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Genetic Characterization of Water Yam (*Dioscorea alata* L.) and Production Constraints of Yam in Pwani Region and Unguja Island, Tanzania

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Abstract

Yam (*Dioscorea* spp.) is a tuber crop cultivated in over 30% of all regions in Tanzania, contributing to the carbohydrate needs of many households. The morphological and genetic diversity of water yam (*D. alata* L.) local cultivars collected from Pwani region and Unguja Island were evaluated using 26 agro-morphological characters and 10 Short Sequence Repeat (SSR) markers. Cluster analysis using morphological characters classified nine water yam local cultivars into two main clusters with three subclusters. Tuber flesh and leaf colour were the main descriptors for clustering. The genetic diversity of 128 water yam genotypes using ten polymorphic SSR markers generated a total of 40 alleles that ranged from 2 to 7 per SSR marker. The mean polymorphic information content (PIC) ranged from 0.3 to 0.9, with a mean of 0.56. Cluster analysis separated 128 yam genotypes into two main clusters with five subclusters. The findings revealed relatively high genetic diversity of *D. alata* in the study areas. A structured questionnaire revealed drought (29.3%) and lack of planting materials (23.3%) as the major constraints to yam production in the study areas. Information reported in this study is useful in improving, reviving and promoting yam through breeding and conservation programs.

Key words: Genetic diversity; morphological descriptors; water yam

Introduction

Yam belongs to the family Dioscoreaceae, genus *Dioscorea*. It is among the tuber crops grown by smallholder farmers as a source of food and income, mainly in Sub-Saharan Africa, especially West Africa, South Eastern Asia and tropical America (Talwana et al. 2009, Rao et al. 2019). The genus *Dioscorea* contains more than 600 species, of which about ten are considered economically important food sources in the tropics (Kumar et al. 2017). The commonly cultivated species grown worldwide include *D. rotundata* (white guinea yam), *D. alata* (water yam), *D. bulbifera* (aerial yam), *D. esculenta* (Chinese yam) and *D. Dumetorum* [trifoliate yam] (Mignouna et al. 2007, Kumar et al. 2017).

West Africa accounts for more than 65% of the total yam produced globally, with

Nigeria being the leading country (FAOSTAT 2021). In East Africa, Sudan is the leading producer of yam, accounting for 166,843 tons annually, while Tanzania accounts for only 9493.32 tons (FAOSTAT, 2021). This production data for Tanzania might have yet to capture the reality since, for many years, yam has contributed to food security for many households in over 30% of regions of Tanzania (Massawe and Temu, 2022). Yam is traditionally cultivated in various parts, including Zanzibar Islands, especially Unguja in the fertile coral rag soils, in the high and mid-highlands, including the Lake Zone (especially Kagera and Mwanza), Arusha, Manyara, Kilimanjaro, Mbeya, Morogoro, Lindi and Mtwara (Massawe and Temu 2022). Although minimal reports are available, the three most common types of yam that are informally grown in Tanzania are*D. rotundata*, *D. bulbifera* and *D. alata*. However, other species like *D. Dumetorum* and *D. cyanosis*have also been reported (Massawe and Temu 2022, 2023).

D. alata(commonly water yam or greater yam) is among the most cultivated *Dioscorea* species worldwide, making it of the greatest economic interest in Africa. In Tanzania,*D. alata* is also the commonly grown yam in almost all regions due to its ability to grow in different agroecological zones on the natural soil fertility for its growth and yield (Guessan et al. 2009, Hgaza et al. 2010). The importance of *D. alata*in food security is accounted for its high-yielding potential and tuber storability, while the bulbils can be used as propagules (Alieu and Robert 2014).

