

Characterization of genetic resources of onion (*Allium cepa* L.) from the Spanish secondary centre of diversity

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Abstract

Onions are the second most-valuable vegetable in the world. Despite its global culinary and economic significance, the knowledge of genetic diversity and resources is limited. The aim of this study was to morphologically and physico-chemically characterize eighty-six onion landraces from Spain, part of the secondary Mediterranean Centre of diversity. The evaluated traits in the bulb included: weight, shape, firmness, soluble solids content (SSC), pungency and sugars content (glucose, fructose and sucrose). The results evidenced a great variability in all the evaluated traits. Correlations were significant between pungency and SSC ($r=0.34$), firmness ($r=0.32$), and sucrose content ($r=0.34$); between fructose and glucose contents ($r=0.79$); between sucrose content and SSC ($r=0.57$); between SSC and weight ($r=-0.35$); between fructose and sucrose contents ($r=-0.22$); and between weight and sucrose content ($r=-0.43$). The combination of cluster and discriminant analyses resulted in a classification into four clusters (95.3% fit). Cluster 1 is represented by firm, pungent and large-sized bulbs; cluster 2 consists of large-sized, mild and sweet onions; cluster 3 is constituted by pungent, high-SSC and small-sized bulbs and cluster 4 is made up by elongated bulbs. The variability in important agronomical traits found point out that these onion accessions could be candidates for future breeding programs. In addition, the clustering could make the initial plant material selection easier and the correlations among the evaluated traits found could help to establish adequate selection strategies.

Additional key words: cluster analysis; firmness; genetic diversity; pungency; soluble solids; sugars.

Resumen

Caracterización de los recursos genéticos de cebolla (*Allium cepa* L.) procedentes del centro secundario de diversificación de España

La cebolla es el segundo cultivo hortícola a nivel mundial. Sin embargo, el conocimiento sobre los recursos y la diversidad genética de esta especie es limitado. Por ello, el objetivo del presente trabajo consistió en el estudio morfológico y físico-químico de una colección de 86 cultivares de cebolla procedentes de España (que forma parte del centro secundario de diversificación). Los caracteres evaluados en el bulbo incluyeron: peso, forma, firmeza, contenido en sólidos solubles (SSC), pungencia y contenido en azúcares (glucosa, fructosa y sacarosa). Los resultados evidenciaron una gran variabilidad en todos los caracteres evaluados. Las correlaciones fueron significativas entre la pungencia y SSC ($r=0,34$), firmeza ($r=0,32$) y contenido en sacarosa ($r=0,34$); entre contenido en fructosa y glucosa ($r=0,79$); entre contenido en sacarosa y SSC ($r=0,57$); entre SSC y peso ($r=-0,35$); entre contenido en fructosa y sacarosa ($r=-0,22$) y entre el peso y contenido en sacarosa ($r=-0,43$). El análisis discriminante dio lugar a la formación de cuatro grupos con un ajuste del 95,3%. El grupo 1 está formado por bulbos grandes, firmes y picantes, el grupo 2 por cebollas grandes, suaves y dulces, el grupo 3 por cebollas pequeñas, picantes y con un alto SSC y el grupo 4 por bulbos de forma alargada. La variabilidad encontrada para caracteres de interés agronómico indica que este material podría ser utilizado en futuros programas de mejora genética de esta especie. Además, la agrupación de los cultivares contribuiría a localizar mejor el material vegetal de interés y las correlaciones halladas entre los caracteres evaluados a establecer una adecuada estrategia de selección.

Palabras clave adicionales: análisis cluster; azúcares; diversidad genética; firmeza; pungencia; sólidos solubles.

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Introduction

Onions are the second most-valuable vegetable in the world, following only tomato. Despite the global culinary and economic significance, genetic research of onion has greatly lagged that of other major vegetable crops (McCallum *et al.*, 2008). The knowledge of onion genetic diversity and resources is limited mainly due to a paucity of public marker and germplasm resources and their out-breeding, biennial habit (McCallum and Havey, 2006).

According to Vavilov (1926) the Southwest Asian gene centre is proposed as the primary centre of domestication and variability of onion. In addition, the secondary centre of origin in the Mediterranean gene centre represents the area from which onions with large bulbs were selected (Castell and Portas, 1994).

Because onions have been cultivated for so long, and their bulb and inflorescence development must be closely adapted to the temperatures and photoperiods that prevail where they are grown, there exists a huge range of cultivars and landraces, developed over the centuries to fit the diverse climates and food preferences of the world (Brewster, 1994). Onions show particular diversity in the Mediterranean countries and these regions are, therefore, most important sources of genetic diversity (Astley *et al.*, 1982).

The availability of genetic variation within crops, present in *ex situ* and *in situ* collections, is of pivotal importance for a sustainable agriculture. The improvement in any crop is proportional to the magnitude of its genetic variability present in the genotypes. A total of 9,000 accessions of *Allium* were reported to be present worldwide and the number of onion accessions was 7,000, by far the largest one (Kik, 2008). As in many other crop species, genetic erosion is taking place also in *Allium cepa* L. Currently around 12,740 onion accessions (some of them duplicates) are held in genebanks worldwide (Kik, 2008). The characterization and evaluation of these accessions would be of importance for the utilization of the germplasm. Modern varieties sold by international seed companies, in particular F1 hybrids which have a narrow genetic base, are replacing old varieties, with a danger that these, and therefore many potentially valuable and adaptive genes they contain, would be lost. Hence, the characterization, collection, preservation and regeneration of seeds and vegetable clones of these old varieties and landraces are very important (Astley, 1990).

In Spain, onions are grown from the arid South to the lush green and mountainous Northwest, passing through the high plains of the interior; and as well in the Balears and Canary Islands, placed in the Mediterranean Sea and the Atlantic Ocean respectively. The Vegetable Germplasm Bank of Zaragoza (BGHZ) and the Centre for the Conservation and Breeding of the Agrobiodiversity (COMAV), both in Spain, hold important *Allium cepa* L. collections where most of the Spanish onion variability is represented (Castell and Diez, 2000; Carravedo and Mallor, 2007). Some of these traditional onion varieties are still grown in some regions of Spain due to their high quality and acceptance by local markets, but others have already disappeared.

Activities related to genetic resources are characterized by high cost and long term return. Introduction and germplasm exchange, collection, characterization, evaluation, documentation and conservation are essential steps that can not be overemphasized. The importance of genetic resources is widely recognized. Besides the conservation of genetic variability for the future, the actual utilization of available accessions is another important goal. However, the low utilization of germplasm banks is a rule world wide (Nass and Paterniani, 2000). The main factors responsible for the low utilization of plant genetic resources are lack of documentation and adequate description of collections, lack of the desired information by breeders, accessions with restricted adaptability, insufficient plant breeders, particularly in developing countries, and lack of collection evaluations. The purpose of the study reported here was, therefore, to improve the knowledge of the Spanish onion landraces in order to reduce the gap between the available genetic resources and onion breeding programs. Measurements were based on a range of morphological and physico-chemical parameters related to onion quality such as bulb weight, shape, colour, firmness, soluble solids, pungency and sugars in eighty-six onion accessions held at the BGHZ.

Material and methods

Plant material

Eighty-six Spanish onion accessions were supplied by the Vegetable Germplasm Bank of Zaragoza (BGHZ) (Spain). These accessions are local onion landraces collected as seeds from farmers in the main growing regions of Spain between 1981 and 2006. Onion seeds

were grown during the same season in a randomized block design with three replications at the *Centro de Investigación y Tecnología Agroalimentaria* (CITA) of Zaragoza, Spain (latitude 41° 39' N), where soils have a loam texture, moderately basic pH and relatively high salt contents.

Field crops were initiated in April being the seedlings cultivated in greenhouse from January. Transplants were done in double-row beds 0.8 m apart where plants were set at 10 × 15 cm. Plants were drip-irrigated and bulb harvest was performed from 21 July to 11 September; when 50% of the leaves of each accession felt over. Curing was made in the field during three days to allow the leaves to dry and turn brown, leaves and roots were then cut off, and 25 bulbs of each plot, made up by 80 plants, placed into plastic storage containers (60 × 40 × 20 cm) previously disinfected.

