



## Selection in segregating populations of ornamental pepper plants (*Capsicum annuum* L.) using multidimensional scaling<sup>1</sup>

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### ABSTRACT

Multidimensional scaling is a multivariate analysis technique that can be used to exploit genetic diversity, aiming at the selection of *Capsicum* genotypes with desirable characteristics for in-pot ornamental purposes. This work aimed to select genotypes with ornamental potential within F<sub>4</sub> populations of ornamental pepper plants. Three F<sub>4</sub> families were used (17.18, 30.16, and 56.8). The genotype distance matrixes between genotypes were estimated based on qualitative and quantitative descriptors, both separately, combining the standardized distances of Gower and Mahalanobis, respectively. The relation of the distance between genotypes was graphically studied through non-metric multidimensional scaling. Kruskal' Stress was used as the measured misadjustment of the *nMDS* solution. There is genetic diversity within the analyzed families, allowing to practice selection. The selection in family 17.18 of genotypes 6 and 32 is recommended, as well as in family 30.16 of genotypes 22 and 4, and family 56.8 of genotypes 15 and 36, since they present important characteristics for ornamental purposes. The selection of genotypes is more efficient when using mixed data since it provides a more complete genetic diversity in an improvement program.

**Keywords:** Multivariate analysis; genetic diversity; Gower; Mahalanobis.

### INTRODUCTION

Pepper (*Capsicum* spp.) is one of the most important and most cultivated horticultural plants in the world (Bosland & Votava, 2012; Silvar & García-González, 2017). They can be used in culinary, pharmaceutical and cosmetic industries, as well as ornamental plants (Rêgo *et al.*, 2015a; Cardoso *et al.*, 2018).

In ornamentation, pepper plants have been highlighted for their growing and continuous receptivity by the consuming market (Finger *et al.*, 2015; Rêgo & Rêgo, 2018). This growing interest for ornamental peppers occurs due to the great diversity observed in several features, such as size, leaves, fruit color, and adequate architecture (Stommel & Bosland, 2006; Padilha *et al.*, 2016).

In spite of the diversity found in this genus, only some ornamental varieties are available in the Brazilian market

(Rêgo *et al.*, 2015a; Neitzke *et al.*, 2016). It is, therefore, important to insert pepper plants in improvement programs aiming at the development of new varieties for ornamental purposes (Rêgo *et al.*, 2015b; Vasconcelos *et al.*, 2012; Silva *et al.*, 2017).

The Federal University of Paraíba (UFPB) has developed in the last few years an improvement program for ornamental pepper plants whose purpose is the development of new varieties, based mostly on hybridization, generating segregating populations and allowing for the improvers to select superior genotypes, with desirable characteristics for in-pot cultivation (Silva Neto *et al.*, 2014; Rêgo *et al.*, 2015a; Mesquita *et al.*, 2016; Lima *et al.*, 2019; Fortunato *et al.*, 2019; Nascimento *et al.*, 2019; Pessoa *et al.*, 2019). In this perspective, the study of the genetic diversity among genotypes in segregating

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*Capsicum* populations can be explored through the use of multivariate analysis.

Multidimensional scaling is a multivariate analysis technique that can be used in the study of the genetic diversity in *Capsicum* since it allows to graphically represent the distances of the genotypes through dimensional reduction. The individuals are, therefore, arranged as points in space, generally bidimensional or tridimensional, in which it is possible to observe the similarity and the dissimilarity between individuals (Borg & Groenen, 2005; Manly, 2008). The use of this analysis in the study of genetic diversity in improvement programs was performed in studies with cocoa (Leal *et al.*, 2008), garlic (Silva *et al.*, 2014), broad bean (Barroso *et al.*, 2019), and a segregating population of *Capsicum* (Pessoa *et al.*, 2019). Then, this analysis may be appropriate for studies on genotypic dispersion, since the conventional improvement program is based mainly on phenotypic data, which are based on qualitative and quantitative characters (Barroso *et al.*, 2019). Furthermore, it may also be important in the selection of more divergent individuals with desirable features for the improvement program.

This work aimed to select genotypes with ornamental potential within populations of the fourth filial generation ( $F_4$ ) of ornamental peppers through multidimensional scaling.

