



Recurrent selection of pumpkin (*Cucurbita moschata*) genotypes to increase fruit quality

Wilyanne Monique Danôa Bonfim · Maria Auxiliadora Coêlho de Lima ·
Cristina dos Santos Ribeiro Costa · Rita Mércia Estigarríbia Borges · Barbara Fernanda Ribeiro da Silva ·
Tamiris Georgia Barros Soares de Oliveira · Antônio Francisco de Mendonça Junior

Received: 30 May 2024 / Accepted: 6 September 2024
© The Author(s), under exclusive licence to Springer Nature B.V. 2024

Abstract The aim of pumpkin breeding programs is to develop cultivars that meet the production, commercial quality, and nutritional value, considering several attributes. Therefore, the objective of this study was to select pumpkin genotypes with high fruit quality for future use in genetic breeding programs. Two production cycles were carried out. The evaluations included the determination of genetic parameters (the additive genetic variance, environmental

variance between residual variances and individual phenotypic variance, accuracy of progeny selection, restricted heritability, coefficient of individual additive genetic variation, coefficient of genotypic variation between progenies, coefficient of residual variation, coefficient of relative variation, variance of the prediction error of the genotypic values of the progeny), the prediction of genetic gains, and the selection of genotypes with fruits of better physical and chemical quality attributes. The carotenoid and β -carotene contents of the pulp were distinguished by high values of additive genetic variance, residual variance and individual variance. Additionally, genetic gains were achieved for the main carotenoid compounds, as xanthophyll (68%), lycopene (58%) and β -carotene (49%). In the second production cycle, using the technique of Mulamba and Mock, a new ranking method was used to select the best genotypes. These genetic parameters, especially restricted heritability, indicate the potential for advancing new selection stages to develop pumpkin cultivars with superior qualities, including fruit length (0.976), skin thickness (0.951) and orange pigments, particularly β -carotene (0.894). Additionally, firmer fruits with high ascorbic acid content and a pulp hue angle representing an orange color were ranked among the best genotypes.

Supplementary Information The online version contains supplementary material available at <https://doi.org/10.1007/s10681-024-03411-5>.

W. M. D. Bonfim (✉) · C. dos Santos Ribeiro Costa ·
A. F. de Mendonça Junior
Department of Agronomy, Federal Rural University
of Pernambuco, Recife, PE 52171-900, Brazil
e-mail: wilyanne.monique@gmail.com

W. M. D. Bonfim
Federal Agency for Support and Evaluation of Graduate
Education, Brasília, DF 70040-031, Brazil

M. A. C. de Lima · R. M. E. Borges
Brazilian Agricultural Research Corporation, Embrapa
Semi-Arid, Petrolina, PE 56302-470, Brazil

B. F. R. da Silva
Postgraduate Program in Plant Genetic Resources,
State University of Feira de Santana, Feira de Santana,
BA 44036-900, Brazil

T. G. B. S. de Oliveira
Federal University of São Francisco Valley, Petrolina,
PE 56300-000, Brazil

Keywords Functional compounds · Genetic parameters · Genetic gain prediction · Selection index · Pumpkin

Introduction

Pumpkin (*Cucurbita moschata*) is an economically important vegetable that has received much attention due to the nutrients and bioactive compounds present in its seeds and fruits (Han et al. 2022). It has fruits with pulp colors ranging from an intense yellow to orange, revealing high carotenoid contents, mainly α - and β -carotene, β -cryptoxanthin, lutein and zeaxanthin (Grassino et al. 2023; Stupar et al. 2021). β -carotene is the most abundant, and its importance is highlighted by its high pro-vitamin A activity (Mokhtar et al. 2021; Stupar et al. 2021).

Vitamin A deficiency is a serious public health problem in many developing or subdeveloped countries, especially among women of reproductive age and children under five years old (Miglioli et al. 2015). Studies have shown that night blindness and xerophthalmia are associated with vitamin A deficiency and protein-energy malnutrition, which generally results from a low intake of carotenoids, especially β -carotene, in the diet (Janani et al. 2018). Therefore, accessible and distinct β -carotene sources increase the dietary intake of vitamin A, resulting in high social and economic benefits.

In Brazil, the pumpkin breeding program developed by Embrapa (Brazilian Agricultural Research Corporation) is based on the variability of the germplasm collected in the northeastern region of the country. By evaluating this germplasm, the chances of obtaining genotypes with high carotenoid contents, particularly β -carotene, increase. The program has already developed progenies with traits of commercial and nutritional interest for future selection (Borges et al. 2019a).

The choice of genitors and knowledge of the mechanisms controlling the main agronomic traits of interest for the species are important breeding strategy decisions that allow for scientific and technological advances. These mechanisms support the selection of more effective crossing methods (Darrudi et al. 2018).

Although pumpkin is an allogamous plant (for which cross-pollination is preferred), it is still possible to perform self-fertilization (Oliveira et al. 2021). Therefore, recurrent selection is recommended to obtain superior results. This method can efficiently and quantitatively improve population characteristics and maintain genetic variation.

Kumar et al. (2022) reported genetic advances improvements of 76% for increase in ascorbic acid content and 101% for increase in β -carotene content in pumpkin genotypes in India. According to the authors, since pumpkin is a cross-pollinated crop and the studied agronomic and fruit quality traits are controlled by additive gene action, breeding procedures such as synthetic breeding, composite breeding and population improvement by recurrent selection for general combining ability (gca) can be utilized for improvement.

Generally, the success in launching and adopting of a new cultivar on the market is related to the presence of multiple superior traits, including product quality (Latheef et al. 2022). This condition demands the use of effective techniques to progress breeding in a reasonable time. Direct selection may not be effective when the breeding program deals with complex traits like fruit yield and fruit quality, which are controlled by multiple genes and influenced by environmental factors (Hedge et al., 2024). Therefore, estimatives of genetic parameters are crucial for understanding the inheritance of traits and enabling effective selection.

Genetic variance combined with genetic parameter estimates can predict genetic gain through selection. In this way, the estimation of genetic parameters associated with the use of selection indices is an efficient strategy to be adopted in breeding programs. For pumpkin, there is an opportunity to explore this approach aiming to select genotypes with a group of superior traits for fruit quality. Some parameters are essential for the successful evaluation and selection of superior genotypes, including the availability of genetic variability in the population, the selection method adopted, genetic correlations between traits, the type of inheritance involved and experimental accuracy (Costa et al. 2023).

The aim of this study was to select pumpkin genotypes with high fruit quality for future use in the genetic breeding programs.

Material and methods

Genetic material and experimental design of the study

The genetic material used in this investigation was 12 pumpkin lines obtained in previous stages of the

genetic breeding program developed at Embrapa Semiárido. The experimental area was located in the Bebedouro Experimental Field at Embrapa Semiárido, in Petrolina, Pernambuco State, Brazil (09°09'S, 40°22'O, 365.5 m altitude). Two production cycles were evaluated, the first of which was carried out from March to July 2022, during which the plants were transplanted to the field on March 15th and harvested on July 28th. The second production cycle occurred during the months of January–May 2023, during which the plants were transplanted into the field on January 20th and harvested on May 25th. In each of the production cycles, the best genotypes were selected, considering a selection index of 10%.

The original population (s_0) evaluated in the first production cycle consisted of 12 lines. These lines were obtained by four successive cycles of self-fertilization and they were selected on the basis of commercial characteristics of interest (Table 1). The best fruit of each on of the 12 lines was selected based on visual aspects and the absence of skin damage.

The variables analyzed in the first cycle were mass (M), length (LF), largest (LD) and smallest diameter (SD), skin (ST) and pulp thickness (PT), cavity diameter (CD) and skin and pulp color (evaluated by luminosity—L, chroma—C and hue angle—H), titratable acidity (TA), soluble solids (SS), carotenoids (Car), β -carotene (β car), γ -carotene (γ car), lycopene (Lyc), cryptoxanthin (Cryp) and xanthophyll (Xan) contents. Based on the results of the variables analyzed, the best fruits were selected and fifty seeds from each selected fruit were mixed and used in the following experiment.

The second production cycle (population s_1) was composed of a random sample of the seed mixture from the previous cycle, with the population consisting of ten lines containing nineteen plants each, without a specific experimental design, aiming at random crossing and genetic recombination within the population. At harvest, the best fruit from each plant was also chosen for evaluation. This selection was based on M, LF, LD, SD, ST, PT, CD, skin and pulp color, pulp firmness (PF), TA, and SS and ascorbic acid contents.

Growing conditions and trial evaluation

In both production cycles, plants were sown in expanded polystyrene (EPS) trays filled with an

appropriate commercial substrate, with one seed per cell. Transplanting to the field took place 13 days after sowing. The soil was previously prepared by plowing to a depth of 30 cm, followed by harrowing and furrowing. A 4 m × 2 m spacing was used, and the area was irrigated using a drip system.

At 120 days after transplanting, fruits were harvested separately per plant for weighing, and according to visual aspects, the best fruit was selected for evaluation. The genotypes were ranked using a 10% selection index, and the 19 best fruits were determined. The seeds of the fruits selected in the first production cycle were mixed together for the new experiment in which the plants were subjected to open pollination. In the second experiment, 190 fruits were evaluated, and a 10% selection index was applied for a new ranking.