Bulb analysis

A sample of ten bulbs per accession was analyzed. Bulbs were evaluated for the following traits:

— Bulb size and shape were estimated by determining the weight, height, and maximum diameter of each bulb; then, the bulb shape index (BSI) was estimated as ratio height/diameter. Colour measurements were done using the Royal Horticultural Society Colour Chart.

— Firmness of the bulb was measured on opposite paired cheeks at the equatorial region using a penetrometer (Tr[®] mod. 53205, Forli, Italy) equipped with an 8 mm cylindrical plunger. Data were expressed in kg cm⁻².

— The soluble solids content (SSC) of onion juice samples was measured using a digital refractometer (Tr[®] mod. 53020; Forli, Italy). Data were expressed as equivalent °Brix (or %SSC).

— Pungency of the bulb was evaluated by quantifying the pyruvic acid produced after cell disruption, using the method proposed by Schwimmer and Weston (1961) as modified by Boyhan *et al.* (1999). Onion juice samples were obtained from equator-transverse sections 1 cm thick. Results for pungency were expressed in micromoles of enzymatically formed pyruvic acid per gram of fresh weight (μmol g⁻¹ FW).

— The separation of different sugars (fructose, glucose and sucrose) was carried out by high performance liquid chromatography (HPLC), and they were detected using a differential refractive index detector model ERC-7512 (Erma Inc., Tokyo, Japan) at 40°C

temperature work. Identification of chromatographic peaks was based on retention times by comparison with known standards (Merck, Darmstadt, Germany). An external calibration curve was prepared for each sugar standard to calculate the concentration of sugars present in the onion juice. Juice samples of 0.5 mL were dissolved in 2 mL of Mill-Q water. Then, 20 μL of the aqueous extracts were filtered by 0.45 μm nylon filters and injected in a HPLC 360 (Kontron, Milan, Italy), equipped with a double piston pump, self-sampler, and regulated furnace at 40°C. The column used was a 250 mm × 4.6 mm i.d., 5 μm, Kromasil Amine. The lineal range for the concentration determination of each sugar was 0.01 to 2 g/100 g.

Data analysis

A clustering of observations was performed to classify onion types into groups. It was applied using the squared of Pearson distance between observations (the sum of square distances divided by variances) and the Ward method as linkage procedure (the distance between two clusters is the sum of squared deviations from points to centroids and its objective is to minimize the within-cluster sum of squares). Analyses of variance (ANOVA) were carried out for all variables to check the differences in their mean values among the groups obtained from the clustering stage and complemented with a post hoc analysis (Duncan test) to detect set of groups with no statistical differences in their means.

After the classification of the onion types in 4 groups, statistically different and with a physical interpretation, a discriminant analysis was executed to obtain a rule to classify new observations (onions) into one of these groups. An observation is classified into a group if the Mahalanobis distance of observation to the group mean is the minimum. There is a unique part of the squared distance formula for each group and that is called the linear discriminant function for that group. For any observation, the group with the smallest squared distance has the largest linear discriminant function and the observation is then classified into this group. Discriminant analysis calculates these discriminant functions. All onions are classified into the groups using these discriminant functions.

Statistical analyses were performed by using the software Minitab[®] 15.1, for cluster and discriminant analyses, and SPSS Statistics 17.0 for the ANOVA and other descriptive analyses.

Results and discussion

Bulb traits

The onion accessions studied here evidenced great intervarietal variability for most of the characters studied (Table 1). Significant differences ($p < 0.001$)

were observed among accessions for all the evaluated traits, and mean coefficients of variation (CV) varied from 20.4% for SSC to 85.7% for sucrose. The variability of genetic resources found in this study agrees with the fact that Spain is part of the secondary centre of onion diversification (Astley *et al.*, 1982).

Table 1. Characterization of Spanish onion accessions held in the Vegetable Germplasm Bank of Zaragoza (Spain) according to weight, shape, skin and flesh colour, firmness, soluble solids, pungency and sugars content. Data are means \pm standard deviations ($n = 10$); for colour the predominant one has been specified following the Royal Horticultural Society's Colour Chart (RHS code)

