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Original Article

Genetic Diversity of Sorghum [*Sorghum bicolor* (L.) Moench] Genotypes in the Central Plateau Zone of Rwanda for Qualitative Traits

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Abstract - *Sorghum is a key cereal in Rwanda, ranking second after maize. However, climate change, pests, and diseases contribute to low yields. To improve its yield, a study was conducted in the central plateau zone of Rwanda to evaluate the genetic diversity of 36 sorghum genotypes based on qualitative traits for the identification of drought-tolerant genotypes to use in future breeding programs. The field experiment was conducted in a simple lattice design at Rubona Research Station, and 10 traits were studied. The descriptive results identified the umuceri genotype with very slight senescence, indicating its possible tolerance to drought. Among collection origins, the Ruhango district recorded the highest diversity index of 0.81 (tillering capacity). For all genotypes, Shannon-Weaver results showed nine polymorphic traits, with tillering capacity (0.96) having a high diversity index. The genotypes showed a diversity index ranging between 0.51 (midrib color) and 0.91 (inflorescence shape) within the collection origins and between 0.09 (inflorescence shape) and 0.49 (midrib color) among the collection origins. Principal component analysis revealed four major components explaining 78.64% of the total variation. Clustering analysis grouped the 36 sorghum genotypes into five main clusters, showing the genotype Umuceri being the most distant and potentially valuable for future sorghum improvement programs.*

***Keywords* -** *Cereals in Rwanda, Diversity index, Drought tolerant, Shannon-Weaver, Sorghum genotypes.*

1. Introduction

Sorghum is a mainly self-pollinated diploid species with $2n = 2x = 20$ chromosomes (Poehlman and Sleper, 1976). It is grown under several biotic and abiotic production constraints, including drought-prone areas where other crops cannot be grown consistently. It is the staple food for the most food-insecure people in the world, mainly in Sub-Saharan Africa and South Asia (Smith and Frederiksen, 2000; Bibi et al., 2010). Besides being an important food, feed, and forage crop, it provides the raw material for the production of starch, fiber, dextrose syrup, biofuels, alcohol, and other products. It also has a number of applications, including those in pharmaceutical diagnosis, medicine, the synthesis of organic chemicals, and utility goods (Mamo et al., 2023). In Africa, sorghum is a traditional food that provides a comparable nutrient value to maize. However, it has high antioxidant and anti-inflammatory properties, and when consumed as a whole grain, it helps to meet requirements for dietary fiber. Sorghum is also more adaptive to climate change, showing tolerance to drought conditions, and has the potential to build resilience in the African food system (Pereira and Hawkes, 2022).

Sorghum can adapt to a wide range of climatic gradients due to its large degree of variation in flowering time and plant and inflorescence morphology (Morris et al., 2013; Lasky et al., 2015; Zhang et al., 2015). According to Harlan and de Wet (1971) and Li et al. (2010), as the continent of domestication and a centre of diversification for cultivated sorghum, it is therefore expected wide diversity within and among the sorghum cultivars at both phenotypic and genotypic levels (Kong and Hart, 2000; Hart et al., 2001). It was reported that the presence of a broad diversity of landraces is mainly grown by farmers and used for sorghum improvement in Africa because the landraces are adapted to harsh conditions and possess stress tolerance genes (Atokple, 2003). Thus, understanding the genetic diversity in sorghum will facilitate further improvement of this crop for its genetic architecture (Jayaramachandran et al., 2011) for effective germplasm management, utilization, and genotype selection for crop improvement (Bucheyeki et al., 2010). The level of species adaptability and survival in unpredictable environmental conditions is commonly measured with the level of genetic diversity within a species (Rao and Hodgkin, 2002; Govindaraj et al., 2015), whereas the level of genetic variation within a population is the basis for germplasm selection in plant breeding and is vital for crop improvement (Mohammadi and Prasanna, 2003). The more diverse genotypes or accessions can be crossed to produce superior hybrids with resistance to abiotic and biotic stresses. Thus, understanding, utilization and conservation of plant genetic variation are crucial to human food security (Rao and Hodgkin, 2002).

In Rwanda, sorghum is used in making local alcoholic beverages, *ikigage,* and non-fermented beverages, *ubushera,* that are frequently utilized in many festival ceremonies such as weddings, baptisms, dowries, and birth ceremonies (Lyumugabe et al., 2014). In many special events, these drinks made from sorghum are used as a symbol of harmony and familial cohesion. Additionally, *impengeri,* which is sorghum boiled in water, used to be a typical staple food during times of severe famines (Ezeanya-Esiobu et al., 2018). Sorghum is grown in all agroecological regions and all districts of the country (NISR, 2023); it is among the key crops that are intercropped with other crops (cassava, legumes, and potatoes) in the Rwandan principal farming system (Cyamweshi et al., 2017). Despite the importance and multipurposeness of the crop in the country, the national average yield is low in comparison to the world average yield of 1,500 kg. ha^{-1} and average yield of Ethiopia (2,697 kg. ha^{-1}) (FAO, 2021). The low yield is caused by stalk borers, leaf bright, cercosporiosis, shoot fly, *Striga hermonthica* (Wortmann et al., 2006; Niyibigira, 2012), and drought-induced by the current global climate change that increases the annual temperatures and intense annual rainfall occurring in very short periods of time (Huggings, 2017). The problem is more aggravated by soil overutilization caused by the high population density in Rwandan (NISR, 2023). Climate change-induced drought affects specifically smallholder farmers who are mainly growing sorghum and practicing subsistence rainfed agriculture. Attention has to be paid to this problem as Rwanda's economy mostly relies on agriculture, contributing almost a third of the national GDP and employing around 68% of all households (Kemen et al., 2020; NISR, 2023). The utilization of genetic diversity existing in the genetic resources is one of the measures to improve the productivity of the crop (Kebbede, 2020; Begna, 2021; Mola and Ejeta, 2021; Yirgu et al., 2022).

