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Morphological Characterization and Estimates of Genetic Parameters in Peppers With Ornamental Potential

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Abstract

The low number of ornamental pepper cultivars available in the market, combined with a high demand for this ornamental product, has boosted breeding programs for this crop. The objective was to morphologically characterize, estimating the genetic parameters of the main variables of ornamental importance in an F_2 population of pepper (*C. annuum*). The experiments were conducted in a greenhouse at the experimental area of the State University of Montes Claros, Janaúba campus, MG, Brazil. The accessions Uni01 and Uni07 were used as parents to obtain F1 generations and, subsequently, a F_2 population; 333 genotypes from the F_2 generation were evaluated using 19 qualitative and five quantitative descriptors correlated to important characteristics for varietal description and ornamental use. Five fruits per plant were used to evaluate fruit characters: mean fruit weight, mean fruit length, mean fruit diameter, mean peduncle length, and mean pericarp thickness. The genetic parameters of quantitative descriptors were estimated using mean square expected values obtained through Anova. The genetic variability found can be explored for most evaluated characteristics. The quantitative descriptors related to fruit characteristics, based on heritability estimates, can be considered for selection.

Keywords: *Capsicum annuum*, heritability, genetic improvement, ornamental pepper

1. Introduction

Brazil is one of the 15 main producing countries of flowers and ornamental plants. In 2020, the sector was composed of 8,300 national producers, 4,565 of them in the state of São Paulo, denoting the potential growth of the sector throughout the country (IBRAFLOR, 2021). According to Pimenta et al. (2020), researches on peppers (*Capsicum* spp.) for ornamental purposes have been intensified in the last years due to the public interest in these varieties, and their main objectives is the obtaining of new cultivars thar combine ornamental and food purposes.

Peppers present a wide genetic diversity for phenotypic characteristics and large versatility of applications and uses, for example, as an ornamental plant (Rêgo & Rêgo, 2018; Person et al., 2019). This genetic variability of the *Capsicum* genus provides information that can be used for the development of new cultivars through breeding programs, which are based on the variability of the characteristic intended to be improved; thus, studies of genetic divergence are important to identify genotypes with characteristics of interest (Person et al., 2018).

Information on genetic variability and genetic parameters of characteristics of interest enable the selection and thus, proceed with the breeding program. The estimating of genetic parameters of descriptors is an important tool for plant breeders, since it enables the obtaining of information on the variability within a population and the mechanisms of action of genes involved with heritage and establishes the bases for selection of better breeding methods (Cardoso et al., 2018).

The characterization of nine accessions identified with ornamental potential at molecular and morphological levels was carried out in 2018 in the State University of Montes Claros (UNIMONTES); the purpose was to use this information in ornamental pepper breeding programs (Pimenta et al., 2020). The parents Uni01 and Uni07 were among these accessions. Given the above, this study aimed to morphologically characterize, estimating the genetic parameters of the main variables of ornamental importance in an F_2 population of pepper (*C. annuum*). The results aim to subsidize the conduction of the segregating population with a breeding method selected after analysis of the variables.

2. Method

2.1 Description of Experimental Conditions

Three experiments were conducted in a greenhouse covered with a 50% shade screen at the experimental area of the State University of Montes Claros (UNIMONTES), Janaúba campus, MG, Brazil (15º48′09″S, 43º18′32″W, and altitude of 533 m) in 2019 and 2020.

2.2 Installation and Conduction of the Experiment

The experiments were classified as Steps I, II, and III. Step I was carried out in the first semester of 2019; the objective was to obtain seeds of F1 generation. The accessions Uni01 and Uni07 were used as parents; they have contrasting ornamental characteristics of interest (Pimenta et al., 2020). The parental accessions were from the *Capsicum* Germplasm Bank of the UNIMONTES. Seeds of the parents were sown and grown for manual hybridization to obtain the F1 seeds.

