Safflower (*Carthamus tinctorius L.)* Characterization in the Pernambuco State Forest Middle Zone

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Authors' contributions

This work was carried out in collaboration between all authors, and all authors read and approved the final manuscript.

Original Research Article

ABSTRACT

This work aims to identify promising safflower genotypes for cultivation in Pernambuco state forest middle zone. The methodology employed evaluated the performance of six genotypes of the specie, growing at an experimental area of Agronomy Department of Federal Rural University of Pernambuco (UFRPE), by a randomized blocks design, between March and May 2017. The performance of six genotypes of the safflower, growing at an experimental area was evaluated. Germination percentage (%) and the emergency speed index were evaluated these at 60 and 80 days after sowing, following variables were observed: plant height (cm), stem diameter (cm), denting of leaf intensity, spinal margin of leaf intensity, number of branches per plant and per stand. Analysis of variance was performed with F test followed by Tukey post-hoc test ($p < 0.05$), using GENES program. All presented adequate height for mechanized harvesting and the genotypes ICA 211, ICA 338 and ICA 400 exhibited a weak or moderate intensity of spinescent margin. Cultivation in winter was not favorable.

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Safflower cultivation during the winter period in the Central Forest Zone of Pernambuco is contraindicated. The genotypes tested can be used in breeding programs to obtain adaptation to high rainfall.

1. INTRODUCTION

Safflower (*Carthamus tinctorius* L.) belongs to the family Asteraceae and genus Carthamus is an annual herbaceous plant, autogamous, oleaginous and plans had versatility to grown in adverse soil and climate conditions [1,2,3]. It has a pivotal root system and their leaves have spinescent margins, characteristics that give it greater tolerance to drought and high temperatures [4]. The culture cycle lasts around 110-150 days, varying according to the genotype, environmental factors and the interaction of these [5].

The seeds of this species stand out among the other oilseeds because they have high oil contents, around 35% to 45%, combined with the excellent quality for human consumption and other industrial uses [6,7]. The oil can also be used to produce biodiesel [8] and the resulting bagasse for protein supplementation in animal feed [9]. The flowers allow the extraction of two dyes [10] and can be exploited in the ornamental market [11]. In addition, seeds, flowers and oil, can be used in medicine to develop new drugs against various diseases [12,13].

This species is native to Asia and Africa, cultivated over two millennia and currently practised in all continents, more than 60 countries. The world seed production of safflower in 2014 refers to approximately 733,852 tons, covering 936,875 hectares of harvested area and world safflower oil production of in the same year corresponds to about 100,751 tons [14]. On the other hand, in Brazil, it is still inexpressive due to the scarcity of information about the culture, management methodologies and market strategies. [15,16]. In this context, studies are being promoted with the objective of evaluating and selecting genotypes with better adaptability to the country's soil and climatic conditions [17].

According to [18] indicates that this oilseed does not show good growth when exposed to soaked soils and high air humidity, evidencing that the safflower presents better development in dry periods and does not respond well to the crop in periods with high precipitation rates. However, studies show that the conduction of an irrigation system in periods of water scarcity can optimize the development of this herbaceous [19,20]. However, research is being developed with the aim of identifying and improving new genotypes and management techniques for the regions of the planet that have high humidity, a preponderant characteristic in Brazil, which presents tropical climate, sometimes dry and sometimes humid [17,4].

This work had the objective of characterizing six safflower genotypes for cultivation in the Pernambuco State Forest Middle Zone.

2. MATERIALS AND METHODS

The work was carried out in the agronomic area of the Department of Agronomy of the Federal Rural University of Pernambuco - UFRPE, Fitotecnia area, Recife - PE, whose geographical coordinates are 8°10'52 " S of latitude, 34°54'47"of longitude, with 2m of altitude. The climate is considered tropical rainy (As'), according to Köppen-Geiger, with average annual rainfall of 2000mm, with a well-defined dry period and rains that occur during the winter [21].

