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Evaluation of okra landraces based on agronomic and biochemical traits

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ABSTRACT

Okra is consumed in several countries and is an important source of nutrients for human food. To promote okra production and consumption is essential the development of new cultivars with high nutritional levels, besides desirable agronomic traits. Thus, this study aimed to evaluate nine okra landraces by agronomic and biochemical traits. The accessions UEL2, UEL3, UEL5, UEL6, UEL7, UEL9, UEL10, UEL13 and UEL15 were grown in open field condition and evaluated in relation to fruit yield, fruit weight, fruit diameter, number of fruits per plant and plant height. The biochemical analysis included mineral composition, crude protein content, vitamin C content, total flavonoid and phenolic content and antioxidant activity. The UEL7 accession stood out for the high levels for biochemical traits such as content of some minerals, vitamin C and antioxidant activity. On the other hand, UEL3 accession had a low performance for biochemical traits, but stood out for agronomic traits such as fruit yield and number of fruits per plant. Hence, the UEL7 and UEL3 landraces may be introduced in breeding programs focused on human food and agronomic performance, beyond to contribute for okra germplasm conservation.

Keywords: Abelmoschus esculentus, mineral composition, biochemical components, genetic diversity.

RESUMO

Avaliação de genótipos de quiabo baseado em características agronômicas e bioquímicas

O quiabo é vegetal consumido em vários países e é importante fonte de nutrientes para a alimentação humana. Para promover a produção e o consumo de quiabo, é essencial o desenvolvimento de novos genótipos com altos níveis nutricionais, além de características agronômicas desejáveis. Assim, este estudo teve como objetivo avaliar nove genótipos de quiabo por características agronômicas e bioquímicas. Os acessos UEL2, UEL3, UEL5, UEL6, UEL7, UEL9, UEL10, UEL13 e UEL15 foram cultivados em condições de campo e avaliados em relação à produtividade, peso do fruto, diâmetro do fruto, número de frutos por planta e altura da planta. As análises bioquímicas incluíram composição mineral, teor de proteína bruta, teor de vitamina C, teor total de flavonoides e fenólicos e atividade antioxidante. O genótipo UEL7 destacou-se pelos altos níveis de características bioquímicas, como conteúdo de alguns minerais, vitamina C e atividade antioxidante. Por outro lado, o UEL3 apresentou baixo desempenho para características bioquímicas, mas destacou-se por características agronômicas, como produtividade e número de frutos por planta. Portanto, as raças UEL7 e UEL3 podem ser introduzidas em programas de melhoramento focados na alimentação humana e no desempenho agronômico, além de contribuir para a conservação de germoplasma de quiabo.

Palavras-chave: Abelmoschus esculentus, composição mineral, componentes bioquímicos. diversidade genética.

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The okra (Abelmoschus esculentus) is a plant of Malvaceae family characterized by high rusticity, tolerance to high temperatures and low production costs (Oliveira et al., 2003). The crop is widely cultivated by Brazilian family farmers, mainly by small producers in the Northeast and Southeast regions (Mota et al., 2005). According to the Registro Nacional de Produção Orgânica (Brasil, 2019), organic okra production increased around 422% between 2016 and 2019, which

highlights the social and economic importance of this culture. In addition, okra has a nutritional importance in the human diet. Okra immature fruits (green seed pods) are an affordable source of carbohydrates, proteins, vitamins A and C, in addition to bioactive compounds such as flavonoids, and minerals (K, Ca, Mg, Zn, Mn and Fe) (Arapitsas, 2008; Petropoulos *et al.*, 2018). Okra fruits, which are consumed as vegetables, can be used in salads, soups and stews, fresh or dried, fried or boiled.

According to the World Health Organization (WHO), low consumption of fruits and vegetables is among the biggest risk factors that contribute to mortality (OMS, 2019). Raise consciousness about the importance of healthy eating to prevent diseases as well as increasing the accessibility of fruits and vegetables for the population are important ways of improving eating habits (Parker *et al.*, 2009). Therefore, promoting the consumption of traditional vegetables such as okra

could provide cheap sources of essential nutrients and also serve as an alternative of dietary diversification (Gemede *et al.*, 2015).