Genetic diversity describes the variation in the amount of genetic information within and among a community, a species, or individuals of a population (UN, 1992; Mangshin et al., 2021). It is the foundation for maintenance and the continuity of any plant population (Mangshin et al., 2021). Genetic diversity in plants can be assessed using agro-morphological, genotypic (molecular), and biochemical markers (Herniwati et al., 2024). Agro-morphological assessment has the privilege of being simple and relatively affordable (Andiku et al., 2022; Herniwati et al., 2024). Appropriate genetic diversity analysis of genotypes would help in the development of new varieties having preferred traits (Gebre et al., 2024). Hence, having information on the genetic diversity among existing genotypes is fundamental for designing effective crop improvement programs intended to select parents with traits of interest (Chaithrashree et al., 2024) and for sustainable conservation of genetic resources (Welderufael et al., 2023).

In Rwanda, the farmers used sorghum landraces for production. Thus, the study of genetic diversity is vital to identify accessions with distinctive features that can be exploited to develop varieties and boost their resistance to environmental challenges. Studies on genetic diversity can also aid in efficient and effective genetic resource conservation and use it when necessary (Afolayan et al., 2019). The introduction of new alleles into locally adapted landraces identified through genetic diversity studies contributes to the improvement of sorghum productivity and environmental stress tolerance, such as drought (Birhan et al., 2022). Morphological characterization with published descriptors is used for genetic diversity studies, and it is also advised to use it as the starting point. This is due to its many benefits, including being easily available for many crops, its ability to be performed in situ, its affordability, and its relative ease of carrying (Watson and Eyzaguirre, 2001; Akatwijuka et al., 2016). In Africa, many studies were conducted using qualitative traits to explore the genetic diversity of sorghum (Gerrano et al., 2015; Akatwijuka et al., 2016). Some of these studies reported the importance of using qualitative traits to identify genotypes with drought tolerance traits (Naoura et al., 2019) and sorghum grain quality preferred by farmers (Akatwijuka et al., 2016). Despite the drought problem and its negative effects on sorghum productivity in Rwanda, there has been no study aimed at discovering drought tolerance sorghum genotypes using morphological characterization with qualitative traits, as was performed in other countries. Thus, this study was carried out to assess the genetic diversity of Rwandan sorghum genotypes and identify potential candidates to use in future breeding programs in the central plateau zone of Rwanda. The results of this study will provide drought-tolerant genotypes that will be used by researchers in future sorghum breeding programs, and farmers will have genotypes that can yield higher in drought environments.

2. Materials and Methods

2.1. Description of Study Area

The research was conducted at the Rwanda Agriculture and Animal Resources Development Board, Rubona station, in the central plateau zone of Rwanda. Rubona is located at 2°48 S latitude and 29°76’ E longitude and is at 1706 m.a.s.l. (Mukamuhirwa et al., 2018), with a normal temperature of 18.7°C and an average annual rainfall of 1200 mm (Shumbusha et al., 2017). The study took place during the 2019 sorghum cropping season, from January to July.

2.2. Experimental Materials and Design

Thirty-six sorghum landraces collected from the Rwanda National Genebank and six varieties obtained from the sorghum research program were evaluated in the study (Table 1). The genotypes obtained from the genebank were selected based on their origin of collection (from five districts of the central plateau zone of Rwanda) and the availability of sufficient seeds. In contrast, the ones acquired from the research program were selected based on their agroecological region suitability (Table 1). In addition to being adaptable to the central plateau zone, the controls were released varieties (Kinyaruka, IS21219, IS8193, and Kat 369) and advanced lines in the final evaluation stages before release (Mabereyingoma and SDL880-160).

The field experiment was set up in a simple lattice design with a plot size of 3 m x 2 m (6 m^2), having four rows with 10 plants in each row. The spacing between rows and between plants within rows was 0.75 m and 0.20 m, respectively.

Table 1. List of sorghum genotypes assessed at Rubona, central plateau zone of Rwanda, in 2019

Number	Accession Number	Landrace/Variety Name	Collection Origin
1	RNGB-00041SGH	Kigosorabaswa	Nyanza District
2	RNGB-00043SGH	Nyirakaganza	Ruhango District
3	RNGB-00047SGH	Rudasakwa	Kamonyi District
4	RNGB-00048SGH	Bukobwa 1	Gisagara District
5	RNGB-00052SGH	Amakoma	Kamonyi District
6	RNGB-00053SGH	Ikinyaruka	Nyanza District
7	RNGB-00055SGH	Nyiragikori y'umweru	Huye District
8	RNGB-00062SGH	Igihove	Ruhango District
9	RNGB-00063SGH	Indinganire	Nyanza District
10	RNGB-00064SGH	Gihove	Kamonyi District
11	RNGB-00068SGH	Nyirakinuma	Huye District
12	RNGB-00072SGH	Bukobwa 2	Huye District
13	RNGB-00073SGH	Ndamirabana	Gisagara District
14	RNGB-00078SGH	Mugabo	Huye District
15	RNGB-00079SGH	Kinanira	Nyanza District
16	RNGB-00080SGH	Gatemwa	Huye District
17	RNGB-00082SGH	Nyiramugufi	Gisagara District
18	RNGB-00086SGH	Nyirakanyamunyo	Nyanza District
19	RNGB-00087SGH	Kebo	Ruhango District
20	RNGB-00089SGH	Amera	Kamonyi District
21	RNGB-00091SGH	Bukobwa 3	Gisagara District
22	RNGB-00095SGH	Cyamwiha	Kamonyi District
23	RNGB-00101SGH	Ntuncurimboga	Nyanza District
24	RNGB-00102SGH	Munebwe	Huye District
25	RNGB-00107SGH	Nyakami	Ruhango District
26	RNGB-00108SGH	Nyiragitenderi	Gisagara District
27	RNGB-00119SGH	Nyiragikori	Gisagara District
28	RNGB-00125SGH	Nyiragahengeri	Ruhango District
29	RNGB-00137SGH	Unkown or amasaka	Ruhango District
30	RNGB-00887SGH	Umuceri	Nyanza District
31	Non-applicable (NA)	SDL880-160	Research Program
32	NA	Kinyaruka	Research Program
33	NA	IS8193	Research Program
34	NA	IS21219	Research Program
35	NA	Kat369	Research Program
36	NA	Mabereyingoma	Research Program

The field was prepared twice using hoes, and the experimental plots were delimitated using a long measuring tape, plastic rope, and wooden pegs. After plots were delimited, two seeds were placed into a hole 25 mm deep, and after germination, one weak seedling was thinned out. Twenty tons of farm manure was applied at planting time and supplemented with mineral fertilizers N17P17K17 at a rate of 250 kg. ha^{-1} (two weeks after germination) and urea (46% N) at a rate of 100 kg. ha^{-1} (six weeks after germination). All recommended sorghum management practices were carried out, and the outbreak of fall armyworms was managed using Profenofos 40% + Cypermetrin 4% EC at a concentration of 2 ml/L of water.