Quality characteristics evaluation













At the laboratory, the fruits were kept at room temperature and evaluated individually. Physical and chemical evaluations were performed. The fruit mass (kg) was determined on a digital semianalytical scale. LF, LD and SD were determined using a digital caliper and a ruler, with the values obtained in cm. STs and PTs were also measured using a digital caliper, and the results are expressed in mm.

A Konica Minolta colorimeter was used to measure the skin and pulp color from L, which ranged from 0 (black) to 100 (white); C, which started at 0 and increased according to the intensity of the color; and H, whose values ranged from 0 to 360°, where 0° = red, 90° = yellow, 180° = green and 270° = blue. The intermediate values between the specified H points correspond to transitions between specific colors.

PF was determined using two slices of each fruit taken from opposite sides. The results were obtained using a digital electronic texturometer (Texture Analyser TA.XT.plus), with values expressed in Newtons (N).

The pulp juice was extracted to determine the chemical parameters. The SS concentration was determined using a refractometer and is expressed in °Brix (AOAC 2010a). TA, expressed as a percentage of citric acid, was determined using 0.1 N sodium hydroxide (AOAC 2010b). In the second production cycle, the ascorbic acid content was also determined

Table 1 Characteristics of elite lines from Embrapa Semiárido's pumpkin breeding program evaluated in the first production cycle

Treatment	GAB origin access	Description
	545	The fruit is flat with a slight chromaticity and predominant orange skin. The secondary skin color is cream with a slight chromaticity. The fruit is marbled and the pedicel is variegated. It has a smooth texture and no warts
Lineage 1		
	569	The fruit is flat with a moderate chromaticity and predominantly orange skin. The secondary skin color is green with a moderate chromaticity. The secondary coloration of the fruit is marbled and the pedicel is yellow. It has a smooth texture and no warts
Lineage 2		
	569	The fruit is flat with a moderate chromaticity and predominantly cream-colored skin. The secondary color is cream and it has a slight chromaticity. The secondary coloration is distributed in dots and the pedicel is yellow. It has a smooth texture, with no warts and no gumminess
Lineage 3		
	545	The fruit is flat with predominantly orange skin of moderate chromaticity. The secondary skin is cream-colored with a slight chromaticity. The secondary color of the fruit is marbled and the pedicel is yellow. It has a smooth texture, with no warts and no gumminess
Lineage 4		
	545	The fruit is flat with predominantly cream-colored skin of moderate chromaticity, with a light-yellow secondary skin color. The secondary color is distributed in spots and the pedicel is yellow. It has a smooth texture, with no warts and no gumminess
Lineage 5		
	545	The fruit is flat and its the skin has a predominantly orange color of a slight chromaticity. The secondary skin color is cream-colored and the chromaticity is slight. The fruit is marbled and the pedicel is variegated. It has a smooth texture, with no warts and medium gumminess
Lineage 6		
	545	The fruit is globular and the skin is predominantly cream-colored with a moderate chromaticity. The secondary skin color is yellow and its chromaticity is slight. The secondary color of the fruit is marbled and the pedicel is variegated. It has a smooth texture, with no warts and a slight gumminess
Lineage 7		
	545	The fruit is flat with a predominantly slight orange skin. The secondary skin color is cream-colored with a slight chromaticity. The fruit is marbled and the pedicel is yellow. It has a smooth texture, with no warts and no gumminess
Lineage 8		
	569	The fruit is flat and the skin is predominantly cream-colored with moderate chromaticity, and a yellow secondary color of slight chromaticity. The secondary color is distributed in dots and the pedicel is yellow. It has a smooth texture, with no warts or gumminess
Lineage 9		
	569	The fruit is flat with predominantly cream-colored skin of moderate chromaticity. The secondary skin color is cream-colored and the chromaticity is slight. The secondary color is distributed in dots and the pedicel is yellow. It has a smooth texture, no warts and no gumminess
Lineage 10		
	569	The fruit is globular with predominantly cream-colored skin of moderate chromaticity, and a cream-colored secondary skin of slight chromaticity. The secondary color is distributed in dots and the pedicel is yellow. It has a smooth texture, with no warts and no gumminess
Lineage 11		
	569	The fruit is pyriform with predominantly dark orange skin. The secondary skin color is a slight orange. The fruit is marbled and the pedicel is variegated. It has a smooth texture, with no warts and a slight gumminess
Lineage 12		

GAB germplasm active bank

by titration in a 0.02% Tilman solution (2, 6 dichlorophenol indophenol). The results are expressed in $\mu\text{g g}^{-1}$ (Strohecker and Henning 1967).

In the first production cycle, the total carotenoid content (Car) was analyzed following the recommendations of Rodriguez-Amaya and Kimura (2004), and the results are expressed in $\mu\text{g.g}^{-1}$. The

contents of β car, γ car, lyc, cryp and xan, expressed in $\mu\text{g}\cdot\text{g}^{-1}$, were determined from the extracts for quantification of total carotenoid content. The concentrations of the compounds were determined by high-performance liquid chromatography using Waters equipment (Model 36 Alliance e2695) coupled to a Diode Array Detector (DAD) 2998 at 450 nm. The extracts were run on a YMC Carotenoid-C 30 column (4.6×150 mm, $3\ \mu\text{m}$) with the following sequence: 0 min, 80% methanol+20% tert-butyl methyl ether; 0.5 min, 75% methanol+25% tert-butyl methyl ether; 15 min, 15% methanol+85% tert-butyl methyl ether; 15.05 min, 10% methanol+90% tert-butyl methyl ether; 16.5 min, 10% methanol+90% tert-butyl methyl ether; 16.55 min, 80% methanol+20% tert-butyl methyl ether; 22 min, 80% methanol+20% tert-butyl methyl ether; and a flow rate of $0.8\ \text{mL}\ \text{min}^{-1}$ and an oven temperature of $33\ ^\circ\text{C}$.

Statistical analysis

Except for heritability, which was estimated using GENES software (Cruz 2013), the variance components were estimated using the restricted residual maximum likelihood (REML) method, and the phenotypic and genotypic values (BLUPs) were predicted using Selegen-REML/BLUP software (Resende 2016). Model 01, which is indicated for the evaluation of individuals in half-sib progenies or open pollination in allogamous species, was adopted (Resende 2007). It is represented in matrix form as follows:

$$y = X_r + Z_a + W_p + e$$

where y is the data vector; r is the vector of replication effects (assumed to be fixed) added to the mean; a is the vector of individual additive genetic effects (random); p is the vector of plot effects (random); e is the vector of errors or residuals (random); and the capital letters represent the incidence matrices for these effects.

Additionally, the additive genetic variance (V_g), environmental variance between residual variances (V_e) and individual phenotypic variance (V_f), accuracy of progeny selection (Ac_{prog}), coefficient of individual additive genetic variation ($CV_{gi}\%$), coefficient of genotypic variation between progenies ($CV_{gp}\%$), coefficient of residual variation ($CV_{e}\%$),

coefficient of relative variation (CV_r), variance of the prediction error of the genotypic values of the progeny, assuming complete survival (PEV), and standard deviation of the predicted genotypic value of the progeny, assuming complete survival (SEP), were estimated.

In the first production cycle, the data were obtained from the genetic values predicted (BLUPs), and the multivariate selection index of Mulamba and Mock (1978) was used to select the superior individuals in the joint evaluation of all the traits. To obtain the selection index, each characteristic was standardized and multiplied by the weighting coefficient (K_x) adopted for each variable and according to their respective importance for pumpkin fruit quality. It was adopted $K_x = 1$ for variables of lesser importance and $K_x = 5$ when the variable has a greater importance (Table 2). In this way, each variable was standardized to the unit so that each had the same relative weight and was multiplied by K_x . Excel software was used to calculate the selection index (SI):

$$SI = K_1 * ((\text{fruit mass} - \text{min}_1)/R_1) + K_2 * ((\text{fruit length} - \text{min}_2)/R_2) + K_3 * ((\text{largest diameter} - \text{min}_3)/R_3) + \dots + K_n * ((\text{xanthophyll content} - \text{min}_n)/R_n),$$

where $K_1, K_2, K_3, \dots, K_n$ are the weighting coefficients assigned to the variables (Table 2); $\text{min}_1, \text{min}_2, \text{min}_3, \dots, \text{min}_n$ are the minimum values assumed by the variables; and $R_1, R_2, R_3, \dots, R_n$ are the ranges of variation of the variables.

