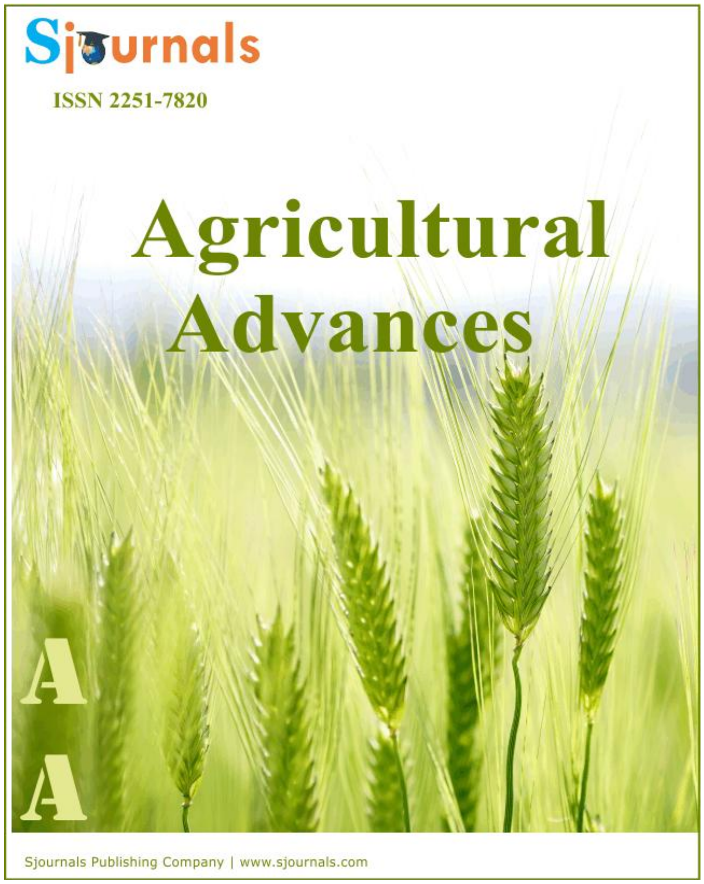
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Original article

Morpho-biometric characterisation of *Brachiaria spp* germplasm in the area of mount Cameroon

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Brachiaria grass has a major potential to relieve the polygastric livestock sector and support its growth in pastoral and extensive mixed systems where ruminants suffer permanent or seasonal nutritional stress from inadequate feed supply as an endemic problem. Morphological description of *Brachiaria spp* germplasm is helpful for the identification and delimitation of taxa that could lead to accession collections and selection of varieties with high agronomy value. The identification of potential accessions of *Brachiaria* in the mount Cameroon area was phenotypically based on twelve qualitative and eleven quantitative descriptors, data been collected using a structured questionnaire. The *Brachiaria spp* have light green coloured leaves (70.9%) which are mostly pubescent (34.5%), intermediate (29.1%) or glabrous (25.5%). The internodes are mostly yellow green (25.5%), the stigma is mostly white (40.0%) while the own panicle is mostly absent. The panicles are mostly open (61.8%) while the grains are mostly round shaped (74.5%). The glumella pubescence is mostly absent while the lemma and palea mostly have a straw colour and the apiculus and/or awn colour are mostly absent (74.5%). There is huge variability for the quantitative traits whatever the locality is. The traits varying most are the number of panicle per plant (146.86%), the grain weight (141.17%) and the number of spikelets per plant (135.10%). The correlation between measurements vary from weak to moderate, the highest positive significant (p<0.01) correlation coefficient (0.637**) was observed between the flag leaf length and the panicle length, while the significant native correlation (-0.283*) was found between the panicle fertility and the flag leaf width. The cumulative variance of the first five principal components explains 73.958% the genetic variability observed within the studied population. The flag leaf length, flag leaf width and culm length are components contributing most to that variability, with respectively 23.10%, 17.16% and 13.16%, giving a total cumulative genetic variability value of 53.43% of the whole *Brachiaria spp* population germplasm in the area of mount Cameroon. The hierarchical clustering revealed that there could exist four main clusters or accessions of *Brachiaria spp* germplasm in the area of mount Cameroon. Accession 4 is the most distant from the three others, while accessions 1 and 2 are the closest. The clustering into groups of accessions with similar morphological within the collection *Brachiaria* germplasm in this study will enable researchers interested in this grass to easily identify subsets of accessions for further evaluation aimed at specific uses. Morphological differences between these accessions and taxonomic differentiation to species level will need molecular characterization for confirmation.

1. Introduction

The livestock sector in developing countries is growing in response to rapidly growing demand for its products. In Cameroon there is great pastoral potential with 30% of the rural population living on livestock sector, which globally represent 16% of the GDP (Pamo, 2006). Ruminants in pastoral and extensive mixed systems suffer permanent or seasonal nutritional stress from inadequate feed supply as an endemic problem in these systems. *Brachiaria spp* has a major potential impact to relieve the polygastric livestock sector and support its growth.

Brachiaria spp is a grass originating from the Eastern and Central African countries such as Uganda, Zambia, Zimbabwe, Burundi, DRC, Kenya etc. (Renvoize et al., 1996). It is now the most widely used tropical grass, especially in Central and South America where forage seed industry has grown to meet the large demand and an expanding export market, placing it in competition with major cereal crops monetary value wise (Santos Filho, 1996). Several perennial *Brachiaria* species of African origin such as *B. brizantha*, *B. decumbens, B. humidicola, B. mutica, B. arrecta, B. dictyoneura* and *B. ruziziensis* have been used for long as fodder of interest in tropical America (Argel and Keller-Grein, 1996). In Cameroon, two species (*B. jubata* and *B. stigmatisata*) have been identified as indigenous, amongst introduced species like *B. ruziziensis, B. mutica* and *B. brizantha* (Pamo et al., 1997; Pamo et al., 1998; Yonkeu and Pamo, 1994).

