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ASSESSING PHENOTYPIC DIVERSITY OF CUCURBITA PORTUGUESE GERMPLASM

SUMMARY

Cucurbita is considered one of the most variable genera in the entire plant kingdom with regard to fruit characteristics (colour, size and shape). The most important species are: *C. maxima*, *C. pepo*, *C. argyrosperma*, *C. ficifolia* and *C. moschata*. In Portugal, the production of *Cucurbita* spp. is based in local populations and is mainly for self-consumption (human food or animal feed) and sale on local markets. The aim of this work was to characterize 108 populations of *Cucurbita* being 55 of *C. pepo*, 32 of *C. maxima* and 21 of *C. moschata*. A total of 20 traits (13 qualitative and seven quantitative) were scored according to the Minimum Descriptors for *Cucurbita* spp. developed by the ECPGR Working Group on Cucurbits were used. The *C. pepo* populations revealed the highest mean values for fruit length, skin thickness and seed weight, and the populations of *C. maxima* showed the highest mean values for fruit width and 100 seed weight. The *C. moschata* populations presented the highest mean values for fruit weight and flesh thickness. ANOVA revealed significant differences for all traits, with exception of skin thickness. Principal Component Analysis showed that the three most informative principal components explained 52.5 % of the total variation and a clear separation of the three species. The results reveal a high variability in this collection of *Cucurbita* populations.

Keywords: *Cucurbitaceas*, morphological traits, germplasm, genetic diversity

INTRODUCTION

The genus *Cucurbita*, belongs to the Cucurbitales order, Cucurbitaceae family, Cucurbitoideae subfamily and Cucurbiteae tribe. The centers of origin and domestication for cultivated *Cucurbita* species can be identified as various

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areas in Central and South America (Jeffrey, 1990) and the earliest domestication of *Cucurbita* dates back 8,000–10,000 years ago (Smith, 2001; Sanjur *et al.*, 2002).

Cucurbita species were brought to Europe in the late 15th century by Spanish and are now used in many parts of the world. There are five domesticated species: *C. maxima*, *C. pepo*, *C. argyrosperma*, *C. ficifolia* and *C. moschata*. The genus *Cucurbita* is one of the most diverse in the plant kingdom, and has more cultivated forms than have been reported for any other crop (Esquinas-Alcázar and Gulick, 1983). The three main cultivated species *Cucurbita moschata*, *C. pepo* and *C. maxima* are widely used for human consumption, as fodder for livestock, and/or for ornamentation. In Portugal the production of *Cucurbita* is based in old local cultivars, landraces, and is mainly for self-consumption (human food or animal feed) and is sale on local markets. The landraces are important genetic resources for plant breeders because of their considerable genotypic variations.

These variations are maintained by deliberate selection for specific traits by farmers. At the research level, the diversity of genetic resources in collections may increase the efficiency of efforts to improve a species (Geleta *et al.*, 2005). Phenotypic characterization in *Cucurbita* has traditionally been based on seed and fruit characteristics (Balkaya *et al.*, 2005; Ferriol and Pico, 2008), which have proved useful in distinguishing related species (Gwanama *et al.*, 2000). The objective of descriptors based on morpho-agronomic characters was considered reliable traits to verify or assess genetic distance or conformity among populations (Hunter, 1993).

Several authors are concordant that genetic diversity within landraces and populations of *Cucurbita* is high, including variation in shape, size and colour of fruits; number and size of seeds; quality, colour and thickness of fruit flesh; tolerance to pests and precocity in fruit production, among other traits (Nerson *et al.*, 2000; Ferriol *et al.*, 2003; Paksoy and Aydin, 2004; Hernandez *et al.*, 2005). Reliable information on character variability within germplasm collections is very useful to breeders in planning crop improvement programs. The aim of the present study was to evaluate the diversity in populations of *C. pepo*, *C. maxima* and *C. moschata* from Northern and inner center of Portugal.

MATERIAL AND METHODS

Cucurbita fruits were obtained from 108 Portuguese populations belonging to three species: 55 *C. pepo*, 32 *C. maxima* and 21 *C. moschata*. These populations were collected between 2011 and 2013 in Northern and inner center of Portugal. The fruits were evaluated by 20 morphological traits: 13 qualitative traits (peduncle transectional shape and binding, fruit shape, ribs, skin texture, predominant and secondary skin colour, secondary fruit skin colour pattern, flesh colour and texture, predominant seed coat colour, seed margin coat colour and seed shape) and seven quantitative traits (fruit length, width, weight, flesh and skin thickness, seed weight and 100 seed weight).

