










## Morphology and genetic divergence of *Stylosanthes scabra* Vog. accesses

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**ABSTRACT:** Germplasm collections for breeding purposes must initially be characterized to evaluate the diversity and allow the selection of materials for specific environments and uses. *Stylosanthes scabra* Vog. is a legume forage from the Brazilian Tropical Semiarid region, which might have potential, but is still underknown. The treatments consisted of eight *S. scabra* accesses (F1 generation) from Bom Jardim Municipality, in the tropical semiarid region in Brazil, on a complete randomized block design, with three replications. Qualitative and quantitative morphological descriptors and anthracnose incidence were evaluated at 30-day intervals, while forage mass was evaluated through cuts every 60 days, totaling three cuts. Frequency analysis by access was performed on qualitative variables and analysis of variance and Friedman on quantitative variables with subsequent Tukey test ( $p < 0.05$ ) when appropriate. Genetic parameters were estimated for qualitative and quantitative descriptors. The accesses of *S. scabra* were different ( $p < 0.05$ ) for all evaluated descriptors except for plant height, with high heritability ( $> 62.8\%$ ) and low repeatability (0.2 to 0.6). The high morphological diversity, even on a relatively small collection from a single tropical semiarid municipality, indicates the possibility of selecting superior materials.

**Key words:** genetic parameters; morphological descriptors; native legume

## Morfologia e divergência genética dos acessos de *Stylosanthes scabra* Vog.

**RESUMO:** Coleções de germoplasma para melhoramento, inicialmente devem ser caracterizadas, para avaliar a diversidade e permitir a seleção de materiais para ambientes e usos específicos. *Stylosanthes scabra* Vog. é uma leguminosa forrageira da região Tropical Semiárida Brasileira que apresenta potencial, mas ainda é sub-estudada. Os tratamentos consistiram em oito acessos de *S. scabra* (geração F1) coletados no município de Bom Jardim, na região tropical semiárida do Brasil em um delineamento em blocos completos casualizados com três repetições. Descritores morfológicos e a incidência de antracnose foram avaliados em intervalos de 30 dias e a massa de forragem foi avaliada por cortes a cada 60 dias, totalizando três cortes. As variáveis qualitativas foram avaliadas por análise de frequência por acesso, e as quantitativas por análise de variância e de Friedman, com teste de Tukey ( $p < 0,05$ ) quando apropriado. Parâmetros genéticos foram estimados para os descritores qualitativos e quantitativos. Os acessos de *S. scabra* foram diferentes ( $p < 0,05$ ) para todos os descritores, exceto altura da planta, com alta heritabilidade ( $> 62,8\%$ ) e baixa repetibilidade (0,2 a 0,6). A alta diversidade morfológica, mesmo em uma coleção relativamente pequena de um único município do trópico semiárido, indica a possibilidade de seleção de materiais superiores.