## MATERIAL AND METHODS

The experiment was performed in a plant nursery, at the Laboratory of Plant Biotechnology of the Center of Agrarian Sciences of the Federal University of Paraíba (CCA/UFPB), in the municipality of Areia-PB, at an elevation of 618 m, 06° 57' 48" S latitude and 35° 41' 30" W longitude.

Three  $F_4$  families were used (17.18, 30.16, and 56.8) each one constituted by 45 individuals originated from the UFPB 77.2 x UFPB 134 crossing, conducted by the genealogical method.

The sowing was performed in expanded polystyrene trays, with 200 cells filled with the Plantmax HT® commercial substrate. Thirty-five days after sowing, when the plantlets presented three pairs of definitive leaves, they were transplanted to plastic pots with volumetric capacity for 900 ml, containing the Plantmax HT® commercial substrate. Whenever necessary, the cultural practices recommended for the crop were performed (Filgueira, 2000).

Twelve quantitative descriptors were used, namely: plant height (PH), canopy diameter (CD), first bifurcation height (FBH), stem diameter (SD), leaf length (LL), leaf width (LW), fruit weight (FW), fruit length (FL), largest fruit diameter (LFD), smallest fruit diameter (SFD), number of seeds (NS), number of fruits (NF). The qualitative descriptors used were: color of the immature fruit (CIF), color of the intermediate fruit (CIF), color of the mature

fruit (CMF), fruit shape (FS), fruit persistence with the pedicel (FPP), pedicel persistence with the stem (PPWS), stem color (SC), stem shape (SS), growth habit (GH), branching density (BD), leaf color (LF), and leaf shape (LS), all based on the *Capsicum* descriptors proposed by IPGRI (1995). The experimental design was completely randomized. Each experimental unit consisted of one plant per pot. three families of an  $F_4$  population were evaluated, each with 45 plants.

The genotypes were represented in a graphic form through the non-metric multidimensional scaling technique based on qualitative, quantitative, and mixed characteristics, with the last one considering the joint analysis of both qualitative and quantitative descriptors.

For the analysis of the quantitative variables, a distance matrix based on the generalized distance of Mahalanobis was obtained through the expression:

$$D_{ii}^2 = \delta^{-1}\Psi^{-1} \quad (1)$$

In which  $D_{ii}^2$  = distance between genotypes  $i$  and  $i'$ ;  $\Psi$  = residual covariance matrix;  $\delta' = [d_1, d_2, \dots, d_n]$ , in which  $d_n = Y_{ij} - Y_{i'j}$  and  $d_n$  = represents the difference between the mean of two genotypes  $i$  and  $i'$  for each evaluated characteristic;  $Y_{ij}$  = mean of the  $i$ -nth genotype in relation to the  $j$ -nth descriptor.

The distance matrix with the qualitative data was constructed using Gower's distance (Gower, 1971), employing the equation:

$$S_{ij} = \frac{\sum_{k=1}^n W_{ijk} S_{ijk}}{\sum_{k=1}^n W_{ijk}} \quad (2)$$

In which  $S_{ij}$  is the similarity between genotypes  $i$  and  $j$ ,  $W_{ijk}$  = weight attributed to the  $i_{jk}$  comparison, attributing the value 1 for the valid comparisons and the value 0 for the invalid comparisons (when the value of the variable is absent in either one or both individuals).  $S_{ijk} = 1$ , the contribution of the  $k$  variable in the similarity between individuals  $i$  and  $j$ , with values between 0 and 1. For a nominal variable, if the value of the  $k$  variable is the same for individuals  $i$  and  $j$ , then = 1, otherwise, it will be equal to 0. Thus,  $d_{ij} = 1 - S_{ij}$ .

The non-metric multidimensional scaling (nMDS) was used for the graphic representation of the distance matrixes in the bidimensional space. The level of adjustment (or lack of adjustment) in the nMDS mapping was calculated through Kruskal's Stress.

$$Stress_I = \left[ \frac{\sum_{i=1}^{n-1} \sum_{j>1}^n (d_{ij} - \tilde{d}_{ij})^2}{\sum_{i=1}^{n-1} \sum_{j>1}^n \tilde{d}_{ij}^2} \right]^{\frac{1}{2}} \quad (3)$$

In which  $d_{ij}$  = distance between genotypes  $i$  and  $j$ ;  $\tilde{d}_{ij}$  = mapped distance between genotypes  $i$  and  $j$ . The table of stress values proposed by Sturrock and Rocha (2000) was used to validate the result obtained with the nMDS

solution, by comparing the value obtained with the stress value generated from the matrixes with the same number of objects and the same number of dimensions. All analyses were performed with the R software, version 4.0.1 (R Core Team, 2020).