Six accessions from germplasm banks of India and Ethiopia were used, which were imported by the Institute of Agrarian Sciences (ICA) in agreement with the Federal University of Minas Gerais (UFMG) and assigned to the Program of Genetic Improvement of Plants of the Federal Rural University of Pernambuco (UFRPE) for the development of that research. The accesses were identified by numbers, these being: ICA 117, ICA 340, ICA 211, ICA 338, ICA 343 and ICA 400.

For the implementation of the experiment, one month before sowing, soil correction was performed, based on the analysis performed (Table 1) and according to the recommendations for cultivation. One week before sowing, the blocks and plots were demarcated and identified according to each genotype and design of the experiment.

Seeding was carried out in March 2017, in a conventional tillage area. The seeds (disinfested the previous day in a 25% hypochlorite solution) were arranged in spacing 0.50 m between rows and 0.20 m between plants, as recommended by [17]. The fertilization of foundation, sowing and other cultural treatments were done manually. The seeding depth was about three centimeters, distributing ten seeds per linear meter and two seeds per pit spaced 0.20 m. The thinning was done 15 days after planting, leaving five plants per meter, corresponding to a population density of 200,000 ha^{-1} plants. Were applied 56 kg ha^{-1} of phosphorus, 32 kg ha^{-1} of potassium and 16 kg ha^{-1} of nitrogen, according to recommendations [22].

The experimental design was a randomized complete block with six replicates. The treatments were composed of six safflower genotypes. The total area comprised 180m^2 (60m x 3m), where 144 plants of each of the six genotypes were distributed, distributed in 36 plots, formed by three lines with 1.5 m each, totalling 24 plants per plot. The evaluations were carried out in the central plants, harvesting twelve representative plants, random, respecting 0.25 m border at the extremities.

The removal of invasive plants was done manually, whenever necessary. Irrigations, also when necessary, were carried out manually and on very sunny days, which was limited to a small number of alternating days, since the cultivation was conducted during the winter period of the region and, therefore, under rainfall. The meteorological data of precipitation and mean air temperature recorded during the period of the experiments were obtained by the National Institute of Meteorology (INMET) and can be seen in Fig. 1.

The number of emerged plants with two cotyledon leaves open were recorded until the ninth day after sowing (before thinning and considering both plants per pit) and used to determine the Emergency Velocity Index (IVE), calculated according to the formula (1) [23]:

$$IVE = \Sigma({En}/{Nn}) \qquad (1)$$

Where: En = number of normal seedlings "n"; Nn = number of days of sowing until the count "n".

After stabilization of the emergence of plants, the final number of emerged plants were useds to calculate the percentage of germination. At sixty and eighty days after sowing, two evaluations (using four plants per plot) were carried out, based on the following characteristics: STAND = Total number of plants in the plot; Height of plant (cm) = performed with a millimeter ruler and corresponding to the measurement of the soil to the apex of the plant; Stem diameter (cm) = corresponding to the measurement performed with a digital caliper at the base of the stem; Intensity of leaf dentin = classified with the aid of a scale of notes: absent or weak (0); moderate (5); strong (10) (adapted from [24]); Intensity of margin of the spinescent leaves = classified by note scale: absent or weak (0); moderate (5); strong (10) (adapted from [24]); and Number of branches per plant = obtained by counting of branches per genotypes.

The data were submitted to analysis of variance, considering the effects of the treatments and the averages as fixed, according to the statistical model (2):

$$Y_{ij} = \mu + t_i + b_j + e_{ij}, \qquad (2)$$

The significance of the average squares obtained was tested by the F test at the 5% probability level and the comparison between the means of the genotypes was performed by the Tukey test at 5% probability, using the GENES program [25].

