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Evaluation of ornamental hybrids of *Capsicum annuum* and their reactions to the fungus *Colletotrichum scovillei*

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Capsicum peppers have long attracted the interest of the ornamental plant market. The demand for plants of this genus in this sector is mainly due to the diversity of shapes and colors of fruits, flowers, and leaves, as well as their compact architecture, which brings charm and harmony to the environment. In 2018, global pepper cultivation reached approximately 40.9 million tons on 3.8 million hectares. In Brazil, data related to pepper cultivation and production are scarce, especially regarding ornamental peppers. However, plants of this genus are frequently affected by pathogens that cause diseases leading to the depreciation of the plant and its fruits, such as anthracnose, caused by the *Colletotrichum* fungal complex. Therefore, there is a need to seek pepper cultivars, especially ornamental ones, that are resistant to these diseases. The objective of this study was to obtain, evaluate, and characterize pepper hybrids from the germplasm bank of the Federal University of Espírito Santo, regarding their ornamental potential and resistance to anthracnose. The hybrids were obtained from seven pepper parentals, previously selected in other studies for their ornamental potential and resistance to anthracnose. The experiment, aimed at evaluating and characterizing the ornamental potential of the hybrids, was conducted in a greenhouse using a completely randomized design with 10 replicates. Seven parentals, seven hybrids, an ornamental control (the cultivars 'Pirâmide Ornamental' and 'Espaguetinho Ornamental'), the cultivar 'Ikeda' (as a positive control for anthracnose), and the UENF1381 accession (as a negative control for the disease) were used, totaling 96 plants. This study identified the hybrids 15x89, 23x40, and 89x15 as having ornamental value and resistance to *Colletotrichum scovillei*, as well as the parents UFES 15 and UFES 23. Compact growth habit was consistently associated with lower anthracnose severity across

evaluation periods, as evidenced by negative correlations between plant height and disease progress in immature and mature fruits. Therefore, the hybrids 15×89, 23×40, and 89×15 are good candidates for the ornamental market, and the parents UFES 15 and UFES 23 can be further explored in future breeding studies focused on ornamental potential.

KEYWORDS

anthracnose, disease resistance, floriculture, ornamental plants, plant breeding

1 Introduction

Plants of the genus *Capsicum* L. (Solanaceae) encompass about 43 species worldwide, being Brazil considered an important center of diversity for this genus. Of the 43 species, only five are domesticated and cultivated: *Capsicum annuum* L.; *Capsicum baccatum* (Wild) Eshbaugh; *Capsicum chinense* Jacq.; *Capsicum frutescens* L.; and *Capsicum pubescens* Ruiz & Pavon. The species *C. pubescens*, which requires low temperatures for its development, has minimal cultivation in Brazil but is well represented in Bolivia and Peru (Ribeiro et al., 2020; Barboza et al., 2022).

As one of the most widely consumed spices, pepper production continues to grow worldwide. Data from 2018 estimate that 40.9 million tons were cultivated globally over an area of 3.8 million hectares (Faostat, 2021). While specific numbers for *Capsicum* production in Brazil are scarce, it is estimated that around 280,000 tons are produced annually, covering a cultivation area of 13,000 hectares (Pinto and Donzele, 2020). In addition to the food industry, peppers are also used in the pharmaceutical and cosmetic industries and are increasingly present in the ornamental plant sector (Zhang et al., 2021; Cunha et al., 2020).

In 2017, the post-gate commerce and service segment for flowers and ornamental plants in Brazil generated a Gross Domestic Product (GDP) of 2.19 billion reais. Of this amount, 178 million were related to wholesale trade, 321 million to supermarkets and hypermarkets, 624 million to florists, 728 million to decoration services, and 335 million to landscaping and gardening services. These figures highlight the significant added value of these products, which undergo considerable appreciation from the time they leave rural properties until they reach the final consumer (Cepea and Ibraflor, 2022).

The growing production and sale of peppers in the ornamental market are mainly due to the diversity of fruit, flower, and leaf shapes and colors, their compact architecture, and the ease of fruit harvesting (Rêgo and Rêgo, 2018). However, like most Solanaceae, plants of the *Capsicum* genus suffer from diseases, which complicates their production and affects the visual quality of the plants, the most appealing factor for consumers in the ornamental sector. Among the main diseases affecting peppers is anthracnose, caused by a complex of fungi from the genus *Colletotrichum* (Sharma et al., 2022; Mohammadbagheri et al., 2021). In many cases, this disease is only controlled with chemical products, which are often applied indiscriminately.

With the advancement of science and the understanding of the harmful effects of chemical products when misused, there is a

growing demand on finding management techniques that are less harmful to the environment and humans, especially when it comes to ornamental plants, which consumers bring into their homes, coming into direct contact with the residents. As the ornamental market grows, so does the demand for more conscious and sustainable control of diseases caused by phytopathogens. Therefore, it becomes essential to seek plants with ornamental potential that are also resistant or at least tolerant to diseases like anthracnose (Baba et al., 2019; Costa et al., 2021).

One way to achieve ornamental characteristics required by the market while also ensuring disease resistance is through plant breeding (Cunha et al., 2020; Ro et al., 2021; Cui et al., 2020). However, existing research on ornamental plants rarely systematically evaluates their ornamental potential and resistance to diseases such as anthracnose. Therefore, this study aims to fill this gap, initiating a breeding program with the goal of eventually releasing a new ornamental pepper cultivar resistant to anthracnose developed in Brazil. It is worth noting that most ornamental pepper cultivars sold in Brazil were not developed in the country, leading to a lack of knowledge and exploitation of our existing pepper genetic resources.

2 Materials and methods

2.1 Plant material

In this study, 14 *C. annuum* genotypes from the *Capsicum* germplasm bank of the Federal University of Espírito Santo (UFES) were used, consisting of seven parental lines (UFES 15, UFES 23, UFES 29, UFES 40, UFES 56, UFES 87, and UFES 89) and seven hybrids (15×89, 23×40, 23×56, 29×40, 87×40, 89×15, and 89×40). The parents were selected from a characterization and evaluation study for ornamental purposes and resistance to anthracnose in preliminary studies. In addition to the 14 genotypes, two ornamental controls were also used, the cultivars 'Pirâmide Ornamental' and 'Espagueteinho Ornamental', along with the cultivar 'Ikeda' as a positive control for anthracnose, and the genotype UENF1381 as a negative control.

2.2 Hybrid development/production

To obtain the hybrids, crosses were made early in the morning and late in the afternoon, when temperatures were milder. Using tweezers, the flower buds of the female parents were emasculated

and marked with colored wool. For pollen extraction from the male parent, flowers were collected and exposed to incandescent lamps to stimulate the opening of the anthers. The pollen was then removed, placed in a gelatin capsule, and stored in a refrigerator at 5 °C.