## RESULTS AND DISCUSSION

The non-metric multidimensional scaling technique (nMDS) presented distances between the genotypes of three families (17.18; 30.16; 56.8) of pepper plants in a

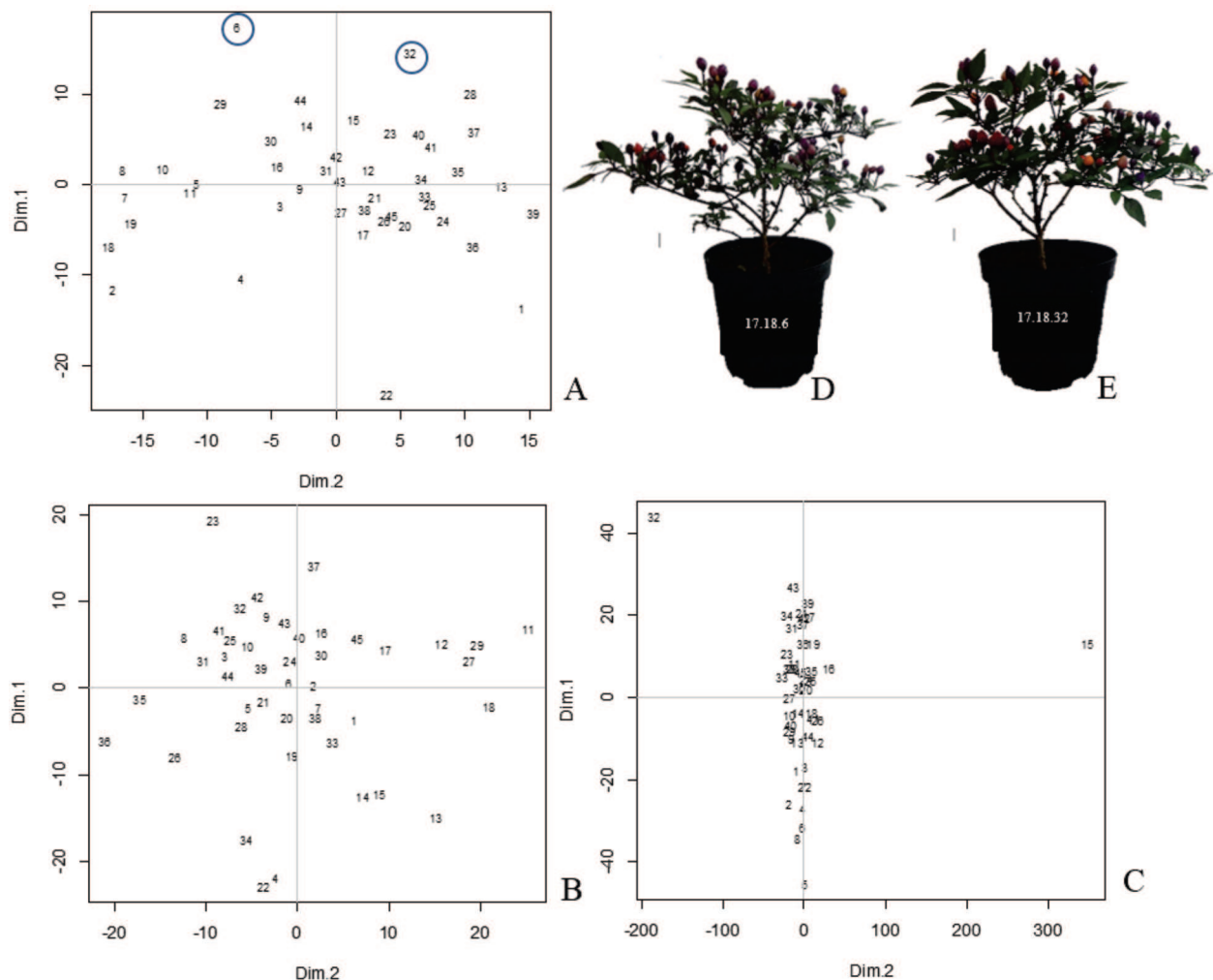
**Table 1:** Stress values (Kruskal,1964) associated to the non-metric multidimensional scaling analysis for 45 genotypes of the 17.18, 30.16 and 56.8 families regarding qualitative, quantitative, and mixed characteristics of ornamental pepper plants (*Capsicum annuum* L.)

