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Multivariate techniques for identifying potential carrot hybrids

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Abstract

The adaptation of carrot cultivars to the environment is a fundamental factor for the cultivation. This study aimed to evaluate the agronomic potential of carrot genotypes cultivated in two periods by the hierarchical and optimization methods. The experiments were set as completely randomized block design. The treatments consisted of four experimental hybrids (Hybrid 8, 12, 14 and 17), three open-pollinated cultivars (Brasilia, BRS Planalto and Suprema), and a commercial hybrid (Verano), carried out at two cropping seasons. The unweighted pair-group using arithmetic averages (UPGMA), Tocher, and graphics of dispersion methods were applied. The UPGMA method and the graphic of dispersion enabled better separation of the genotypes evaluated in the growing seasons. The genotypes hybrid 12, cultivars Suprema and Verano presented a similar performance in both growing seasons. The cultivar Alvorada presented the greatest carrot yield (26.6 t ha^{-1}) and leaf fresh mass (1538.6 g $plant^{-1}$) in the spring-summer season, while hybrid 27 presents great potential for a great production (37.6 t ha^{-1}) of carrots in the autumn season.

Keywords: *Daucus carota* L.; genotype adaptability; UPGMA; Tocher method.
UPGMA: Unweighted Pair Group Method with Arithmetic Mean.

Introduction

Carrot (*Daucus carota* L.) is a plant that presents an edible root rich in bioactive compounds, such as carotenoids, dietary fibers and several useful components with functional, nutritional and therapeutic characteristics (Surbhi et al., 2018). In Brazil, the south, southeast, central-west, and northeast regions are great carrot producers (Anuário Brasileiro de Hortaliças, 2012), indicating how important is the production and consumption of this vegetable (Carvalho et al., 2017a). The average annual yield varies between 30 to 40 t ha^{-1}, depending on the use of technologies and investments during carrot cultivation (Dossa and Denck, 2018).

Despite carrot comes from mild climate regions, its cultivation in tropics can be done throughout the year if adapted cultivars are used and the recommended sowing time is respected (Resende and Braga, 2014). The adaptation of carrot cultivars to the environment (region or growing season) is a fundamental factor for the cultivation of this species, since there are direct and indirect influences of the environment on the production and quality of the carrots produced.

The stress caused by non-adequate choice of genetic (carrot genotype) and environment causes changes in the plants metabolism and oscillations in the levels of genes expression related to the synthesis of bioactives and carotenoids, drastically resulting in reductions of the content of carotenoids and alterations in efficient use of water (Perrin et al., 2016; Perrin et al., 2017).

The interaction between the environment and the plant varieties promotes different results according to the climate oscillations and the degree of sensitivity of the genotypes, as observed by Silva et al. (2012), Carvalho et al. (2014a) and Pereira et al. (2015a). The cultivars and hybrids sown in spring-summer come from the selection of genetic material with heat tolerance, mostly derived from the Brasilia and Kuroda carrot group (Resende and Braga 2014; Carvalho et al., 2014a).

In Brazil, the studies evaluating carrot experimental hybrids are scarce (Pereira et al., 2015b), especially by applying multivariate techniques. The actual tendency is that carrot hybrids dominate the seed market because of competitive advantages, mainly due to high plant stand uniformity and a great percentage of roots reaching commercial standard (Bandeira et al., 2013; Carvalho et al., 2015). This reinforces the need to extend the methodologies of adaptability evaluation.

To establish efficient strategies in identifying superior genotypes, methods of univariate and multivariate analysis are used. The univariate selection considers one or a few characteristics involved in the productive system, while the multivariate analysis uses combinations of multiple information to indicate the most promising genotypes (Costa et al., 2013). The multivariate analysis provides information about growth regions and growing seasons with great accuracy of the results for the production and quality of the commercialized products (Carvalho et al., 2014b).

Therefore, the objective of this study was to evaluate the agronomic potential of open-pollinated cultivars and hybrids of carrots cultivated in two seasons by the hierarchical and optimization methods of evaluation.