The traits were scored according to the Minimum Descriptors for *Cucurbita* spp. developed by the ECPGR Working Group on Cucurbits (2008). Data analyses were performed as analysis of variance, mean separations were made using Duncan Test ($P = 0.05$), designed to allow all possible linear combinations of group means to be tested.

All determinations were performed in triplicate. Correlation coefficients were determined as Spearman's coefficient using the software Statistica 8. Categories registered for each parameter were used to perform the PCA. This statistical procedure was applied to create a correlation matrix from which standardized principal component (PC) scores were extracted. Scatter plots of the first 2 PC scores were created. To determine which of the PC scores accounted for the greatest amount of variation for each trait, the eigenvalues of the three PC scores were compared for each trait. Data processing was performed using the statistical program MVSP 2.33.

RESULTS AND DISCUSSION

Knowledge of the extent of genetic diversity and the identification, differentiation and characterization of genotypes and populations, provide an information tool for the detection of duplicates in collections and a better characterization and utilization in plant breeding programs (Hornokova et al., 2003). The results of the quantitative traits showed high diversity between populations of the three species. The *C. pepo* populations revealed the highest mean values for fruit length, skin thickness and seed weight (40.7cm, 19.0mm and 117.9g, respectively) and the lowest means for flesh thickness (30.1mm). The populations of *C. maxima* showed the highest mean values for fruit width (31.2cm) and the lowest mean values for fruit length and weight and skin thickness (26.7cm, 8.6Kg and 15.0mm, respectively) (Table 1). ANOVA revealed significant differences for all quantitative traits, except for skin thickness trait. The qualitative traits that showed higher variability were: fruit shape, predominant fruit skin colour at maturity, secondary fruit skin colour, secondary fruit skin colour pattern and fruit skin texture.

Regarding fruit shape, in populations of *C. pepo* the oblong/elliptical shape was predominant (64%), followed by globular shape (21%). In most of the *C. maxima*, fruits are characterized by globular shape (55%) and flat shape (17%) and in *C. moschata* the pyriform shape (36%) was the predominant followed by the globular shape (28%). All *C. pepo* populations had fruits flesh colour yellow and in *C. maxima* this was the predominant flesh colour (68%). In *C. moschata*, salmon flesh colour was the most frequent (64%).

These results are in agreement with the ones reported by Ferriol et al (2003), Balkaya et al. (2010), Xiaohua et al. (2011) and Ahamed et al (2011). Principal Component Analysis (PCA) showed that the three most informative principal components explained 52.5 % of the total variation. The first component accounted for 22.41% of the total variation, and is mainly defined, positively, by

skin thickness and seed weight and negatively by seed margin, coat colour and predominant seed coat colour.