Family	Qualitative	Quantitative	Mixed
17.18	22.83 %	17.31 %	23.52 %
30.16	15.62 %	20.00 %	20.00 %
56.8	20.00 %	10.41 %	19.59 %

graphic form, in which a dispersion can be observed in the qualitative, quantitative, and mixed data, indicating the occurrence of genetic variability in the  $F_4$ , with more selection cycles being useful to proceed with the improvement program of ornamental pepper plants aiming to obtain new lineages.

The stress varied from 10.41% to 23.53% in the analyzed families for the qualitative, quantitative, and mixed characteristics (Table 1). According to the table proposed by Sturrock & Rocha (2000), with  $n = 45$  genotypes,  $k = 2$  dimensions, there is a probability ( $p < 0.01$ ) that the genotypes will be randomly arranged in the bidimensional space.

Furthermore, stress values up to 20.00% are acceptable in this type of analysis (Kruskal *et al.*, 1964; Meyer *et al.*, 2004). The stress values obtained indicate a good ordination in the qualitative data for families 17.18; 30.16, and 56.8, and in the quantitative and mixed data for families 30.16 and 56.8. Thus, through this analysis, it was possible to identify the more divergent genotypes, which contain a



**Figure 1:** Graphic representation of the bidimensional scaling based on the quantitative data of the 17.18 (A), 30.16 (B), and 56.8 (C) families of 45 genotypes of *Capsicum annuum*. Genotypes 17.18.6 (D), 56.18.32 (E). Bar corresponding to 2 cm. Areia-PB. CCA-UFPB-2020.

phenotype of in-pot ornamental interest. In this study, the dispersion based on quantitative and mixed data for the 17.18 family was unsatisfactory. This indicates that the representation of the distances in the bidimensional space is not an appropriate choice (Silva, 2016).

According to the quantitative analysis of the 17.18 family, the most distant genotypes in the graphic were the 6, 32, 28, 1, 4, and 2 (Figure 1A). Genotype 6 presented smaller size, fruits, and leaves, Genotype 32 presented a wider canopy diameter and a lower first bifurcation height. Genotype 28 presented a smaller stem diameter and smaller fruits. Genotype 1 presented a large size and larger leaves, contrasting with genotype 2, which, in spite of presenting a large size, presents small leaves. Genotype 4 presented a wider leaf width (Table 2). Among the most distant individuals, those that presented interesting characteristics for the in-pot ornamental pepper were genotypes 6 and 32 (Figure 2D and 2E), for possessing small fruits and leaves allied to a small size, adapting to the pot and able to be recommended for use in the improvement of the ornamental pepper (Büttow *et al.*, 2010; Costa *et al.*, 2019), being attractive to the customer at the time of purchase (Neitzke *et al.*, 2016).

For the family 30.16, in the quantitative data analysis, the genotypes that stood more distant were the 23, 37, 11, 18, 13, 22, 2, 36, and 34 (Figure 1B). Among the more distant genotypes, which presented desirable characteristics for

the in-pot ornamental pepper plant, genotypes 22, 36, and 34 (Figure 1B) are highlighted. Genotype 22 presented the greatest number of fruits. Genotype 36 exhibited low size and a lower first bifurcation height, whereas genotype 34 presented smaller leaves and fruits (Table 2). For ornamental purposes, the selection of plants with these characteristics is recommended (Finger *et al.*, 2012; Silva *et al.*, 2015; Bianchi *et al.*, 2016; Bărcanu *et al.*, 2017) to continue the improvement program with ornamental pepper plants.