Plants of the parent Uni01 were used as recipients (female parents); they were emasculated at the morning and immediately pollinated by introducing pollen grains from the anther of the Uni07 plants (male parent) into their stigmas. Floral buttons in the pre-anthesis were identified for emasculation and the flowers were labeled and covered with aluminum foil to avoid contamination (Nascimento et al., 2012). When the fruits reached the full maturation stage, they were collected and their seeds were used to obtain the F1 generation.

Step II was carried out in the second semester of 2019; it consisted of obtaining the F_2 generation. Floral buttons of F1 plants at the pre-anthesis were protected using paper bags for approximately 5 days to ensure the obtaining of self-fertilized fruits. Then, the self-fertilized fruits were harvested, identified, and taken to the laboratory for processing, drying, and storage of seeds, which were identified as F_2 generation. When the fruits reached the maturation stage, they were collected to obtain the seeds for planting the segregating population (F_2).

Step III consisted in the evaluation of the segregating population; the generations were tested in the first semester of 2020 using the F_2 seeds obtained from the previous step. The sowing was carried out in 128-cell plastic trays filled with a commercial substrate (Biolpant®), using one seed per cell. The trays were maintained in the greenhouse under daily irrigation. When the seedlings reached four to six definitive leaves, they were transplanted to 3-L pots filled with a mixture of clayey soil, coarse sand, and bovine manure (1:1:1 v v^{-1}), constituting the segregating population for evaluation.

A total of 400 plants were sown, obtaining 333 viable plants for evaluation. The plants were subjected to cultural practices throughout the experiment, as recommended for the crop in a conventional system (Filgueira, 2012), making adaptations for protected cultivation in pots.

2.3 Evaluated Descriptors

2.3.1 Evaluation of the Qualitative Descriptors

The genetic variability in the segregating population (F_2) was determined by evaluating qualitative and quantitative characteristics of plants and fruits. Qualitative characteristics included 19 descriptors (anthocyanin pigmentation at the plant node levels, number of flowers per axil, flower peduncle position, corolla color, spot in the corolla, anther color, stigma position, fruit color before maturation, fruit color intensity before maturation and at the maturation stage, position, fruit length and diameter, fruit shape in the longitudinal section, fruit surface texture, fruit color at maturation, brightness and shape of the fruit apex, capsaicin in the fruit placenta), which are recommended for distinguishability, homogeneity, and stability tests for cultivars of the *Capsicum* genus by the National Service for Protection of Cultivars (SNPC) of the Brazilian Ministry of Agriculture, Livestock, and Food Supply (MAPA) (Ministério da Agricultura, 2021).

The presence of capsaicin in the placenta was determined by subjecting part of the placenta to immersion in 3 mL of an ammonium vanadate solution, according to the method adapted by Riva (2006).

Qualitative descriptors were evaluated by side-by-side comparison; the variability was based on direct observations of genotypes through tests in the greenhouse. The characteristics were evaluated visually, using a scale of grades for each descriptor and genotype involved in the assay.

2.3.2 Evaluation of the Quantitative Descriptors

Quantitative descriptors of fruits evaluated were: mean weight (MMF), mean length (CMF), mean diameter (DMF), mean peduncle length (CMP), and mean pericarp thickness (EMP). These evaluations were carried out using a balance and a caliper. These descriptors were evaluated by determining the mean of 5 fruits of each plant; the genotypes were distributed in a completely randomized design, with five fruits representing the replications of each genotype of the F_2 population.

2.4 Statistical Analysis

2.4.1 Statistical Analysis of Qualitative Descriptors

Qualitative descriptors were evaluated through descriptive statistics; the frequency distribution of continuous variables was used to obtain the values fitted into the classes proposed for each descriptor, as described by the SNPC, using the mode as descriptive statistics, which consists in the most frequent value in the data set of the scale of grades attributed to each class.

2.4.2 Statistical Analysis of Quantitative Descriptors

The quantitative descriptors were subjected to the Lilliefors and Bartlett tests ($p < 0.05$) for verification of normality and homogeneity of the data, respectively. When the demands for individual analysis of variance (ANOVA) were confirmed, it was carried out at 5% probability of error. Estimates of variance components and genetic and phenotypic parameters were obtained through mean square expected values. The genetic parameters estimated were: phenotypic variance; genotypic variance; environmental variance, and heritability. All statistical analyses were carried using the Genes program (Cross, 2016).