2.3. Data Collection

Data on ten qualitative traits were recorded using visual observation on a plot basis, and the characterization of genotypes was performed based on the sorghum descriptor (IBPGR and ICRISAT, 1993). Data were collected on inflorescence compactness and shape, inflorescence exsertion, grain color, glume color, threshability, leaf midrib color, senescence, leaf orientation, leaf rolling, and the presence of productive tillers or not (tillering capacity), as presented in Table 2. The 10 traits were selected based on their relevance to drought tolerance, grain yield, and smallholder farmers' preferences.

2.4. Data Analysis

The raw data of the ten qualitative traits collected for 36 sorghum genotypes were subjected to descriptive statistical analysis (frequency distributions), and observation data were grouped by collection origin. Using the frequency distributions observed in various classes, the Shannon-Weaver Index of diversity (H') was calculated for each character as $H' = 1-\Sigma\, pi \log_e pi$; with H'= Shannon Diversity Index; pi = the proportion of accessions in the ithclass of an n–class character; n = the number of phenotypic classes of traits (Shannon and Weaver, 1964). Each diversity index value was divided by its greatest value (logn) and then normalized to preserve the values between 0 and 1. The diversity index for each character was calculated using the entire data set. The average diversity index ($\overline{H}'$) for each collection origin (C1-C6) was estimated as $\overline{H}'=\Sigma H'/n$, for 'n'=number of traits. The average diversity index (Hcl) for each trait was estimated as $\Sigma H'/nc$, where 'nc'=the number of collection origins (six collection origins). To perform analyses, Microsoft Excel was used to generate all those frequencies, as it is a friendly and accessible software that other authors previously utilized to analyze this type of data.

The six collection origins were designated from C1 to C6. Six sorghum landraces were collected from the Huye, Gisagara, and Ruhango districts and were designated C1, C2, and C4, respectively. Then, seven sorghum landraces designated as C3 and five sorghum landraces designated as C5 were collected from the Nyanza and Kamonyi districts, respectively. The six check varieties obtained from the sorghum research program were designated as C6 (Table 1).

Principal component analysis (PCA) was calculated using average values measured from 36 sorghum genotypes. Clustering was also performed based on the Unweighted Pair-Group Method with Arithmetic Means (UPGMA) (Sneath and Sokal, 1973). For PCA and clustering analysis, R was utilized as it is powerful and free software that helps to identify groups and partners within the data set.

Table 2. The qualitative traits of sorghum landraces scored at Rubona in the central plateau zone of Rwanda using sorghum descriptor (IBPGRI/ICRISAT, 1993)

Traits	Traits description
Inflorescence shape and compactness	Very lax panicle:1, very loose erect: 2, very loose drooping: 3, loose erect: 4, loose drooping: 5, semi-loose erect: 6, semi-loose drooping:7, semi-compact elliptic: 8, compact elliptic: 9, compact oval: 10, half broom corns: 11, broom corn:12
Inflorescence exsertion	1. Slightly exserted <2 cm: 1, exserted 2-10 cm: 2, well exserted >10 cm: 3, peduncle recurved: 4
Grain colour	White:1, yellow: 2, red: 3, brown: 4, buff: 5
Glume colour	White:1, sienna: 2, mahogany: 3, red: 4, purple: 5, black: 6, grey: 7
Threshability	Very difficult (<50%): 1, difficult (60-69%): 2, intermediate (80-84%): 3, good (90-94%): 4, excellent (99-100%): 5
Leaf midrib colour	White: 1, dull green: 2, yellow: 3, brown: 4, purple: 5
Senescence	Very slightly senescent: 1, slightly senescent: 2, intermediate (about half of leaves dead): 3, mostly senescent: 4. completely senescent (leaves and stalk dead): 5
Leaf orientation	Erect: 1, dropping: 2, intermediate: 3
Leaf rolling	Non rolled leaf: 1, 25% leaves rolled: 2, 50% leaves rolled: 3, 75% leaves rolled: 4, all leaves rolled: 5
Presence of capacity	Presence of productive tillers: 1, absence of tillers: 0