It is observed that the family 56.8 presented a lower dispersion of genotypes, indicating a greater homogeneity of the plants within this family when compared to the other families. It is then notorious that this family is closest to homozygosis. In spite of the existence of very close genotypes, only genotypes 32, 15, and 5 were distant for the quantitative characteristics (Figure 1C). Genotype 32 presented smaller leaves and larger fruits; conversely, plant 15 presented a smaller size and a lower first bifurcation height, whereas individual 5 presented a smaller stem diameter (Table 2). Among these more distant individuals, the selection of genotype 5 is recommended for presenting desirable descriptors for the in-pot ornamental pepper plant. Santos *et al.* (2014) affirm that is important to select pepper genotypes that present plant and fruit sizes adequate to in-pot cultivation, aiming at the decoration of internal spaces.

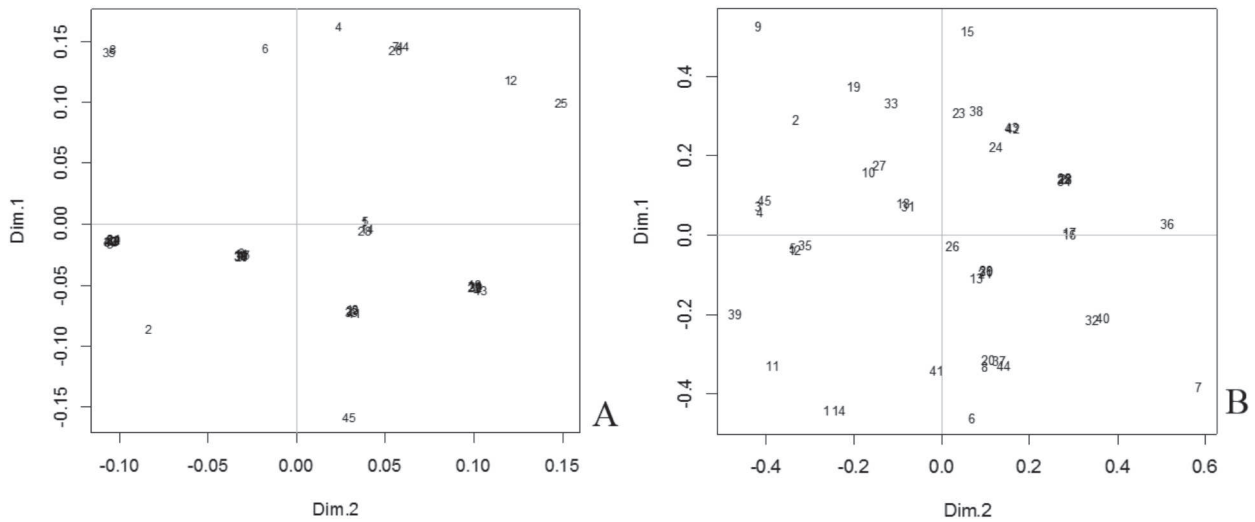
**Table 2:** Quantitative descriptors of families 17.18, 30.16, and 56.8 of pepper plants (*Capsicum annuum* L.)

17.18	PH	CD	FBH	SD	LL	LW	FW	FL	LFD	SFD	NS	NF
1	32.0	26.0	16.0	0.49	3.81	1.53	0.73	1.56	0.97	0.26	30	46
2	31.0	35.0	12.5	0.50	2.00	1.67	0.45	1.05	0.87	0.33	25	46
4	30.0	31.0	15.0	0.47	3.00	1.90	0.40	0.92	0.84	0.36	19	39
6	23.0	28.0	15.0	0.59	2.13	0.88	0.34	0.84	0.74	0.25	15	35
28	23.0	26.0	16.0	0.44	3.52	1.31	0.30	0.79	0.74	0.29	12	51
32	23.0	37.0	11.0	0.64	3.25	1.24	0.69	1.10	0.95	0.34	28	30
30.16	PH	CD	FBH	SD	LL	LW	FW	FL	LFD	SFD	NS	NF
2	24.0	33.0	14.0	0.56	2.06	1.02	0.35	1.17	0.70	0.33	26	60
11	27.0	36.0	14.0	0.64	2.17	1.13	0.73	1.65	0.82	0.39	48	76
13	32.0	35.0	16.0	0.62	2.07	0.93	0.41	1.43	0.75	0.30	22	91
18	28.0	29.0	14.0	0.56	2.49	1.35	0.58	1.48	0.84	0.37	41	83
22	21.0	28.0	12.0	0.52	1.90	0.84	0.16	0.76	0.51	0.31	10	99
23	20.0	19.0	13.0	0.52	1.95	0.81	0.20	0.78	0.60	0.35	30	49
34	24.0	36.0	12.0	0.44	1.64	0.56	0.17	0.77	0.53	0.25	12	86
36	20.0	30.0	9.0	0.27	1.32	0.61	0.38	1.13	0.76	0.33	28	48
37	22.0	28.0	11.0	0.76	2.39	1.18	0.49	1.40	0.79	0.33	25	45
56.8	PH	CD	FBH	SD	LL	LW	FW	FL	LFD	SFD	NS	NF
5	25.0	33.0	13.0	0.42	3.14	1.25	0.61	1.22	0.64	0.32	47	61
15	31.0	31.0	14.0	0.48	3.10	1.54	0.90	1.72	1.03	0.30	30	35
32	22.0	25.0	13.0	0.65	2.76	1.26	1.31	2.25	1.18	0.29	27	38