| Accession number | Weight (g) | Shape (height (\O^{-1})) | Colours (RHS code) | | Firmness (kg cm^{-2}) | Soluble solids ($^{\circ}\text{Brix}$) | Pungency ($\mu\text{mol pyruvic acid g}^{-1}$) | Sugars ($\text{g}/100\text{ g}$) | | | |
|------------------|------------|-------------------------------------|--------------------|-------|----------------------------------|--|--|------------------------------------|---------------|---------------|---------------|
| | | | Skin | Flesh | | | | Fructose | Glucose | Sucrose | |
| 1 | BGHZ-0182 | 442.2 \pm 122.9 | 0.8 \pm 0.5 | 167/B | 155/D | 7.6 \pm 1.2 | 8.3 \pm 0.8 | 7.9 \pm 2.7 | 1.7 \pm 0.3 | 2.0 \pm 0.4 | 0.2 \pm 0.0 |
| 2 | BGHZ-0225 | 59.0 \pm 34.9 | 0.8 \pm 0.2 | 164/D | 155/D | 4.1 \pm 1.0 | 10.3 \pm 1.1 | 7.1 \pm 1.4 | 2.3 \pm 0.4 | 2.6 \pm 0.3 | 1.1 \pm 0.2 |
| 3 | BGHZ-0227 | 269.9 \pm 60.8 | 1.9 \pm 0.3 | 184A | 183/C | 5.5 \pm 1.7 | 8.7 \pm 1.1 | 3.0 \pm 0.8 | 1.1 \pm 0.3 | 1.3 \pm 0.3 | 1.6 \pm 0.0 |
| 4 | BGHZ-0228 | 328.2 \pm 59.1 | 0.6 \pm 0.0 | 171/D | 155/D | 5.2 \pm 0.8 | 8.4 \pm 0.7 | 4.7 \pm 1.3 | 2.1 \pm 0.5 | 2.0 \pm 0.1 | 1.0 \pm 0.3 |
| 5 | BGHZ-0229 | 293.9 \pm 99.1 | 1.2 \pm 0.3 | 185/B | 183/C | 4.8 \pm 1.4 | 8.3 \pm 1.5 | 4.2 \pm 3.1 | 1.0 \pm 0.4 | 1.3 \pm 0.4 | 0.7 \pm 0.7 |
| 6 | BGHZ-0230 | 230.0 \pm 29.6 | 0.8 \pm 0.1 | 182/D | 155/D | 5.6 \pm 0.8 | 5.9 \pm 0.9 | 10.5 \pm 2.1 | 1.8 \pm 0.3 | 2.0 \pm 0.3 | 0.3 \pm 0.0 |
| 7 | BGHZ-0231 | 206.9 \pm 45.9 | 0.7 \pm 0.0 | 155/C | 155/B | 6.1 \pm 1.5 | 12.9 \pm 1.2 | 11.4 \pm 4.3 | 0.9 \pm 0.3 | 1.2 \pm 0.3 | 0.6 \pm 0.1 |
| 8 | BGHZ-0233 | 348.2 \pm 58.9 | 0.9 \pm 0.1 | 173/B | 155/D | 7.7 \pm 2.6 | 9.6 \pm 0.9 | 11.7 \pm 3.7 | 1.9 \pm 0.1 | 2.2 \pm 0.1 | 0.3 \pm 0.1 |
| 9 | BGHZ-0264 | 273.0 \pm 78.4 | 0.9 \pm 0.1 | 180/D | 155/D | 4.6 \pm 1.6 | 7.8 \pm 1.1 | 3.5 \pm 1.3 | 2.2 \pm 0.2 | 2.5 \pm 0.3 | 0.6 \pm 0.3 |
| 10 | BGHZ-0265 | 119.4 \pm 49.4 | 0.7 \pm 0.1 | 168/C | 155/D | 5.7 \pm 1.9 | 9.2 \pm 1.4 | 10.3 \pm 4.7 | 1.0 \pm 0.3 | 1.5 \pm 0.5 | 1.1 \pm 0.6 |
| 11 | BGHZ-0268 | 183.4 \pm 40.7 | 0.6 \pm 0.1 | 186/D | 155/D | 4.9 \pm 0.5 | 8.1 \pm 1.5 | 7.9 \pm 2.7 | 2.1 \pm 0.4 | 2.3 \pm 0.5 | 0.2 \pm 0.0 |
| 12 | BGHZ-0315 | 177.9 \pm 51.8 | 0.8 \pm 0.1 | 181/C | 155/D | 5.2 \pm 1.4 | 11.2 \pm 1.5 | 9.4 \pm 4.0 | 1.4 \pm 0.6 | 1.9 \pm 0.3 | 0.9 \pm 0.3 |
| 13 | BGHZ-0323 | 192.9 \pm 55.1 | 1.1 \pm 0.2 | 168/C | 155/D | 3.0 \pm 1.0 | 8.5 \pm 1.2 | 4.3 \pm 1.5 | 1.9 \pm 0.1 | 2.9 \pm 0.1 | 0.3 \pm 0.0 |
| 14 | BGHZ-0325 | 289.3 \pm 124.2 | 0.9 \pm 0.2 | 170/B | 155/D | 5.2 \pm 0.9 | 7.5 \pm 0.7 | 7.8 \pm 2.8 | 1.8 \pm 0.2 | 2.0 \pm 0.3 | 0.4 \pm 0.2 |
| 15 | BGHZ-0347 | 62.1 \pm 11.3 | 0.7 \pm 0.1 | 187/B | 186/C | 5.6 \pm 0.8 | 11.6 \pm 1.1 | 7.5 \pm 1.9 | 2.0 \pm 0.6 | 2.8 \pm 0.4 | 0.9 \pm 0.2 |
| 16 | BGHZ-0349 | 312.2 \pm 114.0 | 2.3 \pm 0.5 | 168/C | 183/C | 5.8 \pm 1.3 | 8.5 \pm 0.9 | 8.3 \pm 2.3 | 1.3 \pm 0.5 | 1.6 \pm 0.6 | 0.5 \pm 0.3 |
| 17 | BGHZ-0352 | 189.0 \pm 44.4 | 0.6 \pm 0.1 | 155/D | 155/D | 5.3 \pm 0.5 | 5.9 \pm 0.6 | 5.8 \pm 1.3 | 1.8 \pm 0.2 | 2.2 \pm 0.3 | 0.2 \pm 0.0 |
| 18 | BGHZ-0352 | 177.7 \pm 84.8 | 0.6 \pm 0.0 | 186/A | 183/C | 5.5 \pm 2.0 | 8.5 \pm 0.8 | 9.8 \pm 2.8 | 1.4 \pm 0.6 | 2.5 \pm 1.2 | 0.4 \pm 0.1 |
| 19 | BGHZ-0353 | 304.0 \pm 74.4 | 0.9 \pm 0.1 | 183/C | 183/C | 5.9 \pm 2.0 | 8.1 \pm 0.9 | 9.6 \pm 2.8 | 1.7 \pm 0.2 | 2.0 \pm 0.3 | 0.4 \pm 0.1 |
| 20 | BGHZ-0360 | 242.9 \pm 91.6 | 0.9 \pm 0.1 | 167/B | 155/D | 6.4 \pm 2.4 | 7.9 \pm 0.9 | 9.9 \pm 2.0 | 2.5 \pm 1.2 | 2.9 \pm 1.3 | 0.3 \pm 0.1 |
| 21 | BGHZ-0433 | 203.3 \pm 45.6 | 0.6 \pm 0.1 | 170/A | 155/D | 4.9 \pm 1.1 | 7.6 \pm 1.4 | 5.2 \pm 1.6 | 2.1 \pm 0.5 | 3.2 \pm 0.5 | 1.0 \pm 0.4 |
| 22 | BGHZ-0434 | 186.7 \pm 44.6 | 0.8 \pm 0.1 | 172/C | 155/D | 4.8 \pm 1.7 | 8.9 \pm 1.2 | 5.6 \pm 1.4 | 1.6 \pm 0.1 | 1.8 \pm 0.3 | 0.6 \pm 0.3 |
| 23 | BGHZ-0534 | 443.2 \pm 108.7 | 0.9 \pm 0.1 | 173/B | 155/D | 4.3 \pm 0.9 | 7.2 \pm 1.2 | 3.5 \pm 1.8 | 1.9 \pm 0.4 | 2.1 \pm 0.4 | 0.3 \pm 0.1 |
| 24 | BGHZ-0535 | 255.1 \pm 31.5 | 1.0 \pm 0.1 | 183/C | 183/C | 6.3 \pm 1.7 | 7.9 \pm 1.3 | 16.0 \pm 4.1 | 1.0 \pm 0.7 | 1.0 \pm 0.8 | 0.4 \pm 0.3 |
| 25 | BGHZ-0536 | 70.0 \pm 37.4 | 0.8 \pm 0.1 | 174/D | 155/D | 4.5 \pm 1.1 | 11.3 \pm 0.8 | 12.2 \pm 2.9 | 2.4 \pm 0.3 | 1.5 \pm 0.0 | 0.7 \pm 0.1 |
| 26 | BGHZ-0537 | 104.0 \pm 20.1 | 0.7 \pm 0.1 | 171/D | 155/D | 4.7 \pm 0.9 | 10.6 \pm 1.5 | 9.9 \pm 1.9 | 1.5 \pm 0.3 | 1.8 \pm 0.7 | 1.4 \pm 0.4 |
| 27 | BGHZ-0599 | 210.6 \pm 34.8 | 0.8 \pm 0.1 | 174/D | 155/D | 5.1 \pm 0.9 | 5.8 \pm 0.9 | 8.2 \pm 1.9 | 1.9 \pm 0.4 | 2.6 \pm 0.3 | 0.4 \pm 0.1 |
| 28 | BGHZ-0600 | 244.1 \pm 87.3 | 0.8 \pm 0.1 | 168/C | 155/D | 7.9 \pm 2.2 | 9.1 \pm 1.8 | 11.5 \pm 4.8 | 1.3 \pm 0.5 | 1.6 \pm 0.6 | 0.4 \pm 0.3 |
| 29 | BGHZ-0601 | 262.8 \pm 58.8 | 0.6 \pm 0.0 | 168/C | 155/D | 6.5 \pm 1.0 | 8.0 \pm 0.8 | 11.1 \pm 3.5 | 1.6 \pm 0.2 | 1.8 \pm 0.2 | 0.4 \pm 0.1 |
| 30 | BGHZ-0604 | 104.8 \pm 35.0 | 0.7 \pm 0.0 | 155/D | 155/D | 3.8 \pm 0.9 | 10.1 \pm 0.9 | 7.3 \pm 2.7 | 1.8 \pm 0.1 | 1.7 \pm 0.2 | 0.5 \pm 0.3 |
| 31 | BGHZ-0799 | 318.9 \pm 104.1 | 0.8 \pm 0.1 | 181/B | 183/C | 4.9 \pm 0.8 | 8.3 \pm 1.5 | 7.4 \pm 2.7 | 1.3 \pm 0.5 | 1.3 \pm 0.6 | 0.3 \pm 0.1 |
| 32 | BGHZ-0801 | 289.1 \pm 56.1 | 0.6 \pm 0.0 | 168/C | 155/D | 5.4 \pm 2.2 | 8.8 \pm 1.0 | 8.6 \pm 3.0 | 1.8 \pm 0.4 | 2.2 \pm 0.4 | 0.4 \pm 0.3 |
| 33 | BGHZ-0803 | 301.7 \pm 90.7 | 0.8 \pm 0.1 | 168/B | 155/D | 3.3 \pm 1.1 | 7.8 \pm 1.2 | 7.2 \pm 3.5 | 1.5 \pm 0.8 | 1.5 \pm 0.8 | 0.2 \pm 0.1 |
| 34 | BGHZ-0805 | 343.9 \pm 90.7 | 0.6 \pm 0.1 | 182/A | 183/C | 5.4 \pm 1.1 | 8.0 \pm 0.8 | 8.4 \pm 3.5 | 1.6 \pm 0.3 | 1.5 \pm 0.5 | 0.3 \pm 0.2 |
| 35 | BGHZ-0903 | 208.3 \pm 30.1 | 0.7 \pm 0.1 | 155/D | 155/D | 5.0 \pm 0.7 | 7.6 \pm 0.8 | 5.0 \pm 1.1 | 2.7 \pm 0.3 | 3.1 \pm 0.3 | 0.3 \pm 0.1 |
| 36 | BGHZ-0904 | 162.1 \pm 66.6 | 0.6 \pm 0.1 | 182/A | 185/C | 4.4 \pm 1.2 | 9.2 \pm 1.1 | 7.7 \pm 3.0 | 2.0 \pm 0.5 | 3.0 \pm 0.3 | 1.0 \pm 0.2 |
| 37 | BGHZ-0906 | 107.5 \pm 42.2 | 1.0 \pm 0.1 | 168/D | 137/A | 4.0 \pm 0.8 | 7.5 \pm 0.9 | 4.1 \pm 1.3 | 1.3 \pm 0.3 | 1.3 \pm 0.3 | 0.2 \pm 0.0 |
| 38 | BGHZ-0907 | 360.0 \pm 57.1 | 0.7 \pm 0.0 | 173/C | 155/D | 4.3 \pm 0.4 | 7.7 \pm 0.4 | 5.8 \pm 1.6 | 1.8 \pm 0.6 | 2.0 \pm 0.6 | 0.1 \pm 0.1 |
| 39 | BGHZ-1127 | 357.8 \pm 96.8 | 0.6 \pm 0.1 | 173/C | 183/C | 5.9 \pm 1.4 | 10.3 \pm 1.4 | 10.6 \pm 2.8 | 1.5 \pm 2.3 | 2.2 \pm 0.4 | 1.3 \pm 0.1 |
| 40 | BGHZ-1129 | 291.7 \pm 92.9 | 0.7 \pm 0.1 | 183/D | 183/C | 4.2 \pm 0.6 | 7.4 \pm 1.0 | 5.5 \pm 2.2 | 1.7 \pm 0.6 | 1.9 \pm 0.6 | 0.1 \pm 0.0 |