Brachiaria spp is of great economic importance for pastures in the tropics (Miles and Valles, 1996), as it shows rapid regrowth under grazing and good persistence under heavy or frequent defoliation, it is mostly drought tolerant and can withstand water logging stress, it also has high forage quality when fertilized and well managed (Lascano and Euclides, 1996). Species like *B. decumbens* and *B. humidicola* show excellent adaptation to low fertility and acidic soils (Ndikumana and Leeuw, 1996). *Brachiaria* is highly palatable to ruminants, maintain high ground cover until it "crashed" under prolonged and heavy grazing at a high stocking rate and once *Brachiaria* pasture is established it can last for over 20 years and as such can play an important role in the sustainable creation of pastures for livestock feeding. These qualities have opened up a huge seeds market for species like *B.*

brizantha and *B. decumbens* (Fisher and Kerridge, 1996). Despite all the above mentioned qualities, *Brachiaria* is subject to destruction from several insects, fungal and viral diseases.

Valerio et al. (1996) highlighted great variability exists in the available *Brachiaria spp* germplasm and genetic factors for resistance to pathogens which could be exploited in breeding for improved varieties. Morphological characters are helpful in identification and delimitation of taxa. These characters had been recognized as basic criteria for identification and authentication of plants (Sultana et al., 2011). Such research could lead to accession collections to have varieties combining high seed yields with high forage quality, persistence under heavy grazing, high drought tolerance, erect growth habit and high dry matter yields for cut-and-carry forage systems or that display good shade tolerance (Pizarro and Hare, 2014). The mount Cameroon area combines the characteristics of having the most fertile soils of the Guinea gulf, coastal area with altitude varying from 0 (sea level) to 4100 m, bearing the second most rainy area in the world (Debundscha), hot humid environment. This motivates a rapid flow-in of populations, hence need for a continuous improvement in the livestock productivity to meet the demand for livestock-based food products.

According to Nascimento et al. (2011), morphological characterization of germplasm accessions is fundamental in order to provide information for plant breeding programs. This study, therefore will set the stage for selection of *Brachiaria* accessions that are locally adapted and that could be used for pasture development and prospect for seed production. The mount Cameroon area, with its diversity of agro-ecology, altitude, temperature, pluviometry and soil types could be a very fertile land for explorations in *Brachiaria* seed production for the whole Africa. The combination of morphological, agronomic and genetic/genomic data could increase the chances of achieving new superior genetic combinations or even amplifying the population variability in crosses between related morphological types that are genetically distant (Ferreira, 2003). The general goal of this study is to contribute to *Brachiaria spp* germplasm productivity through characterization of its existing local accessions in Cameroon.

2. Materials and methods

2.1. Description of study area

The mount Cameroon area is located in the humid mono-modal rainforest agro-ecological zone of Cameroon (Fig. 1a). It has an equatorial climate characterized by a short dry season (November-March) and a long rainy season (March-November) with abundant rainfalls (2,000-10,299mm). This is one of the most fertile volcanic soils of the whole central Africa with the fastest expansion land use (Fig. 1b), mostly for agriculture. The area rises from sea level (0m) at Limbe and Tiko to 4100m at the mount Cameroon summit in Buea. The temperature varies from 18°C in August to 35°C in March according to altitude and proximity to the Atlantic ocean. The relative humidity is constantly high (75%-80%).

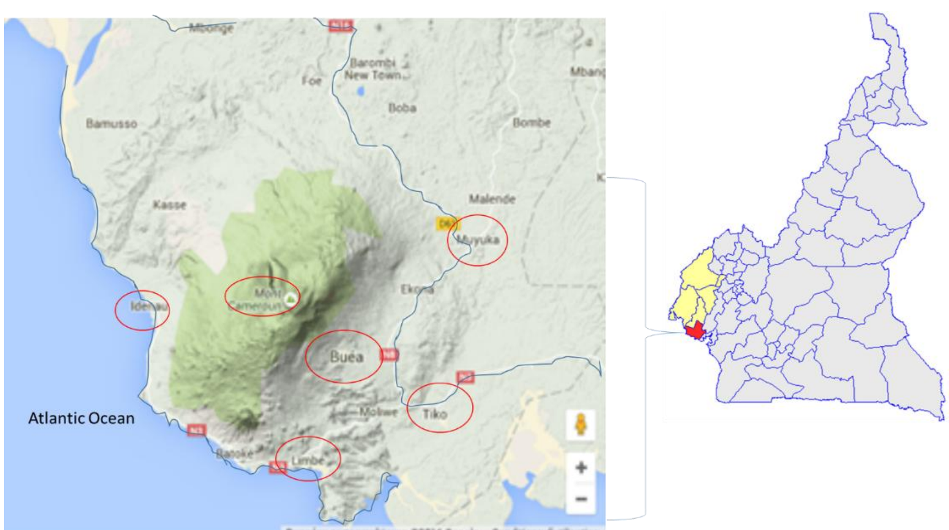

Fig. 1a. Geographical location of the mount Cameroon area and sites of samples collection.