(PH) plant height (cm), (CD) canopy diameter (cm), (FBH) first bifurcation height (cm), (SD) stem diameter (cm), (LL) leaf length (cm), (LW) leaf width (cm), (FW) fruit weight (g), (FL) fruit length (cm), (LFD) largest fruit diameter (cm), (SFD) smallest fruit diameter (cm), (NS) number of seeds, (NF) number of fruits

Other works have demonstrated the distance of the genotypes in a graphic form through the non-metric multidimensional technique using quantitative data, such as that by Silva *et al.* (2014), who used the non-metric multidimensional scaling analysis based on quantitative data for the identification of garlic genotypes based on

different phenotypical characteristics. Barroso *et al.* (2019) used the analysis in the selection of more divergent broad bean genotypes, having quantitative data as a base (Pessoa *et al.*, 2019), and also selecting genotypes in a segregating population of pepper plants based on quantitative data.



**Figure 2:** Graphic representation of the multidimensional scaling of the families based on qualitative data. Families 30.16 (A) and 56.8 (B), each with 45 *Capsicum annuum* genotypes. Areia-PB. CCA-UFPB-2020.

**Table 3:** Qualitative descriptors of families 30.16 and 56.8 of ornamental peppers (*Capsicum annuum* L.)

30.16	CIM	CIF	CMF	FS	FPP	PPWS
2	Light green	Yellow /orange	Red	Triangular	Persistent	Persistent
4	Light green	Yellow	Orange	Triangular	Intermediate	Intermediate
12	Light green	Yellow	Orange	Triangular	Intermediate	Intermediate
25	Light green	Yellow	Orange	Triangular	Intermediate	Intermediate
45	Light green	Yellow/ orange	Red	Triangular	Persistent	Intermediate
30.16	SC	SS	GH	BD	LF	LS
2	Green	Angular	Intermediate	Intermediate	Dark green	Oval
4	Green	Angular	Intermediate	Intermediate	Dark green	Lanceolate
12	Green	Angular	Intermediate	Intermediate	Dark green	Lanceolate
25	Green	Angular	Intermediate	Intermediate	Dark green	Oval
45	Green	Angular	Intermediate	Intermediate	Light green	Oval
56.8	CIM	CIF	CMF	FS	FPP	PPWS
6	Purple	Brown /yellow	Orange	Triangular	Intermediate	Light
7	Green/Purple	Yellow	Orange	Triangular	Intermediate	Light
9	Purple	Brown /yellow	Orange	Triangular	Persistent	Light
15	Purple/Brown	Brown /yellow	Orange	Triangular	Persistent	Light
36	Purple	Yellow	Orange	Triangular	Intermediate	Light
56.8	SC	SS	GH	BD	LF	LS
6	Green/ strips	Cylindrical	Intermediate	Intermediate	Green	Oval
7	Green/ strips	Cylindrical	Intermediate	Intermediate	Green	Lanceolate
9	Green/ strips	Cylindrical	Erect	Scarce	Light green	Lanceolate
15	Green/ strips	Angular	Intermediate	Intermediate	Light green	Lanceolate
36	Green/ strips	Angular	Intermediate	Intermediate	Dark green	Lanceolate

color of the immature fruit (CIM), color of the intermediate fruit (CIF), color of the mature fruit (CMF), fruit shape (FS), fruit persistence with the pedicel (FPP), pedicel persistence with the stem (PPWS), stem color (SC), stem shape (SS), growth habit (GH), branching density (BD), leaf color (LF), and leaf shape (LS).