To promote okra production and consumption the development of new improved cultivars for agronomic and nutritional traits is essential (Vidal et al., 2018). The variability for agronomic and biochemical traits of okra has not yet been fully revealed, which is extremely important resource for plant breeding programs. Several studies have addressed the characterization of the genetic diversity of okra landraces based on phenotypic descriptors in different regions of the world (Reddy et al., 2012; Yonas et al., 2014; Asare et al., 2016; Ramgiry & Singh, 2017) including Brazil (Massucato et al., 2020). However, to the best of our knowledge, there is little information about biochemical characteristics, which is also important for advancing the okra breeding. Thus, the objective of this study was to characterize nine okra landraces using agronomic characteristics and biochemical parameters, aiming the promising genotypes selection for breeding programs.

MATERIAL AND METHODS

Plant material and experimental condition

Nine landraces of okra (UEL2, UEL3, UEL5, UEL6, UEL7, UEL9, UEL10, UEL13 and UEL15) were selected from germplasm bank of the Universidade Estadual de Londrina (UEL), which already were molecularly characterized (Massucato et al., 2020). The genotypes were evaluated in the experimental area of UEL in Londrina, Paraná, Brazil (23°19'42"S, 51°12'11"W), from January to April 2019 in field conditions. The design consisted of randomized blocks with three replications and eight plants per plot. The plots consisted of two rows, spaced 0.8 m between rows and 0.5 m between plants, and for the useful area, the first and last plants of each line were not sampled.

The soil classification in the experiment area is red latosol. The

chemical analysis of the soil presented the following conditions: pH (CaCl₂)= 5.9; P= 17.03 mg dm³; K⁺= 0.94 cmolc dm³; Ca⁺²= 7.26 cmolc dm³; Mg⁺²= 1.9 cmolc dm³; Al⁺³= 0.01 cmolc dm³; H + Al= 3.42 cmolc dm³; OM= 1.5%. A hand seeder was used for sowing in rows and the plants were grown following practices recommended for organic production (normative 17/2011 and Law 10.831/03). It was not necessary to apply chemical control of pests and diseases because economic damage level was not reached. Planting fertilization was carried out with the application of 5.0 t ha⁻¹ of cattle manure. The top-dressing fertilization was carried out after one month of sowing, spraying in total area, 20 L of cow urine in the aerial part of the plants (10 mL L⁻¹ of water) (Brasil, 2016).

Agronomic characterization

One month post-flowering, immature fruits were harvested in three days intervals, resulting in a total of eight harvests throughout the experiment. Only fruits with 6 to 15 cm length were selected, which corresponds to commercial classes (CEAGESP, 1997). The evaluated agronomic characteristics were: fruit yield (Yield, t ha-1), fruit weight (WF, g), fruit diameter (DF, mm), number of fruits per plant (NF) and plant height (PH, m) measured at the end of the harvest.

Biochemical characterization

Biochemical analyzes were carried out using three repetitions, with five fruits randomly sampled from each plant, resulting in a composite sample. The minerals content was quantified using the protocol described by Vizzoto et al. (2018), with atomic absorption spectrometry, flame photometry and spectrophotometry, for magnesium (Mg), calcium (Ca), zinc (Zn), copper (Cu), iron (Fe), phosphorus (P) and potassium (K). The results were expressed in mg 100 g⁻¹.

Vitamin C (VitC, mg 100 g⁻¹) content was quantified by AOAC method (AOAC, 1984) modified by Benassi & Antunes (1988). The total phenolic content (TPC, mg GAE 100 g⁻¹) was quantified using the protocol described by Swain & Hillis (1959). Total flavonoid content (TFC, mg QE

100 g⁻¹) was quantified following the protocol described by Lee *et al.* (1995). The antioxidant activity by sequestration of the 2,2-Diphenyl-1-picryl-hydrazyl (DPPH, μmol 100 g⁻¹) was quantified using the protocol described by Brand-Williams *et al.* (1995). Crude protein content (Prot, g 100 g⁻¹) was quantified using the Kjeldahl method (AOAC, 2012).

Statistical analysis

The data were analyzed via F test (P<0.05) in the analysis of variance (ANOVA). Significant differences in mean values were grouped using Scott-Knott test (P<0.05). Then, the data were analyzed using principal component analysis and visualized by heatmap with clustering using Euclidian distance and UPGMA method. The software R was used for statistical analysis using the ExpDes, FactomineR and pheatmap packages.

