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Diversity of pomegranate (*Punica granatum* L.) germplasm in Spain

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Abstract. Twenty-nine pomegranate (*Punica granatum* L.) accessions were studied to determine the overall degree of polymorphism. Cluster analysis showed a considerable phenotypic and genetic diversity in the local pomegranate germplasm. The cluster analysis produced a dendrogram with four main clusters, showing accessions closely related and simultaneously very different from others. The geographic origin was a determinant criterion for cultivars clustering.

Keywords. Diversity – Pomegranate – *Punica* – Spain – Accession – Characterization.

I – Introduction

There is growing interest in pomegranate not only because it is pleasant to eat, but also because it is considered to be a functional product of great benefit in the human diet, as it contains several groups of substances that are useful in disease prevention (Melgarejo and Martínez, 1992; Melgarejo and Artés, 2000; Melgarejo and Salazar, 2002, Cam *et al.*, 2009). Prospections for local pomegranate germplasm were undertaken since 1992. Many local types were inventoried and described (Melgarejo, 1992). The collection original containing 59 accessions collected from different growing regions in the country were established. They represent about 16 local denominations (Melgarejo, 1992).

This paper describes the diversity observed in local germplasm when evaluated under uniform conditions. The main objectives of this work were to determine the overall degree of polymorphism of the characters used in morphometric studies.

II – Material and methods

1. Plant material

Areas prospected and germplasm collecting procedures adopted were reported in Melgarejo (1992). Twenty-nine accessions, representing 9 denominations, were included in the present study (Table 1). They are represented by adult trees maintained in the same collection at Alicante in Southeast of Spain (Melgarejo, 1993).

2. Characters studied

Studies were based on characteristics of the fruit and of the seeds, as well as of the leaves and flowers. Morphometric measurements and chemical analyses carried out on samples of 20 mature fruits, in 25 seeds, 50 leaves y 25 flowers of every variety. The study was realized for three consecutive years, measuring up the following variables.

Table 1. Names, abbreviations and origin of pomegranate accessions evaluated

Code	Accession	Location†	Code	Accession	Location†
AB1	Albar de Blanca 1	Blanca (Murcia)	ME17	Mollar de Elche 17	Elche (Alicante)
ADO2	Agridulce de Ojós 2	Ojos (Murcia)	ME18	Mollar de Elche 18	Elche (Alicante)
ADO3	Agridulce de Ojós 3	Ojos (Murcia)	ME19	Mollar de Elche 19	Elche (Alicante)
BA1	Borde de Albatera 1	Albatera (Alicante)	ME20	Mollar de Elche 20	Elche (Alicante)
BO1	Borde de Ojós 1	Ojos (Murcia)	ME21	Mollar de Elche 21	Elche (Alicante)
CRO1	Casta del Reino 1	Ojos (Murcia)	MO3	Mollar de Orihuela 3	Orihuela (Alicante)
CRO2	Casta del Reino 2	Ojos (Murcia)	MO4	Mollar de Orihuela 4	Orihuela (Alicante)
MA1	Mollar de Albatera 1	Albatera (Alicante)	MO5	Mollar de Orihuela 5	Orihuela (Alicante)
MA2	Mollar de Albatera 2	Albatera (Alicante)	PTO3	Piñón tierno de Ojós 3	Ojos (Murcia)
MA3	Mollar de Albatera 3	Albatera (Alicante)	PTO4	Piñón tierno de Ojós 4	Ojos (Murcia)
ME11	Mollar de Elche 11	Elche (Alicante)	PTO5	Piñón tierno de Ojós 5	Ojos (Murcia)
ME12	Mollar de Elche 12	Elche (Alicante)	PTO6	Piñón tierno de Ojós 6	Ojos (Murcia)
ME13	Mollar de Elche 13	Elche (Alicante)	PTO7	Piñón tierno de Ojós 7	Ojos (Murcia)
ME14	Mollar de Elche 14	Elche (Alicante)	PTO8	Piñón tierno de Ojós 8	Ojos (Murcia)
ME16	Mollar de Elche 16	Elche (Alicante)			

†Melgarejo (1992).

A. Fruit

Fruit weight (FW), expressed in g; Equatorial diameter (FD1), expressed in mm; Calyx diameter (FD2), expressed in mm; Fruit height without calyx (FL1), expressed in mm; Total fruit height (FL2), expressed in mm; Calyx height (FL3), expressed in mm; Number of carpels (Nc); Rind and carpels weight (PcMc), expressed in g; Skin thickness (Ec), expressed in mm, the measurements were performed on two opposite faces in the equatorial zone; Seeds yield (Rs) = [FW-(PcMc)/FW]x100 (%).