Table 1 (cont.). Characterization of Spanish onion accessions held in the Vegetable Germplasm Bank of Zaragoza (Spain) according to weight, shape, skin and flesh colour, firmness, soluble solids, pungency and sugars content. Data are means \pm standard deviations (n = 10); for colour the predominant one has been specified following the Royal Horticultural Society's Colour Chart (RHS code)

| Accession number | Weight (g) | Shape (height (\O^{-1})) | Colours (RHS code) | | Firmness (kg cm^{-2}) | Soluble solids ($^{\circ}\text{Brix}$) | Pungency ($\mu\text{mol pyruvic acid g}^{-1}$) | Sugars (g /100 g) | | | |
|------------------|---------------------|-------------------------------------|--------------------|-------|----------------------------------|--|--|-------------------|---------------|---------------|---------------|
| | | | Skin | Flesh | | | | Fructose | Glucose | Sucrose | |
| 41 | BGHZ-1132 | 420.7 \pm 73.0 | 0.9 \pm 0.1 | 170/A | 155/D | 9.0 \pm 3.7 | 9.2 \pm 1.3 | 9.3 \pm 2.9 | 2.0 \pm 0.4 | 2.1 \pm 0.4 | 0.8 \pm 0.2 |
| 42 | BGHZ-1134 | 339.2 \pm 134.5 | 0.9 \pm 0.2 | 155/D | 155/D | 4.5 \pm 1.1 | 8.2 \pm 1.0 | 5.7 \pm 2.2 | 1.6 \pm 0.5 | 1.8 \pm 0.4 | 0.1 \pm 0.1 |
| 43 | BGHZ-1352 | 378.6 \pm 38.9 | 0.9 \pm 0.1 | 170/B | 155/D | 6.6 \pm 2.6 | 8.4 \pm 1.3 | 11.7 \pm 4.8 | 1.1 \pm 0.1 | 1.1 \pm 0.2 | 0.2 \pm 0.0 |
| 44 | BGHZ-1354 | 132.5 \pm 22.5 | 0.7 \pm 0.4 | 155/D | 155/D | 5.9 \pm 1.5 | 12.4 \pm 1.1 | 18.1 \pm 3.5 | 1.0 \pm 0.2 | 1.5 \pm 0.6 | 1.2 \pm 0.5 |
| 45 | BGHZ-1355 | 118.8 \pm 53.8 | 0.8 \pm 0.1 | 155/D | 155/D | 6.1 \pm 1.7 | 11.2 \pm 0.7 | 10.0 \pm 3.3 | 2.3 \pm 0.2 | 2.2 \pm 0.5 | 1.3 \pm 0.3 |
| 46 | BGHZ-1356 | 104.9 \pm 46.4 | 0.8 \pm 0.2 | 174/D | 155/D | 5.2 \pm 1.0 | 8.9 \pm 1.6 | 9.4 \pm 2.7 | 1.7 \pm 0.1 | 2.4 \pm 0.1 | 1.0 \pm 0.4 |
| 47 | BGHZ-1358 | 425.4 \pm 105.5 | 1.0 \pm 0.2 | 178/B | 183/C | 6.1 \pm 1.2 | 9.7 \pm 1.2 | 5.1 \pm 2.7 | 2.0 \pm 0.5 | 2.4 \pm 0.6 | 1.2 \pm 0.4 |
| 48 | BGHZ-1449 | 234.1 \pm 60.7 | 0.8 \pm 0.1 | 173/D | 155/D | 3.7 \pm 1.2 | 8.3 \pm 1.4 | 5.6 \pm 2.2 | 2.0 \pm 0.3 | 2.0 \pm 0.4 | 0.4 \pm 0.4 |
| 49 | BGHZ-1450 | 146.7 \pm 32.4 | 0.7 \pm 0.0 | 168/D | 155/D | 4.6 \pm 1.5 | 11.3 \pm 1.2 | 4.0 \pm 1.2 | 1.5 \pm 0.5 | 2.0 \pm 0.0 | 2.0 \pm 0.6 |
| 50 | BGHZ-1451 | 144.6 \pm 55.6 | 0.7 \pm 0.1 | 184/A | 183/C | 6.0 \pm 1.8 | 11.2 \pm 1.2 | 10.9 \pm 3.4 | 1.6 \pm 0.6 | 2.1 \pm 0.2 | 1.3 \pm 0.4 |
| 51 | BGHZ-1452 | 171.6 \pm 48.0 | 0.7 \pm 0.1 | 183/C | 155/D | 4.3 \pm 1.1 | 10.2 \pm 0.7 | 8.8 \pm 4.1 | 1.8 \pm 0.8 | 2.6 \pm 0.5 | 1.2 \pm 0.1 |
| 52 | BGHZ-1453 | 353.9 \pm 130.1 | 1.3 \pm 0.4 | 168/C | 155/D | 4.9 \pm 0.6 | 8.5 \pm 0.7 | 6.8 \pm 1.8 | 2.2 \pm 0.1 | 2.6 \pm 0.2 | 0.5 \pm 0.2 |
| 53 | BGHZ-1454 | 185.2 \pm 44.5 | 1.0 \pm 0.1 | 163/D | 155/C | 4.1 \pm 1.5 | 8.2 \pm 0.7 | 3.3 \pm 1.3 | 2.0 \pm 0.6 | 2.3 \pm 0.8 | 0.3 \pm 0.2 |
| 54 | BGHZ-1728 | 241.9 \pm 85.4 | 1.0 \pm 0.1 | 170/B | 155/D | 7.8 \pm 3.5 | 9.1 \pm 1.5 | 13.2 \pm 4.6 | 2.8 \pm 0.2 | 2.6 \pm 0.2 | 0.3 \pm 0.1 |
| 55 | BGHZ-1733 | 397.1 \pm 108.4 | 0.8 \pm 0.1 | 170/B | 155/D | 6.5 \pm 1.2 | 7.5 \pm 1.3 | 7.2 \pm 1.7 | 2.0 \pm 0.3 | 2.5 \pm 0.4 | 0.4 \pm 0.1 |
| 56 | BGHZ-1870 | 355.5 \pm 78.3 | 0.8 \pm 0.1 | 168/C | 155/D | 7.1 \pm 1.4 | 9.9 \pm 2.0 | 11.4 \pm 4.1 | 1.8 \pm 0.2 | 1.9 \pm 0.2 | 0.8 \pm 0.3 |
| 57 | BGHZ-1877 | 19.1 \pm 7.5 | 0.9 \pm 0.1 | 155/A | 155/D | 3.6 \pm 0.5 | 10.2 \pm 0.9 | 9.0 \pm 2.3 | 2.1 \pm 0.3 | 2.4 \pm 0.5 | 1.9 \pm 0.4 |
| 58 | BGHZ-2085 | 352.6 \pm 131.7 | 0.6 \pm 0.1 | 155/D | 155/D | 3.7 \pm 1.1 | 8.5 \pm 1.8 | 5.3 \pm 1.5 | 2.3 \pm 0.3 | 2.2 \pm 0.2 | 0.3 \pm 0.0 |
| 59 | BGHZ-2087 | 209.2 \pm 48.7 | 0.8 \pm 0.1 | 168/D | 155/D | 5.8 \pm 0.9 | 10.3 \pm 1.6 | 9.9 \pm 3.6 | 1.7 \pm 0.7 | 1.8 \pm 0.2 | 1.6 \pm 0.1 |
| 60 | BGHZ-2089 | 210.4 \pm 40.4 | 0.7 \pm 0.0 | 170/C | 155/D | 4.9 \pm 1.6 | 10.4 \pm 0.8 | 7.7 \pm 1.8 | 1.2 \pm 0.4 | 1.7 \pm 0.1 | 1.4 \pm 0.1 |
| 61 | BGHZ-2090 | 317.0 \pm 80.6 | 0.7 \pm 0.1 | 174/B | 155/D | 5.4 \pm 0.6 | 7.9 \pm 1.1 | 10.9 \pm 3.3 | 2.0 \pm 0.0 | 1.6 \pm 0.1 | 0.4 \pm 0.2 |
