

Journal of Biodiversity and Environmental Sciences (JBES) ISSN: 2220-6663 (Print) 2222-3045 (Online) Vol. 9, No. 1, p. 224-230, 2016 http://www.innspub.net

OPEN ACCESS

Morphological evaluation of *Capsicum* spp. diversity in Iran

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Article published on July 25, 2016

Key words: Pepper improvement, Morphological traits, Multivariate analysis.

Abstract

48 genotypes of *Capsicum* spp. obtained from National Plant Gene Bank of Iran. The experiment was conducted as an alpha lattice design and 11 important morphological traits were measured based on IPGRI descriptor. Analysis of variance, Principle Component Analysis, cluster analysis and stepwise regression were performed for all variables. All studied morphological traits showed significant differences among the genotypes. Principle component analysis revealed the first four components can justify over 79% of total variances. The cluster analysis (UPGMA) based on fruit length and fruit width classified all genotypes into four groups. According to the results of stepwise regression analysis 59.65% of the total variation in fruit fresh weight could be attributed to total plant weight, fruit wall thickness and fruit length. The identified diversity between genotypes showed its important potential for breeding program in *Capsicum* spp.

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Introduction

Capsicum L. (pepper) is a member of the Solanaceae family which is cultivating in temperate and tropical regions (Eshbaugh, 1993; Lippert et al., 1966; Pozzobon et al., 2005; Rohami et al., 2010). Fruits of pepper are used as vegetable, spice and medicinal purposes. Vegetable are among the most important horticultural crops for being good sources of vitamins, minerals, fiber and Antioxidants which required in the human diet (Palafox-Carlos et al., 2011). Pepper is one of the most significant plants which use as vegetable, species and medicinal plant. Pepper has important functions in various aspects of food and pharmaceutics. Pepper fruits have highest content of vitamin C among all plants and have important medicinal profits such as antioxidant characteristics (Rohami et al., 2010). Capsicum L. (pepper) is a member of the Solanaceae family and is cultivated in temperate and tropical regions.

Capsicum has about 20-30 species which five of them are domesticated: *Capsicum annuum*, *C. frutescens*, *C. chinense*, *C. pubescens* and *C. baccatum* (De Teodoro-Pardo *et al.*, 2007; Eshbaugh, 1993; Lanteri, 1993; Lippert, 1966; Pozzobon *et al.*, 2005; Rohami *et al.*, 2010). Studies on *Capsicum* species have shown that they contain 24 chromosomes (2n=2x=24), similar to many species of Solanaceae family (Rohami *et al.*, 2010).

Information about genotypes diversity which is obtained through morphological and molecular studies is widely used for pepper improvement (Gelta *et al.*, 2005). Different researches have classified *Capsicum* genotypes according their morphological distances. The analysis of variance of 38 genotypes of chilli (*Capsicum annuum* L.) in India for different morphometric and qualitative characters recognized important amount of variability among the genotypes (Patel *et al.*, 2009). Also Geleta *et al.* (2005) studied 20 different morphological traits of thirty-nine genotypes of pepper collected from different countries during 2001 and 2002 in South Africa. This study showed high genetic diversity and genetic distance between genotypes.