Table 1. Soil chemical analysis for safflower planting in the edaphoclimatic conditions in the Pernambuco state Forest Middle zone, 2017

Soil sample	SSP	P	pH	Ca	Mg	Na	K	Al	H	S	CEC	V	m
		Mg/dm^3	H$_2$O	Cmolc/dm^3								%	
1	11.3	516	6.8	8.5	1.1	0.1	0.8	0	2.55	10.6	13.1	81	0
2	12.0	492	6.6	7.6	0.9	0.1	0.7	0	2.22	9.3	11.6	81	0
Mean	11.6	504	6.7	8.0	1	0.1	0.7	0	2.38	9.9	12.3	81	0

* SSP- sodium saturation percentage; pH - hydrogenation potential; Ca -Calcium; Mg - magnesium; Na -sodium; K - potassium; P - phosphorus; Al -aluminum; H - hydrogen; S - sulfur; CEC- cation exchange capacity; V - Base Saturation; m - aluminum saturation
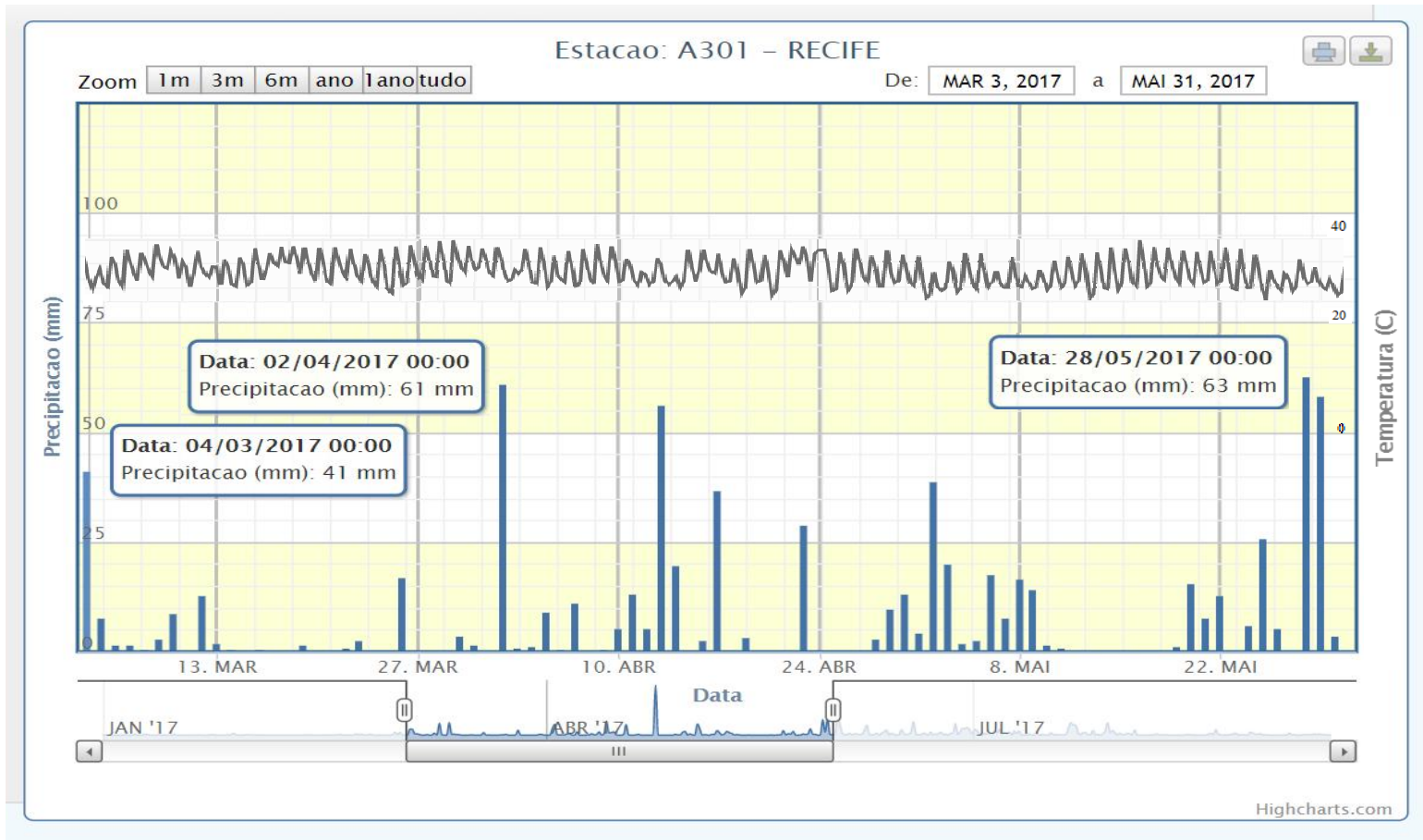
Fig. 1. Behavior of the meteorological variables of precipitation and mean air temperature during the vegetative cycle of the safflower crop in Recife / PE, during the first half of 2017 (Adapted). Source: INMET, 2017

