



Tuber Trait Diversity among Yellow Nutsedge (*Cyperus esculentus* L.) Accessions from East Africa, Egypt, and Spain

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Abstract

Yellow nutsedge (*Cyperus esculentus* L.), a monocotyledonous member of the *Cyperaceae*, is valued for its oil-rich, starchy, protein-containing tubers. The species is cultivated commercially in several countries but occurs mainly as a weed of farmland and uncultivated areas in Kenya and much of East Africa. This study characterized tuber length and width (at the base, mid-point and tip) of 27 *C. esculentus* accessions, comprising 23 wild accessions collected in Kenya, Uganda and Tanzania and four cultivated accessions obtained from Spain and Egypt. A single tuber of each accession was planted in each of three pots in one greenhouse environment at the University of Eldoret, Kenya, and the pots were arranged in a completely randomized design. One mature tuber per pot was measured with a digital caliper, so the pot formed the experimental unit ($n = 3$ per accession). Data were analysed with descriptive statistics, one-way analysis of variance (ANOVA), Pearson correlation, principal component analysis (PCA), and hierarchical cluster analysis (HCA). Mean tuber length ranged three-fold, from 5.00 mm (MAR) to 15.00 mm (EGY), and coefficients of variation among accession means ranged from 27.6 to 30.0%. One-way ANOVA confirmed that accession identity explained a highly significant proportion of variation in every trait ($p < 0.001$). All four traits were positively correlated among accession means ($r = 0.68$ to 0.91). The first principal component (PC1, 86.3%) described overall tuber size and the second (PC2, 9.3%) described elongation, contrasting BIO (large, compact) with EGY (large, elongated). The four cultivated accessions had longer tubers (mean 12.0 mm) than the 23 wild accessions (mean 7.5 mm). The large-tubered branch of the HCA dendrogram contained all four cultivated accessions (BIO, BLACK, EGY, MICRO) and four wild accessions (KHA, BUB, DOM, KARI-E), although bootstrap support for this split was

weak (28%). These greenhouse findings demonstrate that this germplasm collection harbours substantial, statistically significant diversity in tuber size and shape, that tuber size and shape are largely independent traits, and that tuber-based and whole-plant vegetative characterization capture only partially overlapping dimensions of morphological diversity. Because the tuber dimensions were strongly correlated and were measured in one environment and season, the results do not establish the genetic basis of these differences or the independence of tuber size and shape. Multi-environment trials, genotyping, and nutritional analysis are recommended before wild East African accessions, particularly KHA, KARI-E, DOM, and BUB, are selected for crop improvement to advance the region's food and nutritional security agenda.

Subject Areas

Taxonomy, Food and Nutritional Security

Keywords

Cyperus esculentus L., Tubers, Principal Component Analysis, Hierarchical Cluster Analysis, East Africa, Underutilized Crop, Food and Nutritional Security

1. Introduction

Cyperus esculentus L., commonly known as yellow nutsedge, tiger nut, or chufa, is a perennial sedge in the family *Cyperaceae* [1]. This monocotyledon occurs both as a wild weedy species and as a deliberately cultivated food crop (**Figure 1(A)**). The species produces small, calorie-dense underground tubers as its primary storage organ (**Figure 1(B)**). These tubers have sustained human populations since antiquity and continue to support commercial food industries in Spain, China, Nigeria and Ghana [2]. Despite this long history of use and its documented nutritional value, tiger nut remains classified among the neglected and underutilized crop species of Africa. It has received comparatively little formal agronomic research investment relative to major staple crops, and its cultivation and value-chain development in East Africa in particular remain considerably less advanced than in West Africa or Spain.

Tuber size represents one of the most agronomically consequential traits in *Cyperus esculentus* L., since tuber dimensions relate directly to marketable yield, mechanical harvesting efficiency, and propagation performance [2]. Given ongoing efforts to combat malnutrition through nutritious locally sourced foods, understanding the tuber traits of this plant is valuable for potential crop improvement, domestication, and new adoptions [3].

