



GLOBAL JOURNAL OF SCIENCE FRONTIER RESEARCH: C  
BIOLOGICAL SCIENCE  
Volume 24 Issue 1 Version 1.0 Year 2024  
Type: Double Blind Peer Reviewed International Research Journal  
Publisher: Global Journals  
Online ISSN: 2249-4626 & Print ISSN: 0975-5896

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**GJSFR-C Classification:** ASFA: Q1 01403



ASSESSMENT OF MORPHOLOGICAL DIVERSITY IN CASSAVA (*Manihot esculenta* Crantz) CORE COLLECTION: INSIGHTS FOR GERmplasm CONSERVATION AND BREEDING IN TOGO

*Strictly as per the compliance and regulations of:*



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# Assessment of Morphological Diversity in Cassava (*Manihot esculenta* Crantz) Core Collection: Insights for Germplasm Conservation and Breeding in Togo

Gmakouba Tighankoumi<sup>α</sup>, Dzidzienyo K. Daniel<sup>σ</sup>, Some Koussao<sup>ρ</sup>, Tongoona Pangirayi<sup>ϑ</sup> & Asante I. Kwame<sup>¥</sup>

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**Results and implications:** Overall, high morphological diversity was observed among the cultivars for all the traits evaluated. The most diverse traits included petiole color, leaf color, leaf vein color, flowering and seed set abilities, branching levels, end branch color, stem epidermis color, lobe margins, and growth habit of stem. Seven morphotypes with interesting features were identified through cluster analysis. Morphotype 1 is made of unflowering and unbranching cultivars with greenish-red petioles. Morphotype 2 is composed of varieties exhibiting purple petioles, three levels of branching, dichotomous branching habit, good flowering and seed set ability. Morphotype 3 made of only one cultivar was considered as outlier. Morphotype 4 cultivars are characterized by red petioles, compact plants, white root pulp and bad seed set ability; whereas cultivars belonging to morphotype 5 exhibited green petiole, good flowering and seed set abilities. Morphotype 6 is made of cultivars with sessile peduncle root, conical cylindrical root, orange root pulp, short distance between leaf scars, good flowering and seed set ability. Morphotype 7 genotypes exhibited dark green apical leaves, cream stem epidermis, two levels of branching, good flowering and seed set ability. The most diverse traits identified in this study could be used for genetic resources

identification. Parent cultivars could be selected from morphotypes harbouring good flowering and seed set abilities.

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## I. INTRODUCTION

Cassava belongs to the family of Euphorbiaceae and includes 98 species. The crop is native to the American continent, being distributed from the USA to Africa. The main diversity center of cassava (Brazil) posses at least 78 species, approximately 80% of the total number of species. *M. esculenta* is its only domesticated species (Rogers and Appan, 1973).

Cassava plays an essential part in the food security of millions of families in tropical and subtropical regions of Africa. It is one of the main sources of carbohydrates, especially in developing regions, where it is grown as subsistence crop (FAO, 2023). Cassava has a wide range of uses in the so-called '4Fs' of: (i) food for human consumption, (ii) feed for animals, (iii) fuel, which in the form of ethanol is produced from cassava, and (iv) factories, where it is used to make alcohol, citric acid, clothing, medicines, paper, and chemicals. For many years, global demand for cassava has grown strongly due to its many industrial uses and the fact that it has often been cheaper than other starchy crops. This has then lifted it to the status of being the world's 5<sup>th</sup> most important crop, after corn, wheat, rice, and potatoes.

In 2022, 303 million tons of cassava were produced globally worldwide, grown on 23.87 million hectares, with an average yield of 11.24 t ha<sup>-1</sup> (FAO, 2023). In Togo, cassava used to be a crop of the poor for a long time, but of late it is becoming more of a staple crop especially in the areas of production (Sogbedji et al., 2015). Across the country, 10,297 hectares were occupied by cassava plantations, and 38,542 tons of cassava root were harvested in 2022, with an average yield of 4 t ha<sup>-1</sup>, 75% lower than the global productivity (DSID, 2022).

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Cassava is a diploid ( $2n=36$  chromosomes) and monoicous species, with predominantly allogamous fertilization, making it highly heterozygotic (Pootakhan et al. 2014) and giving it high genetic diversity, even though it propagates vegetatively (Costa et al. 2013). Cassava can adapt to different edapho climatic conditions, such as drought and low-fertility soils (Vidal et al. 2015). Because of these characteristics, cassava cultivation is attractive to farmers with limited resources in Togo.

Despite the importance of cassava as a staple crop (Sogbedji et al., 2015), its genetic diversity is poorly documented and consequently the genetic improvement of this crop is limited in Togo (Kombaté et al., 2017). The study of Kombaté et al. (2017) using ethnobotanical survey and morphological descriptors revealed the existence of high diversity. However, ethnobiological studies involving farmers' knowledge in varietal classification have shown large variations according to Agre et al.(2017). Also, there is no consistency in the naming of varieties by farmers. This results in the possibilities of duplicates and mislabelling within the local varieties collected from farmers' fields. Additionally, the number of local varieties with different features and names, most often planted together in a single field, suggest the existence of high diversity within this crop (Siqueira et al., 2009; Rabbi et al., 2015b), which is important for plant breeding and genetic resources programs. This substantial genetic variability is due to the high heterozygosity of the crop, ease of natural cross pollination, fruit dehiscence, and to the volunteer seedlings in farmers fields (Rabbi et al., 2015b; Ceballos et al., 2016). Besides, the informal plant material exchange between farmers promotes a large number of new cultivars and expand cassava genetic diversity (Peprah et al., 2020).

From a point of breeding, small-farm cultivation of cassava is of great importance to the conservation of genetic resources. Exploring the morphological diversity of a given germplasm is fundamental to guide its conservation, management and use in conventional breeding programs (Ceballos et al., 2016).

In West Africa, genetic diversity studies have been carried out for cassava germplasm management and breeding using both morphological descriptors (Adjebeng-Danquah et al., 2016; Agre et al., 2017; Kamanda et al., 2020) and molecular markers (Rabbi et al., 2014; Soro et al., 2024). In addition, multivariate analyses allows for the simultaneous integration of data for multiple traits and has been widely used to quantify the phenotypic diversity in several crops (Kamanda et al., 2020, Soro et al., 2024).

Morphological descriptors are inexpensive and easy to record for most breeders compare to molecular markers. They are strongest determinants of taxonomic classification and agronomic value of varieties (Soyode and Oyetundi, 2009; Rabbi et al., 2015b).

The objective of this study was to explore the phenotypic diversity in a core collection of cassava cultivars based on thirty two (32) morphological traits.

## II. MATERIAL AND METHODS

### a) *Plant Material*

A core collection (Table 1) made of: i. one hundred (100) cultivars obtained from major cassava growing areas across the country, ii. thirty five (35) improved varieties introduced from IITA cassava breeding program, iii. seven (7) cultivars sourced from the gene bank of the Laboratory of Virology and Biotechnology of the University of Lome and iv. two (2) varieties obtained from the cassava gene bank of Embrapa Mandioca Fruticultura (Cruz das Almas, BA, Brazil) was used in this study. Five improved varieties (high yielding and CMD resistant) namely Gbazekoute, TMS 96\_0409, TMS 96\_0166, CRI Sika Bankye and CRI Among bankye, recently released by the national cassava breeding unit were used as checks.

### b) *Experimental Site*

The experiment was run at the Togolese Agronomic Research Institute (ITRA) station of Davié (latitude: 6° 23' 5" North; longitude: 1° 12' 18" East; altitude: 76 meters) located in the cassava production belt. This site is representative of typical cassava-growing conditions in Togo and is characterized by a bimodal rainfall pattern. During the experimentation, a total rainfall of 1231.5 mm was recorded for 80 rainy days. July was the highest monthly rainfall with 207.8 mm for 14 rainy days, while November was the lowest monthly rainfall with 8.7 mm for 4 days rainy days. The annual average temperature was 28.5°C. The egestion is characterized by herbaceous vegetation (Banito et al., 2010). The site's soil, suitable for cassava cultivation (Ezui, 2017) and known as 'Terres de Barre,' is characterized as sandy-clay with 70% sand, 3.8% silt, 8.1% clay, acid pH (H<sub>2</sub>O 1:1) 5.5, 1.05% organic matter, 0.41% total nitrogen (N), 10 ppm available phosphorus (P), and cation exchange capacity (CEC) of 2.89 milliequivalents (meq)/100g of soil in the top 15 cm samples (Sogbedji et al. 2015).