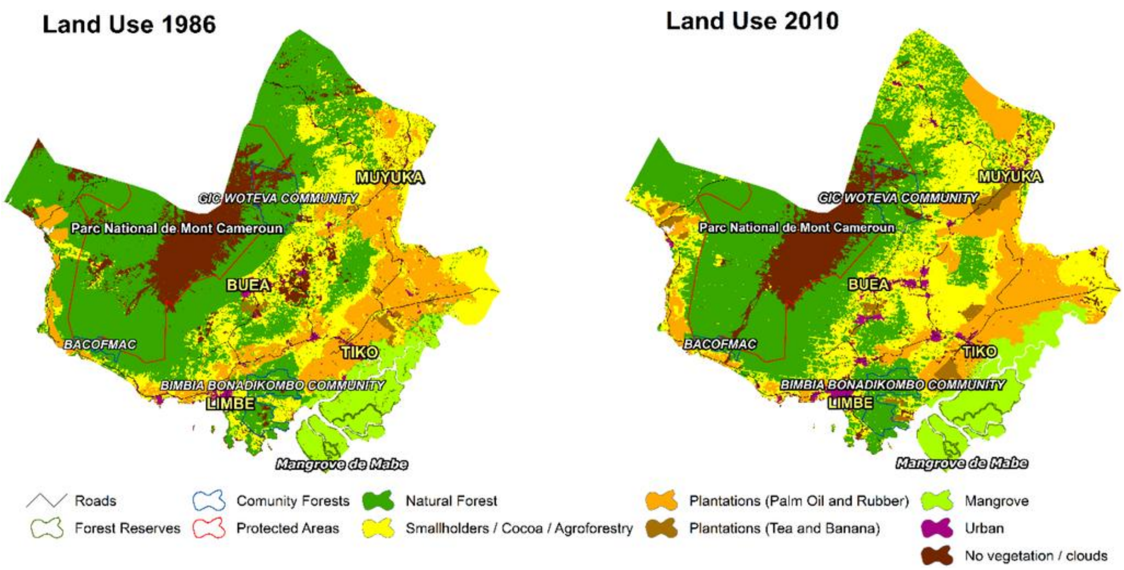

Fig. 1b. Land use in the mount Cameroon area.

2.2. Morphological and biometrical characterization of the different accessions

The identification of potential accessions of *Brachiaria* amongst other forages was based on observation of the panicle for ovate or oblong spikelets arranged on one sided raceme with the lower glume adjacent to the rachis in accordance with Renvoize et al. (1996). The *in situ* morphological characterization of *Brachiaria* accessions was carried out according to descriptors selected from those established by the International Rice Research Institute and the International Board for Plant Genetic Resources (IRRI, 1980; Nascimento et al., 2011). A total of twelve qualitative and eleven quantitative traits were considered (Table 1).

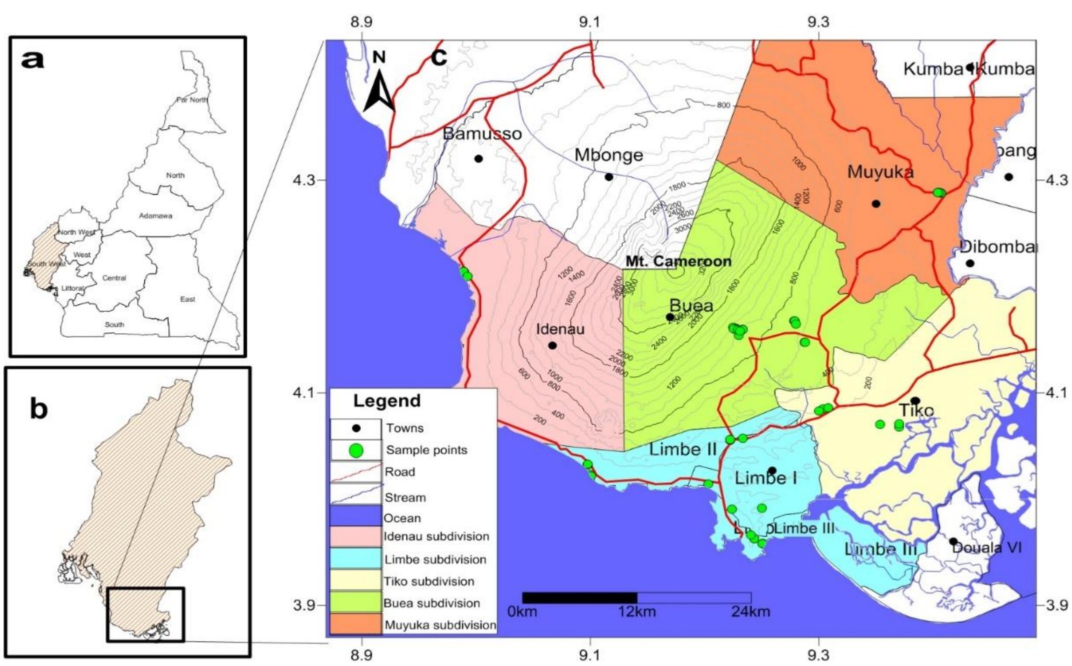

Fig. 2. Map of the mount Cameroon area, showing the various sampling points (round green dots) (Remote Sensing Unit University of Buea, 2016).

2.3. Data collection

Field data collection was done using a structured questionnaire. The GPS coordinates and altitude of sampling locations where data were recorded and are plotted (Fig. 2). The coloured traits were recorded by comparing the plant part's colour with that of a standard colour chart. Microscopic morphological traits like leaf pubescence, presence/distribution of awn per panicle, apiculus and/or awn colour, glumella pubescence and grain shape were recorded with the help of a hand lens of 10x magnification.

Table 1
Descriptors for the evaluation of *Brachiaria* accessions.

Qualitative descriptors	**Phenotypic classes**	**Evaluation phase**
Leaf colour (LC)	1 - light green; 2 - green; 3 - dark green; 4-purple	Early flowering
Leaf pubescence (LP)	1 - glabrous; 2 - intermediate; 3 - pubescent	Early flowering
Lodging resistance (LR)	1 - absent; 2 - plants with moderate lodging; 3 - plants strongly lodged	After maturation
Inter node colour (IC)	1 - green; 2 - gilded; 3 - purple; 4 - purple stripes	Grain filling
Stigma colour (SC)	1 - white; 2 - light green; 3 - yellow; 4 - purple	Flowering
Presence/distribution of awn per panicle (PDAP)	1 - absent; 2 - awns on panicle extremity; 3 - awns throughout the panicle	After grain filling
Apiculus and/or awn colour (AAC)	1 - straw colour; 2 - gilded; 3 - red; 4 - purple; 5 - brown; 6 - green; 7 - black	After grain filling
Lemma and palea colour (LPC)	1 - straw colour; 2 - gilded or with gilded lines; 3 - straw colour with brown stains; 4 -brown; 5 - reddish; 6 - straw colour with purple lines; 7 - purple; 8 - black; 9 - white	Maturation
Glumella pubescence (GP)	1 - absent/very weak; 2 - present	Flowering
Panicle type (PT)	1 - grouped; 2 - intermediate; 3 - open	Maturation
Threshability (Th)	1 - difficult (less than 25 % of the grains were removed); 2-intermediate (from 25 % to 50 % of the grains were removed); 3 - easy (more than 50 % of the grains were removed)	After maturation
Grain shape (GS)	1 - round (length/width ratio was less than 1.50); 2 - semi-round (ratio ranged from 1.50 to 2.00); 3 - semi-elongated (ratio from 2.01 to 2.75); 4 - elongated (ratio from 2.76 to 3.50); 5 - very elongated (ratio larger than 3.50)	After maturation
Quantitative descriptors		
Flag leaf length (FLL)	Arithmetic means of the six random samples	Anthesis
Flag leaf width (FLW)	Arithmetic means of the six random samples	Anthesis
Culm length (CL)	Arithmetic means of the six random samples	Grain filling
Culm diameter (CD)	Arithmetic means of the six random samples	Grain filling
Number of tillers per plant (NTPP)	Arithmetic means of the six samples	Grain filling
Number of panicles per plant (NPP)	Arithmetic means of the six samples	After maturation
Panicle length (PL)	Arithmetic means of the six random samples	After maturation
Number of spikelets per panicle (NSPP)	Arithmetic means of the six random samples	Maturation
Panicle fertility (PF)	Arithmetic means of the six random samples	After maturation
Ratio between length and width of the grain (RGLW)	Arithmetic means of the six random samples	After maturation
1000-grain weight (TGW)	Arithmetic means of the six random samples	After maturation