**Palavras-chave:** parâmetros genéticos; descritores morfológicos; leguminosas nativas



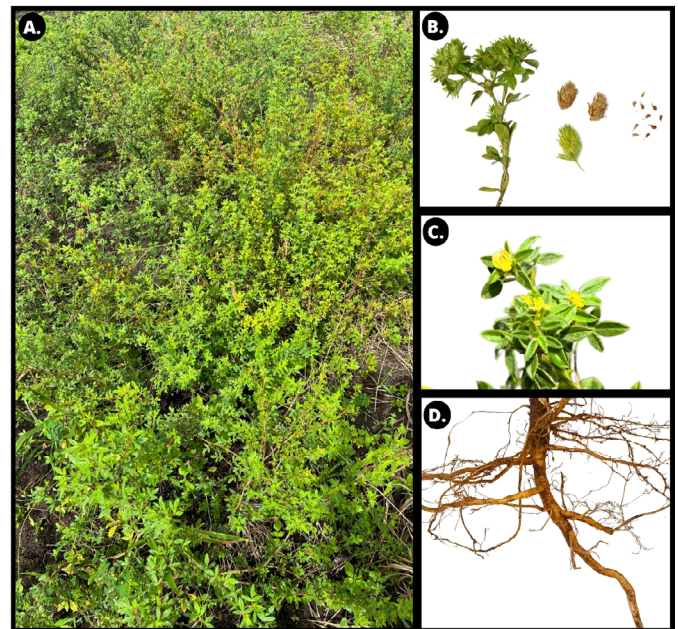
## Introduction

Pasture-based production systems are an economical feed base for ruminants. However, the sustainability of the livestock ecosystem needs forage alternatives that, besides supplying the growing animal protein demands, contribute to the stability of the soil-plant-animal-environment system. Fodder legumes (Fabaceae) have the potential to achieve this balance expected for the ecosystem (Lüscher et al., 2014; Oliveira et al., 2016; Muir et al., 2019). According to Santos et al. (2023), using native legumes besides the forage resource improves and conserves degraded or potentially degradable soils.

The legume family is characterized by wide morphological variability and includes potential forage species with annual and perennial lifecycles and with herbaceous, prostrate, shrubs, sub-shrubs, and arboreal growth habits (Muir et al., 2014; Schultze-Kraft et al., 2018). Generally, legumes have a high crude protein content (Muir et al., 2019); however, part is often bound to fibrous fractions (Santos et al., 2019). Most also have the potential to fix nitrogen in symbiosis with rhizobia and increase N availability in the soil for associated or subsequent crops and forage grasses and soil biota (Schultze-Kraft et al., 2018), supporting non-N<sub>2</sub>-fixing plants in grasslands by transferring symbiotically fixed N (Lüscher et al., 2014) and thus reducing the need for N fertilizer (Lira Junior et al., 2023). Furthermore, they provide multiple environmental benefits, such as restoring degraded lands and mitigating GHG emissions (Muir et al., 2017), thus contributing to the sustainability of the ecosystem in different tropical and subtropical environments).

In recent years, *Stylosanthes* species have been considered among the subshrub, shrub, or prostrate forage legumes of greater economic relevance (Oliveira et al., 2015; Marques et al., 2018; Muir et al., 2019; Silva Neto et al., 2024). Species of this genus (Figure 1) are distributed mainly in South America but also in Africa and Asia and were introduced in Australia (Calles & Schultze-Kraft, 2010; Vanni, 2017) and have wide adaptability to different climatic conditions, with potential for natural occurrence in areas of low precipitation and low soil fertility (Nagaich et al., 2013; Marques et al., 2018). Both high protein value (153 – 230 g kg<sup>-1</sup>) and efficiency in biological nitrogen fixation (~ 25 kg N ha<sup>-1</sup>) are among the main characteristics of *Stylosanthes* species (Mendonça et al., 2017; Diniz et al., 2023). Due to this, they are also recommended for breeding programs and for the recovery of soils and degraded pasture areas (Marques et al., 2018; Muir et al., 2019).

Costa et al. (2018) observed different species of spontaneously distributed *Stylosanthes* in the tropical semiarid region of Pernambuco, including the perennial erect sub-shrub *Stylosanthes scabra* Vog., which is an allotetraploid (AABB), originated by the parent diploids *S. seabrana* (genome A) and *S. viscosa* (genome B). This species has had approximately 600 accesses found in regions of Colombia, Ecuador, Peru, Bolivia, Paraguay, Australia, and,



**Figure 1.** *Stylosanthes scabra* Vog. - General appearance of the plant (A), inflorescences and loment (B), flowers and leaves (C), and nodules on the roots (D).

mainly, Venezuela and Brazil (Calles et al., 2016; Oliveira et al., 2016; Vanni, 2017; Costa et al., 2018).

Some *S. scabra* are found in ecosystems with altitude up to 1900 m, with an annual average temperature between 18.8 to 26.9 °C and annual precipitation of 325 to 1223 mm, with 3 to 5 months raining less than 60 mm (Calles & Schultze-Kraft, 2014, 2016). These areas present characteristic vegetation of savanna, semi deciduous forest; evergreen forest or thorn forest.

Some of this species advantages, associated with morphological and physiological characteristics, are drought tolerance, adaptation to soils with pH between 4.3 to 7.3 and low phosphorus requirement, and its biological nitrogen fixation capacity (Mendonça et al., 2017; Habermann et al., 2021). For crude protein, neutral and acid detergent fiber, values of 177-210, 379-448, and 288-320 g kg<sup>-1</sup>, respectively, have been reported (Mupenzi et al., 2009; Mpanza & Hassen, 2015), suitable for feeding ruminants. Among the disadvantages of this species, some authors mention the low seed production, susceptibility to anthracnose, and low persistence in association with grass (Jank et al., 2011; Singh et al., 2018).