| 62 | BGHZ-2399 | 144.4 \pm 24.0 | 0.7 \pm 0.1 | 164/C | 155/A | 4.8 \pm 2.1 | 11.6 \pm 1.1 | 8.9 \pm 4.8 | 0.9 \pm 0.1 | 1.6 \pm 0.2 | 1.8 \pm 0.1 |
| 63 | BGHZ-2405 | 234.2 \pm 56.0 | 0.6 \pm 0.1 | 155/D | 155/D | 4.7 \pm 0.9 | 8.9 \pm 1.1 | 8.3 \pm 1.6 | 2.1 \pm 0.8 | 1.9 \pm 0.7 | 0.4 \pm 0.3 |
| 64 | BGHZ-2707 | 178.5 \pm 75.3 | 2.0 \pm 0.4 | 182/D | 186/B | 4.8 \pm 0.9 | 7.0 \pm 1.1 | 6.0 \pm 2.1 | 2.0 \pm 0.1 | 2.5 \pm 0.1 | 0.3 \pm 0.1 |
| 65 | BGHZ-2708 | 136.8 \pm 38.0 | 2.5 \pm 0.4 | 155/D | 155/D | 6.5 \pm 1.5 | 9.0 \pm 0.7 | 9.1 \pm 2.6 | 1.2 \pm 0.3 | 1.4 \pm 0.3 | 1.1 \pm 0.6 |
| 66 | BGHZ-3096 | 83.8 \pm 18.6 | 0.7 \pm 0.1 | 174/C | 183/C | 5.3 \pm 2.1 | 11.6 \pm 1.0 | 11.9 \pm 2.8 | 0.8 \pm 0.1 | 1.1 \pm 0.3 | 3.0 \pm 0.6 |
| 67 | BGHZ-3098 | 206.2 \pm 64.2 | 0.7 \pm 0.0 | 168/D | 155/D | 5.0 \pm 0.8 | 8.7 \pm 0.6 | 7.0 \pm 3.5 | 1.6 \pm 0.9 | 1.9 \pm 0.6 | 1.8 \pm 1.6 |
| 68 | BGHZ-3102 | 200.6 \pm 17.3 | 1.9 \pm 0.4 | 173/C | 155/D | 6.1 \pm 0.8 | 10.3 \pm 1.1 | 7.1 \pm 1.9 | 1.3 \pm 0.2 | 1.5 \pm 0.0 | 0.8 \pm 0.1 |
| 69 | BGHZ-3103 | 321.3 \pm 102.9 | 0.5 \pm 0.0 | 158/B | 155/D | 3.5 \pm 0.4 | 5.9 \pm 0.3 | 3.9 \pm 1.2 | 1.7 \pm 0.8 | 1.7 \pm 0.8 | 0.2 \pm 0.1 |
| 70 | BGHZ-3104 | 229.1 \pm 58.9 | 0.6 \pm 0.2 | 155/D | 155/D | 3.7 \pm 0.9 | 8.1 \pm 1.1 | 6.7 \pm 2.4 | 1.6 \pm 0.5 | 1.7 \pm 0.7 | 0.2 \pm 0.0 |
| 71 | BGHZ-3325 | 237.2 \pm 64.5 | 0.7 \pm 0.1 | 173/C | 155/D | 4.2 \pm 1.2 | 8.9 \pm 1.5 | 7.3 \pm 2.1 | 2.4 \pm 0.2 | 2.3 \pm 0.2 | 0.6 \pm 0.2 |
| 72 | BGHZ-3326 | 116.8 \pm 42.2 | 0.8 \pm 0.1 | 186/C | 155/D | 5.2 \pm 0.6 | 9.8 \pm 1.3 | 7.0 \pm 1.9 | 2.5 \pm 0.7 | 3.3 \pm 0.3 | 1.0 \pm 0.6 |
| 73 | BGHZ-3328 | 301.4 \pm 40.6 | 0.6 \pm 0.1 | 183/B | 183/C | 4.3 \pm 0.7 | 6.8 \pm 0.5 | 3.6 \pm 1.6 | 2.0 \pm 0.2 | 2.3 \pm 0.2 | 0.5 \pm 0.2 |
| 74 | BGHZ-3329 | 202.7 \pm 56.8 | 0.8 \pm 0.1 | 168/B | 155/D | 4.0 \pm 0.8 | 8.5 \pm 0.9 | 7.1 \pm 2.4 | 2.1 \pm 0.3 | 2.5 \pm 0.2 | 1.0 \pm 0.3 |
| 75 | BGHZ-3331 | 201.5 \pm 58.7 | 0.6 \pm 0.0 | 155/D | 155/D | 4.7 \pm 0.7 | 8.7 \pm 1.0 | 6.3 \pm 1.5 | 2.4 \pm 0.2 | 2.0 \pm 0.1 | 0.4 \pm 0.3 |
| 76 | BGHZ-3332 | 239.9 \pm 85.5 | 0.7 \pm 0.1 | 168/D | 155/D | 5.9 \pm 1.9 | 8.5 \pm 1.3 | 13.1 \pm 4.3 | 1.7 \pm 0.4 | 1.6 \pm 0.3 | 0.8 \pm 0.7 |
| 77 | BGHZ-3333 | 289.4 \pm 83.6 | 0.6 \pm 0.1 | 167/C | 155/D | 5.4 \pm 1.7 | 8.0 \pm 1.3 | 11.0 \pm 3.1 | 1.6 \pm 0.5 | 1.5 \pm 0.4 | 0.4 \pm 0.2 |
| 78 | BGHZ-3536 | 262.3 \pm 45.2 | 0.7 \pm 0.1 | 167/B | 170/C | 4.4 \pm 1.5 | 8.9 \pm 0.9 | 7.2 \pm 1.6 | 1.1 \pm 0.3 | 1.2 \pm 0.4 | 0.7 \pm 0.1 |
| 79 | BGHZ-3743 | 193.6 \pm 49.0 | 0.7 \pm 0.1 | 173/B | 155/D | 4.7 \pm 0.6 | 8.4 \pm 0.9 | 8.7 \pm 4.3 | 1.5 \pm 0.1 | 1.7 \pm 0.2 | 0.6 \pm 0.3 |
| 80 | BGHZ-3744 | 128.0 \pm 38.1 | 0.7 \pm 0.1 | 185/B | 155/D | 4.3 \pm 1.3 | 8.8 \pm 1.1 | 5.8 \pm 2.8 | 1.3 \pm 0.2 | 1.5 \pm 0.2 | 0.7 \pm 0.5 |
| 81 | BGHZ-3745 | 588.3 \pm 95.9 | 0.8 \pm 0.1 | 168/B | 155/C | 4.0 \pm 0.6 | 6.3 \pm 0.8 | 2.7 \pm 1.8 | 1.3 \pm 0.2 | 1.4 \pm 0.3 | 0.2 \pm 0.1 |
| 82 | BGHZ-4085 | 127.7 \pm 50.4 | 0.7 \pm 0.0 | 185/B | 186/A | 4.7 \pm 1.8 | 10.7 \pm 0.8 | 8.7 \pm 3.4 | 1.5 \pm 0.1 | 1.8 \pm 0.1 | 1.4 \pm 0.2 |
| 83 | BGHZ-4086 | 148.4 \pm 42.1 | 0.6 \pm 0.1 | 155/D | 155/D | 4.7 \pm 0.8 | 10.2 \pm 0.6 | 5.4 \pm 1.0 | 3.0 \pm 0.4 | 3.0 \pm 0.7 | 0.2 \pm 0.1 |
| 84 | BGHZ-4177 | 405.2 \pm 56.7 | 0.8 \pm 0.1 | 182/A | 183/C | 6.5 \pm 2.4 | 10.0 \pm 1.4 | 12.8 \pm 4.9 | 2.4 \pm 0.1 | 2.4 \pm 0.1 | 0.5 \pm 0.1 |
| 85 | BGHZ-4179 | 337.6 \pm 109.5 | 0.7 \pm 0.1 | 168/C | 155/D | 4.9 \pm 1.3 | 7.7 \pm 0.9 | 9.8 \pm 3.2 | 1.3 \pm 0.6 | 1.3 \pm 0.6 | 0.3 \pm 0.1 |
| 86 | BGHZ-4222 | 443.5 \pm 163.3 | 0.9 \pm 0.1 | 167/C | 155/C | 5.7 \pm 1.9 | 6.6 \pm 0.8 | 2.8 \pm 1.2 | 1.9 \pm 0.2 | 2.1 \pm 0.3 | 0.4 \pm 0.2 |
| | Average | 245.0 \pm 124.8 | 0.8 \pm 0.4 | | | 5.2 \pm 1.8 | 8.8 \pm 1.8 | 8.0 \pm 4.0 | 1.7 \pm 0.6 | 2.0 \pm 0.6 | 0.7 \pm 0.6 |
| | CV ¹ (%) | 50.9 | 50.0 | | | 34.6 | 20.4 | 50.0 | 35.3 | 30.0 | 85.7 |