Also Geleta et al. (2005) reported that the cluster analysis divide genotypes into three main clusters. Peppers with similar fruit sizes classified in same groups (Gelta et al., 2005). The genetic diversity of five pepper (Capsicum annuum L.) varieties which are gathered from different collection sites in Mauritius was studied base on morphological and molecular characteristics (Peeraullee, Ranghoo-Sanmukhiya, 2013). The morphological traits used for this study were plant height, plant canopy width, leaf length, leaf width, leaf color, leaf shape, fruit color, fruit length and fruit shape. This study showed a high degree of similarity among the Capsicum varieties which include plant characteristics and leaf characteristics. In contrast, the study revealed great variation in the fruit traits between varieties (Peeraullee and Ranghoo-Sanmukhiya, 2013). A Study about phenotypic diversity of 48 genotypes of Capsicum annuum, including germplasm lines and commercial cultivars in Turkey was done to characterize all genotypes for 67 agro-morphological traits during plants growth (Bozokalfa et al., 2009). The results showed high diversity in the collections. Bozokalfa et al. (2009) used Principle Component Analysis (PCA) to determine the variation of the pepper genotypes. PCA showed the first six components justified over 54% of the variability among the 48 accessions and their lines. Also the Ward's clusters analysis based on fruit shape and fruit agronomic traits classified the 48 pepper genotypes in seven groups (Bozokalfa et al., 2009). Ghazi Zadeh et al. (2010) studied 77 genotypes collected from different areas of Iran and evaluated more than 40 morphological traits. The results of cluster analysis divided genotypes in five groups and PCA analysis showed that the main 13 components can justify 74.59% of total variance. Also stepwise regression depicted best contributing of fruit width, fruit wall thickness, mature leaf length, mature leaf width and fruit length respectively to fruit weight as a dependent variable (Ghazi zadeh et al., 2010).

Capsicum is native to the new world, but this plant is under cultivation in different regions of Iran especially in North regions of this country. Because of several biotic and abiotic stresses in this area, we need to analyze variation of between cultivated crops and using it in pepper's breeding programs. The purpose of this study was to evaluate the morphological diversity between 48 genotypes Collection of pepper obtained from different parts of Iran and recognizing the potential of improvement of these genotypes.

Material and methods

Materials

In this study, 48 genotypes of *Capsicum* spp. including germplasm lines, local ecotypes and commercial cultivars were obtained from National Plant Gene Bank of Seed and Plant Improvement Institute of Iran. Most of these genotypes were collected from different part of Iran and original selected area of seeds had been identified by National Plant Gene Bank of Iran (Table 1).

Table 1. The studied genotypes obtained from National Plant Gene Bank of Iran (NPGBI) and Iranian Seed

 Company.

Genotype	Genotype	Original selected	Genotype	Genotype	Original selected area
Number	Code	area	Number	Code	Original selected area
1	38-2	Unknown	25	0-1	unknown
2	3-2	Australia	26	31-8	China
3	4-9	Australia	27	72-1	Oroumieh
4	57-5	Local-Bonab	28	1-23-2	Local-Varamin
5	3-4	unknown	29	0-2	unknown
6	23-4	Sabzevar	30	3-10-4	unknown
7	27-2	Unknown	31	4-9-2	Australia
8	3-7	unknown	32	13-9-3	South Korea
9	14-4-4	Australia	33	17-1-2	North Korea
10	58-5-3	Local-Bonab	34	33-5	unknown
11	3-3	Australia	35	37-1-2	Kurdistan province
12	37-1-2	Kurdistan province	36	0-3	unknown
13	68-1	Sanandaj	37	56-1-2	Local-Sari
14	1-23-2	Local-Varamin	38	57-5-4	unknown
15	18-1-3	Sabzevar	39	58-9-4	Local-Bonab
16	12-1	Hungary	40	0-4	unknown
17	57-5-4	unknown	41	0-5	unknown
18	3-4	unknown	42	27-2	unknown
19	3-7	unknown	43	0-6	Slender-Hungary
20	5-4	Azerbaijan	44	0-7	unknown
21	10-5	unknown	45	0-8	USA
22	12-3	Hungary	46	0-9	USA
23	13-8	South Korea	47	0-10	USA
24	13-9	unknown	48	0-11	USA

Procedure

The experiment was conducted in 2013 as an alpha lattice design with two replications at the experimental field of the Islamic Azad University, Karaj branch, Iran (51° 6' E, 35° 45' N and 1313m above the sea level). During this study 11 important morphological traits were measured based on IPGRI descriptor (1995) including plant height, plant canopy width, mature leaf length, mature leaf width, fruit length, fruit width, fruit pedicel length, fruit wall thickness, 1000 seed weight, total plant weight and fruit fresh weight.