Another characteristic of *S. scabra* is the high morphological variability between germplasm from different populations (Calles & Schultze-Kraft, 2010; Costa et al., 2018) and the little genetic divergence from *S. hamata*, *S. guianensis* and *S. viscosa*, hampering the correct identification (Nagaich et al., 2013). According to Cook & Schultze-Kraft (2020), *S. seabrana* is clearly different from *S. scabra*. The relationship between genetic diversity and geographic distribution could support genus exploration (Habermann et al., 2021).

Morphological descriptors are inheritable characters with little or no environmental interaction (Porbeni et

al., 2018). Likewise, in assessments based on agronomic, morphological, and physiological variables in accesses of *S. scabra*, it is recommended to associate them with information from molecular markers. Thus, one could evaluate the variability of accesses in specific locations and identify those promising considering the adaptability to agroecological conditions and biomass production (Oliveira et al., 2016; Costa et al., 2018). According to Kadam et al. (2018), selection's effectiveness depends on the germplasm's genetic variability and how much of it is hereditary. This trial, thus, aimed to characterize morphological diversity, production, and genetic parameters of *Stylosanthes scabra* Vog. accesses, collected in the Agreste mesoregion of the Pernambuco state, Brazil under tropical semiarid conditions.

## Materials and Methods

### Site and experimental design

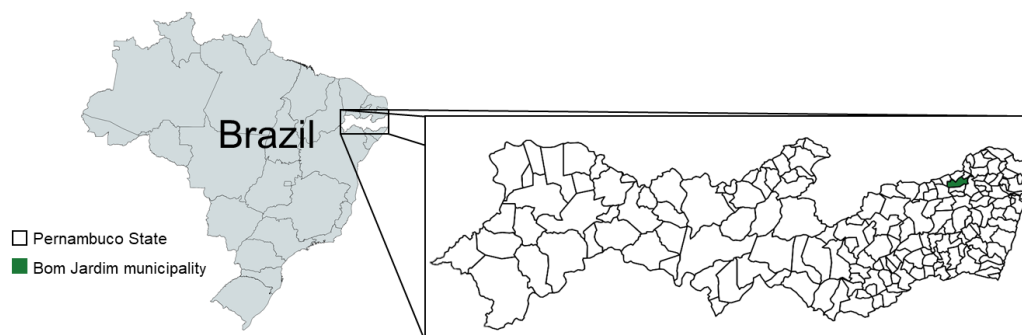
The experiment was carried out in a greenhouse, located at 08°01'13.4"S and 34°57'14.9"W (Recife – Pernambuco, Brazil) with climate classified as tropical dry-summer (As') according to Köppen-Geiger (Peel et al., 2007), evaluating eight accesses of *Stylosanthes scabra* Vog. (F<sub>1</sub> generation), on a randomized block design with three replicates.

Accesses were collected in different locations of the Bom Jardim municipality (Figure 2), in the Northern part of Agreste mesoregion of Pernambuco state (Table 1), at 07°45'45 "S and 35°35'13" W, with climate classified as tropical dry-summer (As') according to Köppen-Geiger (Peel

**Table 1.** Seed collection sites for *Stylosanthes scabra* Vog. accesses, in the Agreste mesoregion Pernambuco, Brazil.

Access	Soil	Geographic coordinates		Site*	Altitude (m)
		Bom Jardim Municipality			
		Longitude	Latitude		
5F	LU	35°33'781"	7°48'066'	1	323
15F	LU	35°33'983"	7°48'586"	3	297
26F	DRA	35°32'684"	7°47'466"	1	297
31F	DRA	35°32'861"	7°47'440'	2	327
33F	DRA	35°32'861'	7°47'440"	2	327
102	DRA	35°33'861'	7°47'440"	3	351
41F	DRA	35°32'614'	7°47'390"	4	282
52F	LN	35°32'206"	7°48'142'	1	225

LU: Luvisolo; DRA: Dark Red Argisol; LN: Litolic Neossol (Santos et al., 2018).  
\* Classification of collection sites according to geographical coordinates and altitude.