Table 2. Summary of variances analysis and genetic parameters estimates offor germination percentage (%G) and the emergency speed index (ESI), plant height at 60/80 days (PH1/PH2); Stem Diameter at 60/80 days (SD1/SD2); total number of plants present in the plot at 60/80 days (STAND1/STAND2); intensity of leaf dentin (DentL); intensity of margin of the spinescent leaves (IMSL) and number of branches per plant (NB), recife, 2017

FV	DF	MS		DF	MS						DF	MS		
		%G	ESI		PH1	SD1	IMSL	NB	DentL	STAND1		PH2	SD2	STAND2
Accessions	5	25.13	11.23	4	112.2	57.96	55.52	31.52	55.52	39.12	3	165.4	35.18	99.44
Blocks	5	315.4	120.6	5	79.78	1.32	5.33	0.15	5.33	38.53	5	64.23	1.47	9.37
Residual	25	200.6	72.45	20	33.26	1.26	1.89	0.09	1.89	8.22	15	36.79	1.57	11.88
F		0.12^{ns}	0.18^{ns}		3.37*	46.1**	29.3**	335.44**	29.3**	4.76**		4.50*	22.4**	8.37**
Mean		65.67	39.30		48.67	8.78	4.83	1.78	4.83	14.53		55.99	10.61	6.37
CV		21.57	21.66		11.85	12.77	28.49	17.19	28.49	19.72		10.83	11.79	49.20
σ^2_g					13.16	9.45	8.94	5.24	8.94			21.44	5.60	
H^2					70.36	97.83	96.58	99.70	96.58			77.76	95.54	
CV_g					7.45	35.01	61.85	128.33	61.85			8.27	22.30	
CV_g/CV_e					0.63	2.74	2.17	7.46	2.17			0.76	1.89	

* and ** significant at the 5% and 1% levels, respectively, of the probability by the F test and "ns" not significant by the F test

The variance components and genetic parameters were estimated from the following expressions (3), (4), and (5):

$$\sigma_g^2 = \frac{QMG - QMR}{r} \qquad (3)$$

Which σ_g^2 corresponds to the genetic variance between means, QMR and GMQ match the variation between the accesses and experimental variation, respectively, and r is the number of repetitions;

$$h^2 = \frac{\sigma_g^2}{QMG/r} \qquad (4)$$

Wherein h^2 refers to heritability coefficient;

$$CV_g = \frac{100\sqrt{\sigma_g^2}}{\mu_a} \qquad (5)$$

Where CV_g matches the genetic coefficient of variation and μ_a is the overall average access.

3. RESULTS AND DISCUSSION

The Table 2 are shown summary of the analysis of variance and estimation of genetic parameters relating to morphological characteristics the accessions. It is possible to identify statistically significant differences among traits, indicating the existence of genetic variability.

The six evaluated safflower accessions showed moderate germination. Seedling emergence began at four days after sowing and lasted until eighth days, resulting in percentage averages that exceeded 60%. The germination capacity allows deduction of the first productive potential (Table 2).