Results and Discussion

Carrot yield

There was a significant interaction between genotypes (cultivars and hybrids) and growing season (spring-summer and autumn) for yield and leaf fresh mass (Table 1). Commercial yield, scrap, diameter and length of carrots did not differ among the genotypes or growing seasons. The height of leaves, number of leaves and leaf dry mass differed between genotypes. The height of leaves also varied between the growing season (Table 1).

Although significant results were found in the present study, Silva et al. (2013) and Carvalho and Silva (2017) pointed out that the production components, such as the length and diameter of the carrot are highly related to the performance of the carrot genotype, defining these characteristics as auxiliaries in the discrimination of genotypes.

The cultivars Brasília, Suprema and Hybrid 12 did not differ between the two growing seasons for yield (t ha^{-1}). Thus, the cultivation of these genotypes may be performed in both seasons, spring-summer and autumn, without damage carrot production (Table 2). The cultivar Brasília also presented great yield of commercial roots in a wide variation of years and places of cultivation in the research performed by Pereira et al. (2015c). On the other hand, there are cultivars with restricted adaptation to different climatic conditions, as occurred with the cultivars BRS Planalto, Verano and Alvorada (Carvalho et al., 2017b).

Resende et al. (2016) observed that the cultivar Brasília, produced higher yield of carrot roots (58.2 t ha^{-1}) at temperatures between 24.4 and 30.4 °C between May and August in Petrolina Pernambuco state. This was greater than the yield observed in the present study, highlighting the responsive capacity of the Brasília cultivar. In the same way, Resende and Braga (2014) also observed greater yield in (96.30 t ha^{-1}) Brasília carrot in the conditions of the São Francisco Valley, in a period of mild temperatures (June to September).

Silva et al. (2013) found a phenotypic divergence between the cultivar Brasília and BRS Planalto related to the interaction genotype x environment, which determined the manifestation of agronomic traits. In the present study, such interaction was observed for the cultivar BRS Planalto, but not for Brasilia.

The yield of other genotypes was higher in 2017 (autumn), which can be explained by the climatic conditions close to the ideal range for the species (temperate climate). Yield was between 6 and 48.5% higher than that observed in 2016 (spring-summer), for the cultivar Alvorada and Hybrid 27, respectively (Table 2). The greatest productivities were observed for the genotypes Alvorada and Hybrid 27, in 2016 and 2017, respectively. In 2016, all yields were lower than the national average yield (39.7 t ha^{-1}), which can be explained by the great precipitation during the period (Figure 1).

In 2017, the hybrid 27 and cultivars BRS Planalto and Alvorada reached the Brazilian average for carrot yield. The hybrid Verano and the cultivar Supreme, should not be grown in conditions like the present study due to the low productive performance, being close to half of the national average yield (Table 2). The leaf fresh mass did not differ between growing seasons for the hybrid 27. In all others, the mass accumulation was greater in cultivation in spring-summer season, a period where great temperatures and humidity leads to great vegetative performance, which was not reflected in the root production.

The best performance of leaf fresh mass in spring-summer season was observed for cultivar Alvorada and hybrid 12. In 2017, the hybrids 14 and 27, and the cultivars BRS Planalto and Alvorada presented the greatest leaf fresh mass. The greatest root productivities were related to great vegetative rates, which were expected due to great photosynthetic rate and accumulation of photoassimilates, generally proportional to yield.

Carvalho et al. (2015) found greater commercial production and uniformity of carrot in hybrids when compared to open-pollinated cultivars in summer planting, opposed to the present study, where the yield of Alvorada was up to 26.8% greater in summer, compared to hybrids.

The changes that have occurred in the performance of the genotypes during the growing seasons highlight the importance of this study, by providing information about genotype and season recommendation, indicating which genetic material is better adapted to the specific environments (Silva et al., 2012). Such knowledge is essential since the standardization of genotypes can generate large losses in performance, highlighting the need for regional research programs as reported by Pereira et al. (2015c).