Since yellow nutsedge propagates through tubers formed at the apical ends of rhizomes [4], rather than through seed, tuber-specific traits may be inherited or expressed somewhat independently of the broader vegetative morphology. Docu-

menting tuber size and shape variation directly therefore serves as a necessary complement to, rather than a substitute for, whole-plant morphological characterization, and provides breeders and germplasm curators with trait-specific information relevant to propagation and yield-oriented selection decisions. This study therefore aimed to characterize and document the tuber trait diversity among *Cyperus esculentus* L. accessions collected from East Africa, Egypt and Spain.



Figure 1. *Cyperus esculentus* L. mature plant (A) and (B) *C. esculentus* L. tubers.

2. Methodology

2.1. Plant Materials

Twenty-seven *Cyperus esculentus* L. accessions, comprising both wild and cultivated material collected across East Africa (**Table 1**), were used in this study.

Table 1. List of *Cyperus esculentus* L. complex used in the study.

Identity Code	Locality	Name of Collector	Type
BLACK	Spain	A. Carlsson	Cultivated
MICRO	Spain	A. Carlsson	Cultivated
BIO	Spain	A. Carlsson	Cultivated
EGY	Egypt	A. Carlsson	Cultivated
KARA	Karatina Tea farm, Kenya	E. Too, A. Onkware, B. Were	Wild
KARI-E	Kari-Embu	E. Too, A. Onkware, B. Were	Wild
EMBU-G	Embu-Gakwelon	E. Too, A. Onkware, B. Were	Wild
NKU-M	Nkubu Meru	E. Too, A. Onkware, B. Were	Wild
SOK	Sokomoko farm	E. Too, A. Onkware, B. Were	Wild

Continued

TURBO J.	Turbo Juakali	E. Too, A. Onkware, B. Were	Wild
CHEP	University of Eldoret (Chepkoilel)	E. Too, A. Onkware, B. Were	Wild
DOM	Dominion Farms	E. Too, A. Onkware, B. Were	Wild
CHU-18	Chuka Karatina	E. Too, A. Onkware, B. Were	Wild
NGA	Ngata Nakuru	E. Too, A. Onkware, B. Were	Wild
OGEMBO	Ogembo Kisii	E. Too, A. Onkware, B. Were	Wild
MAZ	Mazombe, Kilolo, Tanzania	E. Too, A. Onkware, B. Were	Wild
MAR	Marindi, Rodi Kopany	E. Too, A. Onkware, B. Were	Wild
IDO	Idofi, Mafindi, Tanzania	E. Too, A. Onkware, B. Were	Wild
IDU	Iduda, Mbeya, Tanzania	E. Too, A. Onkware, B. Were	Wild
ILU	Ilula, Mbeya, Tanzania	E. Too, A. Onkware, B. Were	Wild
BUMO	Bumoni, Tororo, Uganda	D. Akinyi, E. Too, A. Onkware, B. Were	Wild
BUB	Bubutu, Tororo, Uganda	D. Akinyi, E. Too, A. Onkware, B. Were	Wild
KHA	Khamitaro, Mbale, Uganda	D. Akinyi, E. Too, A. Onkware, B. Were	Wild
BUWA	Buwangola, Tororo, Uganda	E. Too, A. Onkware, B. Were	Wild
KAM	Kamungusa-Ugenya	E. Too, A. Onkware, B. Were	Wild
LWA	Luanda, Kotieno, Kenya	E. Too, A. Onkware, B. Were	Wild
SUK	Sukunanga-Eldoret, Kenya	E. Too, A. Onkware, B. Were	Wild

2.2. Sample Preparation

Tubers of each accession were multiplied in soil-filled plastic bags under standardized nursery conditions prior to data collection, a propagation approach adopted to confine each accession's rhizome system. The resultant tubers were then planted in the greenhouse facilities of the University of Eldoret, located in Uasin Gishu County, Kenya. Soil was collected from the university farm and placed in sample collection bags. These were then sterilized using an autoclave for 1 hour at 121°C then allowed to cool before transfer to clean, sterilized pots.

2.3. Planting

The *C. esculentus* L. accessions were then rehydrated by soaking in water for 24

hours, incubated in an oven at 28°C for 12 hours then planted directly onto pots filled with soil to $\frac{3}{4}$ full. Three pots were established per accession, each planted with a single tuber, giving 81 pots (27 accessions $\times$ 3 pots). The pots were arranged on the greenhouse benches in a completely randomized design, with each pot assigned to a bench position by random numbers. No blocking factor was imposed, and the statistical analysis therefore contains no block term. Plants were watered twice daily and observed until maturity.