Table 2 Weighting coefficient (K_x)^{*} adopted for fruit quality attributes and bioactive compounds evaluated to select the most promising pumpkin genotypes

Features	Kx	Features	Kx
Fruit mass	3	Pulp hue angle	5
Fruit length	3	Soluble solids content	3
Largest diameter	3	Titrateable acidity	3
Smaller diameter	2	Ascorbic acid content	5
Skin thickness	4	Pulp firmness	4
Pulp thickness	5	Total carotenoid content	5
Cavity diameter	4	β -carotene content	5
Skin luminosity	1	γ -carotene content	2
Skin chroma	3	Lycopene content	1
Skin hue angle	5	Cryptoxanthin content	2
Pulp luminosity	1	Xanthophyll content	2
Pulp chroma	3		

^{*} K_x varies from 1 to 5, according to a greater importance of the trait

The selection index was used to classify the genotypes from the first cycle, applying a selection intensity of 10% and selecting the 19 best genotypes. For the selected genotypes, the gains for each trait were predicted by the following equation:

$$GG\% = \frac{\bar{X}_m - \bar{X}_0}{\bar{X}_0} \times 100$$

where GG (%) is the gain from selecting the best genotype in the population for the trait; \bar{X}_m is the average BLUP of the genotype selected from the population; and \bar{X}_0 is the overall BLUP of all genotypes for the trait.

For the second cycle, selection was again carried out using the Mulamba and Mock multivariate selection indices, through M, LF, LD, SD, ST, PT, CD, LS, CS, HS, LP, CP, HP, SS, TA, AA and PF. It was applied a selection intensity of 10%.

Results

All the estimates of genetic variance (V_g) were positive (Table 3). The values ranged from 0.001 for cryptoxanthin content (Cryp) to 10,472.571 for total carotenoid content (Car), and the lowest values were observed for titratable acidity (TA), γ -carotene (γ car), lycopene (Lyc), xanthophyll (Xan) content, largest diameter (LD), skin thickness (ST) and cavity diameter (CD). Some variables had higher estimates for individual phenotypic variance (V_f) than did V_g , namely, mass (M), SD, CD, skin chroma (Cs), pulp luminosity (Lp), pulp hue angle (Hp), soluble solids content (SS), TA, Car, β -carotene content (β car) and γ car. In particular, the V_f and V_g values for TA and γ car were very low.

The highest and lowest heritability, coefficient of individual additive genetic variation, coefficient of genotypic variation between progenies and coefficient of environmental variation values are written in bold and italics, respectively.

Heritability is determined by the ratio between V_g and V_f . Therefore, this approach provides information on the transmission of a character to successive generations of the species. For heritability in the strict sense (h) of the characters evaluated in the first pumpkin production cycle, M had an average estimate of 0.147 (Table 3). Among the physical characteristics,

skin luminosity (Ls) had the lowest h value when compared to length (LF), LD and CD, whose h value averages were greater than 0.90. According to the chemical analyses, the lowest h value was observed for Xan (0.019), while the highest h value was observed for β car (0.894). These higher estimates also showed high h values for other variables, such as Cryp (0.835) and Lyc (0.828).

According to the analysis of the accuracy of selection (Acprog), as shown in Table 3, there were high or very high values for the studied variables, except for SS. This parameter is used to identify the level of statistical confidence in the experimental analyses.

For the characters LF, LD, ST, PT, Lc, Hc, γ car, Lyc and Xan, the coefficient of genotypic variation between progenies (CVgp%) whose values ranged from 6.55% to 41.45%, was greater than the coefficient of environmental variation (CVe%) (Table 3). For all the characters analyzed, the relative coefficient of variation (CVr), which is the ratio between the genetic and environmental coefficients of variation, was the lowest among the CVs determined.

Considering the results obtained for the estimates of genetic parameters in the genotypes evaluated and using a selection intensity of 10%, the 19 best fruits were selected (Tables 4 and 5 and Fig. 1). This selection allowed for the evaluation of a set of characteristics associated with commercial fruit quality, such as M, SS and TA, as well as those associated with functional properties, such as Car and β car. The high PT, Hp, SS, Car and β car values are much greater than those of commercial cultivars or hybrids and even those of most germplasm banks according to the results of Barboza et al. (2023), Bergantin et al. (2018) and Stupar et al. (2021).

In addition to the high values of important quality characteristics observed in the selected genotypes (Tables 4 and 5), there was a prediction of favorable genetic gains for increasing the variables LF, SD, PT, Cp, SS, TA, Car, β car, γ car, Lyc, Cryp and Xan in the next stages of selection (Table 6). Among the variables, LF had the highest percentage of genetic gain, while genetic gain for PT was considered intermediate. For both variables, medium-sized and larger fruits are expected because they may have a greater proportion of pulp.

The percentages of predicted genetic gains for Car, β car, Lyc and Xan were 36, 49, 58 and 68%, respectively (Table 6). For Car and β car, which

Table 3 Genetic parameters of 21 variables, in the first production cycle, from analyses of pumpkin genotypes using Genes software and restricted residual maximum likelihood (REML) and phenotypic and genotypic values (BLUPs) from Selegen software

Variable	Genetic parameters											general average
	Vg	Ve	Vf	Acprog	h	CVg%	CVgp%	CVe%	CVr	PEV	SEP	
M (Kg)	21.45	277.32	300.28	0.46	0.147	77.83	38.91	130.37	0.30	4.23	2.06	3.03
LF (cm)	86.07	-58.24	28.15	0.99	0.976	57.01	28.50	7.71	3.70	0.51	0.72	16.27
LD (cm)	20.53	-10.68	10.51	0.95	0.901	21.46	10.73	6.00	1.79	0.48	0.70	21.11
SD (cm)	1.37	4.17	5.57	0.70	0.792	12.86	6.43	11.40	0.56	0.17	0.42	8.93
ST (mm)	1.11	-0.23	0.93	0.91	0.951	23.63	11.81	9.39	1.26	0.05	0.22	4.46
PT (mm)	110.57	-61.83	48.94	0.97	0.753	38.32	19.16	7.66	2.50	1.40	1.18	27.44
CD (cm)	1.96	4.22	6.20	0.75	0.34	11.49	5.74	8.82	0.65	0.22	0.46	13.81
Ls	119.91	-61.21	58.98	0.97	<i>0.034</i>	14.39	7.20	3.23	2.23	1.88	1.37	76.18
Cs	7.71	29.76	39.56	0.62	0.36	8.11	4.05	8.86	0.46	1.18	1.09	34.25
Hs	116.82	-57.96	60.68	0.96	0.693	13.09	6.55	3.37	1.94	2.37	1.54	82.66
Lp	4.60	8.45	13.09	0.77	0.642	3.23	1.62	2.35	0.69	0.47	0.69	66.31
Cp	14.06	39.68	53.91	0.71	0.607	5.47	2.74	4.66	0.59	1.73	1.32	68.56
Hp	2.51	2.00	4.91	0.78	0.49	2.44	<i>1.22</i>	<i>1.67</i>	0.73	0.24	0.49	64.99
SS (^o Brix)	16.81	2048.29	2100.91	0.17	0.384	26.22	13.11	135.35	0.10	4.09	2.02	15.64
TA (%)	0.000019	0.000027	0.000051	0.72	0.695	<i>0.52</i>	8.12	13.46	0.60	0.000002	0.0015	0.03
Car (µg·g ⁻¹)	10,472.57	1441.85	13,280.36	0.84	0.658	26.71	13.35	14.82	0.90	762.12	27.61	383.09
βcar (µg·g ⁻¹)	10,053.90	889.20	12,874.06	0.82	0.894	35.02	17.51	21.00	0.83	814.80	28.54	286.77
γcar (µg·g ⁻¹)	0.29	0.06	0.35	0.89	0.730	26.53	13.26	11.73	1.13	0.01	0.12	2.02
Lyc (µg·g ⁻¹)	0.85	-0.24	0.72	0.88	0.828	82.90	41.45	39.04	1.06	0.05	0.22	1.13
Cryp (µg·g ⁻¹)	0.0006	0.0024	0.0030	0.66	0.835	13.28	6.64	13.05	0.51	0.00008	0.01	0.18
Xan (µg·g ⁻¹)	0.15	-0.05	0.11	0.91	<i>0.019</i>	60.11	30.05	23.21	1.30	0.01	0.08	0.64

M mass; LF length; LD largest diameter; SD smallest diameter; ST skin thickness; PT pulp thickness; CD cavity diameter; Ls skin luminosity; Cs skin chroma; Hs skin Hue angle; Lp pulp luminosity; Cp pulp chroma; Hp pulp Hue angle; SS soluble solids content; TA titratable acidity; Car carotenoid content; βcar β-carotene content; γcar γ-carotene content; Lyc lycopene content; Cryp cryptoxanthin content; Xan xanthophyll content

Vg additive genetic variance (environmental + non-additive); Vf individual phenotypic variance; Acprog accuracy of progeny selection; h restricted heritability; CVg_t% coefficient of individual additive genetic variation; CVgp% coefficient of genotypic variation between progenies; CVe% coefficient of environmental variation; CVr relative coefficient of variation; PEV prediction error variance of the genotypic values of the progeny; SEP standard error of prediction of genotypic value of the progeny

Table 4 Predicted genetic values (BLUPs) of the physical characteristics of the fruits of the genotypes selected in the first production cycle