Multivariate analysis of carrot

The UPGMA dendrogram was generated from the dissimilarity matrix (Mahalanobis distance). The cophenetic correlation coefficient was 0.88% with distortion of 17.4%, demonstrating a proper relationship between the matrix and the dendrogram generated (Figure 2).

The separation of the groups was performed by the delimitation of a cutting line considering 40% of dissimilarity among the genotypes. The cutting line was established where it was observed abrupt changes in the branches presented in the dendrogram (Cruz et al., 2012), generating twelve distinct groups. The group I was composed of 16.70% of the genotypes; groups II, III, IV, V, IX, X, XI and XII were composed of 5.60%; groups VI and VII presented more than 11%, and group VIII by 16.70% of the genotypes.

The performance analysis of open-pollinated varieties and hybrids in two growing seasons shows that, there is divergence for all treatments, which is indicative of the influence of the environment. The hybrid 27 showed a greater divergence between the environments (Group 1 x Group XI). In contrast, the commercial cultivars Verano and Supreme, and the hybrid 12 showed great similarity when cultivated in two different seasons, indicating superior stability compared to distinct environmental conditions and similar phenotypic response in two seasons for the set of characters evaluated.

It is worth to mention that these results are from a multivariate analysis, involving all response variables of the study while by analysis of variance (F test, 5%). There was a significant interaction between genotypes (open-pollinated varieties and hybrids) and growing season (spring-summer

Table 1. Factors of variation (FV), degrees of freedom (DF), mean square (MS) and coefficient of variation (CV %) of the analysis of variance of the variables commercial yield (%), scrap (%), yield (t ha^{-1}), carrot diameter (cm), carrot length (cm), leaf height (cm), number of leaves, leaf fresh mass (g) and leaf dry mass (g) of nine carrot genotypes and two growing seasons.

FV	DF	Mean Squares - MS								
		Commercial yield	Scrap	yield	D	L	LH	NF	LFM	LDM
		%		(t ha^{-1})	(cm)				(g)	
Genotype (G)	8	59.6^{ns}	59.6^{ns}	130.4^{ns}	0.2^{ns}	0.1^{ns}	44.7 **	4.2 **	210768.1^{ns}	5684.5*
Season (S)	1	670.6^{ns}	670.6^{ns}	530.2^{ns}	0.01^{ns}	0.02^{ns}	2265.9**	1.7^{ns}	2688935.2^{ns}	10380.6^{ns}
G*S	8	18.3^{ns}	18.3^{ns}	49.0 **	0.3^{ns}	0.07^{ns}	14.0^{ns}	0.7^{ns}	91356.6**	2339.3^{ns}
CV (%)		8.8	13.7	9.0	18.5	3.6	5.5	9.2	16.3	36.6

^{ns}: non-significance; *: significance at 5%, and, **: significance at 1% of probability by the F test. D: Diameter, L: Length, LH: Leaf height, NF: Number of leaves, LFM: Leaf fresh mass, LDM: Leaf dry mass.

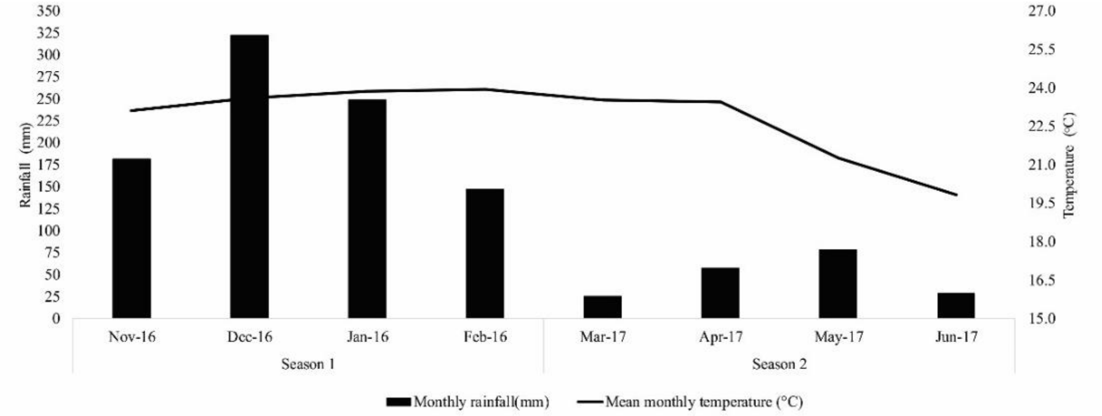

Figure 1. Maximum and minimum temperature (°C) and precipitation (mm) during both seasons.