The quantitative descriptors, their definitions, units of measurement and instrument of measurements are as shown in Table 2 and some are illustrated by Fig. 3.

Table 2
Qualitative traits, their definitions and units of measurement.

	Qualitative traits	Definition	Units	Instrument of measurement
1	Flag leaf length	Length from the ligule to leaf apex	cm	Meter rule
2	Flag leaf width	Distance at widest point of the leaf	cm	Meter rule
3	Culm length	Distance from the surface of the ground to the base of the flag leaf	cm	Centimeter measuring tape
4	Culm diameter	Thickness of culm at lowest internode	mm	Vernier caliper
5	Panicle length	Length of panicle from the base of the flag leaf to the plant apical bud	cm	Tape
6	Panicle fertility	Percentage of fertile seed at maturity	%	Manual counting
7	1000-grain weight	Dry weight of 1000-grains	mg	Milligram-balance

The taxonomic identification up to species level was checked on all accessions using keys from Phillips (1995).

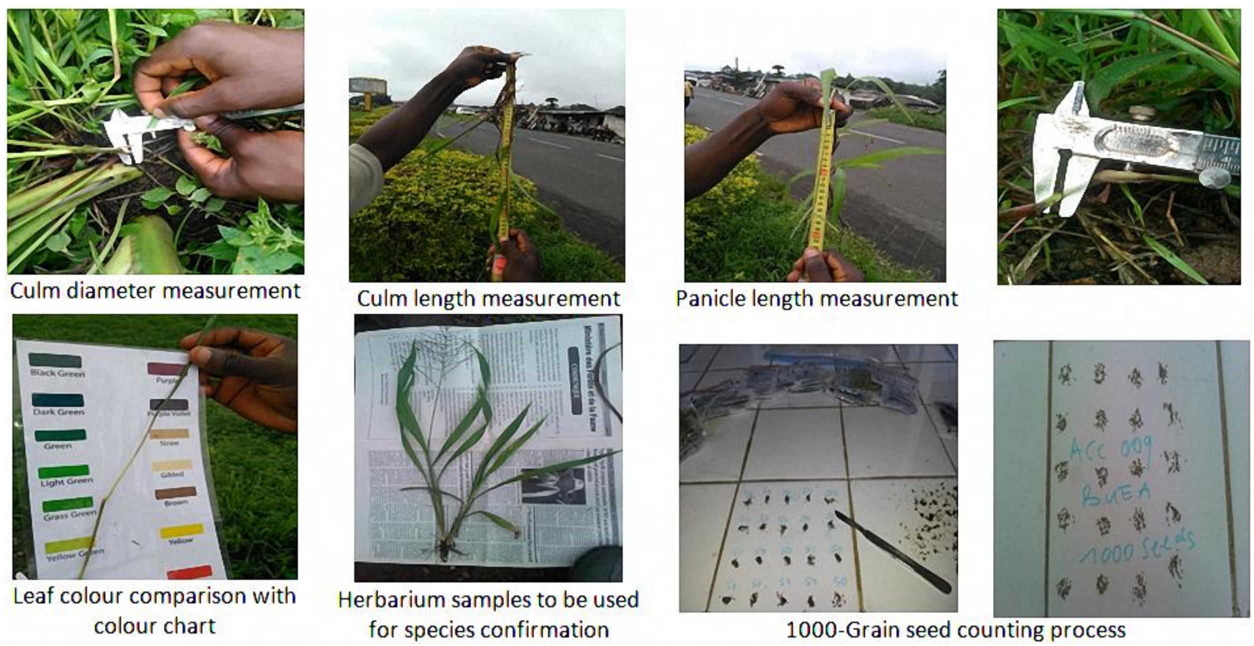

Fig. 3. Qualitative evaluation and quantitative measurements of *Brachiaria spp*.

2.4. Data processing and statistical analysis

2.4.1. Morphological characterization

For data analysis, the statistical significance was set at 5%. The descriptive statistics was use for qualitative data and ANOVA and correlation were done for the quantitative variable. Distances between accessions were carried out using morphology and measurements, calculating dissimilarity and distance using UPGMA (Unweighted Pair Group Method Arithmetic Mean) to generate the dendrogram. The relation between the different accessions was established to construct the phylogenetic tree following the protocol of the hierarchical ascending classification. All these analysis were done using SPSS 20.0 and XLSTAT.

3. Results and discussion

3.1. Morphological characteristics

Table 3 summarises the distribution of qualitative traits of *Brachiaria spp* germplasm in the area of mount Cameroon.

Table 3
Frequency distribution of the qualitative traits of *Brachiaria spp* germplasm in the area of mount Cameroon.