2.4. Data Collection

At maturity, the tubers of each pot were harvested separately and cleaned of adhering soil. One mature tuber was selected at random from each pot and measured, so each replicate value represents a single tuber from a single pot, and the pot is the experimental unit ($n = 3$ per accession). Four linear traits were measured with a digital caliper (resolution 0.01 mm) and recorded to the nearest 0.5 mm:

- Length of the tuber: the maximum longitudinal distance from the proximal to the distal end of the tuber.
- Width at the base: the transverse width measured at the proximal (basal) end of the tuber.
- Width at the mid: the transverse width measured at the midpoint along the tuber's longitudinal axis.
- Width at the tip: the transverse width measured at the distal (tip) end of the tuber.

2.5. Data Analysis

Data were recorded in triplicate, keyed into Microsoft Excel spreadsheets. Genotype means for each trait were subsequently calculated by averaging the three replicate values recorded for each accession. Standard deviation (SD) and coefficient of variation (CV) were also calculated. Results were expressed as mean $\pm$ standard deviation for each trait across the 27 accessions. One-way analysis of variance (ANOVA) was conducted separately for each of the four tuber traits at a 95% confidence level ($p \leq 0.05$). Pearson correlation coefficients were calculated among all pairwise combinations of the four traits, using genotype means ($n = 27$), to characterize the degree and direction of association among the tuber dimensions measured. A p -value of ≤ 0.05 was considered statistically significant.

Principal component analysis (PCA) was then performed on the standardized data matrix. Eigenvalues, percentage variance explained, cumulative variance explained, and trait loadings were extracted for each component, and the first two components were retained for interpretation and visualization on the basis of their cumulative explanatory power. Euclidean distances were then calculated among accessions using the standardized genotype means for the four traits, and hierarchical cluster analysis (HCA) was performed using Ward's minimum-variance linkage method. Node support was estimated by resampling the four traits with replacement over 1,000 bootstrap iterations. Results were then presented in Tables

and Figures.

3. Results and Discussion

Table 2 shows accessions' mean $\pm$ SD Tuber Dimensions (mm) for the four tuber dimensions in 27 *Cyperus esculentus* L. Accessions. Each mean is based on three pots, with one tuber measured per pot.

Table 2. Tuber dimensions (mm) for the selected *Cyperus esculentus* L. accessions.