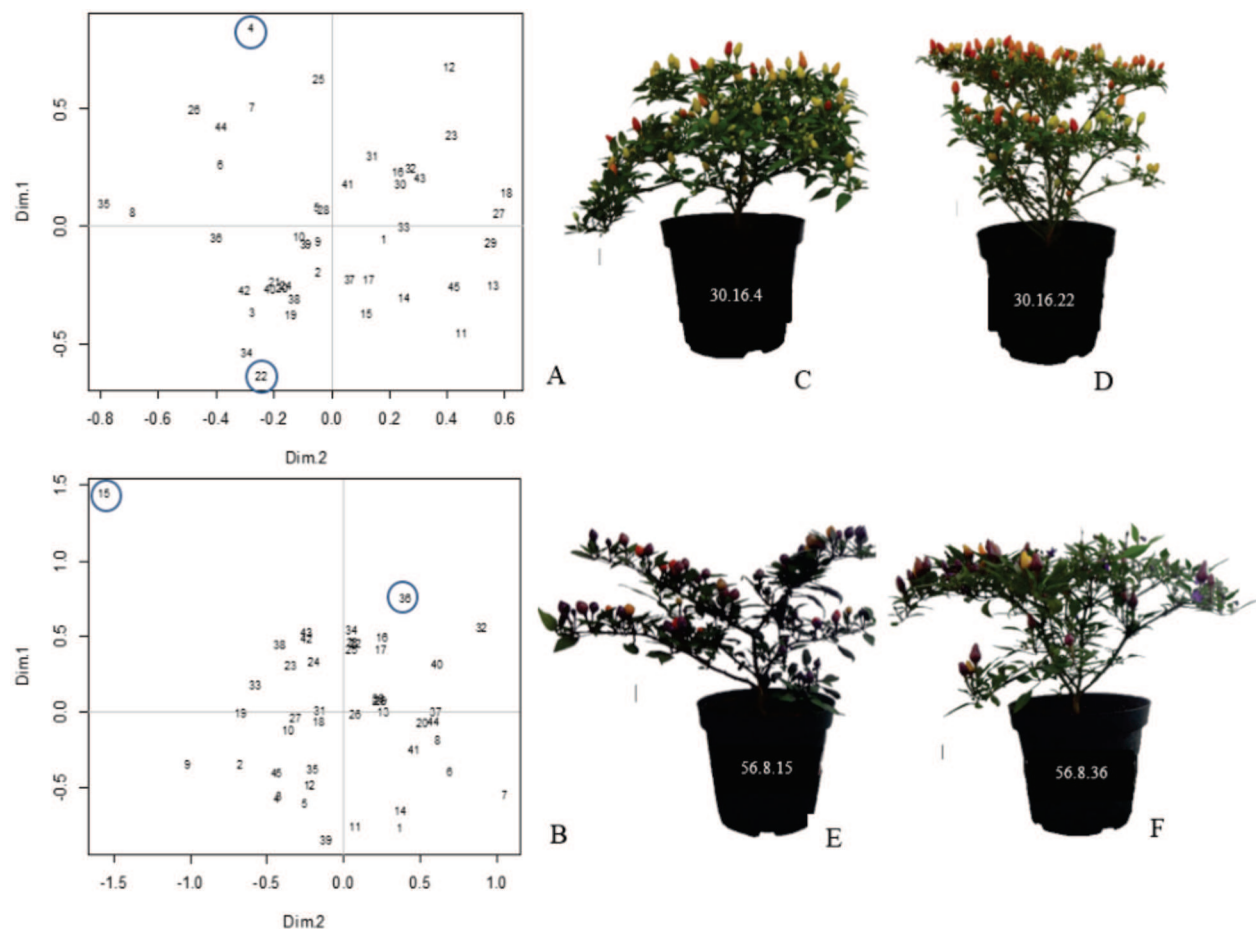
Observing the qualitative data of family 30.16, the more distant genotypes were the 6, 4, 12, 25, 45 and 2 (Figure 2A). These individuals presented differences for the qualitative variables (fruit color in the intermediate and mature stage, fruit persistence with the pedicel, and pedicel persistence with the stem, leaf color, and leaf shape). The genotype with more desirable characteristics for ornamental purposes was genotype 4, presenting fruits with yellow and orange coloration, more persistent fruits with the pedicel, and dark green color of the leaves, with a lanceolate shape (Table 3). The color of fruits and leaves are aesthetical aspects that turn the ornamental pepper plants into good alternatives for the market of in-pot ornamental plants (Stommel & Griesbach, 2008).

