

MORPHOLOGICAL CHARACTERIZATION OF BUFFELGRASS WITH POTENTIAL FOR FORAGE AND SEED PRODUCTION

CARACTERIZACIÓN MORFOLOGICA DE GENOTIPOS DE PASTO BUFFEL CON POTENCIAL PARA PRODUCCIÓN DE FORRAJE Y SEMILLA

Ricardo A. Sánchez-Gutiérrez^{1,4}, Jean Hanson², Chris Jones², Pedro Jurado-Guerra³, Eduardo Santellano-Estrada⁴, Alicia Melgoza-Castillo⁴ and Carlos Morales-Nieto^{4*}

¹Instituto Nacional de Investigaciones Forestales, Agrícolas y Pecuarias (INIFAP), Campo Experimental Zacatecas. Calera de Víctor Rosales, Zacatecas, México. ²International Livestock Research Institute, Addis Ababa, Ethiopia. ³INIFAP, Sitio Experimental Aldama-La Campana, Aldama, Chihuahua, México. ⁴Universidad Autónoma de Chihuahua, Facultad de Zootecnia y Ecología, Chihuahua, Chihuahua, México.

*Autor de correspondencia (morales_nieto_c_r@hotmail.com)

SUMMARY

Buffel grass (*Pennisetum ciliare* L.) is an important forage species. It is necessary to continue exploring the natural variability of this grass and select materials with potential for livestock. Currently, the International Livestock Research Institute (ILRI) in Ethiopia preserves *ex situ* 157 buffel materials; it is important to characterize them to identify materials with productive potential. The objective of this study was to determine the phenotypic diversity of 147 ecotypes and 10 cultivars of buffelgrass to identify materials with potential forage and seed production. The study was conducted in 2014 at the Zwai Experiment Station in Ethiopia. Materials were established in the field with 36 plants per plot. Variables indirectly measured to identify ecotypes with potential for seed yield were: panicles per plant (PP), spikelets per panicle (SP), caryopses per panicle (CP) and days to 50 % flowering (DF), while for forage production variables were plant height (PH, cm) and forage height (FH, cm). Cluster and discriminant analyses were performed. The buffelgrass collection showed large variability in all measured characteristics. The plant and forage height ranges were: from 34 to 139 and from 24 to 118 cm, respectively. Variation of PP ranged from 6 to 322, SP from 13 to 285 and CP from 3 to 113. The standardization cut at 50 % flowering ranged from 48 to 71 days. A high correlation coefficient ($r = 0.98$) was observed between plant height and forage height and there was a negative correlation ($r = -0.62$) between panicles per plant and days to 50 % flowering. Four groups ($P < 0.0001$) were identified for the 157 materials. With the exception of FH, the rest of variables significantly contributed to the separation of groups. The buffelgrass collection presented high variability in the measured characteristics; also, ecotypes 1442, 19448, 19457, 19459, 19462 and 19464 showed forage potential.

Index words: *Pennisetum ciliare*, characterization, diversity, ecotypes, forage, seed.

RESUMEN

El pasto buffel (*Pennisetum ciliare* L.) es una especie forrajera importante. Es necesario continuar con la exploración de la variabilidad natural de este pasto y seleccionar materiales con potencial para la ganadería. Actualmente el Instituto Internacional de Investigaciones Pecuarias (ILRI, por sus siglas en inglés) en Etiopía conserva de manera *ex situ* 157 materiales de buffel; es importante caracterizarlos para identificar materiales con potencial productivo. El objetivo del presente estudio fue determinar la diversidad fenotípica de 147 ecotipos y 10 variedades de pasto buffel e identificar materiales con potencial de producción de forraje y semilla. El estudio se realizó en 2014 en la estación