**Figure 2.** Location of Bom Jardim municipality within the state of Pernambuco, Brazil.

et al., 2007), with an average annual rainfall of 1412 mm., and were identified in the Dárdano de Andrade Herbarium, Agronomic Institute of Pernambuco, Brazil (Galdino, 2014).

### Establishment and maintenance of forage legume

Expanded Polystyrene Styrofoam trays were used for seed propagation, with one seed per cell sown, with previous heat treatment to break dormancy (Alencar et al., 2009). Thirty days after planting, the seedlings were transplanted individually into 20 L pots, with drain and filled with 10 kg of Cohesive Yellow Argisol soil, from Arcoverde municipality, Pernambuco, collected at 0 to 20 cm depth. Soil characteristics were: pH (water, 1: 2,5) = 6,6, Mehlich-I, P = 57 mg dm<sup>-3</sup>, Na<sup>+</sup> = 0,25 cmol<sub>c</sub> dm<sup>-3</sup>, K<sup>+</sup> = 0,41 cmol<sub>c</sub> dm<sup>-3</sup>, Ca<sup>+2</sup>+Mg<sup>+2</sup> = 3,65 cmol<sub>c</sub> dm<sup>-3</sup>, Ca<sup>+2</sup> = 2,9 cmol<sub>c</sub> dm<sup>-3</sup>, Al<sup>+3</sup> = 0,0 cmol<sub>c</sub> dm<sup>-3</sup>, H<sup>+</sup> + Al<sup>+3</sup> = 2,28 cmol<sub>c</sub> dm<sup>-3</sup>, C = 6,11 g kg<sup>-1</sup> and MO = 10,53 g kg<sup>-1</sup>.

Plants were irrigated periodically until drainage, with the drained solution returned to the respective vessel. A uniformization cut was made 60 days after planting at 15 cm from the soil.

### Morphological variables and forage production

After the uniformization cut, qualitative and quantitative morphological descriptors were observed every 30 days for four evaluations. For qualitative descriptors, each plant received grades low (1), medium (2), and high (3) for plant size (PS), leaf pilosity (LP), stem pilosity (SP), and anthracnose incidence (AI). Growth habit (GH) was classified as erect (1), semi-erect (2), open (3), and prostrate (4), and flowering as without flower (1) and with flower (2).

Quantitative morphological descriptors such as central leaflet length (CLL), central leaflet width (CLW), lateral leaflet length (LLL), lateral leaflet width (LLW), stipple length welded to the petiole (SLW), stipple length petiole free (SLF), petiole length (PETL) and stem diameter (SD) were evaluated with the aid of a universal caliper (precision 0.01 mm). Branches (BN) and leaf (LN) numbers were obtained by counting. Plant height (PH) was evaluated with the aid of a measuring tape (precision of 1 mm) to the highest branch without stretching. Forage mass production (FMP) per plant was obtained by cutting at 20 cm from the ground every 60 days, for three cuts in total. Subsequently, the material was weighed and dried in a forced circulation oven at 55 ± 2 °C.

## Genetic parameters

Heritability ( $h^2$ ), repeatability coefficient (R), genetic variation coefficient (CVg) and the ratio between the genetic and environmental variation coefficients (CVg/CVe) were calculated for the qualitative and quantitative descriptors. Repeatability coefficients were estimated using the principal components based on the correlation matrix. The number of measures necessary to predict the real value of the variables was also estimated, considering a coefficient of determination ( $R^2$ ) preset at 0.80.

## Statistical analysis

Qualitative variables were grouped considering the absolute frequency of observations for each access. Analysis of variance was performed for LLL, ELW, PH, with Tukey mean test ( $p < 0.05$ ) when appropriate. Due to the absence of normal distribution and homogeneity of variance for CLL, CLW, LLW, ELF, PETL, SD, BN, and LN, Friedman's analysis was used considering access as the source of variation. Analyses were performed using SAS University Edition and of the genetic parameters Genes software (Cruz, 2013).