Accession	Length	Width Base	Width Mid	Width Tip
BIO	10.33 $\pm$ 1.04	8.67 $\pm$ 1.26	11.67 $\pm$ 1.04	8.00 $\pm$ 0.50
BLACK	13.83 $\pm$ 1.53	8.50 $\pm$ 1.00	10.67 $\pm$ 0.76	8.83 $\pm$ 1.26
BUB	8.50 $\pm$ 1.00	6.00 $\pm$ 0.50	7.17 $\pm$ 1.04	4.83 $\pm$ 0.58
BUMO	7.00 $\pm$ 1.32	4.67 $\pm$ 0.29	6.33 $\pm$ 0.76	4.50 $\pm$ 0.50
BUWA	8.50 $\pm$ 0.50	4.50 $\pm$ 0.50	6.33 $\pm$ 0.76	4.33 $\pm$ 0.29
CHEP	6.67 $\pm$ 1.44	4.00 $\pm$ 0.87	5.33 $\pm$ 1.04	3.67 $\pm$ 0.29
CHU 18	9.33 $\pm$ 0.29	4.50 $\pm$ 0.00	5.33 $\pm$ 0.29	4.00 $\pm$ 0.50
DOM	9.33 $\pm$ 1.44	5.83 $\pm$ 1.04	7.33 $\pm$ 1.44	4.50 $\pm$ 0.00
EGY	15.00 $\pm$ 0.50	8.33 $\pm$ 0.29	7.33 $\pm$ 0.76	5.70 $\pm$ 1.59
EMBU G	5.50 $\pm$ 1.00	3.67 $\pm$ 0.29	5.33 $\pm$ 1.53	4.00 $\pm$ 0.50
IDO	6.83 $\pm$ 1.15	3.67 $\pm$ 0.76	4.67 $\pm$ 0.29	3.00 $\pm$ 0.50
IDU	7.17 $\pm$ 1.26	5.33 $\pm$ 1.04	5.83 $\pm$ 0.29	3.83 $\pm$ 0.76
ILU	6.83 $\pm$ 0.29	4.50 $\pm$ 0.00	5.50 $\pm$ 0.50	4.17 $\pm$ 0.29
KAM	5.33 $\pm$ 0.29	3.83 $\pm$ 0.58	4.50 $\pm$ 0.50	3.17 $\pm$ 0.29
KARA	7.33 $\pm$ 0.76	4.33 $\pm$ 0.76	4.83 $\pm$ 0.76	4.33 $\pm$ 0.29
KARI-E	9.83 $\pm$ 0.29	6.00 $\pm$ 0.50	7.67 $\pm$ 1.61	4.50 $\pm$ 1.00
KHA	10.17 $\pm$ 2.47	6.00 $\pm$ 1.73	6.33 $\pm$ 2.02	5.67 $\pm$ 1.61
LWA	6.17 $\pm$ 0.29	5.33 $\pm$ 0.29	6.33 $\pm$ 0.29	4.00 $\pm$ 0.50
MAR	5.00 $\pm$ 0.50	3.33 $\pm$ 0.29	5.50 $\pm$ 0.50	4.17 $\pm$ 0.29
MAZ	8.50 $\pm$ 0.50	5.17 $\pm$ 0.29	6.17 $\pm$ 0.29	4.00 $\pm$ 0.50
MICRO	8.83 $\pm$ 0.58	7.00 $\pm$ 1.80	8.00 $\pm$ 0.50	5.00 $\pm$ 0.50
NGA	8.83 $\pm$ 1.61	4.17 $\pm$ 1.04	6.00 $\pm$ 2.65	4.00 $\pm$ 1.00
NKU-M	6.33 $\pm$ 1.04	4.50 $\pm$ 0.00	4.33 $\pm$ 0.29	3.50 $\pm$ 0.87
OGEMBO	6.00 $\pm$ 0.50	3.33 $\pm$ 0.29	5.17 $\pm$ 0.29	4.00 $\pm$ 0.50
SOK	5.67 $\pm$ 0.76	3.50 $\pm$ 0.00	4.67 $\pm$ 0.58	3.00 $\pm$ 0.50
SUK	9.50 $\pm$ 0.00	4.67 $\pm$ 0.29	5.33 $\pm$ 0.29	3.50 $\pm$ 0.50
TURBO J	7.00 $\pm$ 0.50	4.33 $\pm$ 0.29	5.33 $\pm$ 0.29	4.33 $\pm$ 0.29
Mean	8.12	5.10	6.26	4.46
SD	2.40	1.53	1.73	1.32
Minimum	5.00	3.33	4.33	3.00
Maximum	15.00	8.67	11.67	8.83
CV (%)	29.5	30.0	27.6	29.6

The mean tuber length across the collection was $8.12 \text{ mm} \pm 2.40 \text{ mm}$, ranging more than three-fold from 5.00 mm in accession MAR to 15.00 mm in accession EGY. Width at the base averaged $5.10 \text{ mm} \pm 1.53 \text{ mm}$ and ranged from 3.33 mm (MAR, OGEMBO) to 8.67 mm (BIO). Width at the mid-point, the broadest dimension recorded on average (mean = 6.26 mm), ranged from 4.33 mm (NKU-M) to a striking 11.67 mm in BIO, the single largest individual trait value recorded anywhere in the dataset. Width at the tip averaged $4.46 \text{ mm} \pm 1.32 \text{ mm}$ and ranged from 3.00 mm (IDO, SOK) to 8.83 mm (BLACK). The four cultivated accessions had larger tubers than the wild accessions, with a mean length of 12.0 mm (range 8.83 to 15.00 mm) against 7.45 mm (range 5.00 to 10.17 mm), and a mean mid-point width of 9.42 mm against 5.71 mm.