experimental Zwai en Etiopía. Los materiales se establecieron en parcelas con 36 plantas cada una. Las variables medidas indirectamente para identificar ecotipos con potencial para la producción de semilla fueron: panículas por planta (PP), espiguillas por panícula (EP) cariósidos por panícula (CP) y días a 50 % de floración (DF), mientras que para producción de forraje las variables fueron altura de planta (AP, cm) y de forraje (AF, cm). Se realizaron análisis de agrupamiento y discriminante. La colección de pasto buffel mostró alta variabilidad en todas las características medidas. Los intervalos de altura de planta y forraje fueron de 34 a 139 y de 24 a 118 cm, respectivamente. La variación de PP fue de 6 a 322, EP de 13 a 285 y para CP fue de 3 a 113. El corte de estandarización al 50 % de floración fue de 48 a 71 días. Se observó una alta correlación ($r = 0.98$) entre altura de planta y de forraje, y una correlación negativa ($r = -0.62$) entre panículas por planta y días al 50 % de floración. Cuatro grupos ($P < 0.0001$) fueron identificados para los 157 materiales. Con excepción de AF, las demás variables contribuyeron significativamente a la separación de grupos. La colección de pasto buffel presentó alta variabilidad en las características medidas; además, los ecotipos 1442, 19448, 19457, 19459, 19462 y 19464 mostraron potencial forrajero.

Palabras clave: *Pennisetum ciliare*, caracterización, diversidad, ecotipos, forraje, semilla.

INTRODUCTION

The main limitation of livestock production in Africa is the instability in yield and low quality of native forage species (Pamo *et al.*, 2007). Buffelgrass [*Pennisetum ciliare* (L.) Link] is a C_4 , apomictic species, common in the Africa subtropics. It has been introduced into arid and semiarid areas around the world, as Argentina (Carlóni-Jarrys *et al.*, 2018) because of its high forage production potential, soil erosion control, and drought tolerance (Quiroga *et al.*, 2013). Genetic diversity for forage traits in buffelgrass offers an opportunity to make selection and to develop new varieties with good production to face the challenges of climate change. There is currently a high demand for forage to feed livestock in areas with low precipitation, thus justifying plant breeding programs (Carlóni-Jarrys *et al.*, 2018) to improve forage production, forage quality and

seed yield. There are morpho-agronomic characteristics associated with forage production and seed yield, which can be measured to get a better understanding of the potential within the species (Ribeiro *et al.*, 2019; Wassie *et al.*, 2018). Exploring the natural variability through a morphological characterization could be used to select the best accessions with some characteristics of interest (Alves *et al.*, 2014). The International Livestock Research Institute (ILRI) holds a collection of 157 accessions of buffelgrass in its field genebank in Ethiopia; thus, it is necessary to perform a phenotypic characterization of this germoplasm to identify novel sources of variation, as well as outstanding plant material. The objectives of this study were to determine phenotypic diversity of 147 accessions and 10 cultivars of buffelgrass in Zwai, Ethiopia, and to identify the best accessions with characteristics for forage and seed production.

Materials and methods

The study was carried out at the Zwai Experiment Station of ILRI in Ethiopia, located at 7° 54' N, 38° 44' E and altitude of 1640 masl. The average annual precipitation is 600 mm and the peak concentration of rainfall is recorded between July and September. The average annual minimum and maximum temperatures are 13 and 27 °C, respectively. Soil is loamy sand with 0.5 m depth and pH from 8.1 to 8.4.

Accessions were planted at least ten years before this study and each accession was established into a 2 × 5 m unreplicated plot, 36 plants per plot with 50 cm separation. The accessions and cultivars maintained in the field genbank came from 10 African countries (Table 1). Some accessions and cultivars were donated by other programs and their origin was unknown. Plots were irrigated five times during the dry season only for maintenance, and 200 kg of nitrogen and 60 kg of phosphorus per hectare were applied.