3. Results and Discussion

3.1 Qualitative Descriptors

The qualitative descriptors evaluated showed no phenotypic variation for spot in the corolla, fruit color before maturation, fruit color intensity before maturation, fruit texture, fruit color intensity at the maturation stage, brightness, and presence of capsaicin. The absence of genetic variability for a descriptor can be positive in a genetic breeding program when the classification is associated to the desirable standard by consumer.

This absence indicates to the program that the gene loci responsible for its expression of the descriptor are or can be fixed. Information of descriptors at the allelic fixation level is an essential subject of study for breeding programs of species of the *Capsicum* genus that assists in decisions regarding the breeding strategy (Pereira-Días et al., 2017).

The absence of spot in the corolla found in the population evaluated is useful for varietal description and, together with the other descriptors, confirms that they are genotypes of the *C. annuum* species. According to Vasconcelos et al. (2012), floral traits like this can be used as morphological markers to determine the genetic divergence between accessions, with the advantage of being evaluated at the beginning of the plant reproduction cycle; in addition, it is attractive characteristic in ornamental peppers, which make them useful for selection procedures. The fruits presented predominantly a greenish-white color before maturation. This result is due to the similar phenotypic characteristic expressed by the parental accessions; the accession Uni01 and Uni07 present greenish-white fruits before the maturation stage (Pimenta et al., 2020).

The fruit surface texture presented no variation within the population, and the surface texture of all individuals were classified as smooth. Several researches on plant breeding found phenotypic divergence between accessions of *Capsicum* spp. based on characterizations through qualitative descriptors, such as fruit texture (Abud et al., 2018). However, in the present work, the absence of variability may indicate a genetic proximity between the parental accessions for this descriptor, which is not so undesirable considering that a smooth fruit surface is commonly preferred by consumers. The evaluations of color intensity in immature and mature fruits, such as brightness, enabled the mean classification for all genotypes evaluated. These results are important for the selection of characteristics of interest, such as fruit color intensity and brightness, for the obtaining of the following populations with no occurrence of segregation.

All genotypes presented capsaicin in the placenta, showing no divergence within the population. Pungency, or spiciness, is the main attribute of peppers, which is connected to the concentration of capsaicinoids (Pinto et al., 2013), which pleases consumers that search for pepper plants with pungent fruits. In this case, this result favors

the development of pepper cultivars that have double purpose (ornamental and food). A high genetic variability was found for anthocyanin pigmentation at the plant node levels (Figure 1A) within the population, with 44% of genotypes presenting high, 22% low, 19% medium, 12% absent or very low, and 3% very high pigmentation. The descriptor number of flowers per axil (Figure 1B) presented low variation within the population; approximately 99% of the plants presented three flowers per axil, whereas 1% of the genotypes presented two flowers per axil within the segregating population.

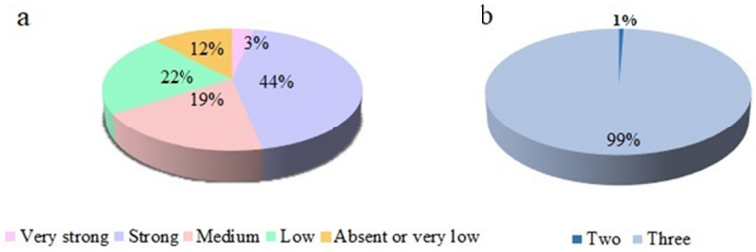

Figure 1. Summary of the percentages found in the F_2 population for qualitative descriptors. a) anthocyanin pigmentation at the plant node levels; and b) number of flowers per axil. Janaúba, MG, Brazil, 2021

Variability is essential in the selection of individuals for the generation advance (Person et al., 2019b), since genetic diversity is an important component for the selection (Anira et al., 2017). In the case of this descriptor, the presence and pigmentation at the internode levels can be explored for ornamental purposes. Ari et al. (2016) reported this descriptor as an ornamental characteristic. According to Silva et al. (2020), this descriptor rises the attention of consumers because its purplish color differs from the green of leaves and the fruit color in pepper plants.