The germination process featuring the capacity of the embryo growth restarting, previously quiescent state, giving rise to a normal seedling, under favorable conditions. The seed features one of the main inputs to the success of production, as they hold all the productive

character of potentiality of the plant, so production can even present inferiority of germination of the seed, imposed on inadequate conditions, but will never exceed [26].

ICA 117 genotype was not evaluated at 60 and 80 days for not presenting the number of plants per plot necessary for statistical analysis. This was caused due to the sensitivity of access to the climatic conditions at the time of cultivation, especially with regard to high rainfall, as can be seen in Fig. 1 which graphically shows the behavior of the meteorological variable precipitation and average air temperature during the vegetative cycle of the culture.

The cultivation period coincided with the rainy period of the region and together with the initial stress observations of the plants due to the intensity of the precipitations, confirmed that the variable STAND was considered an important point to be evaluated, allowing to identify which genotypes presented better behavior to the imposed environmental conditions (Fig. 2 a - d). According to [18] this crop does not present tolerance for development in soaked soils, nor does it respond well to high relative humidity.

The descriptive analysis of the six safflower accesses evaluated in the UFRPE experimental unit is organized in Table 3. The height of the plants varied between 35.75 cm and 71.75 cm, with an average of 48.67 cm, at sixty days after sowing, and 47.00 cm and 81.75 cm, with a mean of 55.98 cm, to eighty days. These values agree with the one commonly observed in safflower, which can assume from 30.00 cm to 150.00 cm, in its minimum and maximum limits, respectively [27]. This variation denotes a differential behavior among the genotypes in order to infer that there is a possibility of selection for this character and more important is the possibility of the use of these genotypes in future breeding programs aimed at raising the adaptation to the climatic conditions of the region.

Table 3. Descriptive analysis of five safflower accesses at 60 days after sowing and four accesses at 80 days, evaluated in the experimental area of UFRPE, Recife / PE, 2017

Description	PH1**	SD1**	STAND1**	PH2**	SD2**	STAND2**	DentL**	IMSL**	NB**
Maximum	71.75	1.53	22.0	81.75	1.62	18.00	10.00	10.00	4.75
Minimum	35.75	0.38	4.00	47.00	0.77	3.00	0.00	0.00	0.00
Mean	48.67	0.88	14.53	55.98	1.06	6.33	4.60	4.83	1.78

* PH1/PH2: Plant Height at 60/80 days; SD1/SD2: Stem Diameter at 60/80 days; STAND1/STAND2: Total number of plants present in the plot at 60/80 days; DentL: Intensity of Leaf Dentin; IMSL: Intensity of Margin of the Spinescent Leaves; and NB: Number of Branches per plant
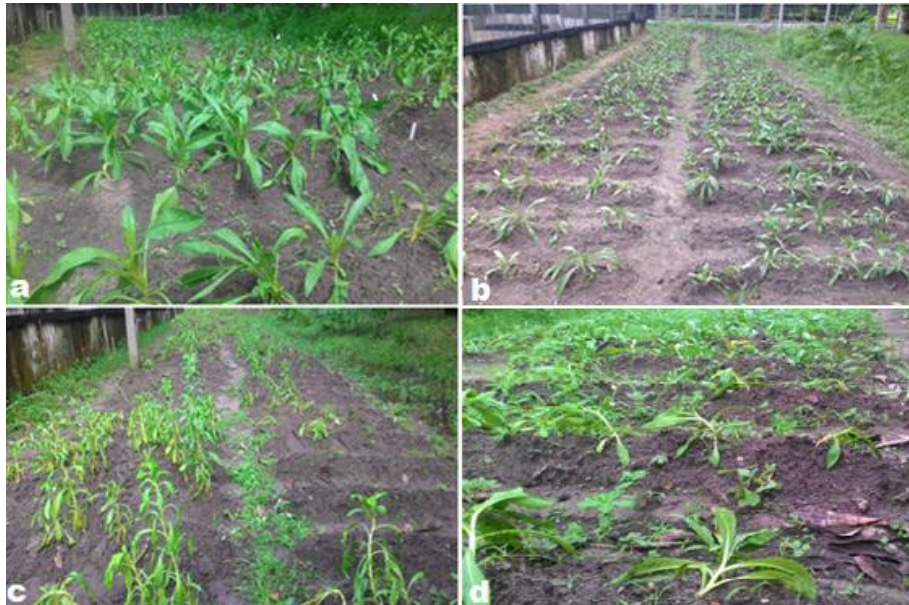
Fig. 2. Safflower plants in experimental area: (a, c) Stand of plant 30 and 80 days after sowing; (b, d) drooping of plants due to heavy rains