On June 16th, 2014, all plants were cut at a height of 150 mm above ground, characterization began on July 16th and finished on December 5th. Six descriptors were recorded from eight plants located at the center of the plot (Van de Wouw *et al.*, 1999); plant height (PH) was measured in cm from the ground level to the tip of all inflorescence, forage height (FH) was also measured in cm from the ground level to the tip of leaves, panicles per plant (PP) was the number of inflorescences per plant. Eight panicles were randomly taken to count the number of spikelets per panicle (SP) and caryopses per panicle (CP). For days to 50 % of flowering (DF), the whole group of plants in the plot was observed and the number of days from cutting until half of the plot reached flowering was registered.

The Pearson correlation coefficient was calculated with the CORR procedure (SAS Institute, 2011) to observe the relationship among variables. Furthermore, a hierarchical cluster analysis using the CLUSTER procedure (SAS Institute, 2011) by the Ward clustering method was performed. Groups were chosen based on PST2 (pseudo- T^2 statistic). To determine the variables with higher discriminating power the STEPDISC procedure (SAS Institute, 2011) was used. The discriminant function for the probability that an accession belonged to a particular group was analyzed with the DISCRIM procedure (SAS Institute, 2011). Also, if a difference between groups was recorded ($P \leq 0.05$), a multivariate analysis of variance (MANOVA) with the Wilks' Lambda statistic was calculated (SAS Institute, 2011).

Results and discussion

The 157 accessions of buffelgrass showed a high degree of diversity in all agro-morphological characteristics measured. Plant and forage height varied from 34 to 138 and from 24 to 118 cm, respectively. Panicles per plant ranged from 6 to 322, days to 50 % flowering were from 48 to 71, spikelets per panicle ranged from 13 to 285 and caryopses per panicle from 3 to 113. The results obtained for plant height were similar to those of previous studies (M'Seddi *et al.*, 2002). In general, our collection showed higher variability in the rest of the traits compared to results reported by Mseddi *et al.* (2004). This discrepancy could partially be attributed to the higher number of accessions in this collection than the 51 accessions in the aforementioned study, perhaps due to the selection of a broader range of eco-geographical locations.

The results obtained reveal a significant potential of this collection to be used in plant breeding programs. High correlation coefficients ($P \leq 0.0001$) were observed between plant height and forage height ($r = 0.98$), and between panicles per plant and days to 50 % flowering ($r = -0.626$). The rest of the correlation coefficients were weak. Most advanced accessions showed a higher number of panicles per plant, which agrees with the result in the previously mentioned study of 52 accessions of buffelgrass (Mseddi *et al.*, 2004). Morales *et al.* (2012) found that in Arizona cottontop [*Digitaria californica* (Benth.) Henr.] forage height is more closely related to yield than plant height. It might be possible to extrapolate from these results and propose that buffelgrass accessions with large plant height could be used for forage production.

Hierarchical cluster analysis assigned the 157 buffelgrass accessions into four groups (Table 1, Figure 1). Also, MANOVA with Wilks' Lambda statistic showed differences among these groups ($P \leq 0.0001$). Group I

included 57 accessions along with Karasberg, Nunbank and Viva cultivars. This group was characterized by early flowering, large plant height, and forage height traits. Group II included 50 accessions and the cultivars Biloela, Boorara, Gayndah, Kongwa and Towoomba. This group was characterized by late flowering, low number of panicles per plant and caryopses per panicle. Group III consisted of 44 accessions and the cultivars American and Molopo, with a larger number of panicles per plant, early flowering and small size. Group IV included six accessions; this group was the tallest one and contained the largest number of spikelets per panicle and caryopses per panicle.

Discriminant analysis demonstrated that most traits, except FH, contributed to separate clusters ($P \leq 0.05$), (Table 2) and the probability that an accession belonged to a particular group was higher than 95 %. The results on days to 50 % flowering and plant height agree with those of previous studies (Griffa *et al.*, 2012; Jorge *et al.*, 2008); however, some groupings among cultivars were different

which could also be related to the location and the period when experiments were performed, as plant characteristics are influenced by environmental as well as by genetic factors. Plant height has been shown to positively correlate with yield forage in buffelgrass (Mansoor *et al.*, 2012; Mseddi *et al.*, 2004); hence, Groups I and IV can potentially be used for silage or hay production.