RESULTS AND DISCUSSION

For the fruit weight variable, the group with the highest averages ranged from 19.47 to 23.03 g and included accesses UEL2, UEL3, UEL6, UEL7 and UEL9 (Table 1). In the second group, which counted the landraces UEL5, UEL10, UEL13 and UEL15, fruit weight ranged from 15.67 to 18.67 g. For the variable fruit diameter, the group with the lowest averages was composed by only two landraces UEL5 and UE15, showing diameters of 17.4 and 17.8 mm, respectively. The second group included the remaining seven accessions, with fruit diameter ranging from 18.47 to 22.27 mm.

The number of fruits per plant ranged from 2.97 to 9.3 and the landraces were arranged in three groups (Table 1). The group with UEL3, UEL5, UEL6, UEL9 and UEL10 had the plants with the highest number of fruits, while UEL7 presented the lowest number of fruits. The plant height ranged from 1.31 to 2.46 m and the landraces were settled in three distinct groups (Table 1). The group formed by UEL3, UEL5 and UEL15 had the tallest plants and the group with UEL6, UEL7 and UEL9 presented the lowest plants. Considering

fruit yield, it was possible to classify the landraces in four different groups (Table 1). UEL3 was the landrace with the highest fruit yield (6.15 t ha⁻¹) while UEL7 (0.95 t ha⁻¹) had the lowest fruit yield. The other landraces were in intermediate groups.

The landrace UEL7, despite being among the plants that produce larger fruits, had less fruits per plant, which may justify the low fruit yield. Results described in the literature also show a wide difference in fruit yield and their components between different genotypes. For example, Saifullah & Rabanni (2009) report fruit yield ranging from 2.8 to 19.20 t ha-1 in a study involving 121 okra genotypes. The variation also extended for fruit weight and diameter, ranging from 15.28 to 26.15 g and 12.6 to 21.2 mm. In another study, the fruit weight and diameter ranged from 9.42 to 37.47 g and 14.45 to 33.16 mm for different commercial cultivars, which also reflected in different fruit yield (Mota et al., 2005). For plant height, landraces ranged from 1.31 to 2.46 m, slightly higher than reported by Passos et al. (2004) who reported plants with 1.3 to 1.9 m height for different cultivars. Plant height is an important variable for ergonomics aspects, mainly related to manual harvesting. Cultivars around 2 m height would be more advantageous for okra crop management (Cividanes et al., 2011).

The content of Mg and Ca ranged from 55.83 to 147.20 and 153.83 to 257.17 mg 100 g⁻¹, respectively (Table 2). The landraces UEL2 and UEL7 had the highest levels of Mg, while accession UEL9 and UEL15 had the highest levels of Ca. The levels of Zn and Cu ranged from 0.53 to 1.19 and 0.18 to 1.34 mg 100 g⁻¹, respectively. The landraces UEL7 and UEL10 showed the highest levels for Zn and Cu, respectively. The Fe content ranged from 2.56 to 5.12 mg 100 g⁻¹ and the landraces UEL9, UEL10 and UEL13 had the highest levels (Table 2). The levels of P and K ranged from 108.60 to 1.348.22 and 215.36 to 448.19 mg 100 g⁻¹, respectively. Potassium, phosphorus and calcium were the main minerals present in okra accessions, as described in other studies (Gemede et al., 2015), although the concentration varied between accessions. Results described in the literature also showed variation in okra mineral composition. For example, Gemede *et al.* (2016) reported K level ranging from 122.59 to 318.20 mg 100 g⁻¹. In other study, Mg and Ca ranged from 34.6 to 64.2 mg 100 g⁻¹ and 273 to 528 mg 100 g⁻¹, respectively (Santos *et al.*, 2013).

Even for elements with a lower concentration, such as iron, the okra landraces had higher levels than foods considered rich in this nutrient, such as kale (*Brassica oleracea* var. *acephala*)

(USDA, 2019) and tomato (Solanum lycopersicum) (Embrapa, 2011). The UEL13 has expressive levels of Fe and VitC, being classified in the groups of highest averages for both characteristics (Table 2). According to Germano (2002), vitamin C is important for the bioavailability of iron, favoring its absorption by humans.