After pollen collection, manual pollination was performed by removing the petals and anthers from the female flower. Using a soft-bristled brush, the stored pollen was transferred from the capsules to the stigma of the female flower. The manually pollinated flowers were covered with paper bags. Once fruit set, the bags were removed to allow the fruits to develop normally until maturity, when they were harvested, and the seeds were extracted and left to dry at room temperature for 48 hours.

2.3 Environmental conditions and experimental design

The trials for obtaining and evaluating the hybrids were conducted in a greenhouse at the Federal University of Espírito Santo (UFES) - Campus Alegre (20°45'48" S, 41°32'2" W, 272 m above sea level). Morphological and agronomic evaluations, focusing on ornamental characteristics and resistance to *Colletotrichum scovillei*, were carried out in the Plant Breeding and Disease Resistance Laboratory (LAMERP) at UFES.

The genotypes were sown in polystyrene trays with 128 cells, containing commercial vegetable substrate from the Provaso® brand. After the emergence of two to four pairs of true leaves, the seedlings were transplanted into 2 L plastic pots filled with a mixture of soil, sand, and bovine manure in a 1:1:1 ratio. The experimental design used was completely randomized blocks with

10 replications, totaling 96 plants. This stage of the experiment was conducted in a greenhouse, where the temperature ranged from 25°C to 30°C and the relative humidity was around 80%.

2.4 Morphological and agronomic evaluation

For the morphological and agronomic evaluations, descriptors proposed by the National Plant Variety Protection Service (SNPC) and the International Plant Genetic Resources Institute, now Bioversity International (IPGRI et al., 1995), were adopted. In addition to the parentals and hybrids, the cultivars 'Pirâmide Ornamental' and 'Espaguetinho Ornamental' from ISLA Sementes Ltda were used as controls for ornamental characteristics. A total of 25 traits were evaluated, comprising 13 quantitative and 12 qualitative characteristics (Table 1).

The evaluated quantitative traits were plant height – ALT (cm); canopy diameter – DCO (cm); stem diameter – DCA (mm); stem length – CH (cm); leaf length – CFO (mm); and leaf width – LF (mm). Measurements taken in centimeters were performed using a tape measure, while those in millimeters were measured with calipers. Regarding the fruits, the following traits were assessed: average fruit weight – MMF (g), calculated as the average weight of fruits from two harvests; total number of fruits – NF, counted as the sum of fruits obtained from two harvests; fruit diameter – DIF (mm); fruit length – CF (mm); pericarp thickness – ESP (mm); number of seeds – NS; and weight of 1000 seeds – MMS. The traits DIF, CF, and ESP were evaluated using the averages of five mature fruits from each plant. For NS, the average number of seeds from

TABLE 1 Qualitative descriptors evaluated in the parentals, hybrids, and ornamental controls of *Capsicum annum*, proposed by the National Plant Variety Protection Service (SNPC, 2006) and the International Plant Genetic Resources Institute, now Bioversity International (IPGRI et al., 1995).

Descriptors	Classes	Evaluation period
Flower		
Number of flowers per axil	1-one; 2-two; 3-three or more	When the flowers were fully open during the early bloomings
Position	1-erect; 3-intermediate; 5-pendant	–
Corolla color	1-white; 2-white with purple base; 3-white with purple margin; 4-greenish white; 5-greenish white with purple margin; 6-light yellow; 7-yellow; 8-yellow-green; 9-5 purple with white base; 10-purple	–
Corolla spot color	1-absent; 2-present	–
Color of the spot	1-white; 2-yellow; 3-yellow-green; 4-green; 5-purple	–
Fruit		
Position	3-erect; 5- intermediate; 7- pendant	When at least 50% of the fruits were mature.
Shape	1-elongate; 2-round; 3-triangular; 4-campanulate; 5-blocky; 6-other	When the fruits were mature.
Predominant number of locules	1-two; 2-two to three; 3-three to four; 4-four or more	–
Color intensity at mature stage	3-light; 5-medium; 7-dark	–
Color at mature stage	1-yellow; 2-orange; 3-red; 4-brown; 5-purple	–
Number of maturing stages	2-two; 3-three; 4-four; 5-five or more	Daily observation from the initial color of the fruit.
Color before mature stage	1-greenish white; 2-yellow; 3-green; 4-purple	–

five fruits was quantified. The MMS was determined by weighing the total seeds extracted from the five fruits.

2.5 Evaluation of resistance to *Colletotrichum scovillei*

In this experiment, the seven parental lines, seven hybrids, ornamental controls, and the cultivar 'Ikeda' (from Feltrin Sementes as a positive control) and the UENF 1381 accession (as a negative control) were utilized.

To obtain the conidial suspension, the isolate of *Colletotrichum scovillei*, collected from pepper fruit in the local market of Alegre, ES, was grown on potato dextrose agar (PDA) in Petri dishes. The plates were maintained in a humid chamber (~90% RH) at a temperature of 28°C for seven days. Following this, the suspension was prepared in autoclaved distilled water, and the conidia were counted using a Neubauer chamber, adjusting the concentration to 1.0×10^6 conidia.mL⁻¹.

For inoculation, the fruits collected from the greenhouse were disinfected using 70% alcohol and 0.1% sodium hypochlorite. Afterward, they were triple washed in autoclaved distilled water, dried, and placed in acrylic boxes (gearbox type). Injuries were made in the central region of the fruits using an insulin needle, and subsequently, 10 µL of the conidial suspension was deposited on the wounds. After inoculation, the fruits were kept in a humid chamber at room temperature (approximately 28°C).

For resistance evaluations, six mature and six immature fruits were randomly inoculated from each plant, totalizing 120 fruits per genotype. Evaluating the fruits at different maturation stages is important because the genetic control of resistance is governed by distinct genes in immature and mature fruits. Assessments were made every 24 hours for ten days, following the methodology proposed by Montri et al. (2009) (Table 2).

The morphoagronomic and resistance evaluations were conducted in two periods of the year. The first sowing occurred in January 2022, with evaluations extending until June of the same

year. The second evaluation began in September 2022 and concluded in February 2023. The average temperatures for the two evaluation periods were 24.08 °C for the first period and 24.12 °C for the second period.

2.6 Statistical analysis

The qualitative traits were evaluated using descriptive statistics, specifically the mode. The data for the quantitative traits were subjected to the Shapiro-Wilk test for normality of residuals at a significance level of 5%. To assess the significance of the effects of seasons and genotypes, as well as their interactions, the quantitative data were analyzed using an F-test (ANOVA) at the 5% and 1% probability levels, according to the following model:

$$y_{ijk} = b/S_{k(i)} + S_i + G_j + GS_{ij} + \epsilon_{ijk}$$

Where y_{ijk} is the observation in the ijk plot; $b/S_{k(i)}$ is the fixed effect of the k -th block within the i -th Season; S_i is the fixed effect of the i -th Season; G_j is the random effect of the j -th Genotype; GS_{ij} is the random effect of the interaction between the j -th Genotype and the i -th Season, and; ϵ_{ijk} is the random effect ($\sim N, 0, \sigma^2$) of the error associated with the ijk observation. The Pearson linear correlation coefficient was estimated for each pair of traits in each planting period.