Parameters	Characteristics	Percentage	Parameters	Characteristics	Percentage
Leaf colour	Grass green	10.9	**Leaf pubescence**	Pubescent	34.5
	Light green	70.9		Adaxialpubescent	1.8
	Green	12.7		Glabrous	25.5
	Light green with brown margins	1.8		Intermediate	29.1
	Yellow green	3.6		Marginal	9.1
	Total	100.0		Total	100.0
Lodging resistance	Plant strongly lodged	47.3	**Presence/distribution of own panicle**	Owns throughout the panicle	25.5
	Absent	7.3		Absent	74.5
	Plant with moderate lodged	45.5		Total	100.0
	Total	100.0			
Internode colour	Yellow green	25.5	**Lemma and palea colour**	Straw colour	41.8
	Gilded	7.3		Gilded	20.0
	Green	12.7		Purple	9.1
	Brown	18.2		Light green	5.5
	White	1.8		Brown	12.7
	Purple stripe	1.8		White	7.3
	Purple brown	3.6		Grass green	3.6
	Purple	7.3		Black	5.5
	Purple vide	1.8		Total	100.0
	Light green	14.5	**Treshability**	Easy	38.2
	Light brown	1.8		Intermediate	9.1
	Whitish green	3.6		Difficult	52.7
	Total	100.0		Total	100.0
Stigma colour	White	40.0	**Apiculus and/or awn colour**	Absent	74.5
	Black	16.4		Gilded	1.8
	Purple	32.7		White	1.8
	Brown	1.8		Purple	10.9
	Yellow	5.5		Straw colour	3.6
	Light purple	3.6		Brown	1.8
	Total	100.0		Total	100.0
Panicle type	Open	61.8	**Glumella pubescence**	Absent	85.5
	Intermediate	38.2		Present	14.5
	Total	100.0		Total	100.0
Grain shape	Round	74.5			
	Semi-round	16.4			
	Elongated	7.3			
	Semi-elongated	1.8			
	Total	100.0			

The majority of the *Brachiaria spp* have light green coloured leaves (70.9%), which are mostly pubescent (34.5%), intermediate (29.1%) or glabrous (25.5%). The internodes are mostly yellow green (25.5%), the stigma is mostly white (40.0%), while the own panicle is mostly absent. The panicles are mostly open (61.8%), while the grains are mostly round shaped (74.5%). The glumella pubescence is mostly absent, while the lemma and palea mostly have a straw colour and the apiculus and/or awn colour are mostly absent (74.5%).

3.2. Biometric characteristics

Table 4 summarizes the means and coefficient of variation of the measurements of *Brachiaria spp* germplasm with respect to localities in the area of mount Cameroon. The first observation is that there is huge variability for the quantitative traits whatever the locality is. The traits varying most are the number of panicle per plant (146.86%), the grain weight (141.17%) and the number of spikelets per plant (135.10%). There is no significant difference between localities for the culm length, panicle length, ratio between length and width of seeds and flag leaf width. However, the flag leaf length are higher in Buea and Idenau, which are the most rainy and humid localities. The culm diameter are significantly greater in the warmest localities (Tiko, Muyuka and Limbe1). It could also be noticed that many other measurements vary significantly according to localities.

3.3. Pearson's correlation between the measurements of *Brachiaria spp* germplasm

Table 5 presents the correlation coefficients between the measurements of *Brachiaria spp* germplasm in the area of mount Cameroon. The correlation in general vary from weak to moderate. The highest positive significant (p<0.01) correlation coefficient (0.637**) is observed between the flag leaf length and the panicle length, while the significant native correlation (-0.283*) was found between the panicle fertility and the flag leaf width.

Table 4

Means and coefficient of variation of the measurements of *Brachiaria spp* germplasm with respect to localities in the area of mount Cameroon.

		localities								
Traits		Tiko	Muyuka	Buea	Mount Cameroon	Limbe 1	Limbe 2	Limbe 3	Idenau	Total
FLL	$\bar{X}$ ± S.D	12.72±4.5^{a}	10.34±2.10^{a}	19.62±19.31^{a}	11.16±7.70^{a}	10.83±6.76^{a}	14.64±5.30^{a}	12.91±6.07^{a}	18.39±10.81^{a}	13.91±9.61
	CV (%)	35.53	20.31	98.42	69.00	62.42	36.20	30.49	58.78	69.09
CL	$\bar{X}$ ± S.D	45.92±15.89^{a}	43.18±37.20^{a}	45.77±37.60^{a}	26.90±11.31^{a}	39.57±10.86^{a}	45.49±22.52^{a}	38.94±50.95^{a}	25.76±12.26^{a}	39.63±29.91
	CV (%)	34.60	86.15	82.15	42.04	27.45	49.51	130.84	47.59	75.47
CD	$\bar{X}$ ± S.D	2.36±0.42^{b}	1.91±0.66^{ab}	1.58±0.91^{ab}	1.39±0.63^{ab}	2.09±0.83a^{ab}	1.78±0.93^{ab}	0.89±0.67^{a}	1.72±0.99^{ab}	1.65±0.82
	CV (%)	17.80	34.55	57.59	45.33	39.71	52.25	75.28	57.56	49.70
NTPP	$\bar{X}$ ± S.D	11.39±10.51^{a}	8.77±6.01^{a}	13.89±6.69^{a}	21.71±22.66^{b}	10.00±3.56^{a}	20.92±20.51^{b}	8.33±4.74^{a}	12.17±5.06^{a}	13.09±11.39
	CV (%)	92.28	68.53	48.16	104.38	0.36	98.04	56.90	41.58	87.01
NPP	$\bar{X}$ ± S.D	4.00±0.33^{a}	19.50±30.11^{c}	8.50±5.94^{b}	10.19±9.37^{b}	10.50±5.91^{b}	2.29±0.61^{a}	5.36±3.27^{a}	3.50±0.50^{a}	8.43±12.38
	CV (%)	8.25	154.41	69.88	91.95	56.29	26.64	61.01	14.29	146.86
PL	$\bar{X}$ ± S.D	34.82±23.27^{a}	33.75±14.45^{a}	41.05±24.62^{a}	32.08±8.74^{a}	25.05±11.38^{a}	29.46±6.94^{a}	35.28±13.23^{a}	28.88±7.25^{a}	33.26±14.83
	CV (%)	66.83	42.81	59.98	27.24	45.43	23.56	37.50	25.10	44.59
NSPP	$\bar{X}$ ± S.D	437.11±146.3^{b}	149.37±167.4^{a}	468.44±427.1^{b}	797.13±124.4^{c}	161.00±146.4^{a}	249.87±196.7^{ab}	368.33±391.3^{a}	223.33±115.5^{ab}	359.45±485.6
	CV (%)	33.47	112.04	91.18	15.60	90.91	78.74	106.23	51.71	135.10
PF	$\bar{X}$ ± S.D	91.67±2.89^{b}	76.00±29.03^{ab}	71.17±25.30^{ab}	72.50±23.63^{ab}	67.00±33.01^{a}	50.00±40.83^{a}	56.33±45.53^{a}	66.67±23.09^{a}	67.94±30.79
	CV (%)	3.15	38.20	35.55	32.59	49.27	81.66	80.83	34.63	45.32
RBLW	$\bar{X}$ ± S.D	1.00±0.00^{a}	1.40±0.55^{a}	1.17±0.41^{a}	1.00±0.00^{a}	1.00±0.00^{a}	1.00±0.00^{a}	1.00±0.00^{a}	1.17±0.00^{a}	1.10±0.29
	CV (%)	0.00	39.29	35.04	0.00	0.00	0.00	0.00	0.00	26.36
GW	$\bar{X}$ ± S.D	1.81±2.67^{b}	0.60±0.54^{a}	0.35±0.37^{a}	0.35±0.37^{a}	1.30±1.17^{b}	0.50±0.35^{a}	0.60±0.84^{a}	0.37±0.29^{a}	0.68±0.96
	CV (%)	147.51	90.00	105.71	105.71	90.00	70.00	140.00	26.13	141.17
FLW	$\bar{X}$ ± S.D	1.12±0.54^{a}	1.04±0.37^{a}	1.31±0.82^{a}	0.96±0.59^{a}	0.67±0.13^{a}	1.08±0.32^{a}	1.79±1.92^{a}	1.11±0.25^{a}	1.18±0.91
	CV (%)	48.21	35.58	62.60	61.46	19.40	29.63	107.26	22.52	77.11