Likewise, a total phenolic content ranged from 32.53 to 60.90 mg 100 g⁻¹ wherein the landraces UEL6 and UEL9 with the highest content presented almost twice as much content as UEL5, the accession with the lowest content

Table 1. Means of agronomic traits evaluated in okra landraces (*A. esculentus*). Londrina, UEL, 2019.

Accessions1/	Fruit weight	Fruit diameter	Number of fruits	Plant height	Fruit yield
	(g)	(mm)	(plant ⁻¹)	(m)	(t ha ⁻¹)
UEL2	21.00 a	19.27 a	5.37 b	1.83 b	1.77 с
UEL3	20.33 a	22.27 a	9.30 a	2.12 a	6.15 a
UEL5	16.33 b	17.80 b	7.50 a	2.19 a	2.34 c
UEL6	22.00 a	19.17 a	8.40 a	1.40 c	3.11 b
UEL7	19.47 a	20.00 a	2.97 с	1.31 c	0.95 d
UEL9	23.03 a	20.87 a	7.00 a	1.31 c	2.35 с
UEL10	18.67 b	18.80 a	6.60 a	1.69 b	2.08 c
UEL13	18.33 b	18.47 a	5.57 b	1.73 b	2.10 c
UEL15	15.67 b	17.40 b	6.20 b	2.46 a	1.78 c
CV (%)	11.79	7.78	20.39	13.70	17.18

 $^{^{1}\!\!}$ Means followed by the same letter in the column are grouped by Scott-Knott test at 5% probability.

Table 2. Mineral content (mg 100 g⁻¹) in okra landraces (A. esculentus). Londrina, UEL, 2019.

Accessions1/	Mg	Ca	Zn	Cu	Fe	P	K
UEL2	131.13 a	170.92 c	0.73 b	0.18 d	4.03 b	122.61 c	223.74 с
UEL3	76.42 c	74.50 d	0.70 b	1.17 b	2.56 d	123.89 с	215.36 с
UEL5	100.17 b	154.58 с	0.54 c	0.47 d	2.97 d	108.60 c	248.86 с
UEL6	55.83 с	23.83 d	0.55 c	0.82 c	3.60 c	110.12 c	238.81 с
UEL7	147.20 a	218.92 b	1.19 a	0.35 d	4.02 b	1348.2 a	448.19 a
UEL9	93.17 b	257.17 a	0.53 с	0.32 d	4.57 a	484.40 b	295.76 b
UEL10	97.33 b	202.50 b	0.72 b	1.34 a	4.84 a	119.30 с	223.74 с
UEL13	89.08 b	153.83 с	0.84 b	0.27 d	5.12 a	125.16 с	225.36 с
UEL15	69.00 c	257.17 a	0.80 b	0.28 d	4.23 b	136.12 с	273.65 b
CV (%)	10.87	15.90	15.76	15.75	5.67	4.85	8.04

¹/Means followed by the same letter in the column are grouped by Scott-Knott test at 5% probability.

(Table 3). Total flavonoid content and total antioxidant activity by DPPH assay showed variations from 24.19 to 56.85 mg 100 g⁻¹ and 471.06 to 722.82 μmol 100 g⁻¹. The landraces with the highest values were UEL2 for total flavonoid content and UEL5, UEL7 and UEL15 for DPPH activity. The levels of crude protein ranged from 2.38% to 3.28% and the landraces UEL15, UEL5 and UEL2 showed the highest levels for this compound.

The principal component analysis,

which explains about 80% of the variability, shows that the higher levels of minerals and vitamin C content contributed to separate the accession UEL7 from others. Besides, the landraces arranged in the left hemisphere of the graph had the best agronomic traits (UEL3, UEL6 and UEL10), while the right hemisphere displays the landraces with higher levels of biochemical content (UEL2, UEL13, UEL9 and UEL7) (Figure 1).

The clustering analysis in heatmap

Table 3. Total phenolic content (TPC, mg 100 g⁻¹), total flavonoid content (TFC, mg 100 g⁻¹), Vitamin C concentration (VitC, mg 100 g⁻¹), antioxidant activity (DPPH, μmol 100 g⁻¹) and crude protein (Prot, %) in okra landraces (*A. esculentus*). Londrina, UEL, 2019.