Coefficients of variation (CV) remained consistently high and closely comparable across all four traits, ranging narrowly from 27.6% (width mid) to 30.0% (width base). This narrow spread in CV values across traits of otherwise quite different magnitude indicates that the four tuber dimensions scale together fairly proportionally across the collection. Accessions with unusually long tubers tend also to have unusually wide tubers, rather than length and width varying independently at markedly different rates.

The observed dimensions fall within the range reported for the species, with the wild accessions toward its lower end. The cultivated Spanish and Egyptian accessions (8.8 to 15.0 mm long and 7.3 to 11.7 mm wide at the mid-point) resemble the morphotypes described from Burkina Faso (9.8 to 13.1 mm long and 9.0 to 11.9 mm wide) [5] and the site means reported from Russia (10.9 to 12.7 mm long) [6]. Most wild East African accessions produced tubers 5 to 9 mm long and 4 to 6 mm wide, in line with descriptions of smaller tubers in weedy than in cultivated forms of the species [7]. The CVs of 27.6 to 30.0% exceed the moderate CVs (15.6 to 19.1%) reported for traits including tuber dimensions among 42 accessions grown in China [8], which likely reflects the inclusion of both wild and cultivated material in the present collection. Length-to-mid-width ratios ranged from 0.89 (BIO) to 2.05 (EGY), spanning shapes comparable to the round, oval, and long classes described by Yang *et al.* [8].

EGY, BLACK, BIO, KHA, and KARI-E were the five longest-tubered accessions, while MAR, KAM, EMBU G, SOK, and OGEMBO formed the five shortest-tubered accessions. One-way ANOVA was conducted separately for each of the four tuber traits to test whether accession identity explained a statistically significant proportion of the observed variation. Accession identity explained a highly statistically significant proportion of variation in all four traits ($p < 0.001$ in every case), with F-statistics ranging from 9.18 (width at the mid) to 17.00 (length) (Table 3). The comparatively larger F-statistic obtained for length indicates that this trait discriminated most sharply among accessions relative to the within-accession replicate variability, consistent with the wide three-fold range and high CV. The smaller, though still highly significant, F-statistic for width at the mid reflects the somewhat greater within-accession replicate variability observed for this trait, particu-

larly evident in accessions such as NGA (SD = 2.65 mm) and KHA (SD = 2.02 mm), where individual tuber measurements varied considerably around the genotype mean. Because accessions were propagated clonally and grown in one environment, repeatability here combines genetic effects, carry-over effects from the mother tuber, and accession-specific responses to the greenhouse. It therefore represents an upper limit for broad-sense heritability rather than an estimate of it [9].

Table 3. One-way ANOVA results for tuber traits across 27 accessions.

Trait	F-Statistic	p-Value	Significance
Length	17.00	<0.001	Significant
Width Base	12.01	<0.001	Significant
Width Mid	9.18	<0.001	Significant
Width Tip	10.33	<0.001	Significant

Note. Significance assessed at $\alpha = 0.05$ (df = 26, 54).

Pearson correlation coefficients were calculated among the four tuber traits using genotype means and presented in **Table 4**.

Table 4. Pearson correlation matrix for tuber traits.

Trait	Length	Width Base	Width Mid	WIDTH TIP
Length	1.000	0.841	0.682	0.718
Width Base	0.841	1.000	0.821	0.851
Width Mid	0.682	0.821	1.000	0.912
Width Tip	0.718	0.851	0.912	1.000

All pairwise correlations were positive and, with the exception of the length-width mid pairing, uniformly strong ($r > 0.82$), consistent with the expectation that linear dimensions of the same physical organ tend to increase together as overall tuber size increases. The strongest single correlation was recorded between width at the mid and width at the tip ($r = 0.912$), indicating that these two width measurements track one another very closely across the collection and may carry substantially overlapping information for classification purposes. Width at the base likewise correlated strongly with both width at the tip ($r = 0.851$) and length ($r = 0.841$), positioning it as a dimension broadly representative of overall tuber girth and size.

The comparatively weaker correlation between length and width at the mid ($r = 0.682$), while still statistically meaningful, represents the weakest pairwise relationship in the matrix and is directly responsible for the emergence of a distinct shape axis in the subsequent PCA. In practical terms, this weaker correlation means that knowing an accession's tuber length alone provides a less reliable prediction of its mid-tuber girth than knowing its base width or tip width would pre-

dict the other width measurements. This is precisely the pattern underlying the length-to-mid-width shape ratio where accessions such as EGY (long, comparatively narrow) and BIO (comparatively short, very wide at the mid-point) diverge most sharply from a simple proportional size relationship.