Table 2. Yield (t ha^{-1}) and leaf fresh mass of (g plant^{-1}) assessed in carrot genotypes in two seasons.

Genotypes	Yield (t ha^{-1})		Leaf fresh mass (g plant^{-1})	
	2016	2017	2016	2017
Hybrid 27	19.36 Bb*	37.60 Aa	941.33 Ac	876.33 Aa
Hybrid 14	18.86 Bb	24.76 Ac	942.00 Ac	660.00 Ba
Hybrid 8	15.70 Bc	26.36 Ac	1055.33 Ac	549.00 Bb
Brasília	20.80 Ab	22.33 Ac	1084.00 Ac	488.00 Bb
Verano	13.90 Bc	18.33 Ad	671.33 Ad	402.00 Bb
Suprema	16.03 Ac	16.53 Ad	675.00 Ad	413.33 Bb
BRS Planalto	21.00 Bb	30.56 Ab	1100.66 Ac	623.00 Ba
Alvorada	26.63 Ba	30.93 Ab	1538.66 Aa	741.66 Ba
Hybrid 12	19.50 Ab	20.76 Ad	1307.00 Ab	545.33 Bb

*Averages followed by the same capital letter in line and lowercase letter in column do not differ at the level of 5% probability by the Scott-Knott test of averages.

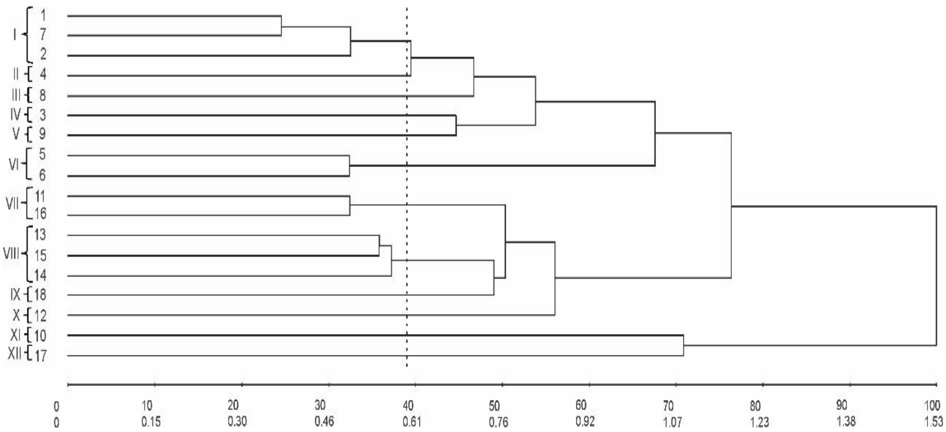

Figure 2. Dendrogram illustrating the analysis of carrot genotypes by the method of average connection between groups (UPGMA) obtained with the Mahalanobis distance generated with nine characters. Crop season 1 (1 = hybrid 27, 2 = hybrid 14, 3 = hybrid 8, 4 = cv. Brasília, 5 = cv. Verano, 6 = cv. Suprema, 7 = cv. BRS Planalto, 8 = cv. Alvorada, 9 = hybrid 12) and crop season 2 (10 = hybrid 27, 11 = hybrid 14, 12 = hybrid 8, 13 = cv. Brasília, 14 = cv. Verano, 15 = cv. Suprema, 16 = cv. BRS Planalto, 17 = cv. Alvorada, 18 = hybrid 12).

Table 3. Grouping by the Tocher optimization method for the carrot genotypes generated with nine characters for two seasons.