### c) *Experimental Design, Field Layout and Maintenance*

The experiment was laid out in an augmented block design with one hundred forty four (144) cultivars as tested genotypes and five (5) checks varieties, distributed in twelve (12) blocks. Each block was delimited after ploughing and harrowing of the site. Distance of 1.5 m separated adjacent blocks and plots. The experimental unit was composed of four rows of 4 m with 16 plants of a genotype. A spacing of 1 m between plants and rows was adopted. The experiment was carried under rainfed conditions. Neither herbicide nor fertilizers were applied. The experiment was kept weed free by regular hand weeding. The trial was harvested twelve months after planting.

Table 1: List of Togo's Cassava Germplasm Cultivars Characterized

N°	Cultivar	Type	Origin	N°	Cultivar	Type	Origin
1	CRI Sika Bankye	Improved	Ghana	27	TMS 92_0326	Improved	Togo
2	CRI Ampong Bankye	Improved	Ghana	28	TMS 96_1708	Improved	Togo
3	TMS 95_0166	Improved	IITA	29	TMS 98_2132	Improved	Togo
4	TMS 96_0409	Improved	IITA	30	TMS 99_0554	Improved	Togo
5	Gbazekoute	Landrace	Togo	31	Agbede	Landrace	Togo
6	TMS 01_0006	Improved	IITA	32	Agou	Landrace	Togo
7	TMS 00_0354	Improved	IITA	33	Aguidagba	Landrace	Togo
8	TMS 00_0364	Improved	IITA	34	Akaleyo	Landrace	Togo
9	TMS 01_0034	Improved	IITA	35	Akebou	Landrace	Togo
10	TMS 01_0046	Improved	IITA	36	Akoss	Landrace	Togo
11	TMS 01_0093	Improved	IITA	37	Ankra atihe	Landrace	Togo
12	TMS 01_0098	Improved	IITA	38	Akpadjin Feto	Landrace	Togo
13	TMS 01_0131	Improved	IITA	39	Alagno	Landrace	Togo
14	TMS 01_0379	Improved	IITA	40	Ankra 3	Landrace	Togo
15	TMS 01_1085	Improved	IITA	41	Ankra Atiyibo	Landrace	Togo
16	TMS 01_1086	Improved	IITA	42	Assiatoe	Landrace	Togo
17	TMS 01_1097	Improved	IITA	43	Atidjin1	Landrace	Togo
18	TMS 01_1206	Improved	IITA	44	Atidjin 2	Landrace	Togo
19	TMS 01_1224	Improved	IITA	45	Atidjin Poli	Landrace	Togo
20	TMS 01_1368	Improved	IITA	46	Atidokpo	Landrace	Togo
21	TMS 01_1368(2)	Improved	IITA	47	Atihe1	Landrace	Togo
22	TMS 01_1371	Improved	IITA	48	Atiyibo 1	Landrace	Togo
23	TMS 01_1610	Improved	IITA	49	Atiyobo2	Landrace	Togo
24	TMS 01_1662	Improved	IITA	50	Awou	Landrace	Togo
25	TMS 01_1797	Improved	IITA	51	Awouye	Landrace	Togo
N°	Cultivar	Type	Origin	N°	Cultivar	Type	Origin
53	Badjogou	Landrace	Togo	79	Kanbom Bantchi	Landrace	Togo
54	Bazoka	Landrace	Togo	80	Kanigbeli 1	Landrace	Togo
55	Bob	Landrace	Togo	81	Kanigbeli 2	Landrace	Togo
56	Bob Assou	Landrace	Togo	82	Kataoli	Landrace	Togo
57	Bob Yegue	Landrace	Togo	83	Katawole	Landrace	Togo
58	BRS Caipira	Landrace	Brazil	84	Kidironi	Landrace	Togo
59	Degaule	Landrace	Togo	85	Kisseimou Koutowou	Landrace	Togo
60	Djakoagni	Landrace	Togo	86	Kola	Landrace	Togo
61	Djeble	Landrace	Togo	87	Kolaoung	Landrace	Togo
62	Djolaoba	Landrace	Togo	88	Kolmon kamkam	Landrace	Togo
63	Djoliba	Landrace	Togo	89	Kossikouma	Landrace	Togo
64	Donmoyibo	Landrace	Togo	90	Koutowou 2	Landrace	Togo
65	Fetonegbodji	Landrace	Togo	91	Kperoung Felgou	Landrace	Togo
66	Flawavi	Landrace	Togo	92	Kperoung Mamougue	Landrace	Togo
67	Gabonvi-ESA	Landrace	Togo	93	Kpla	Landrace	Togo
68	Gbadovi	Landrace	Togo	94	Loki	Landrace	Togo
69	Gbaze- ESA	Landrace	Togo	95	M'beou	Landrace	Togo
70	Vivigbaze	Landrace	Togo	96	MM96/5280	Improved	Togo
71	Ghana spana	Landrace	Togo	97	MM96/JW2	Improved	Togo
72	Gnidou	Landrace	Togo	98	Nigeria Fleur	Landrace	Togo

73	Hogninvo 1	Landrace	Togo	99	Nigeria Kikpaou	Landrace	Togo
74	Hogninvo 2	Landrace	Togo	100	Nigeria Kissaimon	Landrace	Togo
75	Inconnu	Landrace	Togo	101	N'tossou	Landrace	Togo
76	IRAT- Davie	Landrace	Togo	102	Ankra atihe	Landrace	Togo
77	Jhonson	Landrace	Togo	103	Okpoli	Landrace	Togo
78	Kalba	Landrace	Togo	104	Pela	Landrace	Togo
N°	Cultivar	Type	Origin	N°	Cultivar	Type	Origin
105	Peloumkoute	Landrace	Togo	131	D00_126	Improved	IITA
106	Penivi	Landrace	Togo	132	D00_54	Improved	IITA
107	Sabe	Landrace	Togo	133	D00_166	Improved	IITA
108	Sankara	Landrace	Togo	134	Toma 9	Landrace	Togo
109	Sassakawa	Landrace	Togo	135	CVTM4	Landrace	Togo
110	Sorad	Landrace	Togo	136	Toma 162	Landrace	Togo
111	Sawa	Landrace	Togo	137	Unknown 02	Landrace	Togo
112	Spana Assou	Landrace	Togo	138	TMS 96_1317	Improved	Togo
113	Spana Yegue	Landrace	Togo	139	TMS 96_0304	Improved	Togo
114	BRS Tapioqueira	Landrace	Brazil	140	TMS 96_0102	Improved	Togo
115	Tassiodo	Landrace	Togo	141	TMS 96_0869	Improved	Togo
116	Tchigouevi	Landrace	Togo	142	TMS 96_1642	Improved	Togo
117	Tetetidadjin	Landrace	Togo	143	TMS 96_0590	Improved	Togo
118	TME 419	Improved	Togo	144	TMS 96_539	Improved	Togo
119	TM1	Improved	Togo	145	TMS 96_1565	Improved	Togo
120	TME1	Improved	Togo	146	TMS 96_0603	Improved	Togo
121	TME 696	Improved	Togo	147	TMS 30572	Improved	IITA
122	Touwevi	Landrace	Togo	148	KPEM_10_03	Improved	Togo
123	Tuaka Atsu	Landrace	Togo	149	TMS 4(2) 1425	Improved	IITA
124	Tuaka komi Mami	Landrace	Togo	129	D00_208	Improved	IITA
125	Yabaka	Landrace	Togo	130	D00_14	Improved	IITA
126	D00_8300	Improved	IITA	52	Unknown	Landrace	Togo
127	M94_0583	Improved	IITA				
128	D00_137	Improved	IITA				

d) *Phenotypic Data Collection*

Thirty two (32) morphological traits were recorded using the cassava descriptor (Guevara et al., 2010) at three (3), six (6), nine (9) and twelve (12) months after planting (MAP). Data were recorded from the plants within the whole plot, and the most frequent occurrence variant was noted. At 12 MAP, the inner eight (8) plants within each plot were uprooted and observations on roots were taken. The traits assessment date, and method of assessment are summarized in Table 2.