## Results

### Qualitative and measurable variables and forage production

*S. scabra* accesses qualitative descriptors had variable behavior (Table 2). For PS, only access 52F was high, and the remaining accesses in the medium category. Most accesses

had low AI, with 15F access being the most susceptible. Leaf pilosity, in most accesses, was low. However, on the stem, all accesses showed medium pilosity. Growth habit varied between erect (5F, 15F, 33F and 52F) and semi-erect (26F, 31F, 41F and 102F). Flower presence was highly frequent only for access 26F.

Regarding the quantitative morphological descriptors (Table 3), there was a difference between accesses ( $p < 0.05$ ), except for the BN, LN, and PH.

CLL and CLW ranged from 16.5 to 19.9 and 7.3 to 8.8 mm, respectively. Lateral leaflets presented smaller dimensions than central ones, with intervals between 14.0 to 17.8 and 6.0 to 7.6 mm for LLL and LLW, respectively (Table 3). Smaller dimensions in access 26F were observed in all measurable descriptors mentioned above ( $p < 0.05$ ), while higher values were observed in 15F, 52F, and 33F accesses for CLL and in 33F access for LLL. Differences were observed between the accesses for CLW and LLW ( $p < 0.05$ ), with higher averages for the central leaflet in accesses 52F, 33F, 31F, 102F, and 41F and for the lateral leaflet in accesses 15F, 33F, 102F, and 41F. Lower means were observed at access 26F access for both descriptors. The SLW was higher ( $p < 0.05$ ) at the 31F and 41F accesses; and SLF in 15F, 26F and 31F accesses ( $p < 0.05$ ). Only access 5F showed greater PETL, and access 26F greater SD. Smaller SLW, SLF, PETL, and SD were observed for access 52F.

All accesses had two branches, with LN and PH between 123 to 173 and 51 to 64 cm ( $p > 0.05$ ), respectively. DMP

**Table 2.** Qualitative morphological descriptors in the characterization of *Stylosanthes scabra* Vog. accesses.

Descriptor	Access							
	5F	15F	52F	26F	33F	31F	102F	41F
Plant size	M	M	H	M	M	M	M	M
AI	L	M	L	L	L	L	L	L
Leaf pilosity	L	L	L	L	L	L	L	L
Stem pilosity	M	M	M	M	M	M	M	M
Growth habit	E	E	E	SEE	E	SEE	SEE	SEE
Flowering	Not	Not	Not	With	Not	Not	Not	Not

L: low; M: medium; H: high; AI: anthracnose incidence; E: Erect; SEE: semi-erect; Not: without flower; With: with flower.

**Table 3.** Measurable morphological descriptors in the characterization of *Stylosanthes scabra* Vog. accesses.

Descriptor	Access								CV	P-Value
	5F	15F	52F	26F	33F	31F	102F	41F		
CLL (mm)	18.0 ab	19.2 a	18.7 a	16.5 b	19.9 a	18.2 ab	16.7 ab	18.4 ab	51.2	0.0015
CLW (mm)	8.2 ab	8.0 ab	8.4 a	7.3 b	8.8 a	8.3 a	8.4 a	8.6 a	49.4	0.0001
LLW (mm)	6.8 ab	7.0 a	6.9 ab	6.0 b	7.6 a	6.8 ab	7.0 a	7.0 a	51.6	0.0033
SLF (mm)	4.0 ab	4.8 a	2.6 b	4.5 a	2.9 ab	4.5 a	4.3 ab	4.0 ab	48.1	<0.0001
PETL (mm)	11.5 a	9.4 ab	7.5 d	8.9 abc	8.1 cd	8.1 cd	8.7 bc	8.8 bc	43.5	<0.0001
SD (mm)	4.4 d	4.8 cd	4.3 d	6.8 a	4.7 cd	5.6 abc	6.3 ab	5.7 abc	44.1	<0.0001
BN (mm)	1.9	2.0	2.0	2.0	1.9	2.2	2.1	1.8	54.0	0.5139
LN (mm)	123.0	123.9	133.9	172.9	141.0	167.1	160.2	170.9	57.4	0.8577
									SE	
LLL (mm)	15.5 ab	17.1 ab	16.2 ab	14.0 c	17.8 a	15.6 ab	14.6 bc	16.0 ab	0.06	0.0096
SLW (mm)	8.0 ab	7.1 bc	6.1 c	8.8 ab	7.5 abc	9.0 a	8.3 ab	9.0 a	0.04	0.0004
PH (cm)	51.5	59.8	64.7	58.5	56.6	55.8	57.7	59.8	3.67	0.4309
FMP (g plant <sup>-1</sup> )	4.9 b	4.9 b	6.6 ab	8.5 a	5.4 b	7.9 a	8.1 a	7.0 ab	0.51	0.0002