n: Sample size; $\bar{X}$ ± E.S: means ± Standard deviation; CV (%) = Coefficient of variation; $^{a, b, c}$. on the same row, means with the same superscripts are not significantly different (P≤0.05); FLL: Flag leaf length; FLW: Flag leaf width; CL: Culm length; CD: Culm diameter; NTPP: Number of tiles per plant; NPP: Number of panicles per plant; PL: Panicle length; NSPP: Number of spikelet per plant; PF: Panicle fertility; GW: Grain weight; RBLW: Ratio between length and width grain.

Table 5

Correlation coefficient between the measurements of *Brachiaria spp* germplasm in the area of mount Cameroon.

	FFL	FLW	CL	CD	NTPP	NPP	PL	NSPP	PF	RBLW	GW
FFL	1										
FLW	0.371**	1									
CL	0.236	0.243	1								
CD	0.147	-0.119	0.169	1							
NTPP	0.327*	0.094	0.062	0.142	1						
NPP	0.055	0.141	0.127	0.070	0.055	1					
PL	0.637**	0.343*	0.391**	0.243	0.182	0.112	1				
NSPP	0.256	0.110	0.065	0.143	-0.007	-0.084	0.377**	1			
PF	0.001	-0.283*	-0.153	0.463**	-0.008	0.111	0.092	0.351*	1		
RBLW	-0.155	-0.044	0.087	-0.304*	-0.183	0.315*	-0.046	-0.242	-0.250	1	
GW	0.026	-0.003	0.440**	0.146	-0.016	-0.008	-0.032	-0.050	0.069	0.062	1

*=p<0.05: The correlation is significant at 0.05; **=p<0.01: The correlation is significant at 0.01; FLL: Flag leaf length; FLW: Flag leaf width; CL: Culm length; CD: Culm diameter; NTPP: Number of tiles per plant; NPP: Number of panicles per plant; PL: Panicle length; NSPP: Number of spikelet per plant; PF: Panicle fertility; GW: Grain weight; RBLW: Ratio between length and width grain.

3.4. Genetic variability and structure of the population of *Brachiaria spp* germplasm

The genetic variability and population structure based on the principal components of *Brachiaria spp* germplasm in the area of mount Cameroon is presented in Table 6 and illustrated by Fig. 4 and 5. Eigen values are measures of the data variance explained by each of the new coordinate axis. They are used to reduce the dimension of large data sets by selecting only a few modes with significant eigen values and to find new variables that are uncorrelated. They are hence very helpful for least-square regressions of badly conditioned systems.

Table 6
Genetic variability observed within the studied population based on the principal components of *Brachiaria spp* germplasm in the area of mount Cameroon.

	F1	F2	F3	F4	F5	F6	F7	F8	F9	F10	F11
Eigen value	2.541	1.888	1.448	1.187	1.071	0.717	0.652	0.525	0.404	0.324	0.241
Variability (%)	23.104	17.160	13.164	10.791	9.738	6.522	5.930	4.777	3.676	2.944	2.195
Cumulative (%)	23.104	40.265	53.429	64.219	73.958	80.479	86.409	91.186	94.861	97.805	100.00

F1: Flag leaf length; F2: Flag leaf width; F3: Culm length; F4: Culm diameter; F5: Number of tilles per plant; F6: Number of panicles per plant; F7: Panicle length; F8: Number of spikelets per plant; F9: Panicle fertility; F10: Grain weight; F11: Ratio between length and width grain.

The cumulative variance of the first five principal components explains 73.958% the genetic variability observed within the studied population. The first three components, ie the flag leaf length, flag leaf width and culm length are those contributing most to that variability with respectively 23.10%, 17.16% and 13.16% giving a total cumulative genetic variability value of 53.43% of the whole *Brachiaria spp* population germplasm in the area of mount Cameroon.