*Table 2:* List of Morphological Traits recorded in Togo's Cassava Germplasm

N°	Trait	Code	Assessment date	<sup>1</sup> Assessment scale
1	Colour of apical leaves	ColApLea	3 MAP	3, 5, 7 or 9
2	Pubescence on apical leaves	PubApLea	3 MAP	0 or 1
3	Lobe margins	LoMar	6 MAP	3 or 5
4	Colour of leaf vein	ColLeaVe	6 MAP	3, 5, 7 or 9
5	Petiole Colour	PetCol	6 MAP	1, 2, 3, 5, 7 or 9
6	Leaf color	LeaCol	6 MAP	3, 5, 7 or 9
7	Number of leaf lobes	NLeaLo	6 MAP	3, 5, 7, 9 or 11
8	Shape of central leaflet	ShaCeLea	6 MAP	1-10
9	Orientation of petiole	OriPet	6 MAP	1, 3, 5 or 7
10	Flowering ability	FIHa	6 MAP	0 or 1
11	Pollen	Pol	6 MAP	0 or 1
12	Leaf retention	LeaRet	6 MAP	1-5
13	Stipule margin	StiMar	9 MAP	1 or 2
14	Length of stipule	LenSti	9 MAP	3 or 5
15	Color of stem cortex	ColStCor	9 MAP	1-3
16	Colour of stem epidermis	ColStEpi	9 MAP	1, 2, 3, or 4
17	Colour of stem exterior	ColStExt	9 MAP	3, 4, 5, 6, 7, 8 or 9
18	Colour of end branches of adult plant	CoEBran	9 MAP	3, 5, or 7
19	Growth habit of stem	GrHaSt	9 MAP	1 or 2
20	Distance between leaf scars	DisLeaSca	9 MAP	3, 5, or 7
21	Prominence of foliar scars	ProFoSca	9 MAP	3 or 5
22	Fruit	Fr	9 MAP	0 or 1
23	Levels of branching	LeBran	12 MAP	0, 1, 2 or 3
24	Branching habit	BranHab	12 MAP	1, 2, 3 or 4
25	Root constrictions	RoCons	12 MAP	1-3
26	Colour of root cortex	ColRoCor	12 MAP	1-4
27	Colour of root pulp	ColRoPu	12 MAP	1-5
28	External colour of storage root	ExColRo	12 MAP	1-4
29	Extent of root peduncle	ExRoPed	12 MAP	0, 3 or 5
30	Shape of plant	ShaPl	12 MAP	1-4
31	Root shape	RoSha	12 MAP	1-4
32	Texture of root epidermis	TexRoEpi	12 MAP	3, 5, or 7

<sup>1</sup> Each phenotypic trait had distinct phenotypes which were depicted by the values ranging from 0 to 9. Images associated with these scale values can be found in Fukuda et al. (2010). MAP = months after planting

#### e) Phenotypic Diversity Analyses

The morphological diversity of the core collection was assessed following two approaches. Traits distribution was determined using Microsoft Excel (2016) in the first approach. In the second approach, morphological data were subjected to Multiple Correspondence Analysis (MCA) for identification of relevant traits contributing mostly to the germplasm diversity (Giles et al. 2018). From the MCA results, traits that presented the highest variability were used as active variables to perform cluster analysis for morphotypes identification within the germplasm using the Ward's method (Kawuki et al. 2011). The optimal number of clusters was determined using the distribution of the variance function methods. The morphological diversity of the germplasm was visualized by plotting the factors scores for individual genotype in the first factorial plan in order to assess the relationship among cultivars (Selamawit Abebe et al. 2021). Analyses were run in SAS version 9.4.

### III. RESULTS

#### a) Descriptive Analysis of Morphological Traits

The variability observed for qualitative traits among cassava cultivars is given in figure 1. In all 37.58% cultivars showed purplish green colour, 22.82% had purple, 3.36% showed dark green and 36.24% had purplish green colour. About 45% of the cultivars had pubescence on apical leaves, while 55% had not (Figure 1). Approximately 25% of cultivars had yellowish green petioles, 22.82% purple petioles, 16.78% red petioles, 15.54% reddish-green petioles, 11.41% green petioles and 8.72% accessions showed greenish-red petioles (Figure 1). Nearly half of the cultivars (47.65%) had green leaf vein, 28.19% cultivars showed reddish-green leaf vein in less than half of the lobe, 13.42% had reddish-green leaf vein in more than half of the lobe and 10.74% had red leaf vein. Four morphotypes were observed in the germplasm based on the leaves colour. The first morphotype with dark green leaves was

represented by 51.68% of the cultivars, the second morphotype had light green leaves and was represented by 22.82% of the cultivars, the third morphotype exhibited purple green leaves and was represented by 14.76% of the cultivars and the fourth one had purple leaves was represented by 10.74% of the cultivars. The petioles of most cultivars in the collection were horizontal (50.34%), the irregular type was observed in 29.53% cultivars. 12.75% cultivars showed petioles inclined downwards while 7.38% cultivars had petioles inclined upward. About 40.27% accessions had lanceolate central leaflet, 32.88% had elliptic-lanceolate, 10.07% accessions had oblong-lanceolate central leaflet, 10.74% had obovate-lanceolate, 4.03% had ovoid, 0.67% had linear, 0.67% had pandurate and 0.67% had linear pandurate central leaflet (Figure 1). Most cultivars (66.44%) showed a winding lobe margin and 33.56% cultivars in the germplasm had a smooth lobe margin. In the germplasm, 61.07% cultivars had nine leaf lobes, 28.19% had seven leaf lobes, 8.05% had eleven and 2.69% five leaf lobes. For the leaves retention trait, 31.54% cultivars exhibited very poor leaf retention, 33.56% cultivars showed less than the average leaf retention, 19.46% exhibited average leaf retention while 15.44% cultivars exhibited better than average leaf retention (Figure 1).

With regards to stem related traits, 31.54% cultivars showed an upright growth habit of the stems, while the cultivars exhibiting a zigzag growth habit were observed in 68.46% cultivars. About 44% of the characterized cultivars had green end branches, 42.95% cultivars had green-purple and 12.75% had purple end branches. Four morphotypes were observed in the germplasm with regard to the stem epidermis colour (Figure 1). About 33.56% cultivars had light brown stem epidermis, 25.5% had dark brown, 24.83% had orange and 16.11% showed light-green stem epidermis. The colours of stem cortex observed were dark green (61.07% cultivars), orange (22.82% cultivars) and light green (16.11% cultivars). Approximately 30% cultivars had silver stem exterior, 17.45% had greenish-yellowish stem exterior, 14.17% showed gray stem, 12.08% orange stem, 10.74% dark brown stem, 9.39% light brown stem and 6.04% golden stem exterior. Majority of

the cultivars (63.09%) exhibited prominent foliar scars while 36.91% had semi- prominent foliar scars. The distance between leaf scars was short for 92.63% cultivars characterized, medium for 6.04% cultivars and long for 1.34% cultivars. About 69.80% of the cultivars had long stipules while 30.20% cultivars had short stipules. The stipule margin of 61.74% cultivars was split or forked and entire for 38.26% cultivars (Figure 1).

Differences in the flowering ability among cultivars were observed. About 50% of the cultivars produced flowers while remaining did not flower. At harvest, seeds were observed on 42.28% of cultivars. Cultivars exhibiting zero level of branching (47%) and three level of branching (34.23%) were predominant. About 60.40% cultivars showed trichotomous branching habit, 26.85% cultivars showed dichotomous type, 8 cultivars had tetrachotomous type while only 4 cultivars showed an erect type. Cassava cultivars examined phenotypically based on their plant shape exhibited variation with umbrella (39.60% cultivars), compact (33.56%), open (12.64%) and cylindrical (6.04%) shapes. (Figure 1)

At harvest, the external colour of storage root also exhibited variation with dark brown (40.82%), light brown (35.37%), yellow (12.93%), and white or cream (10.88%) colours. The root cortex colour showed high variability among cultivars and four morphotypes were observed. Majority (49.0%) had white or cream root cortex colour, followed by yellow (22.2%), pink (11.1%) and purple (8.9%) colours. Root epidermis was white or cream for 47.62% cultivars, yellow (18%), pink (20.41%) and purple (6.52%). Based on the colour of root pulp cultivars exhibited variation with cream (48.98%), yellow (30.61%), white (14.29%), and pink (6.12%) colours. In the germplasm, 69% cultivars had conical cylindrical root shape, 18.37% cylindrical, 9.52% conical and 3.4% had irregular root shape. Cultivars with few (50.34%) and some (30.61%) root constrictions were predominant. Majority of the cultivars (77.55%) had sessile roots, whereas 14.29% had pedunculate roots. The mixed type was recorded on 8.16% of the cultivars. The texture of root epidermis exhibited variation with rough (33.33%), smooth (34%), and intermediate (32.65%) textures. (Figure 1).