From the means of each genotype, the eigenvalues for the principal components analysis (PCA) were estimated for each evaluation period. To obtain the eigenvalues, the means were standardized in standard deviation units for each trait. The first two principal components were plotted to observe the degree of association between the traits, as well as the dispersion of the genotypes.

Additionally, the Euclidean distance matrix between the genotypes was estimated for each planting period. Based on this distance matrix, the genotypes were grouped into dissimilarity dendrograms using the Unweighted Pair Group Method with Arithmetic Mean (UPGMA). The analyses were conducted using the statistical software RStudio (R Core Team, 2025), utilizing functions from the 'ggplot2', version 3.4.0 'corrplot', 'factoextra', and 'ecodist' packages.

2.7 Obtaining the area under the disease progress curve

From the ratings obtained from the resistance assessments to *Colletotrichum scovillei* during the two periods, the Area Under the Disease Progress Curve (AUDPC) was estimated using the following equation:

$$AUDPC = \sum_{i=1}^n [(Y_{i+1} + Y_i)/2] \times [(T_{i+1} - T_i)]$$

Where, n is total number of observations; Y_i is disease rating at time T_i ; and T_i is time (days) at the i^{th} observation.

After calculating the AUDPC for both green and mature fruits, the resulting values were incorporated into the analyses as CLFV (anthracnose severity in green fruits) and CLFM (anthracnose severity in mature fruits), which were used in all subsequent

TABLE 2 Rating scale for evaluating resistance to *Colletotrichum scovillei* in *Capsicum annuum* fruits (Montri et al., 2009).

Grade	Resistance level	Symptom description
0	Highly Resistant	No symptoms
1	Resistant	1-2% of the fruit area with necrotic lesion or water-soaked lesion around the infection site.
3	Moderately Resistant	>2-5% of the fruit area with necrotic lesion, may or may not have sporulation, or water-soaked lesion exceeding 5% of the fruit surface.
5	Moderately Susceptible	>5-15% of the fruit area with necrotic lesion, presence of sporulation, or water-soaked lesion exceeding 25% of the fruit.
7	Susceptible	>15-25% of the fruit area with necrotic lesion and sporulation
9	Highly Susceptible	>25% of the fruit area with necrotic lesion, lesion surrounding the fruit with abundant sporulation.

evaluations. The F-test (ANOVA) was applied to the AUDPC values ($p < 0.05$).

3 Results and discussion

The comprehensive characterization of *Capsicum* germplasm plays a fundamental role in the conservation and strategic use of genetic resources, particularly in a megadiverse country such as Brazil, which harbors extensive but still underexplored phenotypic and genotypic variability (Cardoso et al., 2018; Constantino et al., 2020). The limited availability of systematic data on plant architecture, ornamental traits, fruit attributes, and disease responses restricts the accurate documentation of this diversity, thereby constraining its effective use in breeding programs (Moo et al., 2023). The lack of consistent characterization efforts may contribute to silent genetic erosion, as poorly described accessions tend to receive less attention in conservation initiatives, ultimately placing unique or locally adapted genotypes at risk.

This situation becomes even more critical under the ongoing pressures of climate change, which have intensified the incidence and severity of biotic stresses, particularly fungal diseases (Seidel et al., 2024) (such as *Colletotrichum scovillei*), further accelerating the loss of genetic variability in underutilized crops. In this context, the integrated evaluation of agronomic, ornamental, and disease-

related traits is essential to identify genotypes capable of functioning as a stable genetic reservoir for present and future demands. A well-characterized germplasm collection enables breeding programs to more efficiently identify and introgress resistance alleles into elite lines or new hybrid combinations, ensuring the maintenance and expansion of a resilient genetic base.

Although a few studies (Nankar et al., 2020; Todorova et al., 2024) have attempted to integrate morphological, ornamental, and disease-resistance assessments in *Capsicum* germplasm, they remain scarce and often limited in scope, frequently exploring these dimensions in isolation – either focusing primarily on pathogenic resistance (e.g. Silvar and García-González, 2017; Mohammadbagheri et al., 2021; Ro et al., 2021) or exclusively on ornamental attributes (e.g. Virga et al., 2020; Brilhante et al., 2021; Ramalho doRêgo et al., 2022) – without providing a unified framework capable of supporting both conservation and breeding applications. Therefore, there is a clear need for more comprehensive investigations such as the present work, which evaluates morpho-agronomic, qualitative, quantitative, and anthracnose-resistance traits, while also estimating the diversity among accessions to define the underlying genetic variability that will guide the strategic use of these genotypes in breeding programs.

Before presenting the qualitative and quantitative results, Figure 1 provides an overview of the phenotypic diversity observed among the accessions, illustrating the morphological contrasts that guided the subsequent analyses.



FIGURE 1

Overall phenotypic diversity of the genotypes evaluated in a greenhouse experiment at the Universidade Federal do Espírito Santo (UFES), Alegre. (A–G) Parental lines: UFES 15, UFES 23, UFES 29, UFES 40, UFES 56, UFES 87, and UFES 89. (H–N) Hybrids: 15x89, 23x40, 23x56, 29x40, 87x40, 89x15, and 89x40. (O, P) Ornamental controls: 'Espaguetinho Ornamental' and 'Pirâmide Ornamental'.

3.1 Qualitative traits

No differences were found in the evaluations of qualitative traits conducted during summer and winter, which resulted in the same outcomes being found in both seasons. This stability is expected for traits predominantly controlled by one or a few genes, where environmental interference is typically low or negligible (Sahid et al., 2022). However, additional mechanisms may also explain this consistency. Our hypothesis is that these traits may be buffered by developmental canalization, which minimizes phenotypic variation even under fluctuating environments (reviewed by Takahashi, 2019). It is also possible that the environmental contrast between the two evaluation periods, although present, was not strong enough to induce detectable plastic responses in traits with simple inheritance. Alternatively, the genotypes evaluated may share common alleles in the major loci controlling these descriptors, thereby reducing the potential for phenotypic divergence across environments.

For the 12 qualitative descriptors evaluated, four were monomorphic, meaning there was no discrimination among the genotypes studied based on these traits. These traits were: flower and fruit position, fruit color at the mature stage, and number of locules. The flower and fruit positions were erect, and all plants had red fruits in the mature stage, with two locules. Knowledge of the flower position allows for the prediction of the fruit position, as many genotypes exhibit the same positions for both flower and fruit (Sahid et al., 2022). Moreover, information about this trait ensures greater success for *Capsicum* spp. producers in the hybridization process and in cross-contamination calculations (Lin et al., 2020).