CLL: central leaflet length; CLW: central leaflet width; LLW: lateral leaflet width; SLF: stipple length petiole free; PETL: petiole length; SD: stem diameter; BN: branches number; LN: leaf number; LLL: lateral leaflet length; SLW: stipple length welded to the petiole; PH: plant height; FMP: forage mass production; CV: Coefficient of variation; SE: Standard error.

varied from 4.9 to 8.5 g plant<sup>-1</sup> with higher yields ( $p < 0.05$ ) for 26F, 31F and 102F accesses (Table 3).

### Genetic parameters

High heritability values were observed for all descriptors (Tables 4 and 5), with the lowest being LN (62.8%), PH (64.4%) and BN (69.2%). Repeatability was low for all characteristics ( $R$  between 0.2 and 0.5), except for the length of the central leaflet ( $R = 0.6$ ).

Genetic variation coefficients were medium to low magnitude, ranging from 4.3% (PH) to 12.6% (LN). However, for most descriptors, the CVg/CVe ratio was greater than one (Tables 4 and 5) with only flowering (CVg/CVe = 0.9), BN (CVg/CVe = 0.9), LN (CVg/CVe = 0.8) and PH (CVg/CVe = 0.8) lower than one.

The minimum number of measurements was variable (Tables 4 and 5), ranging for qualitative descriptors between 4 and 9 measurements and for quantitative descriptors between 1 and 8 measurements.

## Discussion

### Qualitative and measurable variables and forage production

*S. scabra* accesses from the Bom Jardim municipality presented variability (Tables 2 and 3) that is useful for a breeding program. Albuquerque et al. (2024) also observed that the variation in plant height, leaf number, and leaf and total aerial mass in all accesses of the herbaceous native legume *Macroptilium lathyroides* (L.) Urb. indicates that germplasm of this species might support future breeding programs. Américo et al. (2022) observed that *S. scabra* accesses from Embrapa Cerrado's germplasm bank also presented significant genetic diversity.

Medium PS concentrated 87.5% of accesses (Table 2), with the 52F access considered a high size despite not having

a statistically higher PH (Table 3). According to Martuscello et al. (2015), the advantage of high PS is the greater ability to capture light in limited light conditions than those of low size. However, other plant structural and architectural characteristics must be considered both above and below ground, such as specific leaf area, leaf mass fraction, maximum photosynthetic rate and specific root length as described by Freschet et al. (2018).

While *Stylosanthes* spp are generally susceptible to anthracnose, some species have been characterized as resistant even under favorable environmental conditions (Schultze-Kraft et al., 2020). Seven of the accesses had low AI, while access 15F was potentially susceptible, which is a relevant indicator in the selection for the genetic improvement of the species (Assis et al., 2018).

Pilosity was variable between the plant's organs (Table 2); however, it was present in 100% of *S. scabra* accesses both on the leaves and stem. According to Calles & Schultze-Kraft (2010), Medeiros & Flores (2014) and Vanni (2017), its presence is characteristic of the *S. scabra* species. These characteristics are phenotypic adaptations of the plant to increase resistance to drought and reduce transpiration in hot and slightly humid environments (Erice et al., 2013). Moreover, this characteristic, both on the leaf and on the stem, is considered relevant to benefit the register and/or improve the discrimination of specific genotypes in germplasm banks (Martuscello et al., 2015; Borges et al., 2019). However, some authors consider for this descriptor only the categories presence and absence of pilosity (Borges et al., 2019).

*S. scabra* growth habit was evenly split between erect and semi-erect, while the literature categorizes it as erect (Costa et al., 2008; Chandra, 2009; Calles & Schultze-Kraft, 2010; Vanni, 2017). While this growth habit can contribute to

**Table 4.** Genetic parameters, repeatability coefficient and number of observations for qualitative descriptors of *Stylosanthes scabra* Vog. accesses.