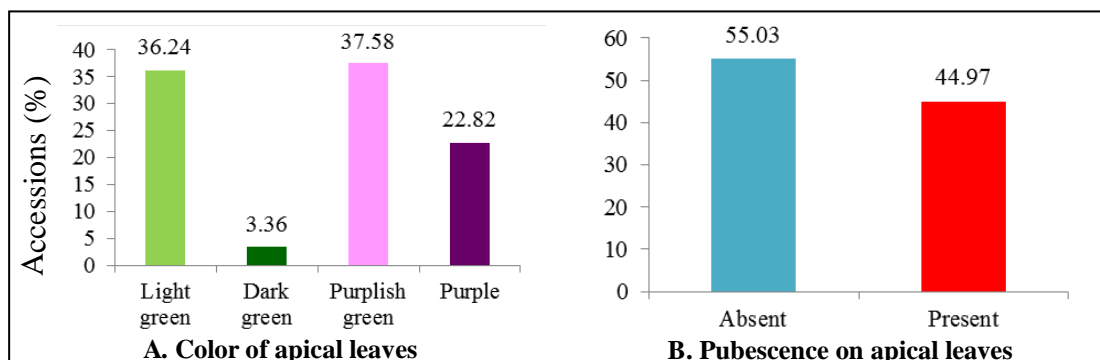


Figure 1: Frequency Distribution of 149 Cassava Cultivars based on Morphological Traits

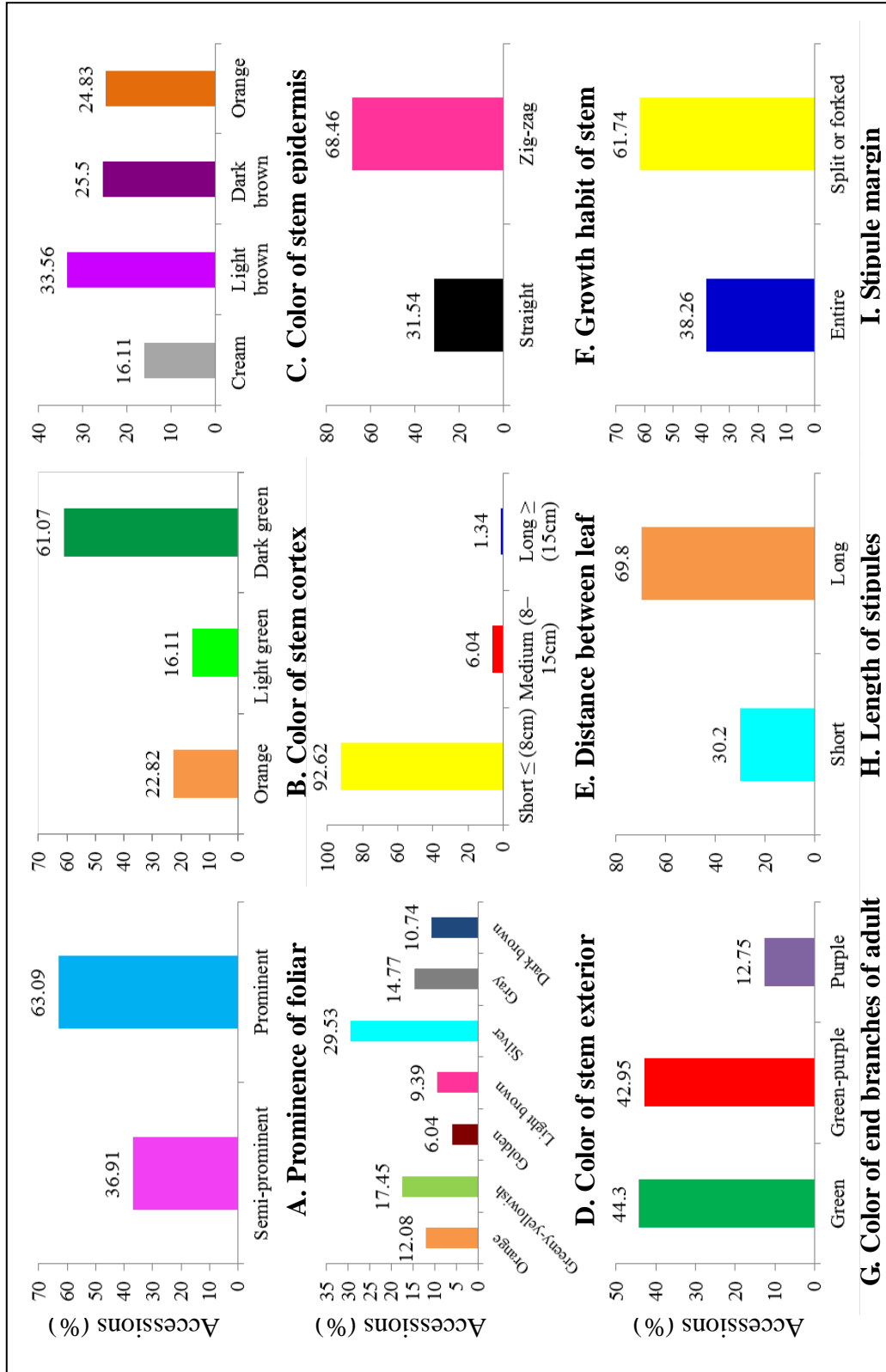


Figure 1. Count Frequency Distribution of 149 Cassava Cultivars based on Morphological Traits



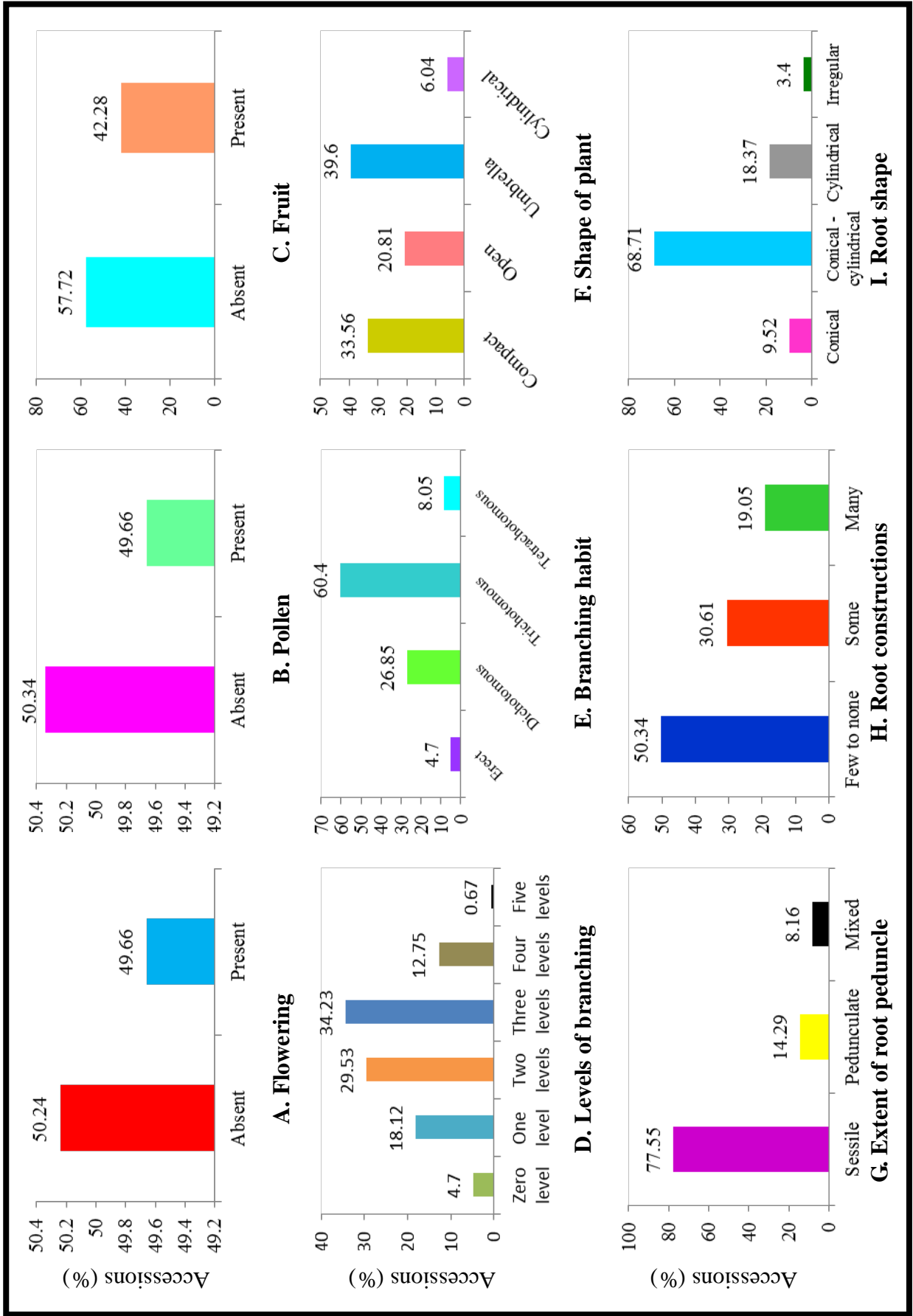


Figure 1. Frequency Distribution of 149 Cassava Cultivars based on Morphological Traits