Statistical methods

Analysis of variance for alpha lattice design and Principle Component Analysis were performed for all variables. The UPGMA (the average-linkage-betweengroups) method was used for cluster analysis based on fruit length and fruit width. Also stepwise regression analysis was done to identify best contributing traits to fruit fresh weight as a dependent variable.

Results

The analysis of variance showed wide differences between pepper collections. All studied morphological traits including plant height, mature leaf length, mature leaf width, fruit length, fruit width, fruit pedicel length, fruit wall thickness, 1000 seed weight, total plant weight, fruit fresh weight (P \leq 0.01) and plant canopy width (P \leq 0.05) have shown significant differences among the genotypes (Table 2).

Principle Component Analysis (PCA) indicated the first four components explained 79.71% (38.57, 19.72, 11.07 and 10.33% by components 1-4 respectively) of the total variance (Table 3). PCA revealed that fruit fresh weight, mature leaf width and mature leaf length had the highest coefficient in the first component. Also 1000 seed weight, total plant weight

and plant height for second component, plant canopy width for third component and fruit length and fruit pedicel length for fourth component showed the highest coefficient.

Table 2. Analysis of Variance for studied morphological traits.

SOV	MS_E	MS_B	\mathbf{MS}_{t}	CV%	
Plant height	21.04	124.96	118.15**	12.1	
Plant canopy width	27.46	23.63	72.04*	15.9	
Mature leaf width	0.24	0.26	1.77**	14.9	
Mature leaf length	0.70	0.009	5.91**	12.9	
Fruit length	1.22	1.06	7.45**	16.1	
Fruit width	0.34	0.30	2.36**	23.9	
Fruit pedicel length	0.15	0.17	0.55**	15.1	
Fruit wall thickness	0.09	0.001	1.10**	16.1	
Total plant weight	9883.3	42078.64	56202.09*	* 28.7	
1000 – seed weight	0.02	0.0072	2.18**	2.7	
Fruit fresh weight	30931.31	95652.05	197591.5**	[•] 20.9	
*, Significant at p≤0.05; **, Significant at p≤0.01.					

Table 3. Eigen value and percentage of variance and cumulative of Principle Component Analysis.

Component	Initial Eigenvalues			Extraction Sums of Squared Loadings		
Component	Total	% of Variance	Cumulative %	Extrac Total 4.243 2.170 1.219 1.137	% of Variance	Cumulative %
1	4.243	38.573	38.573	4.243	38.573	38.573
2	2.170	19.723	58.296	2.170	19.723	58.296
3	1.219	11.079	69.375	1.219	11.079	69.375
4	1.137	10.338	79.714	1.137	10.338	79.714
5	.891	8.099	87.813			
6	.649	5.900	93.713			
7	.280	2.543	96.256			
8	.226	2.058	98.314			
9	.129	1.170	99.484			
10	.057	.516	100.000			
11	-1.001E-013	-1.007E-013	100.000			

The 2D plot of the first and second components revealed dispersion and relationship of 48 studied genotypes which is important for improvement programs (Fig. 1).

The cluster analysis (UPGMA) based on fruit length and fruit width classified all genotypes into four groups (Fig. 2). The largest cluster contains most studied peppers and has 44 genotypes. The second and third cluster showed just one genotype for each group (42 and 44, respectively) and the fourth cluster contains genotypes number 46 and 47 (Fig. 2).



Fig. 1. Dispersion Plot of the first two components from principle component analysis (PCA).



Fig. 2. Dendrogram for the 48 studied genotypes obtained with UPGMA clusters analysis is based on fruit traits.