In some species, characteristics such as number of panicles per plant and total number of seeds per panicle have been shown to be related to seed yield (Youssef and Hansson, 2019). Since Groups III and IV have the largest number of panicles per plant and caryopses per panicle, respectively, accessions in these groups could be used to select for improved seed yield in a plant breeding program. The accessions in Group IV could be selected as new cultivars as they have good productivity and seed yield; nevertheless, it should be noted that it is necessary to perform research focused on seed yield and production (Morales-Nieto *et al.*, 2017), as well as on forage quality. It

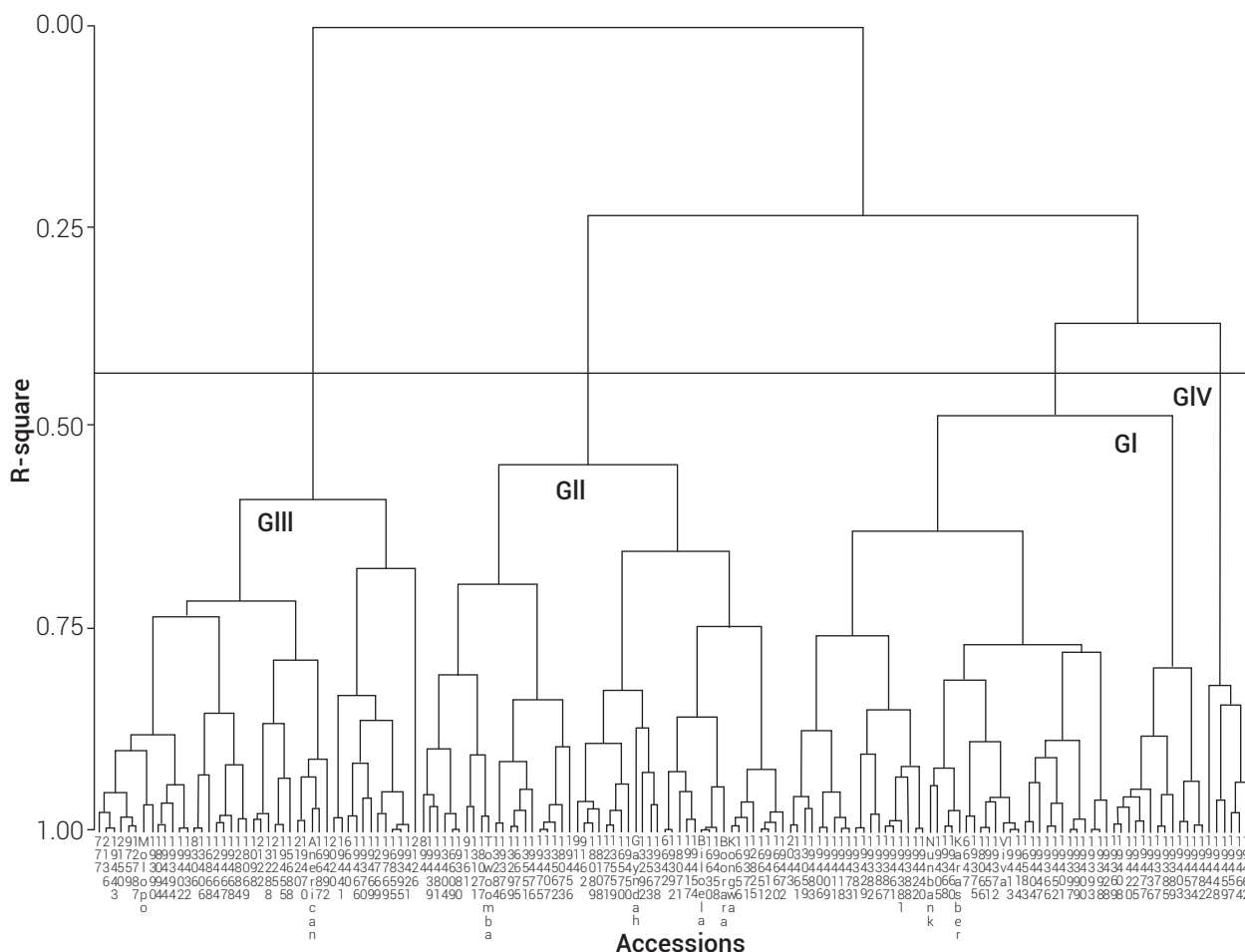


Figure 1. Dendrogram showing the integration of four groups using the Ward method based on agro-morphological characteristics of 157 buffelgrass accessions.

Table 1. List of accessions and cultivars used in the characterization study and the assignment to the corresponding group.

Acc/C	Country	Group	Acc/C	Country	Group	Acc/C	Country	Group	Acc/C	Country	Group
American	Kenya	3	13299	Kenya	2	19381	Ghana	1	19440	Kenya	1
Biloela	Tanzania	2	13404	Ethiopia	2	19382		1	19441	Tanzania	3
Boorara		2	13406	Ethiopia	3	19385	Tanzania	1	19442	Zimbabwe	4
Gayndah	Kenya	2	13407	Ethiopia	2	19386	Tanzania	1	19443	Zimbabwe	3
Karasberg	Nambia	1	13461	Ethiopia	1	19387	Tanzania	1	19444	South A.	3
Kongwa	Tanzania	2	13551	Ethiopia	2	19388	Tanzania	1	19447	South A.	1
Molopo		3	13559	Ethiopia	2	19389	Tanzania	1	19448	South A.	4
Nunbank	Uganda	1	13562	Ethiopia	2	19390	Tanzania	3	19450	South A.	2
Towoomba	South A.	2	13563	Ethiopia	2	19392	Tanzania	2	19451	Botswana	1
Viva	Uganda	1	15687	Mauritania	3	19393	Tanzania	1	19452	Botswana	1