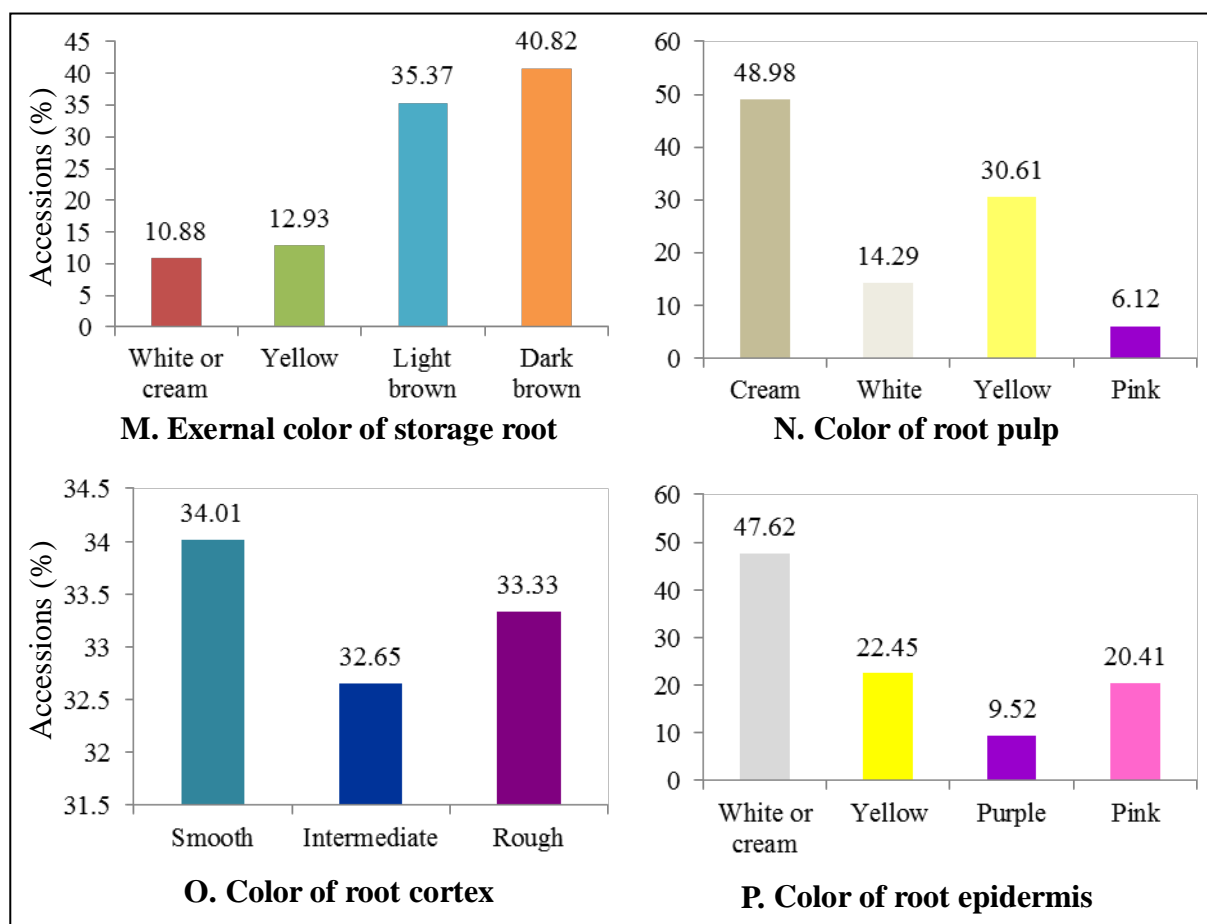


Figure 1: Frequency Distribution of 149 Cassava Cultivars based on Morphological Traits

b) Diversity Among Cultivars and Differentiation based on Morphological Traits

i. Significant traits describing the germplasm diversity

The objective of Multiple Correspondence Analysis (MCA) is to provide interpretable visualization of complex variable space. The meaning given to the axes and analysis of proximities between traits and conditions are usually made from the factorial planes. Thus, the first factorial plan and the factors having eigenvalue greater than one were retained. On the basis of this criterion, the first 11 factors with an eigenvalue greater than one were significant and therefore retained for the subsequent analyses (Table 3). These first eleven factors (Fs) explained 68.14% of the morphological variability among cultivars.

Factor 1 with an eigenvalue of 5.17 and accounted for 16.16% of the morphological variability. This factor was strongly correlated with petiole colour, leaf colour, colour of leaf vein, flowering and seed set ability and the levels of branching. Factor 2 with an eigenvalue of 3, explained 9.40% of the total variation, and was positively defined by leaves colour of end branches, color of stem epidermis, leaf lobe margin and

the growth habit of stem. Factor 3 represented by traits such the colour of stem exterior and color of stem cortex, had an eigenvalue of 2.5, and explained 7.81% of the divergence among cultivars. Factor 4 with an eigenvalue of 1.73 correlated with leaf retention and color of apical leaf. Factor 5 with an eigenvalue of 1.71 was related mainly to the distance between leaf scars. In Factor 6, shape of central leaflet and pubescence on apical leaves were the main traits, while the extent of root peduncle was most important trait in factor 7. In factor 8, the most important traits describing the germplasm variability were the stipule length and the prominence of foliar scars. Factor 9 with an eigenvalue of 1.26, contribute 3.95% of the total variability and was mainly related to the stipule margin. Factor 10 was mainly represented by the orientation of petioles, plante shape and the color of root cortex while factor 11 was represented by the colour of root pulp (Table 3).

Table 3: Eigen values, proportion of variation and contribution associated with the axes of the MCA of 32 qualitative traits

	F1	F2	F3	F4	F5	F6	F7	F8	F9	F10	F11
Eigenvalue	5.172	3.000	2.502	1.734	1.718	1.508	1.394	1.362	1.265	1.148	1.003
Variability (%)	16.161	9.375	7.817	5.419	5.369	4.714	4.356	4.256	3.953	3.586	3.134
Cumulative %	16.161	25.536	33.353	38.773	44.142	48.855	53.211	57.467	61.420	65.007	68.141
ColApLea <sup>1</sup>	0.053	0.122	0.001	<b><u>0.287</u></b>	0.006	0.010	0.016	0.040	0.037	0.051	0.068
PubApLea <sup>2</sup>	0.001	0.019	0.091	0.040	0.001	<b><u>0.180</u></b>	0.047	0.146	0.016	0.003	0.009
LeaRet <sup>3</sup>	0.024	0.122	0.000	<b><u>0.298</u></b>	0.028	0.019	0.117	0.018	0.029	0.012	0.027
ShaCeLea <sup>4</sup>	0.001	0.000	0.049	0.047	0.020	<b><u>0.230</u></b>	0.034	0.053	0.052	0.025	0.021
PetCol <sup>5</sup>	<b><u>0.494</u></b>	0.235	0.028	0.002	0.005	0.003	0.039	0.001	0.001	0.012	0.002
LeaCol <sup>6</sup>	<b><u>0.426</u></b>	0.282	0.003	0.000	0.001	0.043	0.013	0.010	0.015	0.031	0.000
NuLeaLob <sup>7</sup>	<b><u>0.352</u></b>	0.005	0.036	0.001	0.018	0.159	0.038	0.012	0.058	0.000	0.006
LobMar <sup>8</sup>	0.003	<b><u>0.187</u></b>	0.000	0.016	0.142	0.023	0.007	0.021	0.149	0.003	0.001
ColLeaVei <sup>9</sup>	<b><u>0.409</u></b>	0.352	0.002	0.000	0.015	0.027	0.012	0.008	0.017	0.016	0.008
OrPet <sup>10</sup>	0.014	0.142	0.061	0.023	0.001	0.027	0.146	0.008	0.000	<b><u>0.215</u></b>	0.013
Flow <sup>11</sup>	<b><u>0.714</u></b>	0.095	0.013	0.042	0.000	0.004	0.003	0.013	0.000	0.007	0.017
Pol <sup>12</sup>	<b><u>0.714</u></b>	0.095	0.013	0.042	0.000	0.004	0.003	0.013	0.000	0.007	0.017
Fru <sup>13</sup>	<b><u>0.635</u></b>	0.082	0.012	0.065	0.002	0.001	0.002	0.022	0.000	0.001	0.007
ProFoSca <sup>14</sup>	0.104	0.001	0.001	0.000	0.072	0.148	0.000	<b><u>0.224</u></b>	0.036	0.065	0.003
ColSteCor <sup>15</sup>	0.113	0.084	<b><u>0.232</u></b>	0.028	0.101	0.107	0.047	0.047	0.034	0.001	0.000
ColSteEpi <sup>16</sup>	0.014	<b><u>0.187</u></b>	0.018	0.021	0.086	0.173	0.001	0.005	0.009	0.057	0.013
ColSteExt <sup>17</sup>	0.016	0.079	<b><u>0.301</u></b>	0.039	0.154	0.038	0.017	0.036	0.012	0.010	0.001