Selected genotype	M (kg)	LF (cm)	LD (cm)	SD (cm)	ST (mm)	PT (mm)	CD (cm)	Lc	Cc	Hc	Lp	Cp	Hp
T12P2B1	5.18	76.29	16.88	8.78	3.33	74.97	12.80	65.62	33.10	59.72	64.54	68.37	63.23
T11P3B1	6.07	159.05	2.84	9.35	5.71	67.85	12.84	23.33	34.38	60.41	65.12	68.35	63.64
T12P4B3	4.88	40.75	6.40	8.58	1.72	70.68	11.73	58.47	34.72	78.04	63.60	68.11	62.01
T10P1B1	5.45	61.17	8.83	9.58	5.51	28.88	12.66	35.66	35.17	53.58	66.80	66.98	63.78
T7P5B2	5.10	4.72	15.53		5.00	10.53	11.20	90.92	32.84	93.90	66.27	70.46	65.07
T2P3B2	5.43	129.48	2.04	11.51	4.61	43.65	11.04	50.24	33.96	63.40	66.78	65.53	65.40
T12P5B2	5.05	1.23	18.91	8.72	3.79	25.58	12.40	68.58	33.57	65.11	65.28	67.53	63.37
T6P3B3	5.15	18.01	14.30	9.17	5.94	23.78	11.89	68.75	32.10	60.10	64.91	69.94	64.31
T12P1B3	4.89	37.68	9.01	8.58	2.42	100.00	11.63	71.97	33.88	73.82	65.60	68.30	63.48
T12P4B1	5.17	30.30	17.54	8.94	4.18	42.07	12.75	45.73	34.78	65.95	64.38	67.46	62.38
T3P3B3	6.96	7.33	19.09		4.29	28.59	11.81	56.43	35.32	71.33	66.76	68.48	64.76
T4P2B3	6.10	2.57	18.81		4.24	16.20	11.36	101.87	33.83	117.59	66.24	68.19	64.23
T7P1B2	5.09	-2.43	15.20		2.97	6.48	11.37	79.65	32.46	80.31	65.80	69.75	64.44
T8P5B3	4.98	4.61	16.70	4.17	12.81	12.84	80.28	34.66	82.22	66.16	69.29	64.53	
T5P5B3	8.10	15.51	17.14	8.51	4.02	17.94	12.45	79.78	36.08	90.17	65.11	68.41	63.68
T6P4B3	5.16	5.74	19.53	2.91	32.67	11.96	83.41	33.52	92.03	65.17	69.71	64.11	
T7P4B2	5.09	-5.50	11.61	3.95	18.12	11.03	87.45	33.26	92.57	66.28	70.14	64.56	
T8P2B1	5.20	6.70	21.81	5.95	25.19	12.57	77.54	35.44	105.63	67.93	70.30	66.67	
T12P5B3	4.90	-3.20	13.58	8.83	3.28	30.74	11.84	73.47	34.55	78.49	64.50	68.48	63.19
Averages	5.47	31.05	13.99	9.14	4.10	35.62	12.01	68.38	34.09	78.13	65.64	68.62	64.04

M mass; *LF* length; *LD* largest diameter; *SD* smallest diameter; *ST* skin thickness; *PT* pulp thickness; *CD* cavity diameter; *Ls* skin luminosity; *Cs* skin chroma; *Hs* skin Hue Angle; *Lp* pulp luminosity; *Cp* pulp chroma; *Hp* pulp Hue Angle

are the two main objectives of the pumpkin breeding program in this study, the responses represent prospects for progress in the next stages using the selected genotypes.

In the second production cycle, according to the variables analyzed and by applying the Mulamba and Mock method, the 19 best individuals were ranked. Considering a selection intensity of 10%, a new ranking was obtained based on the variables M, LF, LD, SD, ST, PT, CC, Lc, Cc, Hc, Lp, Cp, Hp, SS, TA, ascorbic acid (AA) content and pulp firmness (PF) to obtain genotypes with better functional and commercial qualities (Table 7). The contribution of ascorbic acid, a bioactive molecule, to the fruit quality of the selected genotypes was determined.

Among the variables analyzed, genotype 15 had the best individual genotype (Table 7 and Fig. 2). In particular, it had greater average pulp thickness, pulp firmness and soluble solids content, as well as orange pulp.

In turn, genotypes 4 and 131 were characterized by low fruit mass, cavity diameter and pulp hue angle values. Furthermore, genotype 4 had a distinct pulp firmness (119.0 N), while genotype 131 had the highest ascorbic acid content (1411.8 $\mu\text{g g}^{-1}$). The most intense orange pulp color was observed in genotype 124, corresponding to the lowest H value (61.4).

Discussion

A species that exhibits superiority in terms of various attributes of interest while still exhibiting an increase in yield, nutritional value, and availability to consumers and producers is highly important in breeding programs (Dwivedi et al. 2019). Pumpkin has several of these attributes, and other distinguished agronomic and fruit quality characteristics could be added to its current performance through consistent and long-lasting breeding. These actions increase its inclusion in the population's diet.

Table 5 Predicted genetic values (BLUPs) of the chemical characteristics of the fruits of the genotypes selected in the first production cycle

Selected genotype	SS (°Brix)	TA (%)	Car (µg.g ⁻¹)	βcar (µg.g ⁻¹)	γcar (µg.g ⁻¹)	Lyc (µg.g ⁻¹)	Cryp (µg.g ⁻¹)	Xan (µg.g ⁻¹)
T12P2B1	15.60	0.03	558.65	413.10	1.80	nd	0.19	1.02
T11P3B1	15.56	0.03	683.13	591.99	2.94	1.94	0.19	0.79
T12P4B3	15.55	0.03	720.44	544.13	1.76	2.82	0.19	1.07
T10P1B1	15.53	0.03	542.33	442.28	2.56	2.27	0.18	0.56
T7P5B2	15.56	0.03	574.97	513.80	3.21	2.24	0.20	1.99
T2P3B2	15.53	0.02	490.82	346.34	2.91	1.56	0.17	1.06
T12P5B2	15.62	0.03	518.13	452.58	2.04	3.72	0.19	1.99
T6P3B3	15.49	0.03	488.48	390.45	3.10	1.62	0.18	0.96
T12P1B3	15.53	0.03	469.35	392.33	1.69	0.43	0.17	1.86
T12P4B1	15.61	0.04	565.46	529.64	2.14	3.19	nd	0.98
T3P3B3	15.51	0.03	407.66	368.57	2.11	0.76	0.18	0.49
T4P2B3	15.50	0.03	491.66	413.72	3.14	1.09	0.23	0.72
T7P1B2	15.56	0.02	453.71	383.57	2.14	2.24	0.17	1.20
T8P5B3	15.48	0.02	438.98	395.82	2.56	1.71	0.21	1.05
T5P5B3	20.23	0.03	586.61	456.66	2.09	1.03	0.20	0.77
T6P4B3	15.50	0.02	474.71	369.74	2.71	1.36	0.20	0.67
T7P4B2	15.56	0.02	421.77	291.68	2.21	2.15	0.19	1.57
T8P2B1	15.53	0.03	527.48	428.83	2.42	1.35	0.19	0.40
T12P5B3	15.52	0.03	547.39	407.67	2.04	1.04	0.18	1.47
Averages	15.79	0.03	524.30	428.05	2.40	1.81	0.19	1.08

SS soluble solids content; TA titratable acidity; Car carotenoid content; βcar β-carotene content; γcar γ-carotene content; Lyc lycopene content; Cryp cryptoxanthin content; Xan xanthophyll content
nd not detected

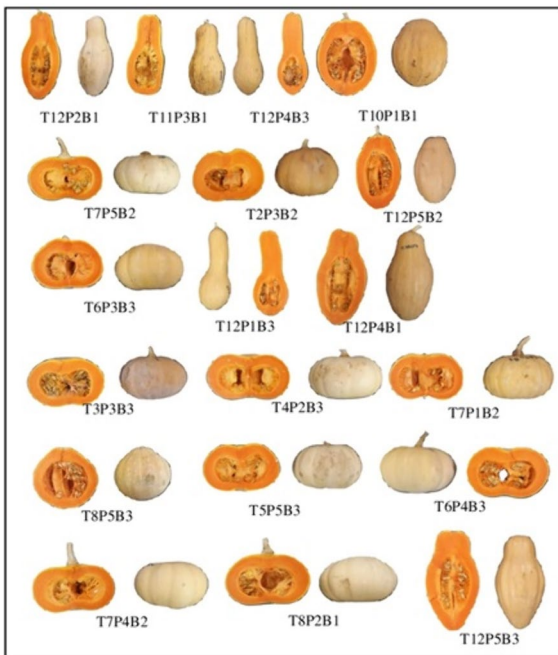


Fig. 1 Visual appearance of the 19 best pumpkin fruits based on the ranking carried out using selegen and excel software

By using the recurrent selection method, gains can be made in attributes with quantitative inheritance, resulting in a population that is superior to the original population and providing better performance for individuals who have been selected for a new production cycle (Ribeiro et al. 2020). In plant breeding programs, the use of selection indices is a very common strategy, with the aim of mitigating negative correlations between characters (Costa et al. 2023; Cruz and Regazzi 2002). In addition to the selection index, the use of genetic parameters, such as h, Vf and Vg, is important because they provide information on the degree to which a character can be transmitted in successive generations of the species (Bello et al. 2012).