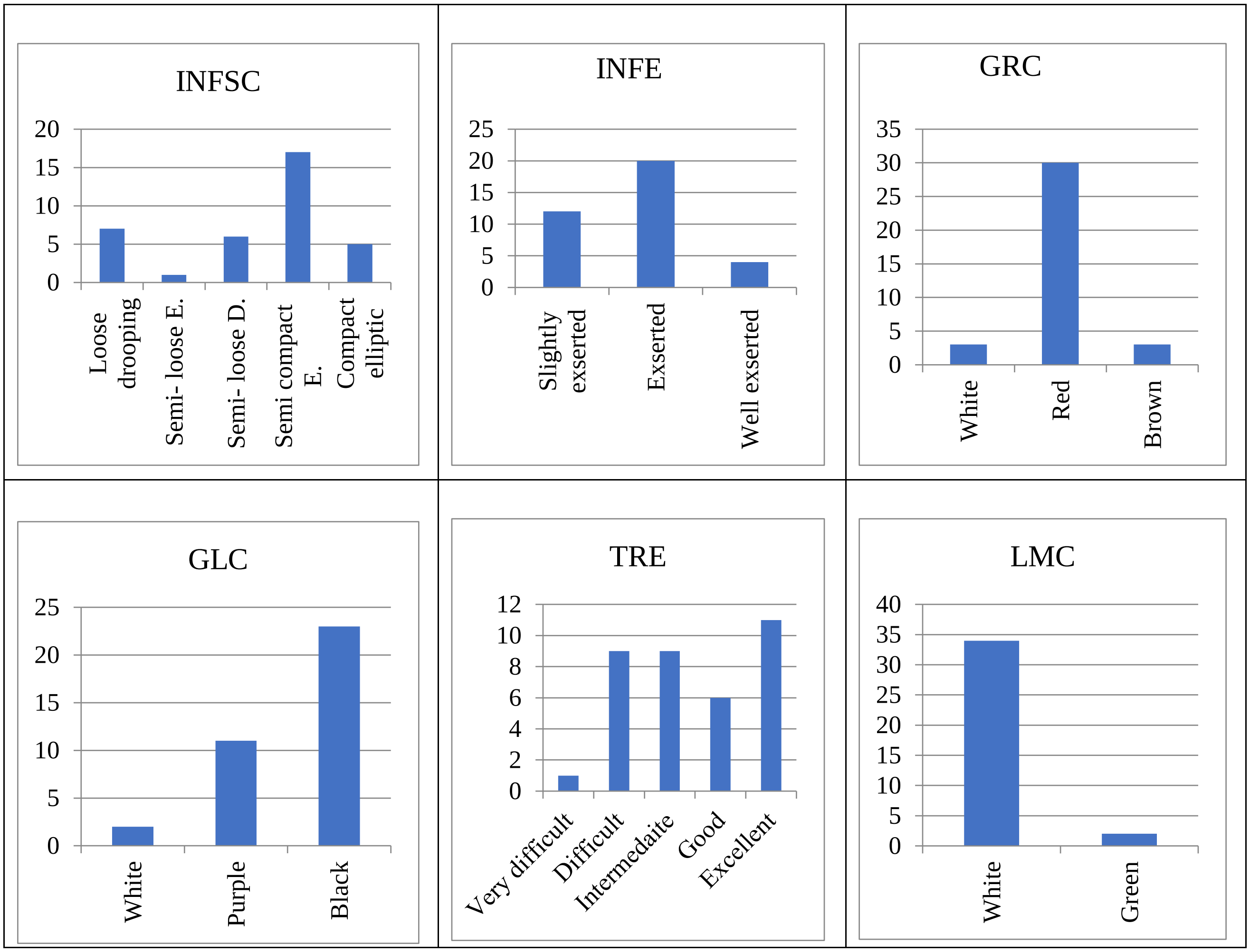
3. Results and Discussion

3.1. Variation of Qualitative Traits

All genotypes had a non-rolled leaf out of the five categories of leaf-rolling traits. The variation of inflorescence shape and compactness, glume colour and leaf midrib colour was less than 50% of the categories of the traits scored by 36 genotypes. The genotypes scored greater than 50% in the categories of inflorescence exsertion, grain colour, and leaf orientation of the phenotype traits. In contrast, a varied number of genotypes scored traits of all categories for threshability, senescence and tillering capacity (Figure 1). This showed that the leaf rolling trait had no variation in 36 sorghum genotypes, whereas the threshability, senescence, and tillering capacity phenotypic traits showed variations in sorghum genotypes more than the other phenotypic traits.

The largest proportion of genotypes had semi-compact erect (47.5%), exserted inflorescence (56%), red grain color (83%), black glume color (63%), excellent threshable grains (32%), white leaf midrib color (94.4%), intermediate senescence (33%), dropping leaf (67%), and no productive tillers (54%). A small proportion of genotypes had semi-loose erect inflorescence (2.8%), well-exserted inflorescence (11.1%), white and brown grain color (8.3%), white glume color (5.6%), very difficult threshable grains (2.8%), green midrib color (5.6%), very slightly senescence (2.8%), intermediate oriented leaf (33%), and productive tillers (46%).

Five of the six check sorghum varieties had semi-compact elliptic inflorescence and productive tillers, while four had red grain and white leaf midrib color. Only two or three of the six varieties scored two or three categories of exserted inflorescence, white grain color, purple and black glume colors, difficult and excellent threshable grains, green midrib color, completely senescent plants, and intermediate and dropping leaf traits (Figure 1 and Table 3). This showed that check varieties and the 30 landraces of sorghum in Rwanda were selected purposely by breeders and unknowingly by farmers for phenotypic traits that a large proportion of varieties and landraces scored. As a preference of sorghum farmers, the predominance of a great number of sorghum genotypes with compact panicles was reported by Assefa (2012) and Apunyo et al. (2022), while the majority of easy threshing panicles were also reported in previous studies by Abdi et al. (2002); Akatwijuka et al. (2016).