Group	Genotypes
I	1; 7; 2; 4; 8; 9; 3; 18; 6; 11; 5
II	13; 15; 14; 16; 12
III	10; 17

Crop season 1 (1 = hybrid 27, 2 = hybrid 14, 3 = hybrid 8, 4 = cv. Brasília, 5 = cv. Verano, 6 = cv. Suprema, 7 = cv. BRS Planalto, 8 = cv. Alvorada, 9 = hybrid 12) and crop season 2 (10 = hybrid 27, 11 = hybrid 14, 12 = hybrid 8, 13 = cv. Brasília, 14 = cv. Verano, 15 = cv. Suprema, 16 = cv. BRS Planalto, 17 = cv. Alvorada, 18 = hybrid 12).

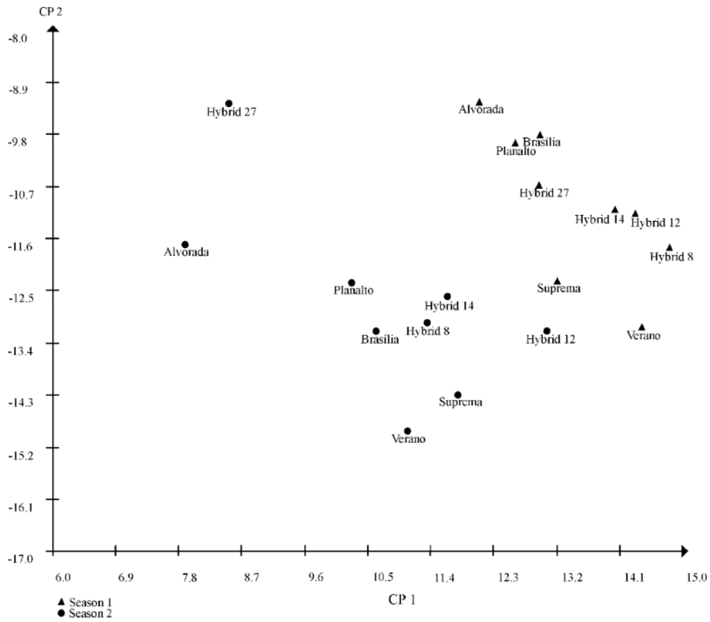

Figure 3. Graphic dispersion for carrot genotypes generated with nine characters for two seasons.

Table 4. Chemical characterization of soils in the experimental areas in two seasons (in the spring-summer of 2016 and in the autumn of 2017).

Soil characteristics	2016	2017
pH water	5.9	4
Phosphor (mg dm^{-3})	30.1	61.3
Potassium (cmolc dm^{-3})	0.22	0.15
Calcium (cmolc dm^{-3})	2.8	2.2
Magnesium (cmolc dm^{-3})	1.0	0.4
Copper (mg dm^{-3})	2.3	6.9
Zinc (mg dm^{-3})	6.6	9.4
Manganese (mg dm^{-3})	6.6	39.3
H+Al (cmolc dm^{-3})	3.40	4.2
Cationic exchange capacity (cmolc dm^{-3})	7.42	6.9
Organic matter (dag kg^{-1})	4.2	3.0
Base saturation (%)	54	40

and autumn) only for yield and leaf fresh mass (Table 1), demonstrating that multivariate analysis increases the range of information for carrot breeders, assisting the decision-making process. The grouping of the treatments in two growing seasons by Tocher's optimization method enables the formation of only 3 distinct groups (Table 3), revealing the non-existence of coincidence in relation to the UPGMA method. However, the method of UPGMA better distinguished the genotypes according to the effect of the environment in different seasons. Similarity, the agronomic performance of the hybrid 12 and Supreme cultivar in both growing seasons by Tocher method (Group I) was observed, agreeing with the result of the UPGMA method, except for the Verano cultivar (Table 3).

Graphs of dispersion can also facilitate the visualization of the group formation, strategically assisting the selection of diverging genotypes that are useful for breeding programs. According to Cruz et al. (2012), the interpretation of the dendrogram can be subjective as to the number of groups formed, being proposed a visual examination of the regions that result in the visualization of distinct groups. However, this characterization can only identify similar and divergent genotypes through the fixing of a genotype of interest.