777	Tanzania	3	15688	Mauritania	3	19394	Tanzania	3	19453	South A.	1
894	Tanzania	2	16570	Namibia	2	19395	Tanzania	3	19454	Zimbabwe	2
914	Tanzania	2	16583	Namibia	1	19397	Tanzania	1	19455	Namibia	3
1098	Dr Congo	3	16609	Namibia	2	19398	Tanzania	1	19456		2
2020	Ethiopia	3	16630	Namibia	2	19399	Tanzania	1	19457		4
2043	Ethiopia	1	16651	Namibia	2	19400	Tanzania	1	19458		2
2120	Ethiopia	3	16656	Namibia	2	19401	Tanzania	1	19459		4
2122	Ethiopia	3	16660	Namibia	2	19402	Tanzania	3	19460		1
2125	Ethiopia	3	16675	Namibia	2	19403	Tanzania	1	19461		1
2126	Ethiopia	3	16855	Niger	3	19404	Tanzania	1	19462		4
2136	Ethiopia	3	16868	Niger	3	19405	Tanzania	1	19464	South A.	4
2150	Ethiopia	3	18066	Botswana	1	19406	Tanzania	1	19465	Somalia	3
6640	Tanzania	3	18069	Botswana	3	19408	Tanzania	1	19466	Somalia	3
6642	Tanzania	2	18071	Botswana	2	19409	Tanzania	1	19467	Somalia	3
6647	Zimbabwe	1	18073	Botswana	2	19411	Tanzania	1	19468	Somalia	3
8306	Ethiopia	3	18077	Botswana	2	19412	Tanzania	2	19469	Somalia	3
9161	Ethiopia	2	18089	Botswana	2	19413	Tanzania	1	19470	Somalia	3
9162	Ethiopia	2	18094	Botswana	3	19414	Tanzania	1	19472		2
9759	Ethiopia	3	18108	Botswana	2	19417	Tanzania	2	19473	South A.	1
12464	Antigua	3	19366	Dr Congo	1	19418	Tanzania	1	19474	South A.	1
12769	Kenya	3	19368	Botswana	1	19420	Tanzania	1	19475	South A.	2
12771	Kenya	2	19369	Botswana	1	19421	Tanzania	3	19476	South A.	2
12787	Kenya	3	19370	Botswana	3	19422	Tanzania	1	19477		1
12825	Kenya	2	19371	Botswana	2	19425	Tanzania	1	19481		2
12884	Kenya	3	19372	Botswana	1	19428	Tanzania	1	19482		1
13059	Kenya	1	19375	Botswana	1	19429	Tanzania	1	19483	South A.	1
13121	Kenya	2	19376	Botswana	2	19431		1	19492		3
13284	Kenya	2	19377		1	19432	Uganda	3			
13288	Kenya	3	19378		2	19436	India	1			
13292	Kenya	2	19380		2	19439	Kenya	2			