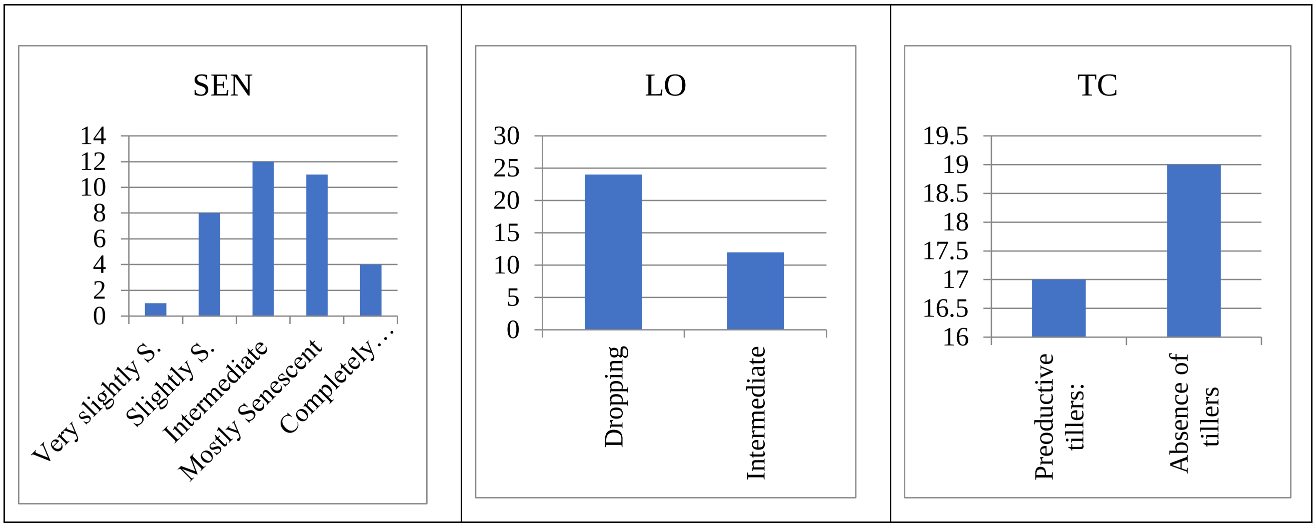


Fig. 1 The distribution of 36 sorghum genotypes is divided into different categories of nine qualitative traits. INFSC = Inflorescence shape and compactness, INFE = Inflorescence exsertion, GC = Grain colour, GLC = Glume colour, TRE = Threshability, LMC = Leaf midrib colour, SEN = Senescence, LO = Leaf orientation and TC = Tillering capacity

3.2. Frequency Distribution of Qualitative Traits in Six Groups of Genotypes

The frequency distribution of each qualitative trait of sorghum genotypes by their origins of collection is presented in Table 3. The results showed that most of the sorghum genotypes with compact inflorescence were collected from the Nyanza and Ruhango districts (two genotypes each). For semi-compact elliptic inflorescence, there were 17 genotypes, and the majority of them were collected from the sorghum research program (five genotypes). In comparison, the others originated from the Huye and Gisagara districts (three genotypes in each district).

No sorghum genotypes were reported to have very lax, very loose erect, very loose drooping, loose erect, loose drooping, compact oval, half broom corns, or broom corn inflorescence. The density and shape of sorghum inflorescence determine seed yield, with compact panicles being preferred by farmers due to their ability to hold more seeds than loose panicles (Fitrahtunnisa et al., 2020). This indicates that the presence of compact and semi-compact panicles may be related to farmer preferences.

In addition to the majority of sorghum genotypes with exserted inflorescence, there were also four genotypes with well-exserted inflorescences, while no peduncle recurved landrace was observed in this study. Gisagara and Ruhango districts were found to have many landraces with exserted inflorescence, while three out of four genotypes with well-exserted inflorescence originated from the Nyanza district. A well-exerted sorghum inflorescence characteristic is associated with post-flowering drought tolerance. Sorghum genotypes with increased inflorescence exertion are better able to withstand post-flowering drought, causing early plant death, lodging, seed size reduction, and final yield losses (Derese et al., 2018). The results of this study are in agreement with Assefa (2012) and Andiku et al. (2022), who reported the predominant number of sorghum genotypes with good exserted inflorescence.

The results of the study showed that Ruhango and Nyanza are the districts with the most red grain sorghum landraces. All sorghum genotypes that originated in the Ruhango district had red grain color. Also, 83.3% of genotypes in the Huye and Gisagara districts and 85.7% and 80% of the genotypes originated in the Nyanza and Kamonyi districts possess red grain color. Only sorghum collected from the research program had a more diverse grain color with 33.3 and 66.7% white and red grain color, respectively. Sorghum grain color is linked to the concentration of tannin in the pericarp, which affects its feeding value (Sedghi et al., 2012). These tannins, or the phenolic substances, help the sorghum plant develop defense mechanisms against pests and fungi (Diouf, 2016). The red color of sorghum may be due to its use in alcoholic local beer production and yeast fabrication in Rwanda. Farmers often choose landraces with desired features, making red grain sorghum more prevalent than other landraces (Akatwijuka et al., 2016). This is consistent with studies in Burundi and Ethiopia, where the majority of sorghum genotypes have red grain color (Habindavyi et al., 2009) and (Assefa, 2012). The preference of red grain sorghum genotypes may also be influenced by birds' non-preference, affecting the preferences of sorghum farmers.

The majority of genotypes (63%) had black glumes, and many of these genotypes originated from the Huye, Gisagara, and Ruhango districts (five genotypes in each district). In agreement with these results, Naoura et al. (2019) reported 66.7%, while Apunyo et al. (2022) reported 53.2% of sorghum accessions with black glume. Concerning threshability, the Kamonyi district showed many genotypes with excellent threshable grains (three genotypes). At the same time, a large number of varieties with difficult treshability were observed among varieties obtained from the research program. Threshability is an important trait, especially for smallholder sorghum farmers who normally perform manual threshing. Very difficult threshing causes significant postharvest loss or lower market value and processability, while excellent threshing results in time and energy savings and clean and high grain quality (Adeyanju et al., 2015). The majority of landraces with excellent threshability may be related to the class of sorghum farmers in Rwanda, who are almost all smallholder farmers, as reported by Niyibigira et al. (2013).

Table 3. The frequency distribution of 10 qualitative traits among 36 sorghum genotypes scored with respect to collection origins at Rubona in the central plateau zone of Rwanda in 2019