Parameters	Plant size	Anthracnose incidence	Leaf pilosity	Stem pilosity	Growth habit	Flowering
$h^2$ (%)	90.2	71.3	92.7	97.7	84.5	72.0
$R$	0.3	0.3	0.5	0.4	0.5	0.4
CVg (%)	7.9	5.2	13.9	8.3	12.4	7.9
CVg/CVe	1.8	0.9	2.1	3.8	1.3	0.9
$N_0$	9	7	4	5	3	7

$h^2$ : heritability;  $R$ : repeatability coefficient; CVg/CVe: genetic variation coefficient (CVg) and environmental variation coefficient (CVe) ratio;  $N_0$ : number of measurements with  $R^2$  of 0.80.

**Table 5.** Genetic parameters, repeatability coefficient and number of observations for quantitative descriptors of *Stylosanthes scabra* Vog. accesses.

Parameters	CLL	CLW	LLL	LLW	SLW	SLF	PETL	SD	BN	LN	PH
$h^2$ (%)	89.7	77.6	84.0	83.5	93.5	85.1	88.2	85.0	69.2	62.8	64.4
$R$	0.6	0.4	0.5	0.4	0.3	0.3	0.4	0.4	0.2	0.4	0.3
CVg (%)	5.9	4.4	6.4	5.5	9.0	14.2	10.3	11.2	5.5	12.6	4.3
CVg/CVe	1.7	1.1	1.3	1.3	2.2	1.4	1.6	1.4	0.9	0.8	0.8
$N_0$	2	7	3	5	3	2	1	4	8	2	6

CLL: central leaflet length; CLW: central leaflet width; LLL: lateral leaflet length; LLW: lateral leaflet width; SLW: stipple length welded to the petiole; SLF: stipple length petiole free; PETL: petiole length; SD: stem diameter; BN: branches number; LN: leaf number; PH: plant height;  $h^2$ : heritability;  $R$ : repeatability coefficient; CVg/CVe: genetic variation coefficient (CVg) and environmental variation coefficient (CVe) ratio;  $N_0$ : number of measurements with  $R^2$  of 0.80.

greater light interception, especially in consortium conditions (Borges et al., 2019), according to Muir et al. (2014), legumes with a prostrate growth habit will be more likely to persist in the long term because of lower accessibility to ruminants, when they are used in grazing and intercropped with grasses. Likewise, Barrios-Maestre et al. (2011) observed that under field conditions, special attention should be paid to erect growth species due to the poor coverage and competition with other species, leading to low persistence, especially when intercropped with fast-growing grasses.

The absence of flowering was frequent, except for access 26F. Generally, late flowering is among the most outstanding characteristics of the *S. scabra* (Chandra, 2009). According to (Weller & Ortega, 2015), some legumes require exposure to photoperiods and/or specific temperatures to flower, and late flowering can be an advantage due to increased biomass accumulation before flowering. Furthermore, flowering time is coordinately controlled by factors such as genetics, plant nutrition, plant age, vernalization, and hormones (gibberellins) (Yun et al., 2023). However, greenhouse results are limited because they probably do not represent varying soil and environmental conditions, as observed by (Kawaletz et al., 2014).

The taxonomic classification of *S. scabra* describes leaflet dimensions 8.0-17.0 mm × 3.0-4.3 mm for length and width, respectively (Calles & Schultze-Kraft, 2010; Vanni, 2017). Likewise, Fortuna-Perez et al. (2011) report intervals 8.0 to 21.0 and 2.0 to 7.0 mm, respectively, closer to the averages of accesses in the present experiment, both for the length and width of leaflets. This variability and association of characters observed could contribute to characterizing the genetic diversity of *S. scabra* accesses in a germplasm bank. Américo et al. (2022) observed there is genetic diversity for forage quality traits among *S. scabra*.

SLW and SLF variability correspond with ranges reported by some authors. Taxonomic information of *Stylosanthes* describes the stipple as a pubescent and/or sinuous structure with length and width ranges of 4-8 × 3-5 mm (Fortuna-Perez et al., 2011). The observed PETL interval was larger than those reported in different taxonomic studies of this species, with measurements between 1 to 8 mm (Costa et al., 2008), 3 to 8 mm (Calles & Schultze-Kraft, 2010; Fortuna-Perez et al., 2011) and 4 to 6 mm (Vanni, 2017).