A peculiarity was found for the descriptor number of flowers per axil. Considering that the parents are *C. annuum* L. (Pimenta et al., 2020), the genotypes should have no more than two flowers per node, which is a characteristic of this species. Carvalho et al. (2006) used this characteristic to identify peppers of the *Capsicum* genus and reported that *C. annuum* peppers present one flower per node and *C. baccatum* peppers present two or more flowers per node. The emergence of more than one flower per node is rare for the *C. annuum* species; however, the species has very short internodes and ornamental varieties present low sizes, which can induce errors during the evaluation, causing a perception that there are more than two flowers per node. Thus, the most genetically probable hypothesis is that this character should be evaluated at the fruiting stage and with caution to properly identify whether the peduncle belongs to the same axil.

Considering the 333 genotypes evaluated in the F_2 population, the flower position in the peduncle were 88% erect, 11% intermediate, and 1% hanging (Figure 2A); 331 genotypes presented white corolla (Figure 2B); and the variability of anther color was low (Figure 2C); 89% of the genotypes presented pale blue anthers, whereas the other genotypes presented violet corolla (11%).

Figure 2. Summary of percentages found in the F_2 population for qualitative descriptors. a) position of the flower in the peduncle; b) corolla color; and c) flower anther color. Janaúba, MG, Brazil, 2021

Pepper fruit position in the peduncle recommended for ornamental purposes are usually erect (Melo et al., 2014). The results showed that most genotypes presented this desirable standard for the market. This characteristic is favorable in breeding programs for selections based on several characteristics, enabling more possibilities of combining this descriptor with other characteristics of interest for selection.

According to Pimenta et al. (2020), the accessions Uni01 and Uni07 have white corolla and the corolla color is an ornamental characteristic of interest, since it makes the plant to be more attractive. These accessions were used to obtain the population studied, denoting that the predominance of white corolla in the population is a characteristic inherited from their parents. Taxonomists evaluated mainly the flowers, considering morphological characteristics, such as number of flowers per node, position of flower, and corolla and anther colors, to identify domesticated species of the *Capsicum genus* (Longatti, 2019). The emergence of a purple corolla in plants of this population may indicate that their parents were not homozygous for this characteristic.

Silva (2019) evaluated F6 populations of ornamental peppers and found variation in anther color (purple and blue) within populations. The anther colors found (Figure 2C) enabled the exploration of the characteristic for ornamental purpose, since it contrasts with the flower, fruits, and leaf colors, making the ornamental pepper genotypes more attractive.

The descriptor stigma position (Figure 3A) presented variations within the population, presenting positions within all classes for the descriptor. 98% of individuals presented stigma position above the anther, 1% below, and 1% at the same level. In addition to stigma position, other descriptors are common, such as anthocyanin pigmentation in the hypocotyl and nodes, fruit surface texture, number of loci, and calyx constriction, although they are not exclusive to the *C. annuum* species (Rodrigues et al., 2016).

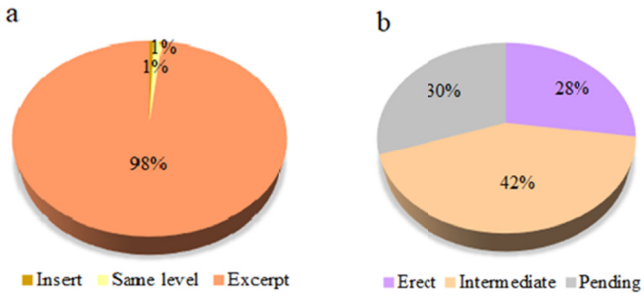

Figure 3. Summary of percentages found in the F_2 population for qualitative descriptors. a) flower stigma position; and b) fruit position. Janaúba, MG, Brazil, 2021

The position of the fruits formed presented variation (Figure 3B). The fruit positions found in the segregating population were: 42% intermediate, 30% hanging position, and 28% erect. Fruit position is other characteristic of this plant that was target for domestication (Pikersgill, 2016).