Trait	Phenotypic class	C1	C2	C3	C4	C5	C6	All
INFSC	Loose drooping	2	1	2	1	1	0	7
	Semi- loose erect	0	0	0	0	1	0	1
	Semi-loose D.	1	2	1	1	1	0	6
	Semi-compact E.	3	3	2	2	2	5	17
	Compact elliptic	0	0	2	2	0	1	5
INFE	Slightly exserted	3	2	1	2	2	2	12
	Exserted	3	4	3	4	3	3	20
	Well exserted	0	0	3	0	0	1	4
GC	White	0	0	1	0	0	2	3
	Red	5	5	6	6	4	4	30
	Brown	1	1	0	0	1	0	3
GLC	White	0	0	1	0	0	1	2
	Purple	1	1	2	1	3	3	11
	Black	5	5	4	5	2	2	23
TRE	Very difficult	0	0	1	0	0	0	1
	Difficult	0	0	3	1	2	3	9
	Intermediate	2	2	2	2	0	1	9
	Good	2	2	0	2	0	0	6
	Excellent	2	2	1	1	3	2	11
LMC	White	6	6	7	6	5	4	34
	Green	0	0	0	0	0	2	2
SEN	Very slightly senescent	0	0	1	0	0	0	1
	Slightly senescent	1	1	1	1	3	1	8
	Intermediate	4	1	1	5	0	1	12
	Mostly senescent	1	4	3	0	2	1	11
	Completely senescent	0	0	1	0	0	3	4
LO	Dropping	5	4	4	5	3	3	24
	Intermediate	1	2	3	1	2	3	12
LR	Non rolled leaf	6	6	7	6	5	6	36
TC	Presence of tillers	1	2	5	3	1	5	17
	Absence of tillers	5	4	2	3	4	1	19

(Note: Frequency distribution of genotypes of all collection origins for the corresponding qualitative traits. C1 = Huye district, C2 = Gisagara district, C3 = Nyanza district, C4 = Ruhango district, C5 = Kamonyi district and C6 = sorghum research program, INFSC = Inflorescence shape and compactness, INFE = Inflorescence exsertion, GC = Grain colour, GLC = Glume colour, TRE = Threshability, LMC = Leaf midrib color, SEN = Senescence, LO = Leaf orientation, LR = Leaf rolling and TC = Tillering capacity)

The results of the study indicated that the Kamonyi district registered a large number of slightly senescent genotypes (three genotypes), while many completely senescent genotypes were found in the sorghum research program, implying the need for a new breeding plan targeting the improvement of this important trait. According to Reddy et al. (2007), senescence is linked to drought tolerance in sorghum plants, as demonstrated by maintaining green stems and higher leaves during grain filling when there is insufficient water. This trait is essential for boosting grain yield in water-scarce situations (Reddy et al., 2007). The results of this study were in line with the findings of Assefa (2012), who reported 52% of sorghum genotypes with intermediate senescence, and Mulima (2017), who found the majority of sorghum genotypes to have slight senescence. Even if many landraces were not good in terms of this trait, one landrace and seven landraces that showed to be very and slightly senescent, respectively, are good parental candidates to consider in the breeding of drought-tolerant genotypes.

The leaf orientation was dropping in most of the landraces (67%), while no erect leaf was observed in the study. As for leaf rolling, there was no variation observed in this study, as all the landraces showed non leaf rolling. The majority of genotypes with intermediate leaves originated from the Nyanza district and the sorghum research program (three genotypes each), while many genotypes with dropping leaves originated from the Huye and Ruhango districts (five genotypes in each district). Leaf rolling and leaf orientation are good indicators for drought-tolerant sorghum genotypes when they are more upright and show rapid rolling, respectively (Truong et al., 2015; Mwamahonje et al., 2021). In the present study, the evaluated landraces did not show those good characteristics. However, it may be attributed to the abundance of rainfall during the same cultural season, as in total, the crop had a well-distributed rainfall of 604.5 mm, while sorghum requires 450–650 mm (Assefa et al., 2010). Similar to the results of this study, Mulima (2017) reported 61.5% and 42.3% of genotypes with dropping leaves and no rolled leaves, respectively.

The results of the study showed that the majority of genotypes with at least one productive tiller originated from the sorghum research program (five genotypes) and the Nyanza district (five genotypes), while the majority of genotypes with no-tillers originated from the Huye district. Sorghum grain yield is significantly influenced by tillering, which is directly related to the number of panicles that are produced (Li et al., 2003). Because of the higher leaf area associated with tillering, which increases intercepted radiation, tillering is also crucial for the formation of biomass (Alam et al., 2009).

3.3. Shannon-Weaver Index of Diversity

Understanding how germplasm varies genetically is key to making use of most of these genetic resources and coming up with the right plans for their conservation (Assefa, 2012). The results of the Shannon-Weaver Index of diversity estimated for each trait in each collection origin are presented in Table 4. Each collection origin's diversity indices showed variations in traits within each area of collections. In the Huye district (collection origin one), leaf midrib color had a low (0.09) diversity index value, whereas threshability had a high value of 0.65 diversity Index. These obtained results indicate that some phenotypic classes of leaf midrib color were more common than others, whereas the phenotypic classes of threshability and tillering capacity were evenly distributed within the Huye district.

Among all collection origins, the lowest diversity index was found in collection origin three (Nyanza district) with 0.08 (leaf midrib color), while the highest diversity index was recorded in collection origin four (Ruhango district) with a diversity index of 0.81 (tillering capacity). Tillering capacity recorded a high average diversity index across all six collection origins of 0.65, probably due to the few phenotypic classes (two). In the past, Abdi et al. (2002) reported that traits with two or three phenotypic classes generally record a higher diversity index than those with more than three. For the genotypes obtained from the research program (collection origin six), inflorescence compactness recorded the lowest diversity index (0.12), while tillering capacity showed the highest diversity index of 0.70 (Table 4).

Table 4. Diversity indices (H') Estimates of nine qualitative traits for each collection origin

	Collection Origins						
Traits	C1	C2	C3	C4	C5	C6	Hcl
Inflorescence compactness	0.27	0.27	0.47	0.47	0.47	0.12	0.34
Inflorescence exsertion	0.41	0.30	0.48	0.30	0.40	0.48	0.39
Grain colour	0.21	0.21	0.17	0.12	0.27	0.30	0.21
Glume colour	0.14	0.14	0.24	0.14	0.27	0.20	0.19
Threshability	0.65	0.65	0.47	0.72	0.35	0.31	0.53
Leaf midrib colour	0.09	0.09	0.08	0.09	0.11	0.23	0.12
Senescence	0.29	0.29	0.77	0.18	0.35	0.46	0.39
Leaf orientation	0.26	0.37	0.38	0.26	0.51	0.51	0.38
Tillering capacity	0.52	0.59	0.58	0.81	0.72	0.70	0.65
H'	0.32	0.29	0.36	0.31	0.34	0.33	0.32
Stdev	0.18	0.19	0.21	0.27	0.17	0.18	0.17