In this study, the absence of negative values for Vg, as shown in Table 3, indicates the possibility of success in selecting superior individuals for the characters studied (Hallauer et al. 2010). However, carrying out more production cycles should result in uniform lines exhibiting characters with lower values of genetic variance. On the other hand, environmental control of variables with high Vf values should determine the potential for expression of the trait.

Table 6 Prediction of genetic gains (GG) in the first production cycle of pumpkin genotypes

Characteristic	GG (%)
Mass—M (kg)	-7.8735
Length—LF (cm)	90.84635
Largest diameter—LD (cm)	-33.7191
Smallest diameter—SD (cm)	1.512999
Skin thickness—ST (mm)	-7.82802
Pulp thickness—PT (mm)	29.78796
Cavity diameter—CD (cm)	-1.49919
Skin luminosity—Ls	-10.2297
Skin chroma—Cs	-0.49203
Skin Hue angle—Hs	-5.45927
Pulp luminosity—Lp	-1.01002
Pulp chroma—Cp	0.108729
Pulp Hue Angle—Hp	-1.4591
Soluble solids content—SS (°Brix)	1.027316
Titrate acidity – TA (% citric acid)	3.367349
Carotenoid content—Car ($\mu\text{g.g}^{-1}$)	36.67167
β -carotene content— βcar ($\mu\text{g.g}^{-1}$)	49.34908
γ -carotene content— γcar ($\mu\text{g.g}^{-1}$)	18.4968
Lycopene content—Lyc ($\mu\text{g.g}^{-1}$)	58.58262
Cryptoxanthin content—Cryp ($\mu\text{g.g}^{-1}$)	1.720028
Xanthophyll content—Xan ($\mu\text{g.g}^{-1}$)	68.96616

According to Borges (2019a), h reflects the proportion of Vf that is inherited and determines how reliably genetic values affect phenotypic values. Heritability in the strict sense, which is of most interest to breeders, considers only the additive genetic variance fixed by selection (Dudley and Moll 1969). A high estimate of heritability combined with high genetic advance provides a good approach for further improvement in future selections (Singh et al. 2019). Studying melon accessions, Amorim et al. (2023) related high heritabilities to attributes such as fruit mass, fruit length and cavity length, denoting the potential for success in future selections.

According to the reported restricted heritability (h) for the pumpkin genotypes, Ls and Xan had values lower than 0.1, indicating that the environment had a greater influence on the heritability (Table 3). For the variables M, CD, Cs and SS, h values ranging from 0.15 to 0.38 still indicate the predominant influence of the environment. However, for variables such as LF, LD, SD, ST, PT, Hs, Lp, Cp, Hp, TA, Car, βcar , γcar , Lyc and Cryp, the genotypic influence was

greater than the environmental influence (Table 3). Restricted heritability values greater than 0.5 are considered high (Resende 1995), thus suggesting that it is possible to carry out new selections with good genetic gains. Studying the genetic variability in *C. maxima* accessions, Barboza et al. (2023) also highlighted the potential for advances in genetic breeding, including programs aimed at biofortification.

The quality of a genotypic assessment is assessed using the accuracy parameter (Acprog). This parameter expresses the correlation between the real genotypic value and the predicted value according to information resulting from field experiments. The smaller absolute deviations in the values of these two variables represent a greater precision (Resende and Duarte 2007). According to Paranhos et al. (2022), the higher the accuracy value, the greater the confidence in the assessment and genetic value of the individual.

To be considered accurate, the Acprog value must be greater than 0.5 (Cortes et al. 2019). In this study, except for the variables M and SS, the other variables had Acprog values greater than 0.5, indicating the secondary influence of environmental conditions on these traits (Table 3).

The phenotypic coefficient of variation and genotypic coefficient of variation values indicated that the influence of the environment was greater for the M, SD, DC, Cs, Lp, TA, Car, βcar and Cryp attributes (Table 3). In this case, it can be concluded that the traits exhibit large variation, and genetic gains can be achieved through a greater number of selection cycles (Ramjan 2021). In contrast to the results obtained in this first production cycle, Ramjan (2021) mentioned slight differences in the environmental effects on the physical characteristics and SS, carbohydrate and carotenoid contents of pumpkins. For example, the coefficients of environmental variation (C_{Ve}) and genotypic variation between progenies (C_{Vgp}) for fruit mass observed by the authors were 24.42% and 23.58%, respectively. In our study, values of 130.37% and 38.91% for C_{Ve} and C_{Vgp}, respectively, were estimated. Further selections are necessary to obtain the desired results.

Singh et al. (2019) mentioned that the variables analyzed in pumpkin plants had a greater coefficient of phenotypic variation than genotypic variation, indicating differences in the experimental material and the environment in which the experiment was

Table 7 Average values of the physical and chemical characteristics of the fruits of the genotypes selected in the second pumpkin production cycle obtained using the method recommended by Mulamba and Mock and a selection index of 10%

Geno-type	M (kg)	LF (cm)	LD (cm)	SD (cm)	ST (mm)	PT (mm)	CD (cm)	Ls	Cs	Hs	Lp	Cp	Hp	SS (°Brix)	TA (%)	AA ($\mu\text{g g}^{-1}$)	PF (N)
15	7.5	26.1	25.18	0	3.6	50.0	13.4	72.1	38.2	80.2	66.9	71.8	67.6	13.4	0.02	821.8	99.1
5	4.6	26.3	17	12.6	4.0	36.7	13.4	63.3	35.7	72.2	66.9	67.3	67.1	13.5	0.02	971.7	112.4
62	3.5	22.5	18.3	12.1	4.6	35.8	11.7	71.0	39.2	79.9	65.3	76.1	66.0	11.1	0.01	1059.3	92.5
83	2.8	25.2	17.6	10.6	4.6	44.8	11.3	57.9	28.5	65.4	66.4	68.1	64.8	12.5	0.02	1058.8	72.7
116	2.7	23	16.6	10.1	8.1	40.5	10.8	69.8	38.7	75.5	65.9	70.7	64.1	11.5	0.01	1306.5	85.9
131	2.6	18.8	17.5	10	4.6	32.3	10.2	61.0	27.8	62.7	68.0	71.7	66.8	11.6	0.01	1411.8	52.9
173	6	13	26.5	0	4.0	37.5	11.1	83.2	30.2	88.9	70.4	65.5	68.2	8.8	0.02	788.0	59.5
53	4	26.7	18.5	12.9	3.3	45.8	12.0	71.4	25.9	70.3	65.3	71.3	66.1	9.5	0.01	961.0	46.2
141	2.9	26	16.9	10.1	4.6	40.1	12.0	69.4	32.2	71.8	66.9	71.3	65.9	10.5	0.01	1212.3	59.5
115	3.4	21.8	18.5	12.5	4.6	44.4	10.8	70.1	33.2	74.1	68.9	71.3	67.6	11.3	0.02	1177.3	46.2
124	2.9	23.4	17	11.9	3.8	31.0	13.3	70.3	23.2	67.3	61.9	70.0	61.4	12.5	0.02	984.3	105.7
107	2	23	15	8.6	3.8	39.8	10.4	74.2	30.1	75.6	65.0	70.7	63.6	13.0	0.03	987.5	99.1
4	2.4	19.5	18.4	0	4.1	41.4	10.9	70.8	40.8	76.4	67.6	72.1	66.5	8.5	0.01	753.3	119.0
165	3.7	22.8	18.4	12	3.1	32.7	13.0	78.0	27.7	78.5	64.9	73.8	65.4	12.2	0.02	964.0	85.9
42	3.2	23.5	16.3	12	4.6	36.6	12.3	65.4	32.6	71.8	65.4	73.0	66.3	14.3	0.02	1036.8	72.7
144	3.1	25.3	17.4	11	5.8	40.4	11.5	65.4	36.0	72.6	64.6	71.1	64.1	12.3	0.02	1067.0	66.1
109	4.3	22.5	21.3	0	4.0	43.5	11.8	67.5	36.1	72.5	69.3	69.4	68.7	9.9	0.01	976.3	59.5
76	2.9	23.5	17	10.2	4.5	28.5	13.5	68.5	35.5	76.1	63.9	69.0	63.3	15.6	0.02	889.5	112.3
103	2.8	18.2	19.6	0	3.5	27.3	12.0	71.5	35.4	76.7	64.0	71.5	64.3	13.0	0.02	1098.8	99.2
Avera-ge	3.6	22.9	18.5	8.7	4.4	39.0	11.8	69.4	32.9	74.0	66.3	70.8	62.4	11.8	0.02	1027.7	80.4

M mass; *L* length; *LD* largest diameter; *SD* smallest diameter; *ST* skin thickness; *PT* pulp thickness; *CD* cavity diameter; *Ls* skin luminosity; *Cs* skin chroma; *Hs* skin Hue Angle; *Lp* pulp luminosity; *Cp* pulp chroma; *Hp* pulp Hue Angle; *SS* soluble solids content; *TA* titratable acidity; *AA* ascorbic acid content; *PF* pulp firmness

conducted. Sahoo and Singh (2020) and Kumari et al. (2020) also observed a dominant environmental influence over the genetic influence in cucumbers, although the difference was considered to be restricted. Therefore, if selection was based on phenotypic performance, the results were considered reliable.