The smaller SD of accesses 5F, 15F, 52F, and 33F could favor their nutritional value. According to Zi et al. (2017), SD in *Stylosanthes* is linked to a decrease in nutritional value due to increased neutral and acid-detergent fiber contents and decreased digestibility. These authors observed a decrease of 27% in digestibility (35.41 to 25.91%), with increments of 1.25 mm for SD (2.66 to 3.91 mm).

For PH, values are considered characteristic of *S. scabra* according to taxonomic studies (Calles & Schultze-Kraft, 2010; Medeiros & Flores, 2014; Vanni, 2017). The lack of variability for BN might be due to the cuts at 20 cm height, while Ogunbode & Akinlade (2012) suggest cutting 25 cm to promote branching and thus BN. Leaf number presented

averages in a wider range, and even without differences between accesses, some authors indicate that this descriptor should be considered as an important characteristic for its association with environmental services such as, accumulation of forage mass (Silva et al., 2010; Barbosa et al., 2019) potential for biological nitrogen fixation (Ogedegbe & Falodun, 2016), forage consumption preferences (leaf/stem ratio) and forage quality (Silva et al., 2010).

Forage mass production was variable ( $p < 0.05$ ), but lowest (4.9 to 5.3 g plant<sup>-1</sup>) for accesses 5F, 15F and 33F accesses (Table 3). This lower production could result from a greater soil nutritional requirement of these accesses, the environmental conditions of the greenhouse, or a smaller accumulation of forage mass characteristic of them at this cutting age.

### Genetic parameters

Most of the descriptors had high heritability ( $h^2 > 70\%$ ), and the character's most susceptible to environmental influence were LN, PH and BN (Tables 4 and 5), which allows *S. scabra* genetic selection, considering the high heritability descriptors (Ogunniyan & Olakojo, 2014; Oliveira et al., 2015; Assis et al., 2018).

However, based on CVg/CVe ratio (Tables 4 and 5), selection for AI, BN, LN, flowering, and PH is unfavorable due to higher environmental than genetic variation. When both genetic indicators are associated, it is possible to identify a better descriptor selection process through a heritability coefficient (Vargas et al., 2020) with recommended values greater than 80% (Ogunniyan & Olakojo, 2014; Assis et al., 2018).

However, there was low repeatability, which indicates a higher measurement number would be required for all descriptors. The minimum number of measurements estimated for each access, with R<sup>2</sup> of 0.80, was very variable between descriptors, and PS, AI, SP, flowering, BN, CLW, LLW, and PH had a higher minimum number of repetitions than actually measured (Tables 4 and 5). This high variability might be due to this index being a highly variable genetic indicator between characters, species, and accesses, depending on environmental conditions, among other factors (Assis et al., 2018). Due to the little information referenced for *S. scabra* accesses, it is recommended to carry out successive assessments that allow finding the appropriate number of measurements for a more constant expression of the evaluated descriptors.

### Conclusions

Morphological characteristics of *Stylosanthes scabra* Vog. show great diversity among accesses from the Agreste mesoregion of Pernambuco, Brazil. Accesses 26F, 31F, and 102F stand out for forage biomass production. Genetic parameters show environmental influence for anthracnose incidence, flowering, width of the central leaflet, branch number, and plant height. For these descriptors plus plant

height, stem pilosity, and width of the lateral leaflet, it is necessary to increase the number of measurements to enhance data reliability.

## Compliance with Ethical Standards

**Author contributions:** Conceptualization: MAL, MVFS; Data curation: SBM; Formal analysis: SBM, MVC, AMHA, NVS; Funding acquisition: MAL, MVFS; Investigation: SBM, MAL, MVFS; Methodology: SBM, MAL, MVFS; Project administration: MAL, MVFS; Resources: SBM, MAL, MVFS; Supervision: MAL, MVFS; Validation: SBM, MAL, MVFS, RLCF, MVC, AMHA, TNO, NVS, MALJ; Visualization: SBM, MAL, MVFS, AMHA, TNO, NVS; Writing – original draft: SBM, MAL, MVFS; Writing – review & editing: MAL, MVFS, RLCF, MVC, AMHA, TNO, NVS, MALJ.

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