<sup>1</sup>Colour of apical leaves, <sup>2</sup>Pubescence on apical leaves, <sup>3</sup>Leaf retention, <sup>4</sup>Shape of central leaflet, <sup>5</sup>Petiole Colour, <sup>6</sup>Leaf color, <sup>7</sup>Number of leaf lobes, <sup>8</sup>Lobe margin, <sup>9</sup>Colour of leaf vein, <sup>10</sup>Orientation of petiole, <sup>11</sup>Flowering ability, <sup>12</sup>Pollen, <sup>13</sup>Fruit, <sup>14</sup>Prominence of foliar scars, <sup>15</sup>Color of stem cortex, <sup>16</sup> Colour of stem epidermis, <sup>17</sup>Colour of stem exterior. Traits that contributed most to the morphological variation of a particular factor are in bold and underlined

Table 3: Continued: Eigen values, proportion of variation and contribution associated with the axes of the MCA of 32 qualitative traits

	F1	F2	F3	F4	F5	F6	F7	F8	F9	F10	F11
Eigenvalue	5.172	3	2.502	1.734	1.718	1.508	1.394	1.362	1.265	1.148	1.003
Variability (%)	16.161	9.375	7.817	5.419	5.369	4.714	4.356	4.256	3.953	3.586	3.134
Cumulative %	16.161	25.536	33.353	38.773	44.142	48.855	53.211	57.467	61.420	65.007	68.141
DisLeaSca <sup>18</sup>	0.105	0.001	0.010	0.061	<b><u>0.189</u></b>	0.009	0.023	0.052	0.085	0.052	0.042
GroHaSte <sup>19</sup>	0.036	<b><u>0.179</u></b>	0.025	0.002	0.028	0.011	0.105	0.070	0.031	0.000	0.031
ColBrAdPI <sup>20</sup>	0.275	<b><u>0.319</u></b>	0.006	0.074	0.045	0.011	0.048	0.007	0.002	0.001	0.012
LenSti <sup>21</sup>	0.058	0.020	0.061	0.096	0.005	0.002	0.019	<b><u>0.243</u></b>	0.011	0.002	0.081
StiMar <sup>22</sup>	0.007	0.000	0.021	0.031	0.018	0.005	0.110	0.001	<b><u>0.489</u></b>	0.055	0.016
LevBran <sup>23</sup>	<b><u>0.211</u></b>	0.193	0.022	0.073	0.143	0.000	0.029	0.002	0.039	0.000	0.002
BraHab <sup>24</sup>	0.048	0.015	<b><u>0.179</u></b>	0.052	0.054	0.139	0.008	0.007	0.001	0.017	0.076
ShaPI <sup>25</sup>	0.085	0.087	0.044	0.161	0.016	0.011	0.004	0.018	0.000	<b><u>0.198</u></b>	0.044
ExRoPed <sup>26</sup>	0.004	0.002	0.121	0.000	0.093	0.044	<b><u>0.233</u></b>	0.046	0.000	0.003	0.028
RoConst <sup>27</sup>	0.003	0.035	0.082	<b><u>0.179</u></b>	0.016	0.009	0.048	0.054	0.000	0.025	0.103
RoSha <sup>28</sup>	0.003	0.008	<b><u>0.244</u></b>	0.011	0.000	0.025	0.013	0.008	0.054	0.026	0.000
ExtColRo <sup>29</sup>	0.001	0.022	<b><u>0.428</u></b>	0.000	0.135	0.003	0.081	0.032	0.009	0.049	0.001

ColRoPul <sup>30</sup>	0.120	0.001	0.051	0.036	0.006	0.038	0.008	0.017	0.038	0.051	<b>0.239</b>
ColRoCor <sup>31</sup>	0.117	0.020	0.041	0.005	0.148	0.004	0.111	0.009	0.036	<b>0.151</b>	0.092
TexRoEpi <sup>32</sup>	0.014	0.009	<b>0.308</b>	0.001	0.168	0.000	0.009	0.117	0.005	0.003	0.021

<sup>18</sup>Distance between leaf scars, <sup>19</sup>Growth habit of stem, <sup>20</sup>Colour of end branches of adult plant, <sup>21</sup>Length of stipule, <sup>22</sup>Stipule margin, <sup>23</sup>Levels of branching, <sup>24</sup>Branching habit, <sup>25</sup>Shape of plant, <sup>26</sup>Extent of root peduncle, <sup>27</sup>Root constrictions, <sup>28</sup>Root shape, <sup>29</sup>External colour of storage root, <sup>30</sup>Colour of root pulp, <sup>31</sup>Colour of root cortex, <sup>32</sup>Texture of root epidermis. Traits that contributed most to the morphological variation of a particular component are in bold and underlined.

c) Structure of the Germplasm Diversity

Factor 1 is positively correlated to the flowering and seed set ability and negatively correlated to the leaf colour, while factor 2 is positively correlated to the end branches colour, stem epidermis colour, leaf lobe margin, and the growth habit of stem (Figure 2). With regard to figure 2, four morphotypes were distinguished.

Morphotypes 1 and 2 exhibited good flowering and seed set ability but differed in terms of the end branches colour, stem epidermis colour, and growth habit of stem. The varieties belonging to morphotype 3 and 4 did not flower and differed in terms of the colour of the end branches, stem epidermis and the leaf lobe margin.

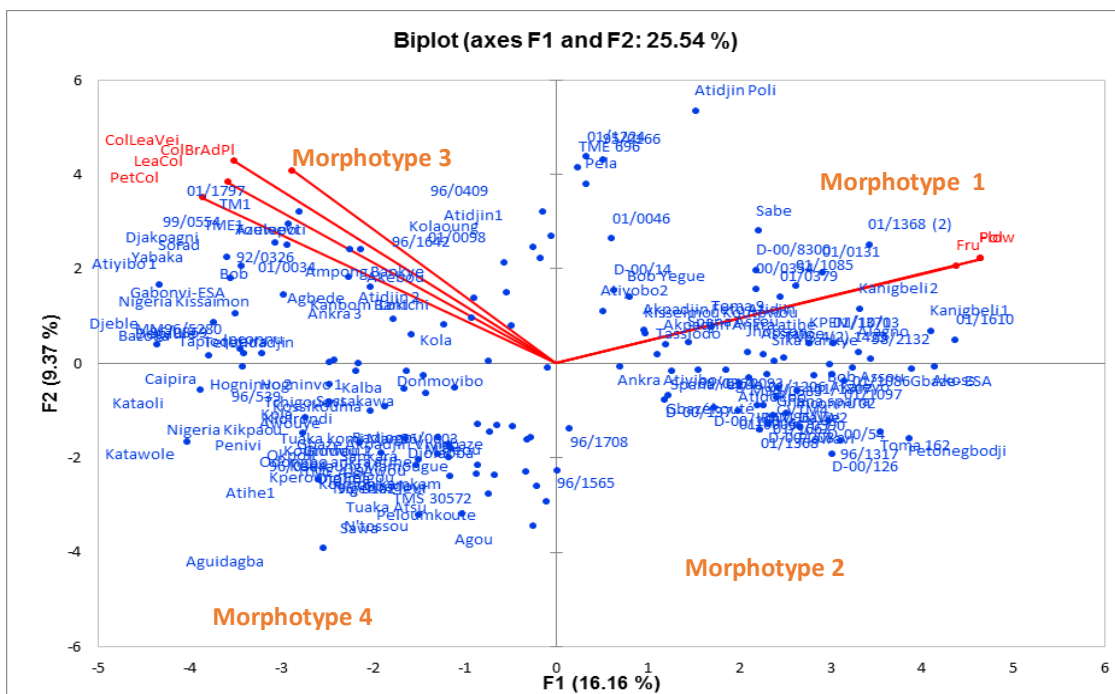


Figure 2: Component patterns of the MCA based upon factor 1 and 2 using significant phenotypic traits observed on 149 cassava cultivars of Togo