PCA was performed on standardized (z-score) genotype means for the four tuber traits to summarize the correlation structure into a smaller number of interpretable axes. PCA of the standardized accession means showed the first two principal components together explained 95.6% of total variation among accessions (Table 5), an unusually high cumulative proportion that follows directly from the strong positive intercorrelation among all four traits and the small number of variables analysed. With only four highly correlated linear measurements available, most of the meaningful variation in the dataset is necessarily concentrated into very few dimensions.

Table 5. Percentage variance explained and trait loadings for PC 1 and PC 2.

Component/Trait	PC1	PC2
Variance explained (%)	86.3	9.3
Cumulative variance (%)	86.3	95.6
Length loading	0.467	0.793
Width base loading	0.519	0.108
Width mid loading	0.506	−0.465
Width tip loading	0.506	−0.378

PC1, accounting for 86.3% of total variation on its own, carried positive loadings of broadly similar magnitude from all four traits (0.467 to 0.519), confirming its interpretation as a general tuber size axis rather than a contrast between specific dimensions. Accessions positioned toward the positive end of PC1, most prominently BIO, BLACK, and EGY, followed by KHA, KARI-E, DOM, BUB, and MICRO, are distinguished by consistently larger measurements across all four traits simultaneously. Accessions positioned toward the negative end of PC1, including MAR, KAM, SOK, EMBU G, OGEMBO, NKU-M, and IDO, are correspondingly smaller across all four traits. This size gradient along PC1 aligns closely with the raw ranking of accessions by mean tuber length reported earlier, confirming that PC1 essentially recovers and formalizes the size ordering already evident from the descriptive statistics.

PC2, accounting for a further 9.3% of variation, presented a markedly different loading pattern: length loaded strongly and positively (0.793), while width at the mid and width at the tip loaded moderately and negatively (−0.465 and −0.378, respectively), with width at the base contributing only a small positive loading (0.108). This pattern identifies PC2 as a tuber elongation or shape axis, orthogonal to the general size axis captured by PC1. Accessions scoring highly on PC2, namely EGY, SUK and CHU 18, combine substantial length with comparatively narrow mid-

and tip-width measurements, producing visibly elongated tuber shapes. BIO, despite its high PC1 score reflecting large overall size, scored the lowest of any accession on PC2, reflecting its unusually compact, rounded tuber shape already noted in the length-to-mid-width ratio analysis. This separation between size (PC1) and shape (PC2) is visualized directly in the PCA biplot (**Figure 2**), where BIO and EGY occupy near-opposite vertical positions despite both scoring toward the positive end of the horizontal PC1 axis. The orthogonality of PC1 and PC2 does not show that tuber size and shape are biologically independent. This is because principal components are uncorrelated by construction, regardless of the underlying biology [10].