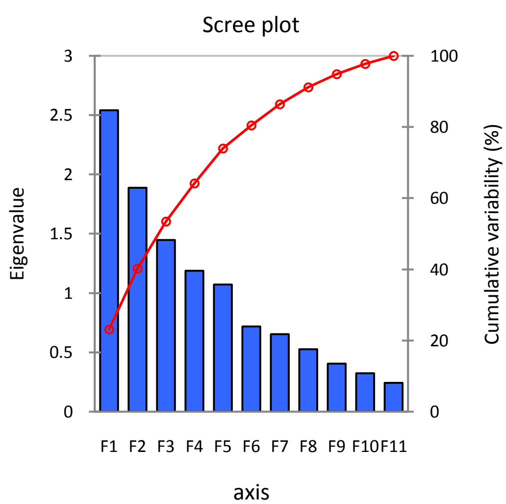

Fig. 4. Contribution of principal components to the genetic variability of *Brachiaria spp* germplasm in the area of mount Cameroon.

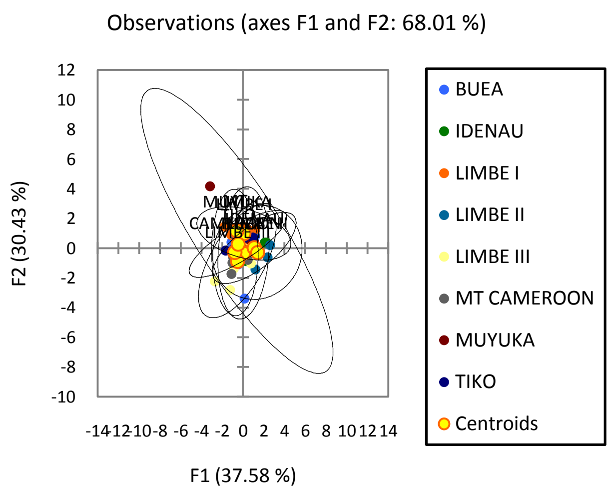

Fig. 5. Population structure of *Brachiaria spp* germplasm in the area of mount Cameroon.

Fig. 4 and 5 illustrating respectively the contribution of the different principal components to the variability within the population and the population structure of *Brachiaria spp* germplasm in the area of mount Cameroon lets appreciate how closed are the different populations assigned considering the localities.

3.5. Phylogenetic relation between the different accessions of *Brachiaria spp* germplasm in the area of mount Cameroon

Fig. 6 and Table 6 illustrate respectively the dendrogram and the genetic distance between and within the different accessions of *Brachiaria spp* germplasm in the area of mount Cameroon.

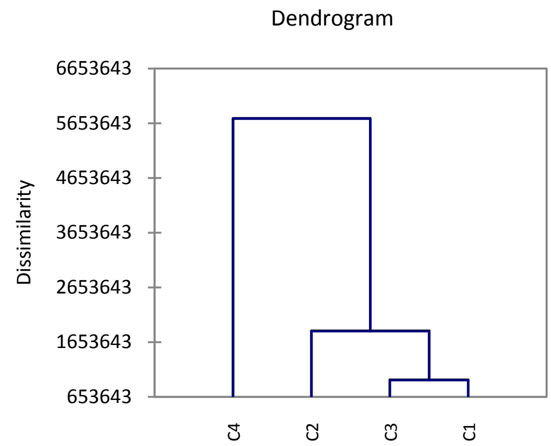

Fig. 6. Hierarchical clustering displaying the potential four clusters/accessions of *Brachiaria spp* germplasm in the area of mount Cameroon.

It comes from the hierarchical clustering that there could exist four main clusters or accessions of *Brachiaria spp* germplasm in the area of mount Cameroon.

Table 7
Matrix of distances between the different accessions of *Brachiaria spp* germplasm in the area of mount Cameroon.

	1	2	3	4
1	0			
2	312.252	0		
3	734.987	1045.802	0	
4	2238.808	2549.305	1506.100	0

It comes from Table 7 that accession 4 is the most distant from the three others, while accessions 1 and 2 are the closest. This brings the necessity of phenotyping and carrying out the molecular characterization of these forage genetic resources. The variability of *Brachiaria spp* genetic resources observed on the field are illustrated by Fig. 7.

Natural plot with numerous species of grasses and legume sinter cropped

Some observed wild variants of *Brachiaria spp* in natural environment

Fig. 7. Typical plot in the mount Cameroon area with numerous species of grasses and legumes intercropped.

These are mostly primary populations, based on the fat that no history of forage programme is known in the area of study. Such resources should be carefully explored for their potential contribution to the genetic improvement programme of *Brachiaria* germplasm in Africa.

The taxonomic identification of the herbarium specimens up to the variety level was not possible due to the inaccessibility of a suitable Identification key. The accessions were simply compared with literature references, taking into consideration the major characteristics that separate the genus *Brachiaria* from other members of the Paniceae (ovate or oblong spikelets, arranged in one-sided racemes with the lower glume adjacent to the rachis). The observed phenotypic variability could be attributed to water deficiency before flowering which changes the density of the panicle branches and the density of the grains (Bonow et al., 2007). In general, plants morphology is affected by both their geographical situation and the physical factors of the environment like the soil type, humidity and temperature, rainfalls etc., and by the intrinsic genetic factors of the plants themselves (Nascimento et al., 2011). Further, Siddiqui et al. (2007) is of the opinion that grain shape should be considered as a quantitative trait, which are probably due to the actions of many genes and the environment.

The high variability in qualitative and quantitative variable is in line with the work of Veasey et al. (2008) who reported variations within and amongst population, but differ from that of Nascimento et al. (2011) where it was classed amongst the least variable. The internode colour varies with the age of the plant and the fertility condition of the soil. The colour goes from dark green to light green on other. The high variability as mentioned earlier is both by genetic and environmental forces which if taken care of will give us a good stand to selecting superior germplasm for breeding programmes. The correlation analysis of the quantitative descriptors showed some degree of relatedness amongst the traits, which could limits the number of traits use in analysis to avoid indirect weighting and the usage of not reliable characters in measurement. With the leaf descriptors; the flag leaf length (FLL) and the Flag leaf width (FLW) showed weak but positive significant correlation (r=0.371). This implies that in subsequent analysis using one of the descriptors could be enough serve to predict the other. Both leaf aspects and leaf growth rate are key factors in selecting new cultivars for higher biomass production. The flag leaf is very instrumental component in seed-setting or grain filling.