Most genotypes presented only one flower per axil, except for genotype UFES 89 and hybrid 89×40, which presented three or more. This unusual pattern raises the hypothesis that alleles controlling multi-flowering may be recessive or conditionally expressed, emerging only in specific parental combinations. It

may also indicate a heterotic response for floral induction or even a physiological trade-off, in which the production of multiple flowers per axil could influence resource allocation and potentially affect fruit development – an aspect that merits investigation in future studies. The corolla color varied between white (UFES 40, UFES 87, and UFES 89), white with violet margins (UFES 29, 15×89, 23×40, and 89×15), violet with a white base (UFES 23 and 29×40), and violet (UFES 15, UFES 56, and 23×56). Progenitor UFES 15 and hybrid 23×56 exhibited spots on the corolla, which was white in both genotypes (Figure 2). It is interesting to note that in hybrid combinations where crossings occurred between genotypes with violet flowers and genotypes with white flowers, both colors were present, as can be seen in hybrids 15×89, 23×40, 29×40, and 89×15. According to Nascimento et al. (2013) this color combination makes ornamental peppers even more attractive, as flower color – alongside fruit traits – is an important characteristic for ornamental purposes and also plays a relevant role in taxonomic identification.

In terms of the number of maturation stages, genotype 29×40 presented the highest number, with five maturation stages. The genotypes UFES 29, UFES 40, UFES 56, UFES 87, UFES 89, 23×56, 87×40, and 89×40 had three stages, while genotypes UFES 15, UFES 23, 15×89, 23×40, 87×40, and 89×15 presented four maturation stages.

For the trait of fruit color before mature stage, there was variation between green, orange, and purple. Only UFES 56 presented a green color before maturity; genotypes UFES 15, UFES 23, UFES 40, UFES 89, 15×89, 23×40, 29×40, 89×15, 89×40, and the two ornamental controls had an orange color, while UFES 29, UFES 87, 23×56, and 87×40 were purple (Figure 3). One of the most important qualitative traits linked to the ornamental market in *Capsicum* is related to the change in fruit color at immature and mature stages. The color variation found at different maturation stages attracts consumers, as it allows for more

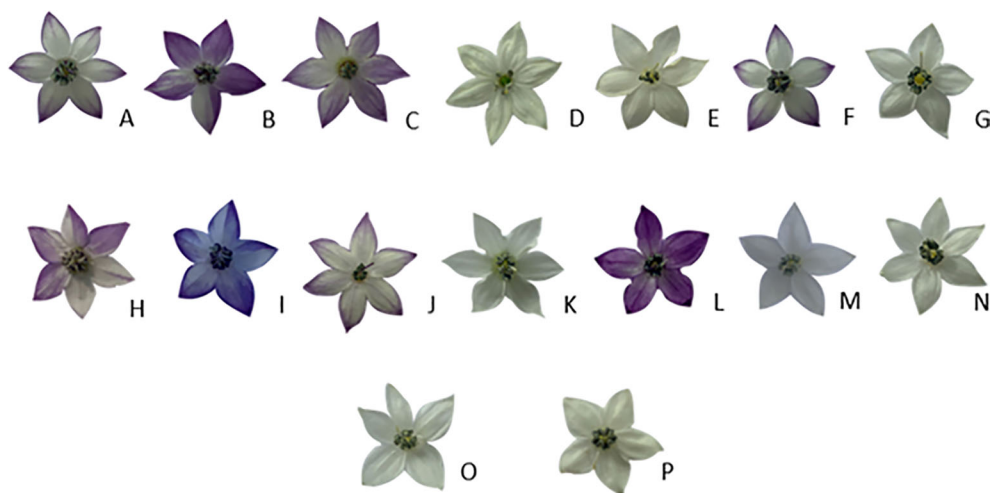


FIGURE 2

Floral variation of the sixteen genotypes grown exclusively in a greenhouse experiment at UFES, Alegre. The panel highlights differences in corolla pigmentation, margin coloration, and presence of corolla spots among parental lines, hybrids, and ornamental controls. All genotypes depicted were assessed for their morphological and ornamental attributes alongside disease response to *Colletotrichum scovillei*. (A–G) 15×89, 23×40, 23×56, 29×40, 87×40, 89×15, and 89×40; (H–N) UFES 15, UFES 23, UFES 29, UFES 40, UFES 56, UFES 87, and UFES 89; and (O, P) 'Espagueteinho Ornamental' and 'Pirâmide Ornamental'.

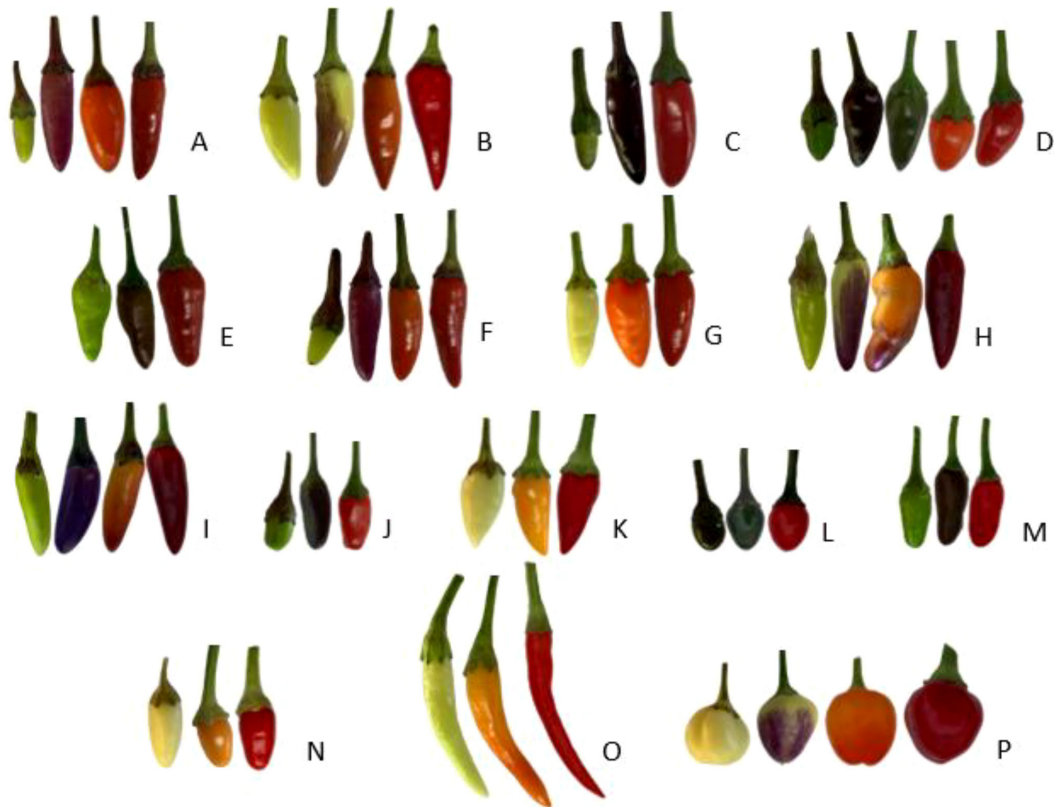


FIGURE 3

Fruit diversity of the sixteen genotypes cultivated in greenhouse conditions at UFES, Alegre. The panel illustrates variation in fruit maturation stages, color transitions, color intensity, and fruit shape. All genotypes shown were simultaneously evaluated for fruit-related traits and for anthracnose severity in immature and mature fruits following inoculation with *Colletotrichum scovillei*. (A–G) 15×89, 23×40, 23×56, 29×40, 87×40, 89×15, and 89×40; (H–N) UFES 15, UFES 23, UFES 29, UFES 40, UFES 56, UFES 87, and UFES 89; and (O, P) ‘Espagueteinho Ornamental’ and ‘Pirâmide Ornamental’.

than one fruit color to be present on the same plant (Silva et al., 2016), adding value to the product.