All accesses showed a significant difference between them for the height character of plants at 60 days after sowing, except for ICA 211 and ICA 338. In this respect, the genotypes ICA 343 and ICA 211 were the ones with the highest and lowest height of 54.25 and 44.33, respectively (Table 4). So it was observed that materials with larger stem diameters present a greater possibility of success for cultivation in periods with higher precipitation index and this characteristic can be used as an important selection criterion for breeding, in this sense the accessions ICA 340, ICA 343 and ICA 338 showed high averages.

The height of plants comprises one of the relevant characteristics to be considered by the breeder when selecting promising genotypes, preferably having a favorable height for the mechanized harvesting process [28]. Plants with very low stature can cause problems during the harvesting process, causing the winding of the windlass of the machine responsible for the collection. Also, very tall plants are directly related to resistance to lodging, presenting a greater tendency for drooping in the field [29].

The genotype ICA 338 differed from the other genotypes for the stem diameter, corresponding to the access that obtained the largest diameter (0.94 cm). On the other hand, ICA 400 had the smallest diameter of 0.17 cm. These values are like those found by [22] and [28] in their studies evaluating agronomic characters of safflower suggesting that the climatic conditions of the region did not interfere drastically in the stem diameter character.

Table 4. Average of plant height at 60 days (PH1); stem diameter at 60 days (SD1); total number of plants present in the plot at 60 days (STAND1); intensity of leaf dentin (DentL); and intensity of margin of the spinescent leaves (IMSL), evaluated in five accesses of safflower, Recife / PE, 2017

Accesses	PH1 (cm)	SD1 (cm)	STAND1	DentL	IMSL
ICA 340	52,12b	0,41b	15,00b	4,67b	7,50a
ICA 211	44,33d	0,31b	13,17c	9,17a	2,50b
ICA 338	45,33d	0,94a	18,00a	1,67c	2,92b
ICA 343	54,25a	0,35b	15,33b	5,83b	8,75a
ICA 400	47,33c	0,17c	11,17d	1,67c	2,50b

Means followed by the same letter do not differ by Tukey test at 5%

Throughout the cultivation, the lodging of many plants was observed (Fig. 2b, d). The genotypes ICA 211 and ICA 400 were the ones that presented the highest incidence of droped plants, leading to the death of several of these and leading to low STANDs, from the 60 days, as can be seen in table 4. These accesses differed statistically from each other and corresponded to the lowest values of a number of plants per plot. The best result corresponded to ICA 338 access with 18 surviving plants. It is noteworthy that in this period, the plants were in rosette phase, where the vegetative growth speed is quite slow and for this reason, corresponds to a critical stage of development [1].

The rosette phase coincided with intense rainy days, marked by some precipitation peaks that were configured around 60 mm, as can be verified in Figure 1, justifying the difficulty of survival and poor development of the plants. In addition, it is worth mentioning that the genotypes ICA 211 and ICA 400, described as less resistant to lodging and with lower values of STAND, are those that present the smallest diameters and, in turn, lower rainfall resistance (Table 4).