Among the variables with high genetic coefficients, such as those obtained for LF, LD, ST, PT, Ls, Hs, γcar , Lyc and Xan, there is a greater possibility of fixing the trait through selection (Georgieva et al. 2016). For the coefficient of residual variation, a value of 1 indicates that the genetic variance within the progeny is zero. In turn, coefficient values greater than 1, as observed for LF, LD, ST, PT, Ls, Hs, γcar , Lyc and Xan (Table 3), indicate favorable conditions for selection (Vencovsky 1987).

With the information obtained from the selected 19 best individuals, some negative predicted genetic gains were observed for some attributes, such as M,

LD, ST, CD, Ls, Cs, Hs, Lp and Hp (Table 6). In some cases, negative gains are desirable. For example, small pumpkin fruits could be obtained to meet consumer demand. This condition was also observed in the study carried out by Amaro et al. (2022), in which the pumpkins had a mass between 2 and 4 kg, which is considered the closest to the standard of Maranhão type pumpkin fruits (4–8 kg, external orange-cream color and orangish internal color). Among the fruits of the genotypes selected from the second production cycle (population s1), even genotype 15, which was characterized by the highest average fruit mass (7.5 kg), fell within the Maranhão type standard (Table 7). However, eleven of the selected genotypes had masses between 2 and 3 kg, corresponding to the Jacararezinho type (2–4 kg, with dark green skin and orangish pulp), which is of interest to current consumers.

According to Barbosa (2009), fruits with thinner skin are more susceptible to mass loss during

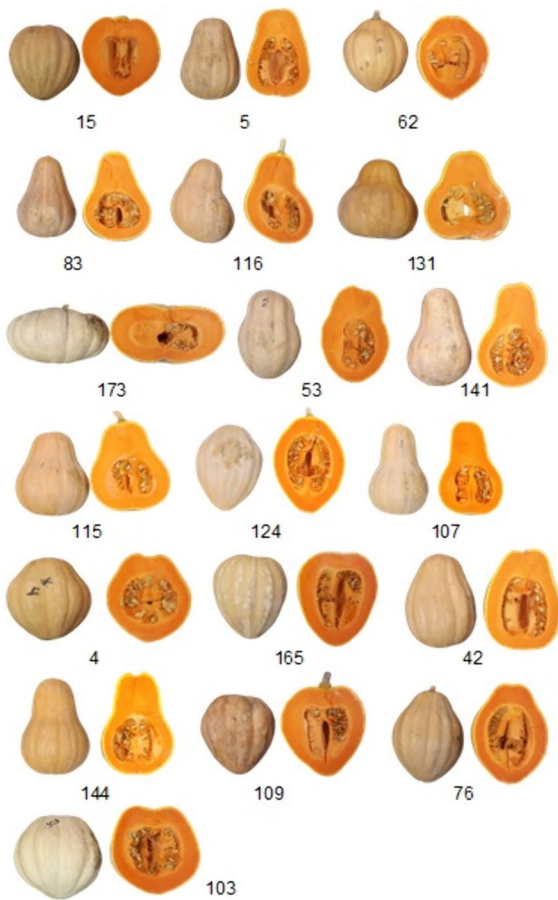


Fig. 2 Visual appearance of the 19 best pumpkin fruits based on ranking via the mulamba and mock method

storage due to their susceptibility to damage and water loss. A skin thickness ranging from 8.4 to 9.3 mm can be more advantageous for longer storage periods and does not affect the pulp yield (Amaro et al. 2014; Priori et al. 2018). In the second production cycle, the ST values ranged from 3.1 to 8.1 mm (Table 7), suggesting the possibility of obtaining new populations to meet these standards.

In the case of CD, the smaller the cavity was, the greater the presence of pulp. If selection is based on fruit with more seeds, then a larger cavity would be appropriate. However, the purpose of selection in this study was to obtain fruit with smaller cavities, based on consumer interest (Barbosa 2009). Among the genotypes evaluated in this study, the CD values ranged from 10.4 to 13.5 cm and were similar to those observed by Borges et al. (2019b), who

evaluated progenies of pumpkin accessions from the Active Germplasm Bank of Cucurbits at Embrapa Semiárido.

Pulp thickness is an important trait in pumpkin breeding programs (Blank et al. 2013). Fruits with greater pulp thickness have a higher pulp yield, which is an important commercial characteristic. In the case of SD, pumpkins destined for the fresh market must have a shape that meets consumer preferences, such as flat fruits, corresponding to a lower SD, and the absence of a neck.

According to Chitarra and Chitarra (2005), there is a relationship between fruit length and diameter, such that values less than 1 indicate globular, ellipsoid and spherical shapes, while higher values indicate elongated fruit shapes. According to the data in Table 7, the LF measurements of the selected genotypes ranged from 13 to 26.7 cm, the LD measurements ranged from 15 to 25.18 cm, and the SD measurements ranged from 0 to 12.6 cm.

In *C. pepo*, El-maaty and Ashour (2021) observed M and PT ranging from 512.4 to 733.1 g, with an average of 590.74 g, and from 8 to 12 mm, with an average of 9.75 mm, respectively. The PT of the fruits produced in the 2nd cycle ranged from 28.5 to 50 mm, representing genotypes 76 and 15, respectively (Table 7). Borges et al. (2019b) observed values ranging from 21.30 to 35.10 mm among the selected pumpkin genotypes, which is considered desirable due to the high pulp yield.

Environmental variation was responsible for most of the phenotypic variation in fruit color (Table 6). According to Costa et al. (2023), the environmental influence on these attributes is determined mainly by quantitative variables. However, for Hs and Hp, negative genetic gains represented lower values of the variables in subsequent stages of selection, indicating the generation of fruits with an intense orange color. This color is represented by H values close to 30°.

The orange color of pumpkin fruit is well appreciated by consumers and is associated with the presence of carotenoids. Table 5 shows an average carotenoid content of 524.30 $\mu\text{g}\cdot\text{g}^{-1}$ in the selected fruits, with the highest values being 720.44 $\mu\text{g}\cdot\text{g}^{-1}$ and 683.13 $\mu\text{g}\cdot\text{g}^{-1}$, which indicate a high presence of carotenoids. Ribeiro et al. (2020) reported total carotenoid contents ranging from 52.65 to 155.78 $\mu\text{g}\cdot\text{g}^{-1}$ in 51 accessions of *C. moschata*.

Kostecka-Gugała et al. (2020) reported that the highest concentrations of β -carotene ranged from 125–146 $\mu\text{g g}^{-1}$ f.w. in pumpkin cultivars, such as ‘Indomatrone’, ‘Australian Butter’, ‘Chicago Warded Hubbard’ (*C. maxima*), ‘Sweet Dumpling’ and ‘Table Gold’ (*C. pepo*), as well as ‘Kogigu’ and ‘Shishigatani’ (*C. moschata*). Among them, ‘Musquée de Provence’ (*C. moschata*) and ‘KamoKamo’ (*C. pepo*) cultivars had the lowest β -carotene contents, showing 5 and 19 $\mu\text{g g}^{-1}$ f.w., respectively. Considering the results from scientific literature, it is possible to recognize wide opportunities for future selections of pumpkin fruits of superior quality, including functional properties, using the selected genotypes in this study.

Fruits with lower TA contents are generally preferred by consumers, and pumpkin is not considered an acidic vegetable. In the second productive cycle, the TA values ranged from 0.012 to 0.025% citric acid (Table 5), representing a lower acidity than that of the first cycle. The results observed in this study are considered acceptable and coherent for the species since it has a nonacidic flavor.

Otherwise, high values of SS are more desirable if the fruits will be processed (Amaro et al. 2018). For the fresh market, pumpkins with SS contents between 11 and 13° Brix are considered to be of high quality (Loy 2004). The results observed in the second production cycle of this study ranged from 8.5 to 15.6°Brix (Table 7). However, it is possible to observe pumpkin fruits with 18.4°Brix, for example (Amaro et al. 2017).

The mean ascorbic acid (vitamin C) contents of the fruits of the genotypes selected from population s1 ranged from 753.3 to 1411.8 $\mu\text{g g}^{-1}$ (Table 7). This vitamin is one of the most sensitive to losses in food under high temperature and oxidation conditions. Therefore, preserving antioxidant properties is a challenge, especially after fruit harvest. According to the studies by De Assis Lima et al. (2020), the values of this variable ranged from 1143.2 to 1263.0 $\mu\text{g g}^{-1}$ during the two production cycles. In addition, Oloyede and Ogunsina (2023) highlighted that the longer the fruit was stored, the lower the ascorbic acid content was.

The pulp firmness of the genotypes selected in the second cycle ranged from 46.2 to 119.0 N (Table 7). These results are similar to those obtained by Monalisa et al. (2020), who reported values of 73.1 N, and

El-maaty and Ashour (2021), who reported values of 78 N cm^{-2} . According to the authors, pulp firmness plays a key role in consumer acceptance and market value. There have been reports of pulp firmness values up to 165,8 N, as reported by Theanjumpol and Maniwaru (2022). To guarantee pulp firmness and meet consumer preferences, storage at mild temperatures is recommended in addition to careful handling (De Moura Guerra et al. 2020).

