sup> |                     |                     |                          |                          |                         |                                   |                               |
|                     | 0.51***             | 0.90***             | 0.94***                  | 0.72***                  | −0.40***                 | −0.78***                         | 0.38***                        |
| Reducing sugars<sup>b</sup> |                     |                     |                          |                          |                         |                                   |                               |
|                     | 0.49***             | 0.99***             | 0.94***                  | 0.73***                  | −0.14 ns                 | −0.83***                         | 0.43**                         |
| Total sugars<sup>b</sup> |                     |                     |                          |                          |                         |                                   |                               |
|                     | 0.94***             | 0.72***             | 0.72***                  | 0.73***                  | −0.13 ns                 | −0.01 ns                         | 0.43**                         |
| Glucose/fructose<sup>c</sup> | −0.10 ns           | −0.05 ns            | −0.40***                 | −0.14 ns                 | −0.13 ns                 | −0.13 ns                         | 0.04 ns                        |
| Sucrose/reducing sugars<sup>c</sup> | −0.02 ns           | −0.85***            | −0.78***                 | −0.85***                 | −0.31***                 | −0.11 ns                         | −0.32**                        |
| Chip darkening index<sup>d</sup> | 0.34**             | 0.44***             | 0.38***                  | 0.43***                  | 0.43***                  | 0.04 ns                          | −0.31**                        |

<sup>a</sup> CCC denotes the accessions from the Colombian Central Collection that constitute the Working Collection from the Potato Breeding Program at the National University of Colombia.

<sup>b</sup> Average sugar content is expressed in g kg<sup>−1</sup> dry weight tuber.

<sup>c</sup> Average ratios are given in mol mol<sup>−1</sup>.

<sup>d</sup> The chip darkening index from 108 genotypes from CCC derives from a Bayesian estimation that resulted from the assessment of a multi-environmental trial as described by Ñústez-López.15

P-values: ***P ≤ 0.0001; **P ≤ 0.001; *P ≤ 0.05; ns, P > 0.05, the coefficient is not significant.
reducing sugar content and chip darkening indexes of contrasting genotypes. The sucrose content analysis provides some insights into the potential greater accumulation of reducing sugars in the subsequent storage of tubers from the different genotypes, thus indicating which genotypes could be more useful for further analyses in cold storage. The analysis of glucose/fructose and sucrose/reducing sugar ratios disclosed also a wide range that might be related to variations in the enzymatic activity, protein structure, expression levels and nucleotide sequences of fructokinases, hexokinases and vacuolar invertases in accessions of Group Phureja, which needs to be proven.

Multivariate analysis revealed five clusters of genotypes with biological relevance, each one influenced to a higher degree by a specific variable. Groups 3 and 4 are related clusters that include 76.8% of the Phureja genotypes with an average sucrose content of $12.52 \pm 2.62\, \text{g}\,\text{kg}^{-1}$ tuber DW and an average reducing sugar content of $2.43 \pm 1.16\, \text{g}\,\text{kg}^{-1}$ tuber DW. The clusters with the lowest amount of genotypes include rare phenotypes that are of great interest in understanding the variability of sugar content at harvest. A deep study of the genotypes with low sugar content is necessary to establish their stability across environments and to recognize their potential mechanisms for low sugar concentrations. The current results complement the frying color analysis data and represent a quantitative approach to comprehend the frying quality trait by studying these precursors of the Maillard reaction. Therefore, these results are relevant to advance in the frying quality trait by studying these precursors of the Maillard reaction. The analysis of glucose/fructose and sucrose/reducing sugar ratios disclosed also a wide range that might be related to variations in the enzymatic activity, protein structure, expression levels and nucleotide sequences of fructokinases, hexokinases and vacuolar invertases in accessions of Group Phureja, which needs to be proven.

**SUPPORTING INFORMATION**
Supporting information may be found in the online version of this article.

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