Through the multidimensional scaling for the qualitative data of family 56.8, the genotypes that were more disperse were the 9, 15, 36, 7, and 6 (Figure 2B). These plants were distanced for presenting different coloration stages of fruits and leaves, growth habit and leaf shape. Genotype 9 presented an erect growth habit and light green leaves, whereas genotype 15 presented fruits with purple

and brown coloration, and genotype 36 presented fruits with yellow coloration and dark green leaves. Genotype 7 presented green and purple coloration, whereas genotype 6 presented leaves with an oval shape (Table 3). The most interesting genotypes for ornamental purposes were the 15, 36, and 7 (Figure 3E and 3F).

In the joint or mixed data analysis for the family 30.16, the most divergent genotypes were the 4, 12, 11, 22, and 34 (Figure 3A), among which only the 22 and 4 presented desirable characteristics for ornamental purposes (Figure 3C and 3D). It is verified that these genotypes, when the quantitative and qualitative data were analyzed separately, also remained distant and presented important characteristics for in-pot ornamental pepper plants. These quantitative and qualitative characteristics found in these genotypes are important regarding the aesthetical aspect of the plants. In this perspective, there is a demand in Brazil for new pepper cultivars that present features of ornamental interest, destined for in-pot cultivation (Nascimento *et al.*, 2014; Neitzke *et al.*, 2016).

Based on the mixed data of family 56.8, the most distant genotypes were the 15, 36, 32, 7, and 9, and can be



**Figure 3:** Graphic representation of the multidimensional scaling of the families based on mixed data (qualitative and quantitative). Families 30.16 (A) and 56.8 (B), each with 45 *Capsicum annuum* genotypes. Genotypes 30.16.4 (C), 30.16.22 (D), 56.8.15 (E), 56.8.36 (F). Bar corresponding to 2 cm. Areia-PB. CCA-UFPB-2020.

considered the most divergent (Figure 3B). All these genotypes were also distant when separately observing the qualitative and quantitative analyses. Therefore, the selection of genotypes 15 and 36 is recommended, which also stood distant in this analysis. These genotypes, as previously reported, presented desirable characteristics for in-pot ornamental purposes. Furthermore, the fruit coloration of the pepper plants found in the selected genotypes is of extreme importance in the selection of individuals due to the color variation in the maturation stages, making them more attractive and arousing the interest of the consumers for decorative purposes (Silva *et al.*, 2015).

The non-metric multidimensional scaling analysis showed to be efficient to study the distances of the pepper genotypes. Some authors observed the importance of using the mixed variable analysis, employing Gower's distance to analyze the genetic diversity in *Capsicum* (Sudré *et al.*, 2010; Moura *et al.*, 2010; Rocha *et al.*, 2010; Cardoso *et al.*, 2018). It is worth noting that when using the joint analysis since there is an acceptable stress level, it is possible to better discriminate desirable genotypes for selection. Therefore, this technique is a strategy that must be considered for the study of diversity in segregating populations of *Capsicum*, especially when combined with the joint analysis of qualitative and quantitative characters. Therefore, a joint analysis of variables can provide a more complete indicator of the diversity in improvement programs (Gomes *et al.*, 2019).

## CONCLUSION

There is genetic diversity within the analyzed families based on the multidimensional scaling using the distances of the genotypes, allowing the practice of selection.

In the quantitative and mixed variable data, family 30.16 is the most divergent; in the qualitative data, it is family 56.8.

The selection in family 17.18 of genotypes 6 and 32 is recommended, as well as in family 30.16 of genotypes 22 and 4, and family 56.8 of genotypes 15 and 36, since they present important characteristics for ornamental purposes, allowing the continuity of the improvement program for in-pot ornamental pepper plants.

The selection of genotypes is more efficient when using mixed data since it provides a more complete genetic diversity in an improvement program.

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