From the cluster analysis, the variance distribution function revealed that the optimal number of clusters was seven, with within class variance of 53.7 (Figure 3). Thus, the varieties were clustered into seven morphotypes (Figure 4). Morphotype 1 was composed of 137 varieties among which 53 were improved varieties, while remaining were landraces. Morphotype 1 is made of unflowering and unbranching cultivars with greenish-red petioles (Figure 4 and 5). Morphotype 2 was composed of 3 varieties (TMS 01\_0379, Akoss, Kolaoung) exhibiting ovoid central leaflet, purple leaves, reddish green leaf vein, light green stem cortex, greeny-yellowish stem exterior, three levels of branching, dichotomous branching habit, conical root, cream root pulp, good flowering and seed set ability. Morphotype 3

composed of cultivar Akaleyo was considered as outlier. The fourth morphotype comprised of cultivars Akebou, Akpadjin and Tassiado is characterized by red petioles, obovate-lanceolate central leaflet, compact plants, pink root cortex, white root pulp and bad seed set ability; whereas cultivars belonging to morphotype 5 (Alagno, Pela) exhibited good leaf retention, irregularly shaped roots, many root constrictions, good flowering and seed set abilities (Figure 4 and 5). In morphotype 6, there were 8 cultivars (D00\_126, Inconnu 2, D00\_137, D00\_208, D00\_14, D00\_54, D00\_166 and D00\_8300) with sessile peduncle root, conical cylindrical root, orange root pulp, short distance between leaf scars, good flowering and seed set ability. The morphotype 7

composed of cultivar TMS 96\_0590 exhibited dark green apical leaves, cream stem epidermis, two levels of branching, good flowering and seed set ability (Figure 4 and 5).

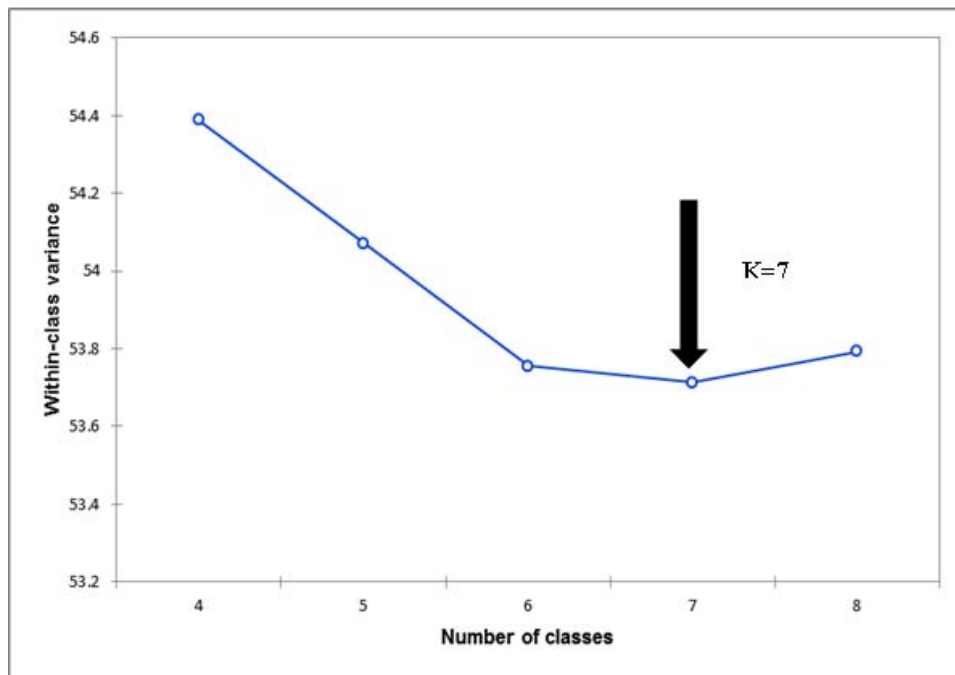


Figure 3: Distribution of the variance function according to the number of clusters obtained from cluster analysis based on significant morphological traits





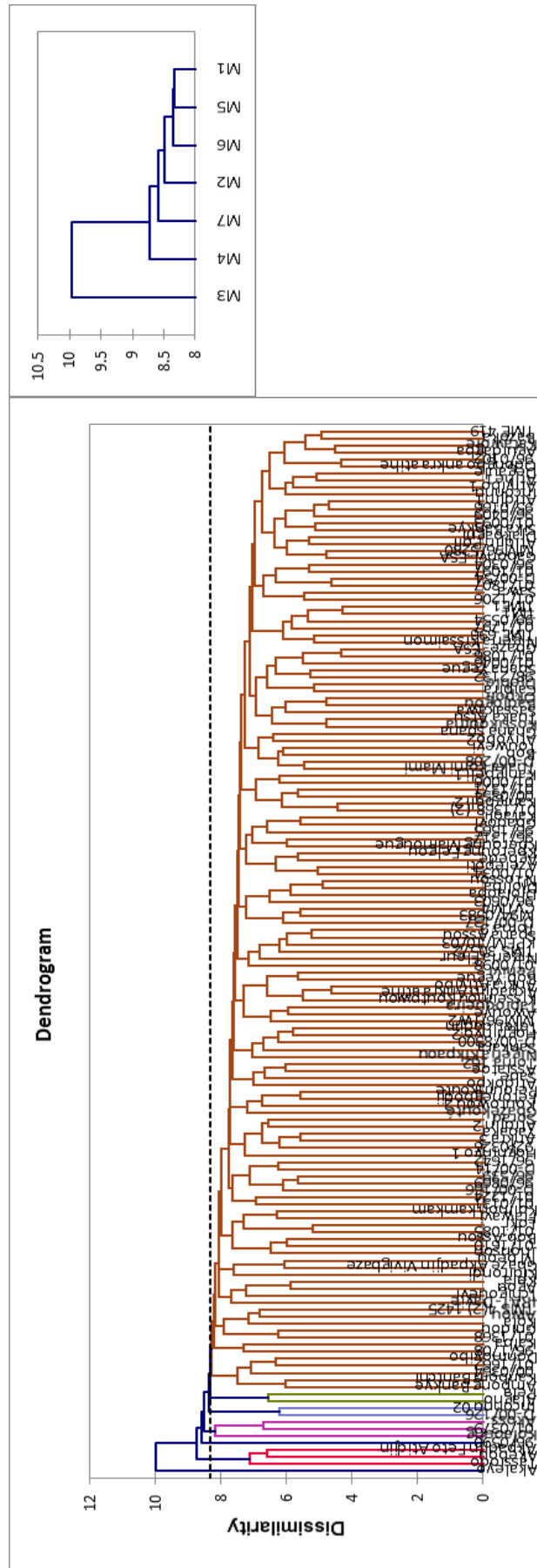


Figure 4: Dendrogram of 149 cassava genotypes revealed by the Wards method based on significant morphological traits

From left to right of the dendrogram the Morphotype 3, Morphotype 4, Morphotype 7, Morphotype 2, Morphotype 6, and Morphotype 5 are respectively represented.