The strong positive correlations recorded between the panicle length (PL) with major agronomic descriptors like the FLL (r=0.637** highest recorded), FLW (r=0.343*) and Culm length, CL, (r=0.391**) could be attributed to the fact that the resources for the panicle development are supplied mostly by the flag leaf which is nearest to the panicle and the culm. It is reasonable to drop less reliable traits like the CL with are difficult to measure in decumbent accession. The number of spikelets per panicle (NSPP)'s strong relatedness to the panicle fertility (PF) could easily be accounted for by the fact that the seeds originate from the spikelets. As such, without them the plant will not be fertile. The positive significant correlation also showed between the PF and CD could be understood from the stand point that the nutrients like water needed for the establishment of the panicle are supplied by the culm, whose diameter determine the quantity sent at even flowering. Also PF and NSPP correlation could be justified from the fact that only spikelets can be fertilized and the greater its number the greater chance of fertility. This as well allow for the elimination of estimated traits like the panicle fertility. The positive relatedness of both seed aspects (1000-grain weight, TGW and ratio of grain length to width, RGLW) enables the RGLW to be used, avoiding the tedious TGW process of even lesser accuracy. Total grain weight is dependent on panicle fertility and grain filling which is highly dependent on environmental effects and shows highly significant variation among accessions and locations, making it a less suitable character for use as a stable trait in characterization of germplasm (van de Wouw et al., 2008). The PF show a negative relation (r= -0.271) with the FLW. The relatedness of the traits helps in elimination of traits with difficult to measure to avoid indirect weighting and have better traits for subsequent analysis. This is in line with the work of van de Wouw et al. (2008).

When the regression analysis was considered on the 11 quantitative traits in the eight different locations of Fako, significant differences among accessions ($p<0.05$) were observed for three of the traits. The accessions for the different locations were almost similar for 8 of the 11. The mean FLW for the accessions divided locations into 2 clusters (Buea, Idenau, Limbe II, Muyuka, Mount Cameroon) and the (Tiko, Limbe I and Limbe III) clusters. This could be accounted for by the availability of different species between the clusters. The CL showed a significant difference ($P<0.05$) that divided the locations into two cluster as well (Buea, Idenau, Limbe I, Limbe II, Limbe III, Muyuka, Mount Caneroon and Tiko) and Limbe I. NSPP showed significant difference ($P<0.05$) on its part divides the location into 3 clusters (Buea, Limbe I, Limbe III, Mount Cameroon and Tiko), (Idenau and Limbe II) and Muyuka; this could be accounted for by the fact that there exist a great distance between the accessions of the various locations for NPP. The significant differences (indications of large distances) between locations for the

FLW, CL and NSPP that indirectly were correlated (Table 3) by the PL, demonstrate a certain degree of interspecific diversity. Though according to Fukuoka et al. (2006) and Nascimento et al. (2011) significant variation maybe found amongst genotypes with the same name. This difference is as a result of difference in locational characteristics that may favour some species and not others or may favour polymorphism within species.

Comparing the means, standard error and confidence interval computed for the qualitative traits in the different locations shows that at least one of the locations was highest in a character. Limbe III had accessions with the largest leaf aspects. That is, longest and widest FLL and FLW respectively, confirmed by the positive correlation they demonstrated earlier, while Limbe I seating at a higher altitude than II and III had the shortest and narrowest FLL and FLW. Limbe III again recorded accession with the tallest average culms, taller than the average stem length for Buea accessions which occupied the last position. About the thickness of the stem at lowest point, Limbe I that recorded the lowest leaf aspects, recorded accessions with the thickest average culm, way thicker than those of Tiko accessions that occupied the bottom position. The highest mean number of tillers per plant value was recorded in the Limbe II accessions, while Idenau recorded the least value. Also, the accession in Muyuka on average showed the highest number of panicles per plant, while the Idenau accessions had very small number of panicles per plant. As such seed production rate in them could be negatively affected. Limbe III once more had accessions, which had averagely tallest panicles than the rest of the locations, while Tiko had plants with the shortest panicles. Mount Cameroon recorded accessions with the highest number of mean spikelets, far above the average number showed by Limbe I occupying the last position. This could be explained by the differences in environmental factors and altitude of these localtions, despite de fact that they are not too distant from each other. The shape of the grains too, another trait of agronomic interest saw Idenau having the most elongated grains, while Limbe III had accessions with the greatest number of rounded seeds on average. Grain shape is mostly affected by polygene action and by environmental forces (Nascimento et al., 2011). The average weight of 1000-Grains indicates that Limbe III has best heavy and well grain filled grains while Idenau at a relatively lower altitude having accessions with the lightest grains or unfilled grains. Muyuka accessions had the highest mean PF value, far larger than the value for Idenua recording the lowest mean value. This could be explained by the fact that Muyuka seats has a higher environmental temperature, conditions that could favour fertility of the panicles. The variability in these traits complements the work of Veasey et al. (2008) who also found significant differences and high variability for 11 populations of *O. glumaepatula*, mainly for the characters number of tillers, plant height at flowering, leaf length and width, culm length, panicle number, panicle height, flag leaf length and spikelet length amongst others.

4. Conclusion

The great variability and distances both in quantitative and qualitative descriptors leaves us with the conclusion that there are different variants of *Brachiaria spp* in the mount Cameroon area. Also, looking at the variability of the qualitative traits that are influenced by the environment, it can be said that the existing accessions varied with ecotype. Seed setting as a very important agronomic feature of *Brachiaria* production also varied with the environmental conditions, particularly temperature as Tiko that has the highest temperature range record the highest TGW. In the nut shell, the successful clustering into groups of accessions with similar morphological within the collection *Brachiaria* germplasm in this study will enable researchers interested in this genus to easily identify subsets of accessions for further evaluation aimed at specific uses. Morphological differences between these accessions and taxonomic differentiation to species level will need molecular characterization for confirmation.

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