Table 1. Morphological traits of the three species of *Cucurbita*

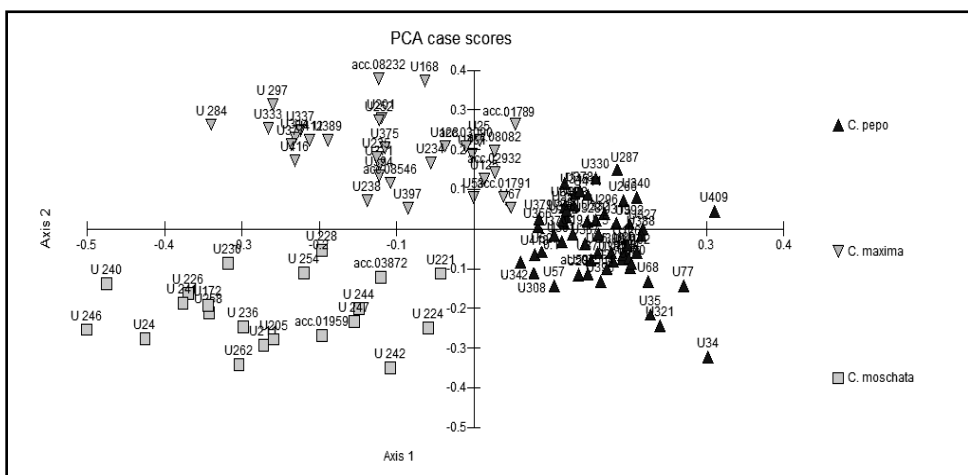
Traits	<i>C. pepo</i>			<i>C. maxima</i>			<i>C. moschata</i>		
	N	Average \pm SD	Min-Max	N	Average \pm SD	Min-Max	N	Average \pm SD	Min-Max
Fruit length (cm)	5	40.7a* \pm	27.7 -	32	26.7b \pm	15.5 -	21	38.7a \pm	18.0 -
	5	1.2	75.4		1.5	44.5		1.9	76.5
Fruit width (cm)	5	29.9ab \pm	24.2 -	32	31.2b \pm	24.3 -	21	28.1a \pm	14.3 -
	5	0.6	41.3		0.8	38.3		1.0	40.6
Fruit weight (Kg)	5	10.1ab \pm	5.0 -	32	8.6a \pm	4.0 -	21	11.7b \pm	2.0 -
	5	0.5	24.7		0.7	15.4		0.8	23.4
Flesh thickness (mm)	5	30.1a \pm	2.6 -	32	34.4a \pm	4.0 -	21	50.4a \pm	4.8 -
	5	2.7	58.5		3.5	61.9		4.3	80.4
Skin thickness (mm)	5	19.0a \pm	2.0 -	32	15.0a \pm	2.0 -	21	18.0a \pm	3.0 -
	5	1.0	46.0		2.0	28.0		2.0	27.0
Seed weight (g)	5	117.9c \pm	72.2 -	32	97.4a \pm	51.0 -	21	70.6a \pm	36.4 -
	5	4.0	261.5		5.2	227.0		6.5	111.3
100 seed weight (g)	5	27.8b \pm	18.9 -	32	35.5c \pm	25.9 -	21	20.8a \pm	11.8 -
	5	0.8	39.3		1.1	70.4		1.3	38.1

* In the line means followed by same letter are not different at $p=0.05$

The second component explains 16.04% of the total variation and is correlated positively by 100 seed weight and secondary fruit skin colour pattern and negatively by fruit length and fruit ribs. The third component accounted for 14.04% and is defined by flesh thickness, fruit width and fruit weight (Table 2). The PCA scatter plot shows a clear three species separation spreading the 108 *Cucurbita* populations into three groups (Figure 1). The first group comprise the populations of *C. pepo*, the second group the populations of *C. moschata* and the third group the populations of *C. maxima*. The genetic diversity of landraces is part of the economic value of global biodiversity and is considered of paramount importance for future world production (Wood and Jenne, 1997; Stoilova *et al.*, 2005). The conservation and maintenance of these valuable genetic resources are important because they are a source of diversity for use in breeding programs (Balkaya *et al.*, 2005; Bettencourt, 2012).

Table 2. Vector loadings and percentage of variation explained by the first three principal components

Variable	PC1	PC2	PC3
Fruit length	0.203	-0.351	0.014
Fruit width	0.181	0.108	0.467
Fruit weight	0.138	-0.186	0.452
Flesh thickness	-0.013	-0.047	0.487
Skin thickness	0.322	-0.063	-0.051
Seed weight	0.321	0.063	0.100
100 Seed weight	0.103	0.338	0.119
Peduncle transectional shape	0.311	-0.269	-0.134
Fruit shape	-0.066	-0.223	-0.332
Fruit ribs	-0.049	-0.325	0.241
Peduncle binding	-0.259	-0.247	-0.161
Predominant fruit skin colour at maturity	-0.112	0.258	-0.019
Secondary fruit skin colour	0.042	0.241	0.026
Secondary fruit skin colour pattern	0.01	0.304	0.073
Fruit skin texture	0.109	-0.033	-0.024
Flesh colour	-0.34	-0.272	0.173
Flesh texture	0.268	-0.108	-0.032
Predominant seed coat colour	-0.361	-0.141	0.224
seed margin coat colour	-0.388	0.043	0.106
Seed shape	0.157	-0.302	0.044
Eigenvalues	4.481	3.201	2.807
Percentage	22.405	16.004	14.037
Cum. Percentage	22.405	38.408	52.446

Figure 1. Projection of the 108 *Cucurbita* populations in a two-dimensional graph defined by PC1 and PC2.

CONCLUSIONS

The genetic resources characterization is a key for the management of genebanks and plant breeding. The three *Cucurbita* species evaluated in this study showed a high variability for some fruit parameters such as weight, fruit shape and predominant and secondary skin colour at maturity. Nevertheless, others fruit parameters such as peduncle transectional shape the variability was nonexistent. The high genetic diversity found could be used in breeding programs to obtain new cultivars and provide relevant information for the diversity conservation.

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