The fruit diameter (Figure 4A) varied, presenting four different classes, with predominance of the medium diameter class, corresponding to 74% of the genotypes and the very small diameter class (2%). The results for predominant fruit shape in the longitudinal section (Figure 4B) presented high variation, showing horn-shaped (49%); narrow-triangular (25%); triangular (25%), and elliptical (1%) fruits.

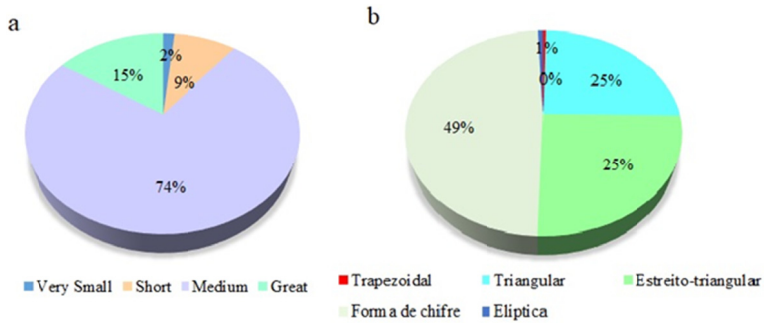

Figure 4. Summary of percentages found in the F_2 population for qualitative descriptors. a) fruit diameter, and b) fruit shape in the longitudinal section. Janaúba, MG, Brazil, 2021

Fortunato et al. (2019) reported that plants that produce fruits with medium diameter are recommended for ornamental use, since they present a balance between plant architecture and fruit size. Fruit diameter is a component that enables indirect selection of more productive genotypes.

The fruit shape in the longitudinal section showed the existence of genetic variability within the population for this essential fruit characteristic associated to fruit shape. Ornamental pepper cultivars can be differentiated from each other by simple observations of this characteristic, which also has esthetic value. Fruit shape is among the most important qualitative descriptors for evaluation of ornamental potential of peppers regarding esthetic aspects, and affects the interest of consumers (Neitzke et al., 2010). De Melo et al. (2014) reported that plants with small fruits present better acceptance in the market of ornamental peppers.

Fruit length presented a high variability (Figure 5A) within the F_2 population; fruits with medium, short, long, very short, and very long lengths were found. Regarding the fruit color at the maturation stage (Figure 5B) of the genotypes evaluated, 95% were red and 5% were orange. The descriptor fruit apex shape (Figure 5C) within the segregating population showed high variability, presenting acute (67%), rounded (37%), depressed shape (3%). Fruit apex shape should be considered, visually, together with fruit shape to establish a favorable esthetic combination, which can be evaluated in breeding programs for ornamental peppers.

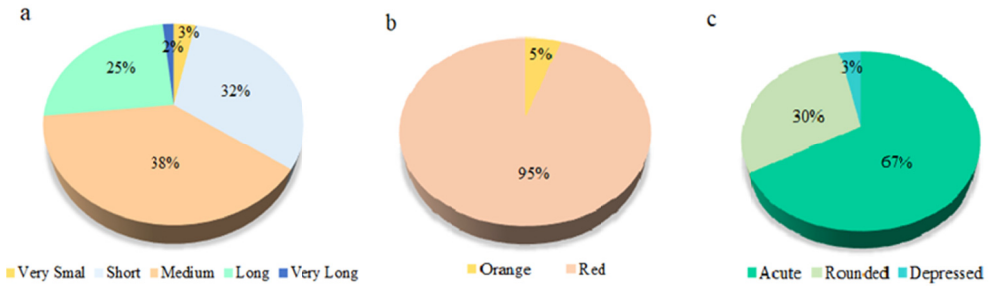

Figure 5. Summary of percentages found in the F_2 population for qualitative descriptors. a) fruit length; b) fruit color at the maturation stage; and c) fruit apex shape. Janaúba, MG, Brazil, 2021

Fruit length is a characteristic that can be explored in breeding programs for ornamental peppers. Long or short fruits can be esthetically explored according to other plant characteristics. Person et al. (2018) studied genetic divergence among pepper genotypes and recommended the selection of genotypes that present short fruits.