¹ Coefficient of variation.

Bulb weight, shape and colour

Accessions BGHZ-1877 and BGHZ-0225 presented the smallest bulbs, with mean values of 19.1 ± 7.5 g and 59.0 ± 34.9 g, respectively. Both accessions were collected in the Canary Islands (Spain) hence they are closely adapted to the environmental conditions of these islands, very different from those prevailing in our experimental fields. On the other hand, accessions BGHZ-4222 and BGHZ-3745 presented the biggest bulbs, with means values of 443.5 ± 163.3 and 588.3 ± 95.9 respectively. Both accessions are from the same region where the assay has been carried out, corroborating that they are well-adapted to this area, where traditionally big bulbs are preferred. The range of the bulb shape index (BSI) was 0.5–2.5. Most of the accessions (87.2%) presented flat, flat globe, rhomboid or broad oval bulbs, with a BSI value lower than 1, while the 3.5% and 9.3% corresponded to globe bulbs (BSI = 1) and elongated bulbs (BSI > 1), respectively. The lower value for the BSI of 0.5 ± 0.0 corresponded to BGHZ-3103. This accession is characterized by its flat shape, hence its local name ‘Chata’, Spanish for flat. The value of 2.5 ± 0.4 corresponded to BHGZ-2708, a landrace with a characteristic spindle shape. There was a great variability in bulb skin colour, appearing white, yellow, brown, and violet bulbs with different shades, as shown in Table 1. All options for onion bulb skin colour that appear in the descriptors for *Allium* (IPGRI *et al.*, 2001) were recorded, except the green one. Most of the bulbs were brown (53.5%), violet (25.6%) and white (16.3%). Regarding flesh colour, most of the accessions presented white (75.6%) and violet and white (22.1%) flesh.

Bulb firmness

Firmness in vegetables is an important textural attribute affecting consumer attitudes toward freshness and quality, which has been also studied in onions (Coolong *et al.*, 2008). Bulb firmness is correlated to mechanical harvest and storage ability of onion cultivars (Larsen *et al.*, 2009). Studies have shown that high dry matter onions had the firmest bulbs at harvest and delayed softening during storage (Coolong *et al.*, 2008). In the collection studied here, bulb firmness ranged from 3 to 9 kg cm^{-2} . Accession BGHZ-0323, known as ‘Liria’, showed the lowest firmness value. This accession comes from Valencia province, where onion is a traditional

crop (Castell and Portas, 1994), and has been reported as the sweetest one among Valencia cultivars (Casallo *et al.*, 1991). The low values for firmness together with the low pungency found ($4.3 \mu\text{mol g}^{-1}$ FW) corroborated the mild flavour and tenderness of the bulbs previously reported. BGHZ-1132, known as ‘Lágrima de Oro’, was the firmest accession; enhance its good storage ability.

Soluble solids content (SSC)

Dry matter content (DMC), consisting mostly of fiber, starch, and sugars (Lin *et al.*, 1995), is an important quality factor commonly estimated indirectly as SSC by refractometry. DMC determine, in part, the end use, storage life, pungency and firmness (Sinclair *et al.*, 1995). DMC exhibits wide genetic variation, from around 6% in sweet onions up to over 25% in dehydration varieties (Jones and Mann, 1963). Our results showed that SSC ranged from 5.8 to 12.8 °Brix. The Brix degree values for fresh onions reported in the literature also varies over a wide range: 4.9–8.6 (Rodríguez-Galdón *et al.*, 2009), 6.8–12.3 (Vågen and Sliemstad, 2008), 11.3–13.3 (Dhumal *et al.*, 2007), 7.2–15.8 (Jaime *et al.*, 2001). The mean CV of SSC (20.4%) was the lower of the studied traits, and was similar to the previously reported by Rodríguez-Galdón *et al.* (2009).