Accessions1/	TPC	TFC	VitC	DPPH	Prot
UEL2	47.65 c	56.85 a	13.58 с	616.94 b	3.08 a
UEL3	48.03 c	32.91 b	17.38 с	611.06 b	2.71 b
UEL5	32.53 e	34.44 b	18.87 c	715.76 a	3.18 a
UEL6	60.90 a	35.08 b	30.46 a	471.06 c	2.38 c
UEL7	51.65 b	34.40 b	31.46 a	722.82 a	2.76 b
UEL9	58.11 a	24.19 c	23.18 b	484.00 c	2.63 b
UEL10	22.19 f	36.96 b	23.84 b	564.00 b	2.42 c
UEL13	50.28 b	36.99 b	29.47 a	565.18 b	2.84 b
UEL15	44.28 d	40.26 b	20.20 b	722.82 a	3.28 a
CV(%)	4.23	8.17	13.01	6.05	15.95

¹/Means followed by the same letter in the column are grouped by Scott-Knott test at 5% probability.

showed the okra landraces into three groups, wherein groups 1 and 3 presented antagonistic traits. Group 1 comprised by the accession UEL7 showed the higher levels of Mg, Zn, P, K, Vitamin C and antioxidant activity and intermediate levels of Ca, Fe, TPC, TFC and crude protein. Group 3 included the landraces with the highest weight and diameter of fruits, number of fruits per plant and fruit yield that is landraces UEL3, UEL6 and UEL9. Accessions UEL2, UEL5, UEL10 and UEL15 that constitute the group 2 presented intermediate means for most of the evaluated traits (Figure 2).

The wide genetic variability among these accessions was also observed by Massucato et al. (2020) from the morpho-agronomical and molecular characterizations. Also, according to the authors, genetic dissimilarity of these accessions is related to different geographic locations of genotypes, as well as to the breeding carried out by small farmers. From the nine okra landraces evaluated for agronomic and biochemical traits, the UEL7 and UEL3 landraces may be introduced in breeding programs focused on human food and great agronomic performance, beyond to contribute for okra germplasm conservation.

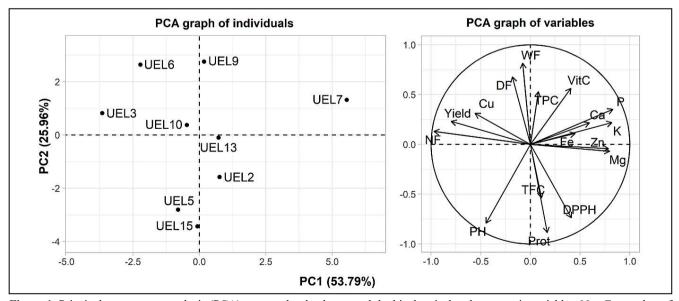


Figure 1. Principal components analysis (PCA) among okra landraces and the biochemical and agronomic variables. NumF= number of fruits per plant; Yield= fruit yield; VitC= vitamin C concentration; TPC= total phenolic content; WF= fruit weight; DF= fruit diameter; TFC= total flavonoid content; PH= plant height; DPPH= antioxidant activity; Prot= crude protein and mineral content (Cu= copper; Mg= magnesium; Ca= calcium; Zn= zinc; P= phosphorus; K= potassium; Fe= iron). Londrina, UEL, 2019.

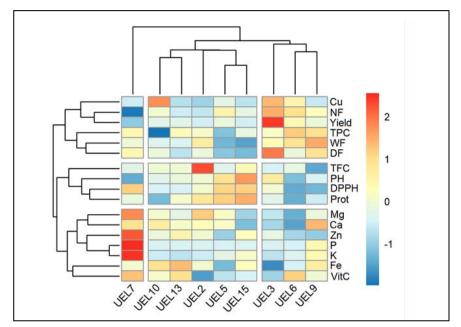


Figure 2. Heatmap relating the okra landraces and the biochemical and agronomic variables. NF= number of fruits per plant; Yield= fruit yield; VitC= vitamin C concentration; TPC= total phenolic content; WF= fruit weight; DF= fruit diameter; TFC= total flavonoid content; PH= plant height; DPPH= antioxidant activity; Prot= crude protein and mineral content (Cu= copper; Mg= magnesium; Ca= calcium; Zn= zinc; P= phosphorus; K= potassium; Fe= iron). The color scale in the right indicates data normalization and intensity related to the hot (red) and cold (blue) colors and the superior and inferior levels compared to the standard mean, respectively. Londrina, UEL, 2019.

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