Note: C1 = Huye district, C2 = Gisagara district, C3 = Nyanza district, C4 = Ruhango district, C5 = Kamonyi district and C6 = Sorghum research program. Hcl = Average diversity index of the trait across all six collection origins. Stdev = Standard deviation

For genotypes, the maximum value of H' was 0.96 for tillering capacity, while the minimum was 0.23 for leaf midrib color, with an average of 0.53 (Table 5). Two of the four traits (senescence = 0.46 and leaf rolling = 0) that are essential for sorghum production under drought conditions showed low polymorphism, as they were below the average mean. For the two remaining traits, inflorescence exertion and leaf orientation had higher H' (0.64 and 0.62, respectively) than the overall mean, suggesting the possibility of selecting drought-tolerant genotypes using these traits. The tillering capacity and threshability traits showed high H' of 0.96 and 0.80 in comparison to the remaining traits, indicating that these traits can be used as visual scoring criteria for selecting sorghum genotypes, considering their importance in sorghum production ability and smallholder farmers' postharvest activities, respectively.

The results obtained in this study seem to be less than what Geleta and Labuschagne (2005) and Assefa (2012) reported in their previous studies. This may be attributed to the small number of genotypes evaluated in this study and to the fact that the genotypes evaluated were all collected from or adapted to the middle agroecological zone of Rwanda.

The collection origins' average diversity index (Hcl) also displayed considerable variation, ranging from 0.12 for leaf midrib color to 0.65 for tillering capacity (Table 5). The genotypes showed a diversity index (Hcl/H') ranging between 0.51 (leaf midrib color) and 0.91 (inflorescence shape and compactness) within collection origins, whereas the diversity index between collection origins [(H'-Hcl)/H'] was between 0.09 (inflorescence shape and compactness) and 0.49 (leaf midrib color) (Table 5). According to (Rao et al., 2021), diversity indices are categorized as high, with values ranging between 0.76- 0.99; moderate, with values ranging between 0.46-0.75; and low, having values ranging between 0.01-0.45. Two traits that are related to drought tolerance (inflorescence exertion and leaf orientation) recorded moderate diversity indices of 0.62 each, while senescence showed a high diversity index of 0.85 within collection origins. As for collection origins, all three drought-tolerant associated traits showed low diversity indices (Table 5). Similar to this study, the low diversity indices found between collection origins were also reported by Assefa (2012) when he studied the genetic diversity among lowland sorghum landraces using qualitative traits. A high proportion of diversity was also reported in previous studies by Ayana and Bekele (1998); Assefa (2012).

Table 5. Diversity index (H') of traits; overall, within and between collection origins

Traits	Collection origins				
	H'	Hcl	Hcl/H'	H'-Hcl	(H'-Hcl)/H'
Inflorescence compactness and shape	0.38	0.34	0.91	0.04	0.09
Inflorescence exsertion	0.64	0.39	0.62	0.25	0.38
Grain colour	0.33	0.21	0.64	0.12	0.36
Glume colour	0.33	0.19	0.56	0.14	0.44
Threshability	0.80	0.53	0.66	0.27	0.34
Leaf midrib colour	0.23	0.12	0.51	0.11	0.49
Senescence	0.46	0.39	0.85	0.07	0.15
Leaf orientation	0.62	0.38	0.62	0.24	0.38
Tillering capacity	0.96	0.65	0.68	0.31	0.32
Average	0.53	0.36	0.67	0.17	0.33
Stdev	0.24	0.17	0.13	0.10	0.13

Note : H'= Diversity Index (H') of each trait calculated from the entire data set (based on the frequency of the classes of the trait against all genotypes in the experiment); Hcl = Average diversity index of the trait across six collection origins; Hcl/H' = Diversity index of each trait within the collection origin, (H'-Hcl)/H' = The diversity index of each trait between collection origin

3.4. Principal Component Analysis

The principal component (PC) analysis was used to determine which traits had the most influence on the differences between the evaluated sorghum genotypes. The results revealed four PCs with eigenvalues greater than 1, accounting for 78.64% of the total variation (Table 6). The first PC was the most influential, making up 31.29% of the total variation, and it was mainly due to high positive loadings of three traits, grain color (0.526), glume color (0.490), and threshability (0.347). The second PC was the second main contributor and was responsible for 20.91% of the variation. It was mostly associated with inflorescence exsertion (0.537).

The third and fourth PCs contributed together to 26.44 % of the total variation, and their influence was due to positive vector loadings of threshability (0.548) and inflorescence compactness and shape (0.339) for the third PC, while for PC4, its influence was due to a nonsignificant positive vector loading of inflorescence exertion (0.163). According to Sejake et al. (2022), component loadings higher than 0.3 are seen as meaningful, while eigenvalues over 1 are significant. It was detected that senescence and midrib color had negative loadings in all four PCs, and this tells us that there was an inverse relation between those factors and those variables.

Table 6. Eigenvalues and contribution of the first four principal components to the variability of 36 sorghum genotypes based on nine qualitative traits

Trait	PC1	PC2	PC3	PC4
Inflorescence compactness and shape	0.035	-0.540	0.339	-0.271
Inflorescence exsertion	-0.144	0.537	0.045	0.163
Grain colour	0.526	0.132	-0.188	0.023
Glum colour	0.490	-0.054	-0.257	-0.084
Threshability	0.347	-0.209	0.548	-0.084
Leaf midrib colour	-0.474	-0.273	-0.105	-0.049
Senescence	-0.103	-0.426	-0.616	-0.028
Leaf orientation	0.126	0.201	-0.243	-0.768
Presence of productive tillers	-0.301	0.241	0.177	-0.540
Eigenvalue	2.816	1.882	1.308	1.071
Variability (%)	31.29	20.91	14.53	11.91
Cumulative (%)	31.29	52.20	66.73	78.64

Similar to the results of this study, Abdi et al. (2002) reported three PCs accounting for 91% of the total variation in the sorghum variability study in Nord Shewa and South Welo, Ethiopia, using qualitative traits. Assefa (2012) found 4 PCs contributing to 82.31% of 12 qualitative traits in Ethiopia. This multivariate analysis helps breeders recognize which traits have the most influence on genetic variability among accessions so they can identify the best parents for crop improvement.