Pungency

Estimation of pungency in bulbs has become necessary as the popularity of low pungency onion has increased (Dhumal *et al.*, 2007). Onion pungency is difficult to measure by taste tests because of the accumulative effect of successive tasting (Platenius and Knott, 1941). It can be more accurately determined by analysis of biochemical components or derivatives (Lin *et al.*, 1995). The determination of pyruvate as an indicator of pungency is perhaps the most established method for pungency assessment in onion. Pyruvate values have been highly and significantly correlated to sensory ratings, indicating that pyruvate analysis can be used as a reliable selection technique for pungency in onion breeding programs (Wall and Corgan, 1992; Pineda *et al.*, 2004). The mean CV of pungency found in this study was higher than previously reported in other open-pollinates lines (50.0% CV vs. 21.3% CV) (Yoo *et al.*, 2006). These results show the high degree of variability in pungency

among the Spanish accessions. It has been shown that more than 80% of onion pungency is determined by genetic factors (Yoo *et al.*, 2006). Thus, choosing milder cultivars with a low CV is essential for producing milder onions with high pungency uniformity. Accessions BGHZ-4086 and BGHZ-0227 with low mean pungency levels ($5.4 \mu\text{mol g}^{-1}$ FW and $3.0 \mu\text{mol g}^{-1}$ FW respectively) and low CV, compared to the other studied accessions, and similar to previously reported for open-pollinated cultivars (Yoo *et al.*, 2006), may be valuable for this purpose. Other accessions showing low mean values for pungency may be used to obtain mild onions after breeding the plant material. Actually, our research team is carrying out a breeding program with this aim. After one cycle of selection, we have obtained progenies with significantly lower and more uniform levels of pungency (Mallor *et al.*, 2011). On the other hand, BGHZ-1354 was the most pungent one ($18.1 \mu\text{mol g}^{-1}$ FW). Highly pungent onions are used for cooking and for manufacturing sauces, canned soups, extracts, and dehydrated products (Galmarini *et al.*, 2001). In contrast, less pungent onion varieties have become preferred for fresh consumption, especially in the USA, Japan and many countries of Europe. Low pungency or mild onions have a pyruvate concentration of $<5 \mu\text{mol g}^{-1}$ FW and command a price premium (Abayomi and Terry, 2009). According to this criterion, 15.1% of the collection studied here is considered as mild onions. Other guideline, used by the sweet onion industry, classifies onions on the basis of pungency as low pungency/sweet ($0-3 \mu\text{mol g}^{-1}$ FW), medium pungency ($3-7 \mu\text{mol g}^{-1}$ FW), and high pungency (above $7 \mu\text{mol g}^{-1}$ FW) (Dhumal *et al.*, 2007). Following this set of values 3.5%, 33.7% and 62.8% of the tested collection are classified as low, medium and high pungency onions respectively.

Sugars

Sweetness in onions is a balance between single sugars and pungency. Pungency and sugars were proposed to be independent traits in onion (Randle, 1997). Therefore, the opportunity to breed high sugar-low pungent cultivars should be possible. In the present study, fructose, glucose and sucrose were detected in all onion samples. Large differences in the relative glucose, fructose and sucrose contents among the studied onion landraces have been found. The contents vary in the ranges 0.8-3.0 g fructose, 1.1-3.3 g glucose and 0.1-

3.0 g sucrose per 100 g FW. Other authors have reported similar values for fructose and higher values for glucose and sucrose (Vågen and Slimestad, 2008) while Rodríguez-Galdón *et al.* (2009) reported lower mean values for all the sugars. Differences may be explained by the different onion varieties studied. In addition, environmental conditions such as climatic conditions or soil type influence the genetic information of the onion cultivars, which produce variations in their chemical composition (Rodríguez-Galdón *et al.*, 2009). McCallum *et al.* (2006) reported that fructose conditions the main differences in bulb carbohydrate phenotype between sweet and storage onion types, since fructose quantitatively dominates sweetness in onion. Accession BGHZ-4086 presented the higher value for fructose content and also low pungency, as described before; both are interesting qualities for sweet onions. Recent works have suggested that the level of glucose concentration is positively correlated with taste preference (Abayomi and Terry, 2009). The red onion accession BGHZ-3326 presented the higher value for glucose and total sugars ($6.73 \text{ g}/100 \text{ g FW}$) but also a high pungency value ($7.0 \mu\text{mol g}^{-1}$ FW).

Relation among the evaluated traits

The significant ($p < 0.05$) correlations between the studied variables are shown in Table 2. Moderate positive correlation between SSC and pungency was found ($r = 0.34$). Soluble solids content was strongly correlated with onion dry weight: $r = 0.95$ (Vågen and Slimestad, 2008) and $r = 0.98$ (Jaime *et al.*, 2001); while, matching up with our results, the correlation between dry weight content and pyruvate content was found to be quite low ($r = 0.38$) (Vågen and Slimestad, 2008). Other authors have also found a medium correlation (r from 0.42 to 0.57) (Schwimmer and Guadagni, 1962; Lin *et al.*, 1995; Galmarini *et al.*, 2001). In contrast, Yoo *et al.* (2006) indicated that there was no consistent trend between these traits in four different clones grown at Weslaco. The relation found in this study may be explained because the compounds responsible for onion pungency also contribute to total dissolved solids. Therefore, in agreement with Lin *et al.* (1995), part of the positive correlation between SSC and pungency is due to the partial identity of the two traits.

In this study, pungency appeared also positively correlated with firmness and sucrose contents and negatively correlated with bulb weight. According to Yoo *et al.*

Table 2. Correlation coefficients for different bulb traits in onion

| | Weight | Shape | Firmness | SSC ¹ | Pungency | Fructose | Glucose |
|------------------|----------|---------|----------|------------------|----------|----------|---------|
| Weight | | | | | | | |
| Shape | | | | | | | |
| Firmness | 0.120** | 0.107** | | | | | |
| SSC ¹ | -0.351** | | 0.190** | | | | |
| Pungency | -0.187** | | 0.318** | 0.338** | | | |
| Fructose | | -0.122* | | | | | |
| Glucose | | | | | | 0.789** | |
| Sucrose | -0.428** | | | 0.574** | 0.156* | -0.220** | |

¹ SSC: soluble solids content. *r*: Pearson coefficients. *** significant at $p < 0.05$ and 0.01 , respectively.

(2006), a possible explanation for a decline in pungency levels in heavier bulbs could be a dilution effect. In that way, the concentration of total solutes in the bulbs is more diluted in the larger bulbs. In contrast, pungency has been strongly correlated with sucrose ($r = -0.924$) and fructose contents ($r = 0.910$) (Rodrigues *et al.*, 2003).

In agreement with McCollum (1968), we found a negative correlation between SSC and bulb size. As described for pungency, the increase in water content, associated with low-solid content onions, could be responsible for diluting solids (Galmarini *et al.*, 2001). These results contrast with those from Rodríguez-Galdón *et al.* (2009), who reported a positive correlation between SSC and bulb size ($r = 0.276$).

Although bulb weight was found to be an important factor correlated with bulb firmness (Larsen *et al.*, 2009), the correlation between weight and firmness was found to be quite low. In the same way, other authors have been reported that onions with high dry matter content tend to be much firmer (Rutherford and Whittle, 1982; Chope *et al.*, 2006), while our results showed a weak positive correlation.

In the present study, positive correlation between fructose and glucose contents and negative correlation between fructose and sucrose contents were found. The contents of different sugars in 15 onion cultivars revealed moderate positive correlations between fructose and glucose ($r = 0.46$) and between glucose and sucrose contents ($r = 0.46$) and a negative correlation was found between fructose and sucrose contents ($r = -0.41$) (Vågen and Slimestad 2008). Rodríguez-Galdón *et al.* (2009) found moderate positive correlations between sucrose with fructose and glucose ($r = 0.511$ and $r = 0.509$, respectively) and, according with our results, high positive correlation between fructose and glucose ($r = 0.798$). The high correlation between fructose and glucose has been observed for

other fruits such as bananas (Foster *et al.*, 2002) and tomatoes (Hernández *et al.*, 2008), and suggest a common origin for both sugars, probably from the sucrose (Rodríguez-Galdón *et al.*, 2009). Jaime *et al.* (2001) and Rodrigues *et al.* (2003) also found significant negative correlations between sucrose and fructose ($r = -0.94$). SSC has been previously correlated with glucose ($r = -0.857$) (Jaime *et al.*, 2001), while our results did not show any correlation.

Cluster analysis

The cluster analysis made possible to identify four main subgroups or clusters within the studied population (Fig. 1).