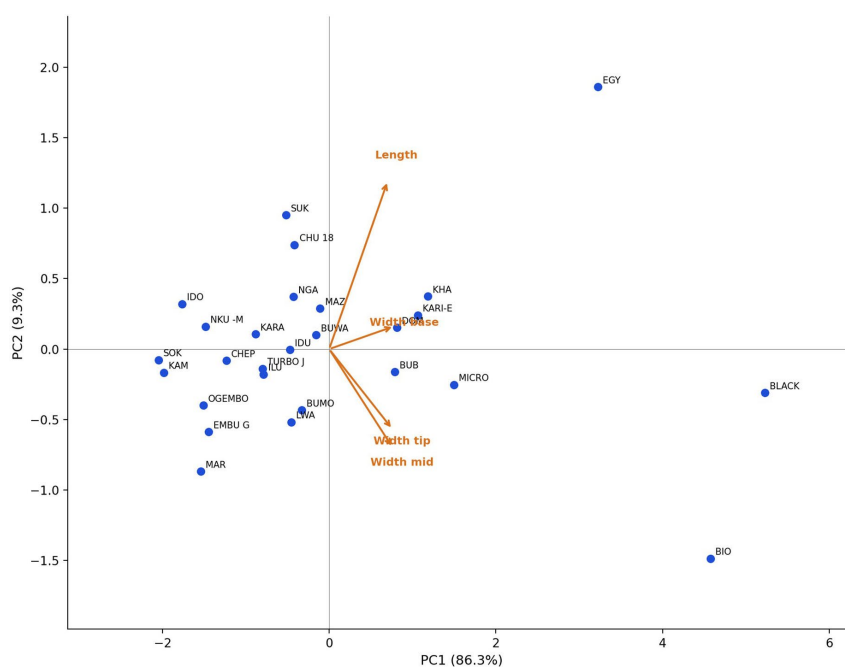


Figure 2. PCA biplot of 27 *Cyperus esculentus* L. accessions based on four standardized tuber morphometric traits.

The remaining two components, PC3 and PC4, together accounted for only 4.4% of total variation and are not interpreted further, since the near-complete summarization achieved by PC1 and PC2 indicates that a two-dimensional size-and-shape representation captures all systematic morphological variation present in this four-trait tuber dataset essentially.

Ward's minimum-variance hierarchical clustering was applied to Euclidean distances calculated from the standardized genotype means. Bootstrap support values were generated by resampling the four available traits with replacement across 1,000 iterations and recomputing the dendrogram topology on each resampled dataset. The resulting dendrogram (**Figure 3**) divided the collection into two principal branches at a Ward distance of approximately 11, though bootstrap support for this primary split was weak (28%), reflecting the limited discriminating power available from only four correlated size-related traits. The smaller of the two prin-

cipal branches, shown in green, grouped eight accessions distinguished by consistently larger tuber dimensions: BIO, BLACK, EGY, KHA, MICRO, BUB, DOM, and KARI-E. This grouping corresponds closely, though not perfectly, to the accessions scoring most positively along PC1 in the preceding PCA, reinforcing that both analytical approaches are detecting the same underlying large-tuber size gradient rather than providing independent structural evidence.

The larger principal branch, shown in orange, grouped the remaining 19 small-to-medium tubered accessions and subdivided further into several smaller sub-clusters at generally low-to-moderate bootstrap support, including a grouping of MAR, EMBU G, and OGEMBO; a grouping of NKU-M, IDO, KAM, and SOK; a grouping built around CHU 18 and SUK; and a grouping built around MAZ, BUWA, and BUWA. Several individual accession pairs within these sub-clusters returned comparatively strong bootstrap support despite the limited trait set available, most notably BIO-BLACK (86%), DOM-KARI-E (84%), MAR-EMBU G (74%), and CHU 18-SUK (52%). These higher-support pairings identify the accession relationships that are most consistently recovered regardless of which subset of the four traits is emphasized in a given bootstrap resample, and thus represent the most reliable tuber-based groupings identified in this analysis.

The close correspondence between the PCA and HCA results in this analysis is an expected consequence of the small number of highly intercorrelated traits examined: with only four variables, most of which correlate at $r > 0.68$, both ordination and clustering methods necessarily converge on describing the same dominant size gradient, offering limited scope for genuinely independent structural insight.