The intensity of ripe fruit coloration in the genotypes UFES 29, UFES 40, UFES 56, UFES 87, UFES 89, 23×40, 29×40, 87×40, 89×40, and ‘Espagueteinho Ornamental’ was light, while in the genotypes UFES 15, UFES 23, 15×89, 23×56, 89×15, and ‘Pirâmide Ornamental’ it was medium (Figure 3). Regarding fruit shape, there was a predominance of the triangular shape (Figure 3). The difference was found only in genotype UFES 56, which had a round shape, and in the controls ‘Espagueteinho Ornamental’ and ‘Pirâmide Ornamental’, which presented elongated and campanulate fruits, respectively (Figure 3). Fruit shape is an important trait for the ornamental sector due to the contrast it can promote in the plant, expressing softness or aggressiveness in the ornamental arrangement (Melo et al., 2014).

3.2 Quantitative traits

The analysis of variance showed that genotype effects were significant for all quantitative traits evaluated, including the area under the disease progress curve (AUDPC) for disease severity in both green and ripe fruits (Table 3). In contrast, the season effect was significant for only seven of the 15 traits: 100-fruit mass (MMF), number of fruits per node (NF), fruit diameter (DIF), fruit length (CF), 100-seed mass (MMS), and those related to

disease severity (CLFV and CLFM). Thus, environmental variation among seasons had a stronger influence on these traits. Genotype × season interaction effects were significant only for MMF, NF, CF, MMS, and CLFM. Nevertheless, the presence of significant interactions highlights the importance of considering multiple seasons to ensure more secure selection of plants exhibiting the desired phenotypic traits.

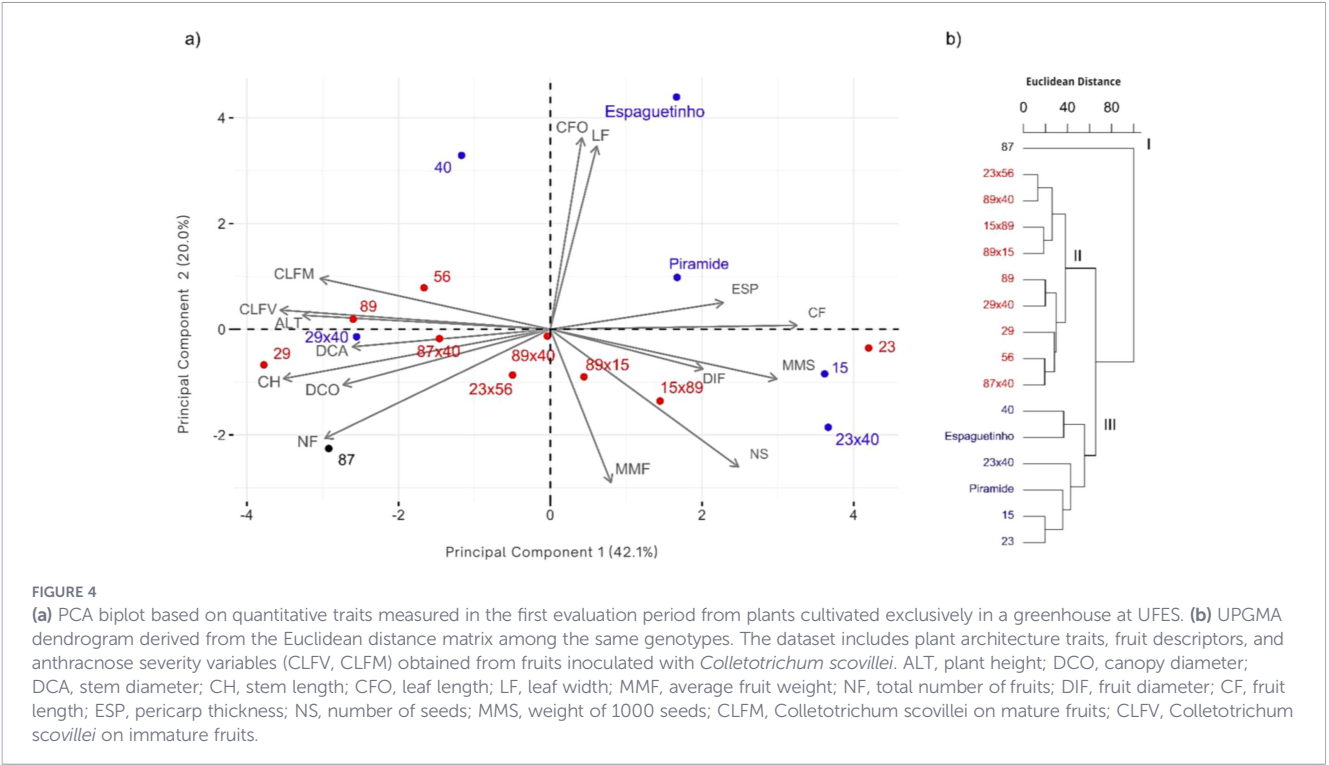
When analyzing the degree of association between the quantitative traits and the genotype dispersion based on principal components analysis (PCA) (Figures 4, 5), it was observed that components 1 (PC1) and 2 (PC2), evaluated during the first period of the experiment, explained 62.1% of the total variation in the quantitative traits assessed, contributing 42.1% to the first axis and 20% to the second axis (Figure 4a). For the data collected during the second evaluation period, PC1 and PC2 explained 54.5% of the variation, contributing 37.6% to the first axis and 16.9% to the second axis (Figure 5a). In PCA-based biplots, it is generally desirable that the first two components together explain 60% of the total variation, as this threshold provides a more reliable projection of the multidimensional structure onto two dimensions. Accordingly, the first evaluation period meets this criterion, allowing greater confidence in the visual representation of trait–genotype relationships. In contrast, the slightly lower cumulative variance in the second period suggests a more complex or diffuse contribution of the quantitative traits to overall variability, although the PCA still captures relevant patterns for interpretation.

TABLE 3 Analyze of variance (F – test) to quantitative traits evaluated in 16 *Capsicum annuum* genotypes in two seasons (winter and summer) in southeast of Brazil.