Acc/C: accession number/cultivar name.

Table 2. Mean and standard deviation of six agro-morphological characteristics of the four groups classified by hierarchical cluster analysis.

Variables	Group I (n = 57)	Group II (n = 50)	Group III (n = 44)	Group IV (n = 6)
PH [†] (cm)	102.9 ± 15	74.3 ± 14	66.3 ± 14	113.8 ± 8
FH (cm)	84.09 ± 14	57.16 ± 13	50.76 ± 13	87.6 ± 11
PP [†]	37 ± 15	21 ± 9	66 ± 53	12 ± 6
SP [†]	86 ± 27	75 ± 19	51 ± 14	217 ± 54
CP [†]	38 ± 19	27 ± 15	21 ± 12	71 ± 21
DF [†] (%)	56 ± 2	61 ± 4	52 ± 3	68 ± 2

[†]Variables with discriminant power ($P \leq 0.05$). PH: plant height, FH: forage height, PP: panicles per plant, SP: spikelets per panicle, CP: caryopses per panicle, DF: days to 50 % flowering.

is concluded that the studied collection of 157 accessions of buffelgrass exhibited high variability in all characteristics measured, highlighting the opportunity to improve this species through a breeding program. Accessions 1442, 19448, 19457, 19459, 19462 and 19464, native to South Africa and Zimbabwe, are the tallest ones and have the largest number of spikelets per spike and caryopses per spike; therefore, they may have potential to be included directly in future plant breeding programs for enhancing seed and forage production of buffelgrass.