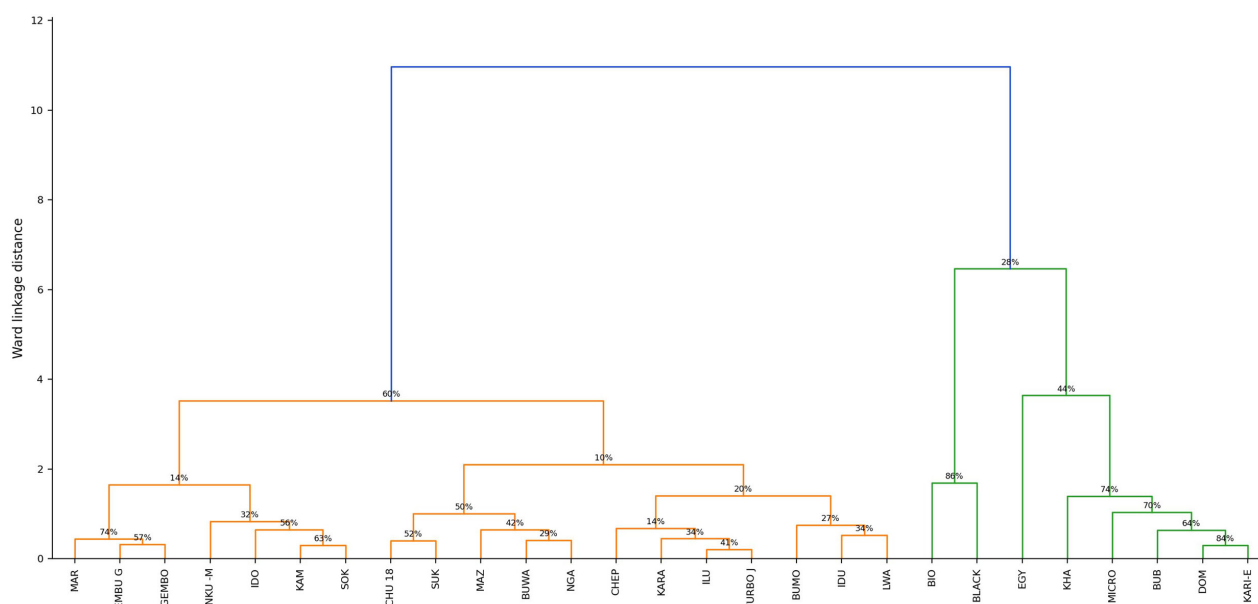


Figure 3. Hierarchical cluster analysis of 27 *C. esculentus* L. accessions based on four tuber morphometric traits.

4. Conclusions and Recommendations

This study concludes that the mean tuber length of the 27 *Cyperus esculentus* L.

accessions collected from wild and cultivated sources across East Africa, Egypt, and Spain ranged more than three-fold, from 5.00 mm (MAR) to 15.00 mm (EGY). All four traits showed consistently high coefficients of variation (27.6% - 30.0%). One-way ANOVA confirmed that accession identity explained a highly significant proportion of variation in every trait ($p < 0.001$), and Pearson correlation showed all four dimensions positively intercorrelated, most strongly between width at the mid and tip ($r = 0.912$), most weakly between length and width at the mid ($r = 0.682$). Principal component analysis (PCA) reduced this correlation structure to two axes explaining 95.6% of total variation: a dominant size axis (PC1, 86.3%) and a secondary elongation axis (PC2, 9.3%). The orthogonality of these axes is a property of PCA and does not demonstrate independent control of tuber size and shape. The four cultivated accessions from Spain and Egypt produced the largest tubers, while KHA, KARI-E, DOM, and BUB were the largest-tubered wild East African accessions. Hierarchical cluster analysis (HCA) grouped eight large-tubered accessions (BIO, BLACK, EGY, KHA, MICRO, BUB, DOM, KARI-E) apart from the remaining nineteen, though with weak bootstrap support (28%). However, because the data come from one environment and from a mixture of wild and imported cultivated material, they establish phenotypic differences under these conditions but not the heritable basis of the diversity or the crop potential of East African accessions.

It is recommended that future work on this *Cyperus esculentus* germplasm collection should: 1) evaluate the accessions in replicated multi-location field trials over more than one season, with several tubers measured per plant, to estimate genotype-by-environment interaction and broad-sense heritability, 2) genotype the accessions to test whether tuber-size groupings reflect genetic relationships, 3) prioritize the largest-tubered wild East African accessions (KHA, KARI-E, DOM, and BUB), alongside the cultivated references BIO, BLACK, and EGY, for nutritional analysis to test whether tuber size predicts oil, starch, protein and mineral content, 4) compare elongated and rounded morphotypes for oil extraction and processing quality, 5) test congruence among tuber, vegetative and nutritional datasets with Mantel tests and combined ordination, and 6) extend collection and characterization to additional East African collection sites to identify populations best suited to advancing food and nutritional security in the region.

Data Availability

Accession-level summaries (mean $\pm$ SD of three pots per accession) are reported in **Table 2**. The individual pot-level measurements (81 records: 27 accessions $\times$ 3 pots, one tuber per pot, four traits) can be provided by the corresponding author if needed. The analyzed data generated are included in this published article.

Author Contributions

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Conflicts of Interest

The authors declare no conflicts of interest.

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