Despite the clear divergence between genotypes, the cataloging of genetic groups was unable due to the inferential character of the analysis, making the principal components of analysis unable to determine with accuracy the genetic groups formed. This analysis was effective only in determining the proximity or separation of a genotype in relation to the other (Figure 3). However, as noted in the UPGMA analysis, there was a clear separation of genotypes in two quadrants, towards the left and right of the figure, indicating the influence of planting seasons in the phenotypic response of the evaluated genotypes.

Material and Methods

Study site

The experiment was carried out at two different periods, or seasons (spring-summer with sown in November 2016, and autumn with sown in March 2017) at the Vegetable Experimental Station of the Federal University of Uberlândia, Campus Monte Carmelo (18º42'43.19"S, 47º29'55.8"; at 873 m above sea level). The environmental conditions of both periods were monitored (Figure 1).

Experimental design

The experimental design was set as completely randomized blocks, with nine treatments (carrot genotypes) and three repetitions. The treatments consisted of four experimental hybrids (Hybrid 8, 12, 14 and 17) developed by the Embrapa program of genetic improvement of carrot, three open-pollinated cultivars (Brasilia, BRS Planalto and Suprema), and a commercial hybrid cultivar (Verano).

The soil preparation was similar in both experiments where two harrowing activities were performed before the mechanized arrangement of the seedbeds for carrot cropping. Before planting, soil sampling was carried out in the 0–20 cm layer, and the sample was chemically analyzed in accordance with the method described by EMBRAPA (2017) (Table 4).

The experimental plots (parcels) were composed of a 3 m long seedbed by 1 m wide, totaling an area of 3 m^2. The sowing was performed manually in double spaced furrows, with the space between rows at 10 cm, and 20 cm between double rows.

Thinning was done at 20 days after sowing leaving one plant every 5 cm, which resulted in a population equivalent to 890.000 plants ha^{-1}. During the cultivation period, the monitoring for pests and diseases and the applications of plant protection products were performed with registered products applied at the doses and frequency recommended by manufacturers, when necessary.

Assessments of plant variables

Yield (t ha^{-1}), commercial yield (%), scrap (% of carrots out of commercial standard), root diameter (cm), root length (cm), plant height (cm), the number of leaves, fresh and dry mass of leaves (g) were estimated. Forty carrots were harvested from each parcel and individually evaluated.

Statistical analysis

After attendance of the presuppositions of analysis of variance by the tests of normality of residues (Kolmogorov-Smirnov test), homogeneity of variances (Levene test) and additivity of block effects and treatments, the analysis of variance was performed at 1 and 5% of probability. In the event of a significant effect, the averages of the treatments were grouped by the Scott-Knott test ($p < 0.05$).

Multivariate analyses were carried out with the objective to determine the similarity between the experimental hybrids and the commercial cultivars at two different growing seasons (spring-summer and autumn). For both seasons the dissimilarity matrix by the Mahalanobis distance was calculated. The genetic divergence was represented by a dendrogram obtained by the hierarchical method of unweighted pair-group using arithmetic averages (UPGMA), graphics of dispersion (scatter plots) and by the Tocher optimization method. The validation of the grouping by the UPGMA method was determined by the cophenetic coefficient of correlation (CCC) calculated by the Mantel test (1967). All data were analyzed using the software v. 2015.5.0 Genes (Cruz et al. 2012).

Conclusions

The methods UPGMA and Tocher, and graphics of dispersion are able to detect differences in agronomic performance in function of growing seasons, differing the dissimilarity and similarity among carrot genotypes. The UPGMA method and the graphic of dispersion enable better separation of the evaluated genotypes and growing seasons. The hybrid 12 and cultivar Suprema especially, and also the cultivar Verano, performed similarly in both growing seasons.

The cultivar Alvorada presents greater carrot yield and leaf fresh mass than the other genotypes evaluated in spring-summer season, while hybrid 27 shows great potential for carrot yield in the autumn season.

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