Cluster 1 is represented by firm, pungent and large size bulbs (Table 3). As bulb firmness and high pungency

Table 3. Mean values for each cluster according with squared Pearson distance. Results in the same row with the same letter were not statistically significant ($p < 0.05$) according to the classification obtained by the Duncan test

| | Cluster | | | |
|---|--------------------|--------------------|--------------------|---------------------|
| | 1 | 2 | 3 | 4 |
| N ¹ | 21 | 34 | 28 | 3 |
| Weight (g) | 283.3 ^a | 280.8 ^a | 141.2 ^b | 207.5 ^{ab} |
| Shape (height Ø ⁻¹) | 0.8 ^b | 0.8 ^b | 0.8 ^b | 2.2 ^a |
| Firmness (kg cm ⁻²) | 6.2 ^a | 4.4 ^b | 4.9 ^b | 5.9 ^a |
| SSC (°Brix) | 8.0 ^b | 8.1 ^b | 10.3 ^a | 8.6 ^b |
| Pungency (µmol pyruvic acid g ⁻¹) | 10.3 ^a | 5.5 ^c | 8.2 ^{ab} | 7.1 ^{bc} |
| Sugars (g/100 g) | | | | |
| Fructose | 1.8 ^{ab} | 2.0 ^a | 1.5 ^{bc} | 1.2 ^c |
| Glucose | 1.9 ^a | 2.2 ^a | 1.8 ^{ab} | 1.4 ^b |
| Sucrose | 0.4 ^b | 0.4 ^b | 1.2 ^a | 1.1 ^a |

¹ Number of accessions per cluster.

levels have been related to storage ability of onion cultivars (Coolong *et al.*, 2008; Larsen *et al.*, 2009), this onion's group has good quality traits for this goal.

Cluster 2 is formed by large size, mild (low pungency) and sweet (high fructose and glucose) onions (Table 3). The onion flavour is a balance between single

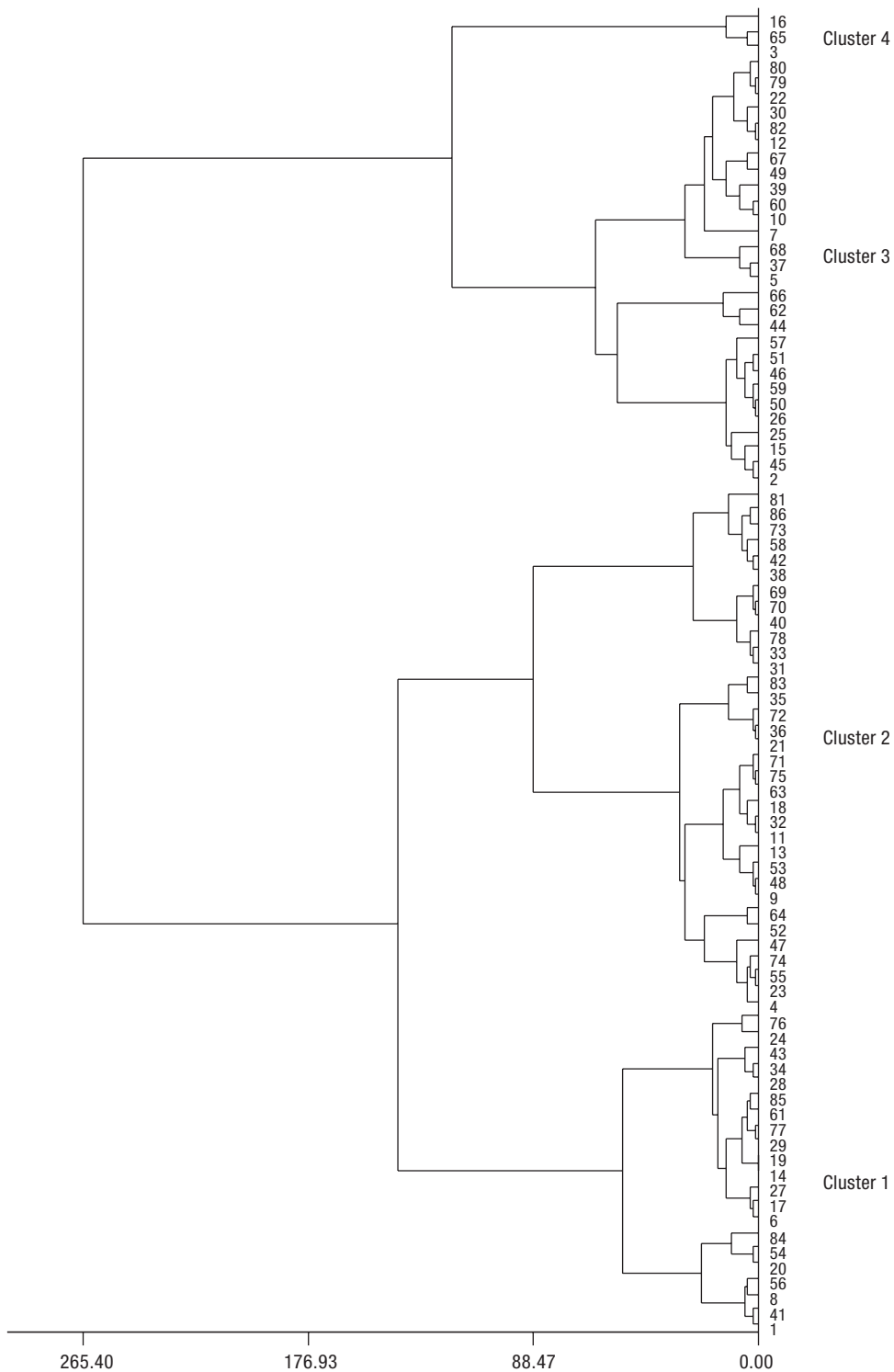


Figure 1. Cluster analysis of 86 Spanish onion accessions (see Table 1) by squared Pearson distance using Ward Linkage.

sugars and pungency. Fructose quantitatively dominates sweetness in onion (McCallum *et al.*, 2006) and higher concentrations of glucose have been positively correlated with taste preference (Abayomi and Terry, 2009). On the other hand, pyruvate values have been highly and significantly correlated with sensory ratings (Wall and Corgan, 1992; Pineda *et al.*, 2004). In that way, sweet-mild onions are characterised by high sugars content and low pungency, like onions in cluster 2.

Cluster 3 mainly includes pungent and small size bulbs with high SSC (Table 3). Pungency and SSC are important attributes of onion bulb quality for processing and storage (Dhumal *et al.*, 2007). Onions with high dry matter, and therefore high soluble solids, tend to store for longer periods before shoot growth and disease incidence deplete the number of marketable bulbs (Chope *et al.*, 2006).

Cluster 4 is mostly characterized by the elongated shape of the bulbs (Table 3). It is only made up by three accessions that presented a characteristic spindle shape.

Discriminant analysis

Linear discriminant analysis was performed to assess the capability of predicting the cluster for a new observation (a new onion accession) with known values in the 8 variables. The coefficients of the four linear discriminant functions are shown in Table 4. The classification of the 86 types provides only 2 errors, while when the classification is made with cross-validation 82 accessions are assigned to the correct cluster. Hence the fit between predicted and experimental values is estimated in 95.3%.

The discriminant functions are also used to classify individual onions for which all variables' values are

Table 4. Linear discriminant functions for onion groups

| Variables | Coefficients of the functions | | | |
|----------------|-------------------------------|--------|--------|---------|
| | 1 | 2 | 3 | 4 |
| Weight | 0.04 | 0.04 | 0.01 | 0.01 |
| Shape | 22.43 | 22.12 | 26.97 | 62.60 |
| Firmness | 4.29 | 1.74 | 2.63 | 3.42 |
| Soluble solids | 4.86 | 5.98 | 7.98 | 7.59 |
| Pungency | 1.41 | 0.47 | 0.37 | 0.51 |
| Fructose | 5.75 | 5.23 | 3.17 | 4.65 |
| Glucose | 2.01 | 4.01 | 0.36 | -4.13 |
| Sucrose | -2.36 | -2.50 | 0.01 | 3.21 |
| Constant | -61.13 | -53.12 | -63.75 | -117.50 |

known. The distribution of the belonging probabilities to each one of the groups also validates the use of the discriminant functions as classifying tool of new onions in one of the 4 clusters. Then this classification procedure can be also useful to study the cluster assignment of the same onion accession in different years or growing conditions, or to classify new onion accessions to the established clusters.

Conclusions

The variability in important agronomical traits found point out that these onion accessions could be candidates for future breeding programs. In addition, the clustering could make the initial plant material selection easier and the correlations among the evaluated traits found could help to establish adequate selection strategies.

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