3.5. Clustering of Sorghum Genotypes Using Qualitative Traits

The nine polymorphic qualitative traits were analyzed based on the Unweighted Pair-Group Method with Arithmetic Means to construct a dendrogram (Figure 2). As a result, the 36 sorghum genotypes were grouped into five main clusters.

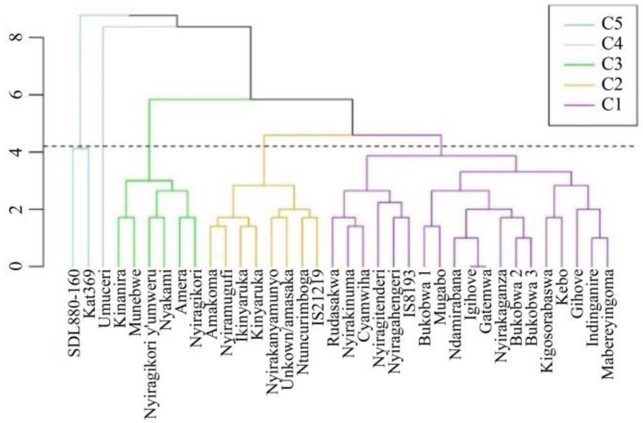


Fig. 2 Dendrogram generated based on UPGMA clustering method depicting genetic dissimilarity among 36 sorghum genotypes based on nine qualitative traits

The first cluster consist of 19 sorghum genotypes, while the second cluster had eight genotypes. The third cluster had six genotypes, while the fourth cluster consisted of a standalone distinct landrace, *Umuceri*. The last cluster had two improved varieties that were obtained from the sorghum research program. Those two genotypes were totally different from the others by their common complete senescence and their green midrib color. Looking at the distance between these genotypes on the dendrogram, it is advised to consider genotype *Umuceri* as a parent in the future improvement of drought tolerance as it is the only one that showed slight senescence among all the 36 genotypes. Lower leaf senescence reflects the sorghum's drought tolerance responses (Dong et al., 2022).

3.6. Distinguishing Phenotype Traits of Genotypes in Three Clusters

The 36 sorghum genotypes were clustered into five major groups based on the Euclidean distance matrix (Figure 2). The first cluster consists of 19 sorghum genotypes, including two identical genotypes (Igihove and Gatemwa) for the evaluated qualitative traits. The genotypes in this cluster are characterized mainly by red grain color (all of them), dropping leaves, black glume color, and no productive tillers (the majority of them) (Table 7). The second cluster had eight genotypes mainly characterized by difficult threshable grains (majority of them) and completely senescent leaves (few of them). The third cluster is composed of six genotypes characterized by brown grain color (half of them), and most of them with loose dropping inflorescence. The fourth cluster has a standalone genotype, *Umuceri*, which is distinguished from the others by its very slight senescence. In contrast, the fifth cluster contains two genotypes, SDL880-160 and Kat369, obtained from the sorghum research program. The two genotypes are distinguished from the others by their slight senescence and their green midrib color. In the past, Abdi et al. (2002) and Mulima (2017) grouped sorghum genotypes into five and four clusters, respectively, using qualitative traits. The findings of this study will help farmers to adapt to climate change-induced drought using affordable and environmentally friendly practices because drought-tolerant genotypes require relatively less water input, less pesticide, fewer labor resources, and less energy, and this, in turn, helps in reducing environmental damage.

Table 7. List of genotypes and distinguishing phenotype traits of sorghum genotypes in three clusters based on nine qualitative traits

Cluster	Genotype	Distinguishing traits of cluster
I	Mabereyingoma, Indinganire, Gihove, Kebo, Kigosorabaswa, Bukobwa 3, Bukobwa 2, Nyirakaganza, Gatemwa, Igihove, Ndamirabana, Mugabo, Bukobwa 1, IS8193, Nyiragahengeri, Nyiragitenderi, Cyamwiha, Nyirakinuma, and Rudasakwa	Semi-loose drooping, semi-compact elliptic and compact elliptic inflorescence. Slightly exserted, exserted and well exserted inflorescence. Red and grain color. Black and purple glume color. Intermediate, good and completely threshable grains. White leaf midrib color. Slightly intermediate and mostly senescent leaves. Dropping and intermediate leaf. Presence and absence of productive tillers
II	IS21219, Ntuncurimboga, Unkown/Amasaka, Nyirakanyamunyo, Kinyaruka, Ikinyaruka, Nyiramugufi, and Amakoma	Difficult threshable grains and completely senescent leaves
III	Nyiragikori, Amera, Nyakami, Nyiragikori y'umweru, Munebwe, and Kinanira	Loose drooping inflorescence, red and brown grain color
IV	Umuceri	Very slightly senescence
V	Kat 369 and SDL880-160	Green leaf midrib color and slightly senescence

4. Conclusion

The results of the study showed significant morphological diversity among the 36 sorghum genotypes based on 10 qualitative traits. The identification of drought-tolerant sorghum genotypes and breeding for drought-tolerant sorghum genotypes will promote agricultural resilience in Rwanda, as drought-tolerant genotypes maintain high yields under drought stress conditions, decrease the risk of yield loss and reduce water use costs, all providing a stable food and financial health for farmers and more stable agricultural systems. The successful identification and development of drought-tolerant genotypes need to be accompanied by local farmers' participation as they guide the selection of genotypes suitable to local conditions. Additionally, farmers' landraces are a precious source of genes to use in the development of drought-tolerant genotypes, and these landraces need to be safeguarded. Thus, the results of this study should also take into consideration the maintenance and conservation of biodiversity, as the evaluated genotypes have to be conserved for the utilization of current and future generations. To validate the present results, it will be good to conduct the same study using molecular markers, as environmental conditions influence the phenotype of the plants, causing variations in agromorphological markers.

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