Trait	Mean square					Mean
	Block/S	Season (S)	Genotype (G)	G x S	Error	
MMF	35.33	157198.21 **	1290.43 **	1276.31 **	24.13	23.17 g
NF	518.26	776885.84 **	39875.12**	4058.67 **	469.38	110.70
DIF	0.73	6.15 **	94.54 **	0.34 ^{ns}	0.26	9.34 mm
CF	3.32	7.15 *	1402.26 **	2.83 *	1.51	23.76 mm
ALT	1.61	2.53 ^{ns}	419.54 **	2.82 ^{ns}	3.11	21.36 cm
DCO	4.65	4.31 ^{ns}	318.43 **	8.93 ^{ns}	5.62	28.05 cm
DCA	0.23	0.02 ^{ns}	6.78 **	0.22 ^{ns}	0.21	4.50 mm
CH	1.10	0.59 ^{ns}	94.54 **	2.30 ^{ns}	1.39	10.12 cm
NS	14.23	241.5 ^{ns}	2068.34 **	66.53 ^{ns}	78.5	31.97
MMS	0.02	0.25**	1.32**	0.06**	0.01	0.70 g
ESP	0.01	0.01 ^{ns}	1.48 **	0.01 ^{ns}	0.01	1.07 mm
CFO	11.04	31.03 ^{ns}	663.45**	32.33 ^{ns}	20.72	46.72 mm
LF	0.66	5.92 ^{ns}	185.93**	2.31 ^{ns}	2.98	20.8 mm
CLFV	121.03	1707.63**	1419.82**	480.43 ^{ns}	285.32	27.86
CLFM	418.38	18453.79**	1509.95**	418.15**	143.22	31.37

**, * indicate significant effects according the F-test at 1% and 5% of probability, respectively (p< 0.01 and p< 0.05). ^{ns} indicate non-significant effects. MMF, average fruit weight; NF, total number of fruits; DIF, fruit diameter; CF, fruit length; ALT, plant height; DCO, canopy diameter; DCA, stem diameter; CH, stem length; NS, number of seeds; MMS, weight of 1000 seeds; ESP, pericarp thickness; CFO, leaf length; LF, leaf width; CLFV, Colletotrichum scovillei on immature fruits; CLFM, Colletotrichum scovillei on mature fruits; Trat, Treatment; Block/S, Blocks; Season (S), Seasons; Genotype(G), Genotypes; GxS, Genotype x Season Interaction; Error, Statistical Error; Mean, Statistical Significance of the F-test.

Three groups were generated in the dendrogram formed from the estimate of the Euclidean distance matrix between the genotypes, in both the first and second evaluation periods. Even if subtle, it is possible to observe group separation within the PCAs. Across different times of the year, group II was responsible for clustering the largest number of genotypes, where almost all hybrid combinations could be found. Group I clustered the smallest number (only genotype UFES 87 in the dendrogram corresponding to the first period analyses, and in the second period analyses, genotype UFES 56 was added to the group) (Figures 4b, 5b).



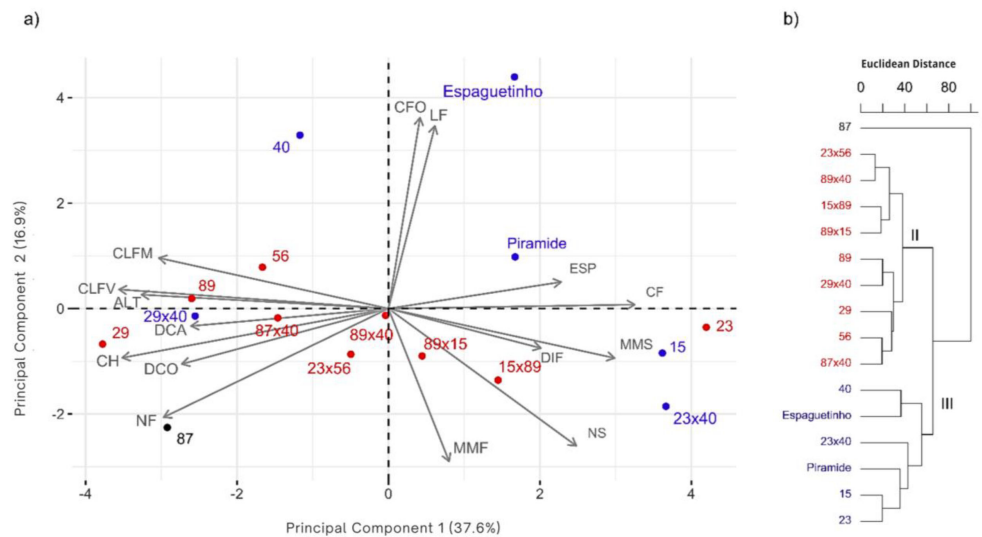


FIGURE 5

(a) PCA biplot based on quantitative traits measured in the second evaluation period from plants grown exclusively in greenhouse conditions at UFES. (b) Corresponding UPGMA dendrogram based on Euclidean distances. The analysis integrates morphological, agronomic, and fruit-related traits together with anthracnose severity in immature and mature fruits (CLFV, CLFM) resulting from controlled inoculation with *Colletotrichum scovillei*. ALT, plant height; DCO, canopy diameter; DCA, stem diameter; CH, stem length; CFO, leaf length; LF, leaf width; MMF, average fruit weight; NF, total number of fruits; DIF, fruit diameter; CF, fruit length; ESP, pericarp thickness; NS, number of seeds; MMS, weight of 1000 seeds; CLFM, *Colletotrichum scovillei* on mature fruits; CLFV, *Colletotrichum scovillei* on immature fruits.

The consistency of cluster formation across periods (Figures 4b, 5b) suggests stable genetic relationships among genotypes, indicating that environmental variation did not substantially alter the underlying structure of quantitative traits. This stability strengthens the reliability of the germplasm classification and suggests that these materials may perform predictably across contrasting conditions, a desirable feature for breeding.

The traits leaf length (CFO) and leaf width (LF) were strongly correlated in both the first and second evaluation periods, though this correlation occurred independently from the other descriptors. The descriptors plant height (ALT), stem diameter (DCA), stem length (CH), canopy diameter (DCO), and number of fruits (NF) were closely related in both evaluation periods, as were fruit thickness (ESP), fruit length (CF), average weight of 1000 seeds (MMS), fruit diameter (DF), number of seeds (NS), and average fruit weight (MMF). These results show that traits related to the plant and fruits were associated separately, as found by Silva et al. (2016). It is reasonable to expect that plants with greater CH would show higher DCO measurements, just as genotypes with higher ESP would have larger MMF.

Compact growth is one of the main attractive traits in ornamental pepper plants (Rêgo and Rêgo, 2018). The genotypes UFES 15, UFES 23, 15×89, 23×40, 89×15, and the two ornamental controls were distantly related to the ALT trait, indicating that these plants have a smaller stature. The average height of these seven genotypes across both evaluation periods was 17.61 cm. Meanwhile, the genotypes UFES 29, UFES 56, UFES 40, UFES 87, UFES 89, 29×40, and 87×40, which were closer to the ALT trait in the PCAs, had an average height of 25.26 cm. Additionally, the smaller genotypes were more closely related to fruit traits such as CF, DIF, and MMF (Figures 4a, 5a). The presence of larger fruits

provides higher fresh mass yield, making them more attractive for the food and pharmaceutical industries (Constantino et al., 2020).