BIBLIOGRAPHY

- Alves G. F., U. J. de Figueiredo, A. D. Pandolfi Filho, S. C. L. Barrios and C. B. do Valle (2014) Breeding strategies for *Brachiaria* spp. to improve productivity – an ongoing project. *Tropical Grasslands – Forrajes Tropicales* 2:1-3, [https://doi.org/10.17138/TGFT\(2\)1-3](https://doi.org/10.17138/TGFT(2)1-3)
- Carlioni-Jarrys E. J., Ma. C. Acosta-Bragato and K. Grunberg-Fraga (2018) Nuclear DNA content and ploidy level in apomictic buffelgrass genotypes. *Revista Fitotecnia Mexicana* 41:23-29, <https://doi.org/10.35196/rfm.2018.1.23-29>
- Griffa S., M. Quiroga, A. Ribotta, E. López C., E. Carlioni, E. Tommasino, C. Luna and K. Grunberg (2012) Relationship between seed yield and its component characters in *Cenchrus* spp. *Electronic Journal of Plant Breeding* 3:701-706.
- Jorge M. A. B., M. Van de Wouw, J. Hanson and J. Mohammed (2008) Characterization of a collection of buffelgrass (*Cenchrus ciliaris*). *Tropical Grasslands* 42:27-39.
- Mansoor U., M. Hameed, A. Wahid and A. R. Rao (2002) Ecotypic variability for drought resistance in *Cenchrus ciliaris* L. Germplasm from Cholistan Desert in Pakistan. *International Journal of Agriculture and Biology* 4:392-397.
- Morales N. C. R., A. Melgoza C., P. Jurado G., M. Martínez S. y C. Avendaño A. (2012) Caracterización fenotípica y molecular de poblaciones de zacate punta blanca (*Digitaria californica* (Benth) Henr. *Revista Mexicana de Ciencias Pecuarias* 3:171-184.
- Morales-Nieto C. R., R. Corrales-Lerma, A. Álvarez-Holguín, F. Villarreal-Guerrero y E. Santellano-Estrada (2017) Caracterización de poblaciones de pasto banderita (*Bouteloua curtipendula*) de México para seleccionar genotipos con potencial para la producción de semilla. *Revista Fitotecnia Mexicana* 40:309-316, <https://doi.org/10.35196/rfm.2017.3.309-316>
- M'Seddi K., M. Visser, M. Neffati, D. Reheul and M. Chaieb (2002) Seed and spike traits from remnant population of *Cenchrus ciliaris* L. in South Tunisia: high distinctiveness, no ecotypes. *Journal of Arid Environments* 50:309-324, <https://doi.org/10.1006/jare.2001.0830>
- Mseddi K., L. Mnif, M. Chaieb, M. Neffati and M. Roux (2004) Aboveground phytomass productivity and morphological variability of Tunisian accessions of *Cenchrus ciliaris* L. *African Journal of Range and Forage Science* 21:49-55, <https://doi.org/10.2989/10220110409485833>
- Pamo E. T., B. Boukila, F. A. Fonteh, F. Tendokeng, J. R. Kana and A. S. Nanda (2007) Nutritive value of some grasses and leguminous tree leaves of the Central region of Africa. *Animal Feed Science and Technology* 135:273-282, <https://doi.org/10.1016/j.anifeeds.2006.07.001>
- Quiroga M., K. Grunberg, A. Ribotta, E. López C., E. Carlioni, E. Tommasino, C. Luna and S. Griffa (2013) Obtaining sexual genotypes for breeding in buffel grass. *South African Journal of Botany* 88:118-123, <https://doi.org/10.1016/j.sajb.2013.04.016>
- Ribeiro S. O., M. C. A. Amaral, W. T. Barros, A. S. Bandeira, A. D. Cardoso and R. C. de Vasconcelos (2019) Quality of top, middle and bottom buffelgrass seeds from different collecting sites. *Journal of Agricultural Science* 11:264-271, <https://doi.org/10.5539/jas.v11n9p264>
- SAS Institute Inc (2011) Base SAS® 9.3 Procedures Guide. Statistical Procedures. SAS Institute Inc. Cary, North Carolina, USA. 528 p.
- Van de Wouw M., J. Hanson and S. Nokoe (1999) Observation strategies for morphological characterisation of forages. *Genetic Resources and Crop Evolution* 46:63-71, <https://doi.org/10.1023/A:1008627527822>
- Wassie W. A., B. A. Tsegay, A. T. Wolde and B. A. Limeneh (2018) Evaluation of morphological characteristics, yield and nutritive value of *Brachiaria* grass ecotypes in northwestern Ethiopia. *Agriculture and Food Security* 7:89, <https://doi.org/10.1186/s40066-018-0239-4>
- Youssef H. M. and M. Hansson (2019) Crosstalk among hormones in barley spike contributes to the yield. *Plant Cell Reports* 38:1013-1016, <https://doi.org/10.1007/s00299-019-02430-0>