Even though it is necessary to carry out others production cycles with reduced environmental influence, the results in this study highlight the high potential of the selected genotypes for the development of cultivars with different quality characteristics. Moreover, there are opportunities to develop cultivars that will serve different markets.

Conclusion

Based on the genetic parameters and predicted gains, promising individuals with the desired fruit qualities were selected. Based on predicted gains, fruits with characteristics of interest, including length and pulp thickness, and especially carotenoid, β -carotene, lycopene and xanthophyll contents, which indicate their functional properties, were selected. In the first production cycle, fruit T12P2B1 was the first in the ranking due to its quality characteristics, such as diameter, pulp hue angle, soluble solids, carotenoids and β -carotene content, although its shape was not considered desirable by the consumer. Nevertheless, other genotypes characterized by fruits with equivalent diameter, pulp hue angle, soluble solids, carotenoids and β -carotene content exhibited a shape suitable for consumer preference. In the second production cycle, genotype 15 had the greatest fruit mass, diameter, pulp thickness and skin luminosity. Additionally, other genotypes produced firmer fruits with high ascorbic acid content and an orange pulp hue angle. All of these genotypes can be used in the next phases of the breeding program.

Acknowledgements The authors are grateful for all the help obtained in research activities. The partnership between the Brazilian Agricultural Research Corporation (EMBRAPA), the Coordination for the Improvement of Higher Education Personnel—Brazil (CAPES) and the Federal Rural University of Pernambuco (UFRPE) was essential for the success of this study.

Author contributions The preparation of the material, data collection and analysis were performed by W.M.D.B., B.F.R.S. and T.G.B.S.O. The first version was developed by W.M.D.B. with the contribution of the authors, M.A.C.L., C.S.R.C., R.M.E.B. and A.F.M.J., both in writing and in correction and editing. All authors have read and agreed to the published version of the manuscript.

Funding This study was supported by the Brazilian Agricultural Research Corporation (EMBRAPA) and the Coordination for the Improvement of Higher Education Personnel—Brazil (CAPES) [research grant for the first author]. The sponsors had no role in the execution of the project and the decision to submit the article for publication.

Data availability No datasets were generated or analysed during the current study.

Declarations

Competing interest The authors declare no competing interests.

References

- Amaro GB, Carmona PAO, Carvalho ADF, Lopes JF, Coimbra KG (2014) Desempenho de híbridos de abóboras e morangas avaliados no Distrito Federal. *Hortic Bras* 31:1916–1923
- Amaro GB, Silva GO, Boiteux LS, Carvalho ADF, Lopes JF (2017) Desempenho agrônomo de híbridos experimentais de abóbora Tetsukabuto para características dos frutos. *Hortic Bras* 35(2):180–185. <https://doi.org/10.1590/s0102-053620170205>
- Amaro, GB, Resende, F, de Carvalho, ADF, Botrel, N, Lopes, J (2018) Desempenho de híbridos de abóbora japonesa no sistema orgânico. *Embrapa Hortaliças*
- Amaro GB, da Silva GO, Maldonado IR, Madeira NR, Faustino RMEB (2022) Desempenho de genótipos de abóboras e morangas para produtividade e qualidade de frutos. *Cadernos De Ciência Tecnol* 39:27075. <https://doi.org/10.35977/0104-1096.cct2022.v39.27075>
- Amorim CC, de Queiroz MA, Barbosa BL, Coutinho MDS, Lima Neto IDS (2023) Genetic parameters in melon sub-accessions from traditional agriculture. *Revista Caatinga* 36:320–328. <https://doi.org/10.1590/1983-21252023v36n209rc>
- AOAC (2010) Association of official agricultural chemists. Official methods of analysis of the Association of the Agricultural Chemists. 18th edn. 3. Gaithersburg: AOAC, pp 957
- de Assis Lima A, Freitas ATC, Sá DMAT, Andrade FJET, Farias MDP, Moraes GMD (2020) Abóbora minimamente processada e revestida com galactomanana extraída da semente de *Caesalpinia pulcherrima*. *Brazilian Journal of Development* 6:12479–12488. <https://doi.org/10.34117/bjdv6n3-202>
- Barbosa, GS (2009) Desempenho agrônomo, caracterização morfológica e polínica de linhagens de abóbora (*C. moschata*) com potencial para o lançamento de cultivares. Pos-Graduate Dissertation Campo dos Goitacazes: Universidade Estadual do Norte Fluminense Darcy Ribeiro – UENF
- Barboza M, Lima FLDS, Ribeiro RMP, Pereira CCA, Santos MGD, Silveira FPD, Nascimento AKQD, Passos ABRDJ, Chagas PSFD, Ribeiro JEDS, Lima JVL, Júnior JTS, Nunes GHS, da Silveira IM, Júnior APB (2023) Determination of Total Carotenoids and β -Carotene in Germplasm of Pumpkin Caboclo (*Cucurbita maxima*). *Agronomy* 13:1109. <https://doi.org/10.3390/agronomy13041109>
- Bello OB, Ige SA, Azeez MA, Afolabi MS, Abdulmalik SY, Mahamood J (2012) Heritability and genetic advance for grain yield and its component characters in maize (*Zea mays* L). *Int J Plant Res* 2:138–145
- Bergantin C, Maietti A, Tedeschi P, Font G, Manes L, Marchetti N (2018) HPLC-UV/Vis-APCI-MS/MS determination of major carotenoids and their bioaccessibility from “Delica” (*Cucurbita maxima*) and “Violina” (*Cucurbita moschata*) pumpkins as food traceability markers. *Molecules* 23:2791. <https://doi.org/10.3390/molecules23112791>
- Blank AF, Silva TB, Matos ML, Carvalho Filho JLS, Silva-Mann R (2013) Parâmetros genotípicos, fenotípicos e ambientais para caracteres morfológicos e agrônomo em abóbora. *Hortic Bras* 31:106–111. <https://doi.org/10.1590/S0102-05362013000100017>
- Borém, A, Miranda, GV (2005) Melhoramento De Plantas, 5nd edn. UFV, Vicosia, pp 525.
- Borges RME, de Lima MAC, da Silva Lima Neto I, de Melo NF (2019a) Prediction of genetic and selection parameters in pumpkin (*Cucurbita moschata* Duch) progenies for morphoagronomic characteristics and pulp quality. *Australian J Crop Sci* 13:199–207
- Borges RME, Coêlho MA, de Lima N, de Melo F (2019b) Correlations between fruit yield and sensory and nutritional quality of pumpkin (*Cucurbita moschata* Duch.). *Australian J Crop Sci* 13(10):1676–1682. <https://doi.org/10.21475/ajcs.19.13.10.p1914>
- Chitarra, MIF, Chitarra, AB (2005) Pós-colheita de frutas e hortaliças: Fisiologia e manuseio. Lavras: Editora UFPA, 1:785
- Cortes DFM, Santa-Catarina R, Vettorazzi JCF, Ramos HCCR, Viana AP, Pereira MG (2019) Development of superior lines of papaya from the Formosa group using the pedigree method and REML/Blup procedure. *Bragantia* 78:350–360. <https://doi.org/10.1590/1678-4499.20180253>
- Costa CDSR, de Lima MAC, Neto FPL, da Silva Costa AE, Vilvert JC, Martins SS, dos Santos Musser R (2023) Genetic parameters and selection of mango genotypes using the FAI-BLUP multitrait index. *Sci Horticulturae* 317:112049. <https://doi.org/10.1016/j.scienta.2023.112049>
- Cruz CD (2013) Genes: software para análise de dados em estatística experimental e em genética quantitativa *Acta Scientiarum. Agronomy* 35:271–276. <https://doi.org/10.4025/actasciagron.v35i3.21251>