The genotypes UFES 40 and the ornamental control “Espaguetinho Ornamental” showed a positive relationship with both traits in the PCAs of both evaluation periods. The smaller CF for the parent genotypes UFES 29, UFES 56, UFES 87, and UFES 89 could also be observed in the PCAs through the opposite positioning of these genotypes in relation to the trait. Longer fruits are important for breeding programs, as they stand out among the leaves and add greater value to the culinary industry (Abud et al., 2018).

The hybrids 89×15 and 15×89 showed a positive relationship with the NS trait. The same was found for the parent UFES 15, and the opposite for UFES 89, suggesting inheritance from the UFES 15 genotype. Multivariate analyses based on quantitative data complement traditional approaches and allow better data interpretation. Through these analyses, it is possible to understand the relationship between the parents, hybrids, and evaluated traits, enabling better selection of the desired trait in breeding programs (Santo et al., 2022).

3.3 Disease resistance

The controls Ikeda and UENF 1381 fulfilled their roles regarding susceptibility and partial resistance to anthracnose but were not included in the analyses since they were not evaluated for ornamental traits. The resistance of mature fruits (CLFM) to the fungus *C. scovillei* was related to the other traits only during the second evaluation period. In that same period, when evaluated in green fruits (CLFV), the resistance did not significantly interact with the genotypes. The genotypes UFES 15 and UFES 23 showed a negative relationship

with the disease in both the first and second evaluation periods, as did the hybrid combinations 15×89, 23×40, and 89×15 (Figures 4a, 5a). It is interesting to note that the parents UFES 15 and UFES 23 likely conferred some resistance to the hybrids, given that UFES 40 and UFES 89 showed a closer relationship with the disease. Part of the genetic variability found in *Capsicum* is linked to intra- or interspecific crosses, meaning that this variability can be increased by crossing contrasting individuals (Mishra et al., 2019; Tripodi et al., 2021).

Among all the genotypes evaluated, none showed resistance or high resistance to *C. scovillei*. The parent UFES 23, the hybrids 23×40 and 23×56, and the control 'Pirâmide Ornamental' displayed moderate resistance in green fruits. In mature fruits, the parents UFES 15, UFES 23, the hybrid 8×15, and the control 'Pirâmide Ornamental' showed moderate resistance. The hybrid combination 23×40 became moderately susceptible, 23×56 highly susceptible, and the control 'Pirâmide Ornamental' susceptible. The other genotypes were either susceptible or highly susceptible. Unlike the results obtained in this study, Baba et al. (2019), when evaluating 59 accessions of *Capsicum* spp. for anthracnose resistance, caused by *C. gloeosporioides*, identified six accessions resistant in both fruit maturation stages (green and ripe). This demonstrates that resistance to anthracnose, in *Capsicum* spp. fruits will depend being evaluated, as the genetic characteristics are different, and on the fungal isolate used, since the pathosystems are distinct. Therefore, these results highlight the importance of studying resistance to *Colletotrichum* spp. in both green and mature fruits,

as the genetic control differs between the two fruit maturity stages (Baba et al., 2019; Giacomini et al., 2020).

When comparing the results obtained during the two periods, similarities were found. The NF trait positively correlated with CH (0.7), as did DIF with ESP (0.7 in the first evaluation and 0.8 in the second), NS with MMS (0.8), and CFO with LF (0.9 in the first evaluation and 0.8 in the second). The CF trait negatively correlated with ALT (-0.7), DCA (-0.8 in the first evaluation and -0.7 in the second), and CH (-0.8) (Figures 6, 7).

It is plausible to consider that the longer the stem of a plant, the higher the number of fruits found on it, which can explain the positive correlation observed between NF and CH during both evaluation periods. Regarding the positive correlation between DIF and ESP, the same was found by Brillhante et al. (2021). However, their study also analyzed fresh mass gains, a trait that positively correlated with both DIF and ESP. Based on these results, it can be concluded that selecting genotypes with larger fruit diameters and thicker pericarps will be advantageous in breeding studies aiming for higher fresh mass yields in peppers.

In the first evaluation period, MMF did not correlate with any traits, as was the case for DCO, CLFV, and CLFM in the second evaluations (Figures 6, 7). However, in the second evaluations, there was a positive correlation between MMF, MMS (0.7), and ESP (0.8) (Figure 7). Ribeiro et al. (2020) found similar results to the positive correlation between MMF, MMS, and ESP observed in this study. According to the authors, ripe fruits with thicker flesh tend to have a

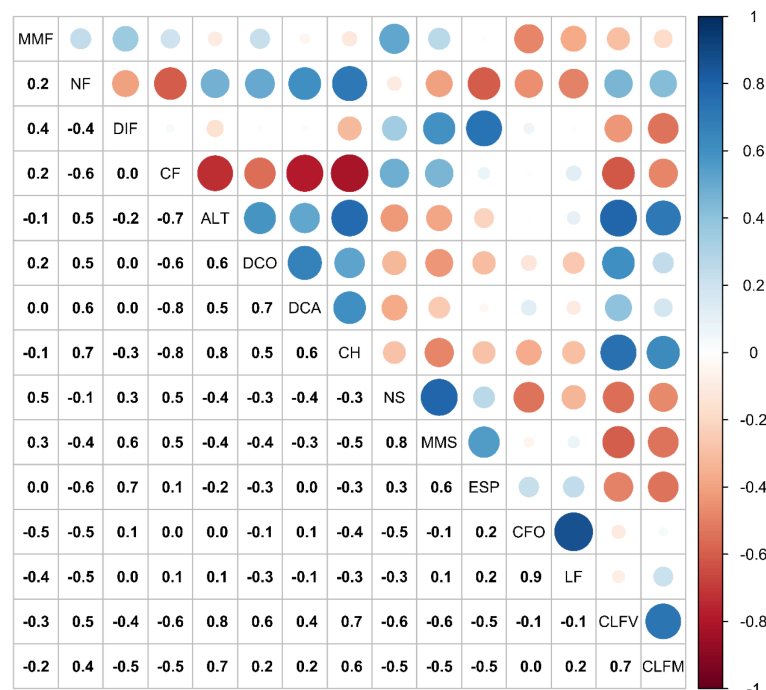


FIGURE 6

Correlation matrix of quantitative traits obtained during the first evaluation period from plants cultivated in a greenhouse experiment at UFES, Alegre. The matrix includes plant growth attributes, fruit characteristics, and disease-related variables CLFV and CLFM, representing anthracnose severity in immature and mature fruits after inoculation with *Colletotrichum scovillei*. Blue and red colors indicate positive and negative correlations, respectively. The size of the spheres is related to the degree of correlation. ALT, plant height; DCO, canopy diameter; DCA, stem diameter; CH, stem length; CFO, leaf length; LF, leaf width; MMF, average fruit mass; NF, total number of fruits; DIF, fruit diameter; CF, fruit length; ESP, pericarp thickness; NS, number of seeds; MMS, mass of 1000 seeds; CLFM, *Colletotrichum scovillei* in mature fruits; CLFV, *Colletotrichum scovillei* in immature fruits.