The stepwise regression analysis with using fruit fresh weight as dependent variable indicated that, total plant weight, fruit wall thickness and fruit length were characteristics which had most effect on fruit fresh weight (Table 4). According to the results, 59.65% of the total variation in fruit fresh weight could be attributed to these 3 morphological traits. The other traits were not included in the analysis because of their low relative contributions. The predicted equation was formulated as follows:

Y= -220.246+1.79X1+156.493X2+5.173X3

Where, Y, X₁, X₂ and X₃ are total plant weight, fruit wall thickness and fruit length respectively.

Table 4. Stepwise regression model for fruit freshweight as dependent variable (Y).

Model	1	2	3
constant	470.350	89.623	-220.246
total plant weight	1.077	1.154	1.79
fruit wall thickness	-	184.777	156.493
fruit length	-	-	5.173
R ²	30.68	50.0	59.65

Discussion

The analysis of variance indicated considerable diversity between genotypes of pepper in Iran. These results are in agreement with the findings of Gelta *et al.* (2005), Patel *et al.* (2009), Bozokalfa *et al.* (2009) and Ghazi Zadeh *et al.* (2010) for same morphological traits. However Peeraullee and Ranghoo-Sanmukhiya (2013) reported less degree of variation among the five *Capsicum* varieties based on phenotypic plant characteristics and leaf characteristics except fruit traits.

In the present study results of principle component analysis revealed that fewer components could justify more than 79% of total variance; however Ghazi Zadeh *et al.* (2010) reported that the first 13 components justified 74.59% of total variance. Also Bozokalfa *et al.* (2009) showed the first six components justified over 54% of the variability among the 48 genotypes collected from different areas of Turkey. Also Bozokalfa *et al.* (2009) used PCA Scatter plot to determine the variation and the relationship of studied genotypes. Cluster analysis showed most studied genotypes does not have significant diversity through fruit length and fruit width. These results are in agreement with the reports of Ghazi Zadeh et al. (2010) which classified Iranian genotypes into 5 groups. Peeraullee and Ranghoo-Sanmukhiya (2013) revealed fruit traits can show great variation between pepper genotypes among all pepper morphological characteristics. Also Gelta et al. (2005) used cluster analysis on the basis of morphological data for 39 genotypes of South Africa and the results showed three main clusters. The largest cluster of Gelta et al. (2005) study contains genotypes from all varietal groups except from the bell peppers. Also Bozokalfa et al. (2009) classified 48 genotypes of collected pepper from Turkey in 7 groups based on fruits traits. The results of cluster analysis and PCA of present study revealed no correlation between genotypes and zones which genotypes were collected from there. This result is similar to reports of Ghazi Zadeh et al. (2010).

The results of stepwise regression is similar to the Ghazizadeh *el al.* (2010) who reported that fruit width, fruit wall thickness, mature leaf length, mature leaf width and fruit length accounted for 73.8% of the genotypes among the varieties in fruit weight.

Results indicated significant diversity between the studied genotypes through morphological traits. UPGMA cluster analysis with using fruit characteristics classified genotypes in four groups and revealed no correlation between genotypes and collection areas of genotypes. Principle Component Analysis (PCA) recognized that the first four components can justify over 79% of total variances and fruit fresh weight, mature leaf width and mature leaf length had the highest coefficient in the first component. Also Stepwise regression analysis depicted that total plant weight, fruit wall thickness and fruit length accounted for 59.65% of the variation in fruit fresh weight among the studied genotypes. As final conclusions this study revealed the high diversity between Capsicum genotypes and its important potential for improving agronomic traits.

During this study most important and commercial pepper genotypes which is cultivating in Iran and Middle East classified based on fruit characteristics. Also the results showed importance of morphological traits include mature leaf width, mature leaf length, total plant weight, fruit wall thickness, fruit length and fruit fresh weight. These results should be considered in pepper improvement programs.

Acknowledgements

We would like to appreciate "the Islamic Azad University-Karaj Branch" for financial supports of this study. The research project number of this study is: 1.216887(1390-11-05).

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