Diameters, fruit height and skin thickness. Fruit weights and Rind and carpels weight were taken (Sartorius Model BL-600) with an accuracy of 0.1 g.

B. Seed

Maximum width (Sw) and length (SL); Seed weight (SW), determined by a precision weighing device (Mettler AJ50) with an accuracy of 0.0001 g; Juice content (JV), using an electric extractor and a seed sample of 100 g; Total soluble solids (TSS) (ºBrix), determined by an Atago N-20 refractometer at 20ºC; Acidity expressed as citric acid (A), determined by acid–base potentiometer and pH; Maturity index (MI = TSS/A). Three repetitions per clone and year were carried out.

The parameters measured in the woody portion of the seeds were: Maximum width (w) and length (l); Weight of the woody portion (wpw) of each seed; Woody portion index (wpi), determined from the wpw/SW ratio 100 (%).

C. Leaf

The surface of the leaves was assessed by an image digital analyzer device (Digital Image Analysis System, Delta-T). The measured variables were: LW: width of the leaf (mm); Ll: length of the blade (mm); Lt: total length of the leaf (mm); Lp: length of the petiole (mm); LS: leaf surface (mm^2).

D. Flower

The measured variables were: FD: diameter of the flower (mm); FL: length of the flower (mm);

NP: number of petals; NS: number of sepals; LP: length of the petals (mm); AP: width of the petals (mm); LE: length of the style (mm); NE: number of stamens.

The lengths were measured by a digital caliper/caliper (Mitutoyo) with a 0.01 mm accuracy. The fruits weight, determined by a precision weighing device (Sartorius BL 600) with an accuracy of 0.01 g.

3. Statistical analysis

Mean values registered for each parameter were used to perform a clustering of cultivars into similarity groups using the Ward´s, Method Squared Euclidean. Data processing was performed using the PASW Statistics 18 (SPSS Inc., Chicago, USA).

III – Results and discussion

The cluster analysis produced a dendrogram with four main clusters (Fig. 1).

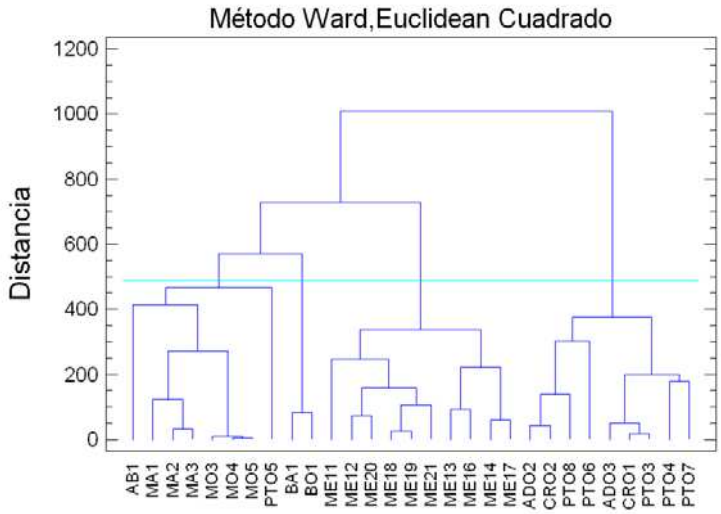

Fig. 1. Cluster analysis grouping of 29 Spanish pomegranate cultivars. See Table 1 for cultivars names abbreviations.

The first cluster (I) is the most heterogeneous group and is composed by 8 varieties proceeding from several localities by fruits of average-big size and sweet juice. The second cluster (II) grouped cultivars BA1 and BO1 characterized by to present fruits of average-big size, high index of woody portion and acid juice. The third cluster (III) includes the varieties of the group varietal ME (10 accessiones). All they have fruits of average size, with low acidity of the juice and in general high indexes of maturity (Fig. 1).

The last group (IV) is composed by 9 varieties, all of them proceeding from the same geographical zone. The varieties of the cluster IV are characterized for presenting fruits and seeds of big size. Also it is interesting to indicate that in the Spanish analyzed varieties, four groups were obtained from an analysis cluster (Fig. 1), remarkably coinciding all grouped varieties with their geographical origins but the PTO5 (Fig. 1). While these results agreed with those reported in for other fruit tree species (Barbagollo *et al.*, 1997), they clearly differed from the classification established by Mars and Marrakchi (1999).

IV – Conclusions

The study revealed considerable phenotypic (and presumably genetic) diversity among pomegranate accessions, showing more proximity between varieties of the same geographical area.

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