Fruit characteristics can increase the interest of consumers. Variations in fruit color at the maturation stage can be explored for ornamental purposes. According to Jang et al. (2020), fruit color is one of the most important aspects for pepper (*C. annuum* L.), since it is the main standard of reference adopted by consumers when selecting and buying peppers.

3.2 Quantitative Descriptors

Descriptors related to fruits were evaluated quantitatively, considering the mean square expected values, to obtain statistical and genetic parameters that can be explored and subsidize breeding programs. According to the experimental precision classification (CV%) proposed by Gomes (1990), the quantitative descriptors mean fruit length, mean fruit diameter, and mean fruit peduncle presented medium experimental precision, which is favorable, since it confirms the results presented in this work (Table 1).

Table 1. Analysis of variance and genetic parameters of quantitative characters related to fruits of ornamental pepper genotypes in the segregating population (F_2). Janaúba, MG, Brazil, 2021

Source of variation	Medium Square					
	GL	MMF	CMF	DMF	CMP	EMP
Population F_2	332	2.37**	5.76**	0.09**	0.57**	0.004**
Residue	1200	0.42	0.57	0.01	0.10	0.001
Overall average		1.93	4.42	0.79	2.47	0.12
Coefficient of variation (%)		33.61	17.12	16.65	12.97	28.91
Phenotypic variance (V_f)		0.51	1.25	0.019	0.12	0.0008
Environmental variance (V_a)		0.092	0.12	0.003	0.02	0.0002
Genotypic variance (V_g)		0.42	1.13	0.016	0.10	0.0005
Heritability (%)		82.12	90.08	80.78	81.81	67.99

Note. **Significant difference at 1% probability of error.

The characteristics mean fruit weight and mean pericarp thickness presented low experimental precisions. In these cases, the CV% can be explained by the fact that they are characters expressed from many genes, which are highly affected by the environment. This effect can be reduced by increasing the number of replications and choosing more rigorous designs for the experiments (Costa et al., 2020).

Highly significant differences were found for the quantitative characters evaluated, indicating presence of genetic variability. Genetic variability is essential for the establishment of breeding programs; however, and efficient selection of superior genotypes is dependent on genetic and environmental parameters correlated to the characteristics of interest (Blind. 2018). The genotypic variance was higher than the environmental variance in all characters evaluated (Table 1), indicating that the phenotypic expressions observed have a genetic origin, they were from genes that are involved in the expression of the characters and can be transmitted to the next generations.

This relationship is essential for decision making about which descriptors could be useful for selection. Therefore, important characteristics for the plants that present high heritability should be emphasized, especially in initial segregating generations, such as the F_2 population evaluated in the present study. A general analysis showed a relatively high coefficient of heritability for the fruit characters evaluated (Table 1). High heritability values denote that the variations observed for the characteristics of study are more connected to genetic than to environmental variation, enabling genetic gains through selection (Silva Neto et al., 2014).

The mean fruit length was the characteristic that presented the highest heritability (90.08%). Mean fruit weight, mean peduncle length, and mean fruit diameter presented heritability of 82.12%, 81.81%, and 80.78% respectively. High heritability results were also found for quantitative characteristics of pepper plants by Silva Neto (2014), Guedes (2018), and Silva (2019).

Considering of results found in the present study and the interest of the market of ornamental peppers, the selection of individuals based on the evaluated characters is possible. These characteristics are part of the preferences considered by consumers when purchasing ornamental pepper plants, mainly variables related to fruit size, such as length and diameter.

4. Conclusion

Genetic variability was found for most descriptors evaluated in the F_2 population from the crossing Uni01 × Uni07. The genetic parameters estimated for fruit characteristics denoted a high genetic contribution to the phenotypic expression. These results allow the selection of genotypes in this population.

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