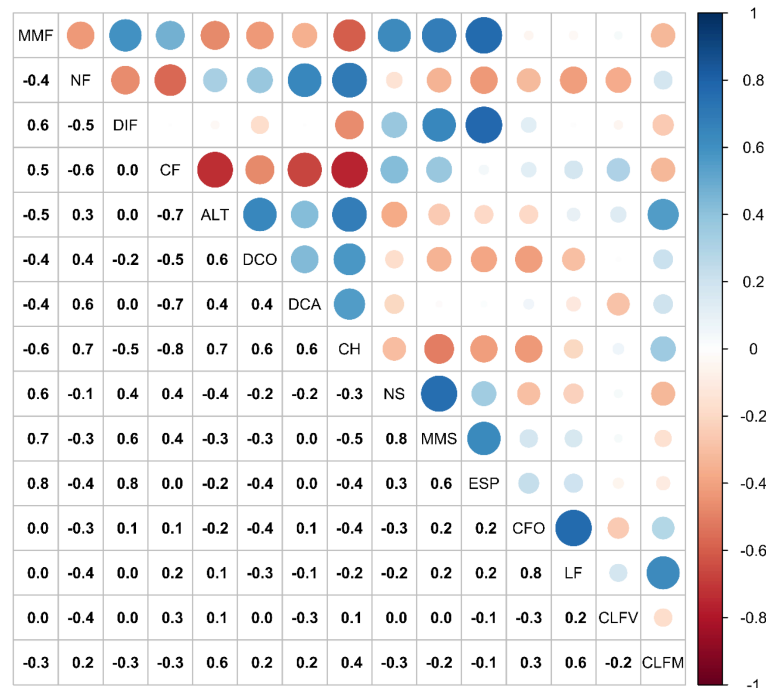


FIGURE 7

Correlation matrix of quantitative traits obtained during the second evaluation period from plants cultivated in a greenhouse experiment at UFES, Alegre. The matrix includes plant growth attributes, fruit characteristics, and disease-related variables CLFV and CLFM, representing anthracnose severity in immature and mature fruits after inoculation with *Colletotrichum scovillei*. Blue and red colors indicate positive and negative correlations, respectively. The size of the spheres is related to the degree of correlation. ALT, plant height; DCO, canopy diameter; DCA, stem diameter; CH, stem length; CFO, leaf length; LF, leaf width; MMF, average fruit mass; NF, total number of fruits; DIF, fruit diameter; CF, fruit length; ESP, pericarp thickness; NS, number of seeds; MMS, mass of 1000 seeds; CLFM, *Colletotrichum scovillei* in mature fruits; CLFV, *Colletotrichum scovillei* in immature fruits.

higher number of seeds, whereas ripe fruits with less flesh contain fewer seeds.

The ALT trait showed a positive correlation with CH, CLFV, and CLFM in the first evaluation period, and these associations were maintained – although at a lower magnitude – between ALT, CH, and CLFM in the second evaluation (Figures 6, 7). In addition, CLFV and CLFM were positively correlated, indicating that genotypes susceptible at the green-fruit stage tended to maintain susceptibility at the mature stage. When the breeder's goal is to obtain ornamental peppers, preference is given to shorter plants, as compact growth is more suitable for pot cultivation and enhances visual appeal (Rêgo et al., 2012; Cunha et al., 2020). In this context, the positive correlations between growth traits (ALT, DCO, DCA, CH) and anthracnose severity are particularly relevant: taller plants, with larger canopies and thicker stems, were more affected by *C. scovillei*, while the shorter genotypes showed comparatively lower disease severity.

This pattern suggests that selecting compact plants for ornamental purposes may simultaneously promote the selection of genotypes with improved resistance to anthracnose. Several hypotheses may explain why compact plants appear more resilient: reduced investment in vertical and radial growth may allow greater allocation of resources to defense-related pathways; architectural traits typical of compact plants – such as shorter internodes, denser canopy structure, or more robust epidermal tissues – may limit pathogen penetration or dispersal; and microclimatic effects associated with smaller canopies may reduce

splash dispersal or direct fruit exposure to inoculum. Altogether, these findings reinforce the potential of compact genotypes for ornamental breeding programs, as they combine desirable plant architecture with enhanced disease resilience.

4 Conclusion

All hybrids exhibited favorable traits for ornamental purposes. The parent plants UFES 15 and UFES 23, along with the hybrid combinations 15×89, 23×40, and 89×15, showed a higher number of characteristics demanded by the ornamental pepper market, particularly emphasizing the shorter stature of these genotypes. These same hybrid combinations also displayed lower susceptibility to *C. scovillei* compared to the other hybrids. Therefore, the hybrids 15×89, 23×40, and 89×15 are promising for future registration, and the parent plants UFES 15 and UFES 23 can be further explored in subsequent studies focusing on the ornamental sector.

Building on these findings, it is worth noting that the simultaneous expression of desirable ornamental traits and reduced disease susceptibility represents a highly advantageous combination for breeding programs. Compact growth habit – a central attribute for pot cultivation – was consistently associated with lower anthracnose severity, reinforcing the strategic value of these genotypes. Moreover, the parental lines UFES 15 and UFES 23 demonstrated strong potential as donors of both ornamental and resistance-related traits, indicating

that they may serve as important genetic sources for the development of new cultivars. Multilocation and field trials under varying inoculum pressures, as well as assessments of consumer acceptance, will be essential next steps to confirm the stability and commercial potential of the selected hybrids.

Nevertheless, the results generated in this study already provide important evidence for the characterization of *Capsicum* germplasm and contribute to strengthening the still-limited body of knowledge on integrated evaluations that simultaneously encompass ornamental traits, morphoagronomic descriptors, and disease resistance an evident gap in the current literature.

Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

Author contributions

MASP: Investigation, Methodology, Writing – original draft, Writing – review & editing. RNA: Investigation, Methodology, Writing – original draft, Writing – review & editing. TOS: Investigation, Methodology, Writing – original draft, Writing – review & editing. MRM: Investigation, Methodology, Writing – original draft, Writing – review & editing. CPS: Investigation, Methodology, Writing – original draft, Writing – review & editing. MMM: Investigation, Methodology, Writing – original draft, Writing – review & editing. FLO: Investigation, Methodology, Writing – original draft, Writing – review & editing. TFMC: Investigation, Methodology, Writing – original draft, Writing – review & editing. SP: Investigation, Writing – original draft, Writing – review & editing. EOCJ: Investigation, Methodology, Writing – original draft, Writing – review & editing. MVBMS: Investigation, Methodology, Writing – original draft, Writing – review & editing. CSB: Investigation, Writing – original draft, Writing – review & editing, Methodology, Project administration.

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Conflict of interest

The author(s) declared that this work was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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