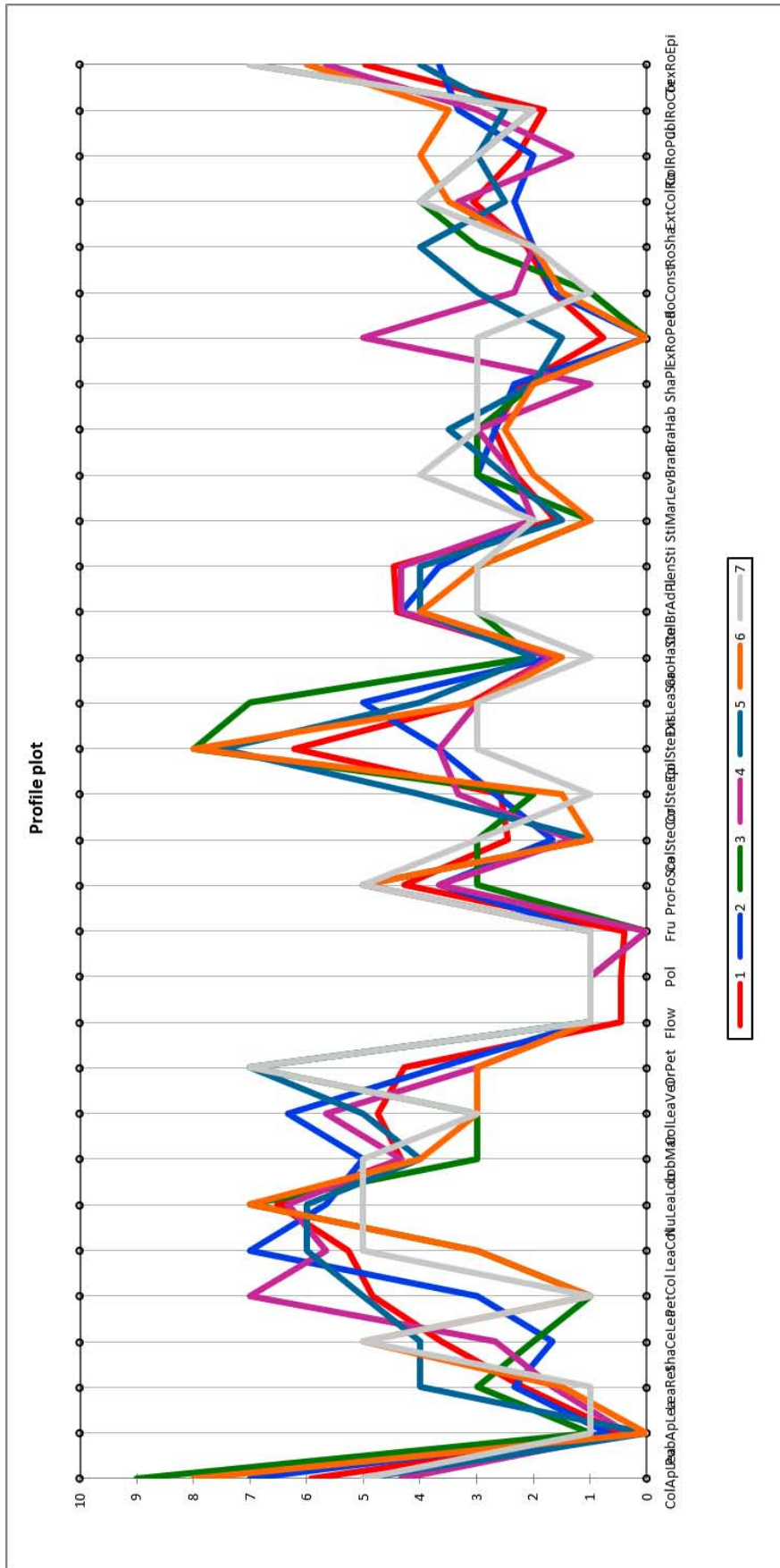


Figure 5: Profile plot describing the seven morphotypes obtained from the cluster analysis based on 32 morphological traits evaluated in 149 cassava cultivars of Togo

## IV. DISCUSSION

### a) *Morphological Diversity of the Germplasm*

Phenotyping of plant materials based on morphological traits has been used to determine the phenotypic variability among cultivars (Avijala et al., 2015; Agre et al., 2015; Adjebeng-Danquah & Gracen, 2020). The use of these traits allows rapid identification of cultivars. In addition, morphological traits are found to be stable, highly heritable and independent from the environment (Fukuda et al., 2010). However, molecular characterization may allow a more accurate detection of differences between germplasm bank cultivars than morphological characterization.

In our study, substantial variation was observed within the germplasm. Traits such as petiole colour, leaf colour, colour of leaf vein, flowering ability, seed set ability, levels of branching, colour of end branches, colour of stem epidermis, leaf lobe margins, growth habit of stem, and root flesh colour were underscored as the most relevant traits for cultivars discrimination. Genetic variability for morphological traits has been reported in different studies in Ghana (Asare et al., 2011; Adjebeng-Danquah & Gracen, 2017); in Benin (Agre et al., 2017), in Burkina Faso (Gmakouba et al., 2018) and in Brazil (Oliveira et al., 2015).

Root flesh colour is a trait with great importance for cassava because of dietary habits of Togolese. Moreover, this trait is directly related to the presence of vitamin. Orange varieties are riched carotenoids (provitamin A) (Kamanda et al., 2020). Low occurrence was found for yellow root colour and pink root colour varieties, which possibly have lycopene in their roots.

### b) *Structure of the Germplasm Diversity*

Cluster analysis classified the varieties into seven morphotypes, showing random distribution of the varieties. The fact that cassava is an outcrossing crop which can propagate vegetatively could explain this result. This facilitates the dispersion of varieties the exchange among farmers and, consequently, the exchange of genes (Agre et al., 2017). The main factor involved in the high diversity found may be gene flow promoted by farmers, who have acted as a dispersing agents for the species. An intense exchange system of varieties has been documented among farmers growing cassava.

The germplasm bank cultivars were not grouped based on the geographical origin distribution. Cultivars collected from place such as Vogan, Wetlope, Akebou, Danyi, Aouda, Davié, and Assoukoko were clustered in morphotype 1. Likewise cultivars from Bafilo, Assoukoko, Danyi, and Bourondè were also clustered together in morphotype 2. The remaining clusters also included cultivars from different collection regions. The informal farmers to farmers seed supply system practiced in the country could explained this

result. This agrees with earlier studies on cassava (Ojulung et al., 2010; Sing et al., 2015; Adjebeng-Danquah & Gracen, 2017, Gmakouba et al., 2018). In addition, the cassava cultivars collected from the same region were clustered into different morphotypes which suggest a high genetic diversity within each collection area. Similar findings were also reported by Agre et al. (2017) in Bénin. Moreover, there was no clear differentiation and real structuring between local and improved varieties in this study as also reported by Kombo et al. (2012).

Morphotypes identified may be valuable in cassava germplasm management and cultivars identification. Especially, the varieties belonging to morphotypes 5, 6 and morphotype 7 might be most desirable for breeding due to their good flowering and seed set ability and adaptability to environmental conditions. These cultivars could be used to set up crossing blocks in order to develop segregating breeding lines with farmers desired traits.

## V. CONCLUSIONS

The study revealed that the cassava germplasm of Togo exhibited high phenotypic diversity. Morphological traits such as petiole colour, leaf colour, colour of leaf vein, flowering ability, seed set ability, levels of branching, colour of end branches, colour of stem epidermis, leaf lobe margins and the growth habit of stem were the most diverse and could be used for cultivar identification in the field. The varieties of morphotypes 5, 6 and 7 harboured interesting features such as flowering and seed set ability and may be usefull for the national breeding programme. For breeding purpose, superior parental clones could be selected from these morphotypes for crossing and generating a breeding population.

## ACKNOWLEDGEMENTS

The authors are thankful to farmers and the agricultural extension agents who participated in the germplasm collection, as well as the Togolese Agronomic Research Institute (ITRA-CRAL) for offering the logistical assistance.

*Funding:* This manuscript is extracted from PhD. thesis, which was co-funded by ECONET-Foundation and the Germany Academic Exchange Service (WACCI/DAAD) at West Africa Centre for Crop Improment (University of Ghana).

*Authors' Contributions:* GMAKOUBA Tighankoumi carried out the study, analyzed the data and drafted the manuscript. DZIDZIENYO K. Daniel, SOME Koussao, TONGOONA Pangirayi and ASANTE I. Kwame participated in the study design and were major contributors in writing and correcting the manuscript. All authors read and approved the final manuscript.

*Consent for Publication:* The authors declare that they obtained an informed consent for publication from people involved in this study.

*Availability of Data and Materials:* Data are within the paper and its supporting information files. The datasets are fully available without restriction on reasonable request from the corresponding author.

*Competing Interests:* The authors declare that they have no competing interests.

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