As for the intensity of the leaves 'dentin and the intensity of margin of the spinescent leaves, according to the Tukey test (Table 4), the genotypes can be described as: ICA 211, strong dentin and weak or moderate spinescent margin intensity; ICA 340 and ICA 343, medium dentin and strong spinescent margin intensity; and ICA 338 and ICA 400 as the weaker ones dentin, in addition to weak or moderate spinescent margin intensity. One of the current objectives of safflower breeding programs is the development of varieties with no spiny margins, not only for inclusion in the ornamental market but also for cultivars with high oil productivity [30]. Mainly in regions where the harvest still predominates manually, due to the discomfort of the collect caused by the intensity of thorns on the leaf and bracts.

It is worth mentioning that the ICA 400 genotype was not evaluated at 80 days because of the number of plants per plot inadequate for statistical analysis. The evaluations performed at 80 days after sowing obtained concordant and complementary results to those obtained at 60 days.

The genotypes presented a statistical difference for the characteristic height of plants. In the same way, ICA 338 access differed statistically from the others, however, ICA 340 access had a growth recovery capacity when compared to the result obtained at 60 days. Also on this aspect, ICA 211 access presented the highest sensitivity to high rainfall. It is noteworthy that at this stage of development, ICA 343 access continued to present the highest plant height, corresponding to 61.71 cm. This may have been due to the less competition between plants in the plot. For the stem diameter, only the ICA 338 presented statistical difference of the other accesses and with a development superior to double of the others, even with the upper STAND (Table 5). This may suggest a plausible justification for the differential behavior and greater ability to survive.

For the variable number of branches per plant, it is evident that ICA 340 and ICA 343 did not differ among themselves and presented the highest values, of 4.41 and 4.17, respectively (Table 5). Similar values were found by [22] and [17] in safflower surveys, referring to accesses with smaller numbers of branches, although they correspond to genotypes different from those evaluated in this study. The genotypes ICA 211 and ICA 338 did not differ among themselves and had low mean number of branches, of 0.17 and 0.12, respectively. These values diverge from those commonly observed in safflower, however, it may be justified by the environmental conditions that are inappropriate to conduct the crop, which probably limited the potential of these accesses. Or that a middle ground must be found to balance the behavior of different accesses.

Table 5. Average of plant height at 80 days (PH2); stem diameter at 80 days (SD2); Total number of plants present in the plot at 80 days (STAND2); and number of branches per plant, evaluated in four accesses of safflower, Recife / PE, 2017

Accesses	AP2 (cm)	DC2 (cm)	STAND2	NR (nº/pl)
ICA 340	59,17b	0,47b	9,17a	4,41a
ICA 211	51,96c	0,40b	1,00c	0,17b
ICA 338	51,12c	0,92a	9,83a	0,12b
ICA 343	61,71a	0,44b	5,33b	4,17a

Means followed by the same letter do not differ by Tukey test at 5%.

The estimates of genetic parameters of heritability and the ratio between genetic and experimental coefficients of variation indicate favorable conditions for selection [31]. The characteristics evaluated had a heritability coefficient higher than 70%. The heritability coefficient (h^2) indicates how much of the total phenotypic variation is due to genetic variance and this information is highly valued by the breeders, since it allows to distinguish how much of that demonstrated by the phenotype offers reliability in relation to the reproductive value [32].

The variables stem diameter, leaf thorn intensity and number of branches per plant had the highest heritability coefficients, higher than 90%. When this value approaches 1.0 or 100% indicates that these characteristics are little influenced by the environment [32] and allows predicting that the average of the characters in the chosen accessions, will present increases close to the selection differential, thus resulting in gain genetics.

It should be pointed out that these same characteristics presented a ratio between the coefficients of genetic and experimental variation greater than 1.0, indicating that the genetic variation was superior to environmental, which in turn favors the selection to be performed in a more efficient [31].

4. CONCLUSION

Safflower cultivation during the winter period in the Central Forest Zone of Pernambuco is contraindicated. The ICA 340 and ICA 338 accessions presented greater chances of success for cultivation in Pernambuco State Forest Middle Zone. The genotypes tested can be used in breeding programs to obtain adaptation to high rainfall.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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