- Cruz CD, Regazzi AJ (2002) Modelos biométricos aplicados ao melhoramento genético, 2nd edn. Imprensa Universitária, Viçosa, p 390
- Darrudi R, Nazeri V, Soltani F, Shokrpour M, Ercolano MR (2018) Evaluation of combining ability in *Cucurbita pepo* L and *Cucurbita moschata* Duchesne accessions for fruit and seed quantitative traits. *Journal of Appl Res Med Aromatic Plants* 9:70–77. <https://doi.org/10.1016/j.jarmap.2018.02.006>
- Dudley JW, Moll RH (1969) Interpretation and use of estimates of heritability and genetic variances in plant breeding. *Crop Sci* 9:257–262
- Dwivedi S, Goldman I, Ortiz R (2019) Pursuing the potential of heirloom cultivars to improve adaptation, nutritional, and culinary features of food crops. *Agronomy* 9:441. <https://doi.org/10.3390/agronomy9080441>
- El-maaty A, Ashour E (2021) Determination of some physical and mechanical properties of summer squash (*Cucurbita pepo*) related to post-harvest processes. *Misr J Agric Eng* 38(4):363–376
- EMBRAPA SEMIÁRIDO (2023) Estação Agrometeorológica de Bebedouro - Estação Bebedouro (accessed by July 15, 2023)
- Georgieva, N, Nikolova, I, Kosev, V (2016) Evaluation of genetic divergence and heritability in pea (*Pisum sativum* L.). *Journal of BioScience & Biotechnology* 5
- Grassino AN, Brčić SR, Sabolović MB, Sicžlabur J, Marović R, Brčić M (2023) Carotenoid content and profiles of pumpkin products and by-products. *Molecules* 28:858
- Hallauer AR, Miranda Filho JB, Carena MJ (2010) Breeding Plans. In: Carena MJ, Hallauer AR, Miranda Filho JB (eds) *Quantitative Genetics in Maize Breeding*. Springer New York, New York, pp 577–653. https://doi.org/10.1007/978-1-4419-0766-0_12
- Han X, Min Z, Wei M, Li Y, Wang D, Zhang Z, Hu X, Kong Q (2022) QTL mapping for pumpkin fruit traits using a GBS-based high-density genetic map. *Euphytica* 218:106. <https://doi.org/10.1007/s10681-022-03057-1>
- Hegde SG, Duhan DS, Nayana KR (2024) Genetic variability, correlation and path analysis in f3 and f4 generation of bottle gourd [*Lagenaria siceraria* (Mol.) Standl.]. *J Adv Biol Biotechnol* 27:1028–1041. <https://doi.org/10.9734/jabb/2024/v27i71063>
- Janani N, Patel MD, Nihaluddin SMD, Sharma S (2018) A review of xerophthalmia due to vitamin-A deficiency with ayurvedic management. *World J Pharm Res* 8:1685–1691
- Kostecka-Gugała A, Kruczek M, Ledwożyw-Smoleń I, Kaszycki P (2020) Antioxidants and health-beneficial nutrients in fruits of eighteen *Cucurbita* cultivars: analysis of diversity and dietary implications. *Molecules* 25:1792. <https://doi.org/10.3390/molecules25081792>
- Kumar R, Rajasree V, Sagar I, Hameedi A, Sheera A, Naga VK, Ferooz K, Pir HA (2022) Genetic variability study for yield and quality traits in pumpkin (*Cucurbita moschata* Duch Ex Poir). *The Pharm Innovat J* 11:1019–1021
- Kumari M, Ram CN, Nath S, Maurya N, Kumar S (2020) Studies on genetic variability, heritability and genetic advance in cucumber (*Cucumis sativus*). *J Pharm Phytochem* 9:481–484
- Latheef A, Pugalendhi L, Rani A, Jeyakumar P, Kumar M, Devi M (2022) Genetic analysis of mango (*Mangifera indica* L) genotypes for year-round flowering and yield characters. *Madras Agric J* 109:23–27. <https://doi.org/10.29321/MAJ.10.000573>
- Loy JB (2004) Morpho-physiological aspects of productivity and quality in squash and pumpkins (*Cucurbita* spp). *Crit Rev Plant Sci* 23:337–363. <https://doi.org/10.1080/07352680490490733>
- Martínez-Valdivieso D, Gómez P, Font R, Alonso-Moraga A, Del Rio-Celestino M (2015) Physical and chemical characterization in fruit from 22 summer squash (*Cucurbita pepo* L) cultivars. *LWT-Food Sci Technol* 64:1225–1233. <https://doi.org/10.1016/j.lwt.2015.07.023>
- Miglioli TC, Fonseca VM, Gomes Junior SC, da Silva KS, de Lira PIC, Batista Filho M (2015) Factors associated with the nutritional status of children less than 5 years of age. *Rev Saude Publica* 49:49–59. <https://doi.org/10.1590/S0034-8910.2015049005441>
- Mokhtar M, Bouamar S, Di Lorenzo A, Temporini C, Daglia M, Riazi A (2021) The influence of ripeness on the phenolic content, antioxidant and antimicrobial activities of pumpkins (*Cucurbita moschata* Duchesne). *Molecules* 26:3623. <https://doi.org/10.3390/molecules26123623>
- Monalisa K, Bhuiyan J, Islam M, Sayem A (2020) Boiling-induced changes on physicochemical, bioactive compounds, color, and texture properties of pumpkin (*Cucurbita maxima*). *Food Sci Technol Int* 26:333–343. <https://doi.org/10.1177/1082013219894402>
- de Moura Guerra AMN, dos Santos DS, Silva MGM, Evangelista RS, Maia AG, da Silva IG, Medeiros AC, Maracaja PB (2020) PVC film coatings promote post-harvest conservation of Italian zucchini fruits (*Cucurbita pepo* L). *Res Soc Dev* 9:e550985530–e550985530. <https://doi.org/10.33448/rsd-v9i8.5530>
- Mulamba, NN, Mock, JJ (1978) Improvement of yield potential of the ETO blanco maize (*Zea mays* L) population by breeding for plant traits [Mexico]. *Egyptian Journal of genetics and Cytology*
- Oliveira RL, Gomes RS, Almeida CF, Júnior RM, Rocha JRASC, da Silva DJH, Carneiro PCS (2021) Multi-trait selection of pumpkin genotypes aimed at reducing the growth habit and improving seed production. *Crop Sci* 61:1620–1629. <https://doi.org/10.1002/csc2.20386>
- Oloyede FM, Ogunsina AG (2023) The influences of storage period and morphotypes on the proximate and selected vitamins of pumpkin (*Cucurbita pepo* L) fruits Notulae. *Sci Biol* 15:11441. <https://doi.org/10.55779/nsb15311441>
- Paranhos JG, Ishikawa FH, de Lima MAC, Lima Neto FP (2022) Estimation of genetic parameters and prediction of breeding values for fruit-quality traits in hybrid mangoes. *Int J Fruit Sci* 22:608–617. <https://doi.org/10.1080/15538362.2022.2080791>
- Priori D, Barbieri RL, Mistura CC, Villela JCB (2018) Caracterização morfológica de variedades crioulas de abóboras (*Cucurbita maxima*) do sul do Brasil. *Revista Ceres* 65:337–345. <https://doi.org/10.1590/0034-737X201865040006>
- Ramjan MD (2021) Characterization of pumpkin (*Cucurbita moschata* Duch Ex Poir) germplasm through genetic variability, heritability and genetic advance. *Electron J Plant Breed* 12:91–96. <https://doi.org/10.37992/2021.1201.014>

- Resende MDV, Duarte JB (2007) Precisão e controle de qualidade em experimentos de avaliação de cultivares. *Pesquisa Agropecuária Tropical* 37:182–194
- Resende MDV (1995) Delineamento de experimentos de seleção para maximização da acurácia seletiva e do progresso genético. *Revista Árvore* 19:479–500
- Resende MDV (2016) Software selegen-REML/BLUP: a useful tool for plant breeding. *Crop Breed Appl Biotechnol* 16:330–339. <https://doi.org/10.1590/1984-70332016v16n4a49>
- Resende, MDV de (2007) SELEGEN-REML/BLUP: Sistema Estatístico e Seleção Genética Computadorizada via Modelos Lineares Mistos Embrapa Florestas: Colombo, Brazil
- Ribeiro RMP, Pereira CCA, da Silveira IM, de Sousa Nunes GH, Ramos SRR, de Queiroz MA, Souza MF, Lins HA, Júnior APB (2020) Indirect selection for content of carotenoid in pumpkin accessions. *Revista De La Facultad De Ciencias Agrarias UNCuyo* 52:12–21
- Rodriguez-Amaya, DB, Kimura, M (2004) *HarvestPlus handbook for carotenoid analysis* Washington: HarvestPlus. (accessed by August 27, 2023)
- Sahoo TR, Singh DK (2020) Estimation of genetic variability, heritability and genetic advance in cucumber (*Cucumis sativus* L) for yield and its components under protected structure. *Int J Current Microbiol Appl Sci* 9:2756–2764. <https://doi.org/10.20546/ijcmas.2020.904.325>
- Singh MK, Singh VB, Yadav GC, Kumar P (2019) Studies on variability, heritability (narrow sense) and genetic advance analysis for growth, yield and quality traits in pumpkin (*Cucurbita moschata* Duch ex Poir). *J Pharm Phytochem* 8:3621–3624
- Strohecker, R, Henning, HM (1967) Analisis de vitaminas: métodos comprobados Paz Montalvo Madrid pp 428
- Stupar A, Šeregelj V, Ribeiro BD, Pezo L, Cvetanović A, Mišan A, Marrucho I (2021) Recovery of β -carotene from pumpkin using switchable natural deep eutectic solvents. *Ultrasonics Sonochem* 76:105638. <https://doi.org/10.1016/j.ultsonch.2021.105638>
- Theanjumol P, Maniwaru P (2022) Physico-chemical and cooking qualities of fresh and stores pumpkins. *Hortic Environ Biotechnol* 63:101–113. <https://doi.org/10.1007/s13580-021-00385-4>
- Vencovsky, R (1987) Herança quantitativa In: Paterniani, E, Viegas, GP (Eds) *Melhoramento genético do milho* 2nd Campinas: Fundação Cargill 135–214

Publisher's Note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

Springer Nature or its licensor (e.g. a society or other partner) holds exclusive rights to this article under a publishing agreement with the author(s) or other rightsholder(s); author self-archiving of the accepted manuscript version of this article is solely governed by the terms of such publishing agreement and applicable law.