There has yet to be a national breeding or conservation program for yam in Tanzania. Production and management practices have been mainly from indigenous experience with scant or no efforts at all from researchers. Consequently, yam production constraints still need to be revealed or well documented. Global climate change is currently threatening various crop production, including those entrusted as staples such as rice, maize and wheat. Yam could be adopted as an alternative or complimentary source of carbohydrates. However, due to a lack of research, very little is known about the production constraints of yam, while the morphological and genetic diversity of *Dioscorea* spp in some major producing regions was documented recently (Massawe and Temu 2022, 2023). Without research, some yam genotypes might get lost due to severe drought and diseases caused by the increasing effects of climate change. The genetic diversity data is crucial for breeders to work on crop improvement via breeding programs. Similarly, information on the yam production constraints is vital to understand the challenges yam farmers face for proper intervention strategies, especially through breeding for resistance and promotion of the crop. Therefore, there is an urgent need for genotyping yam germplasms available on farmers' fields to conserve sources of elite genotypes and plan to establish national breeding programs.

Morphological descriptors have been resourceful in the characterization and identification of yam, especially where biochemical and molecular tools are limited. The IPGRI-IITA (1997) manual for yam description is widely used successfully to preliminary phenotype yam using agro-morphological traits (Mahalakshmi et al. 2007). Some morpho-traits are well known to distinctly characterize and identify *Dioscorea* species (Girma et al. 2018, Massawe and Temu 2022). Similarly, microsatellites are useful for molecular characterization and genetic diversity studies since they are cheaper than advanced techniques such as Next Generation Sequencing (NGS) and Diversity Array Technology (DArT) markers.

Microsatellites are also good enough (powerful) to capture genetic diversity information and provide a highlight of genetic resources available in any germplasm. Other researchers also adopted SSR markers to assess the genetic diversity of yam from different geographical regions (Obidiegwu et al. 2009, Siqueira et al. 2012, Muthamia et al. 2013, Arnau et al. 2017). This study aimed to assess the morphological and genetic diversity of water yam (*D. alata*) and the general production constraints of yam in Pwani and Unguja island. Information on

the variabilities available in the locally grown water yam cultivars and the main production challenges are crucial for further yam improvement in the country via breeding.

Materials and Methods

Sample collection for morphological characterization and DNA work

Water yam samples were collected in Pwani region (Mkuranga, Kibiti and Rufiji districts) and Unguja Island (South, North and West districts) [Figure 1]. Location coordinates were measured using GPS (Garmin etrex 20, USA) and recorded in the datasheet. The sampling map was drawn using ArcGIS 10.3 software (Johnston et al. 2001). Random sampling was employed based on the availability of yams in the field, whereby preliminary identification of water yams was made with assistance from both farmers and a botanist. Nine tubers of *D. alata* local cultivars considered popular to farmers were collected for morphological characterization (Table 1).

For genetic characterization, five young leaves of each water yam genotype (including the nine local cultivars mentioned above) observed in 46 fields surveyed were collected from a single plant. The total samples of water yam genotypes collected were 128. Each leaf and tuber sample was packed in a dry-labelled envelope. The leaf samples were transported to Tanzania Agricultural Research Institute (TARI)-Mikocheni laboratory and stored at -20 °C before DNA extraction, while the tubers were sent to TARI-Kibaha, Pwani for planting. The tubers were kept in a semi-dark room to break the dormancy as we waited for the next planting season. The following season, large yam tubers were sliced to a miniset of about 50 gm and were planted in an experimental field at a spacing of 1 m x 1 m in a randomized complete block design. All tuber minisetts were planted on ridges with three replications. Standard cultural agronomic practices were employed, such as adding cow dung manure before planting, hand weeding and stacking. All plants were manually irrigated using a field sprinkler. Irrigation was done thrice a week using a field sprinkler for the first three months, and after that, the plants were left to grow to maturity.

Table 1: Description of ninepopular water yam local cultivars collected and used for morphological studies.

Sn	Local name	Experimental name	Popular location	Phenotypic characters
1.	Cheupe /Mwendachi	Cheupe A	Kibiti, Mkuranga, Rufiji, Central-Unguja	Long tubers with white flesh
2.	Cheupe/Maganja	Cheupe B	Kibiti	Cream flesh
3.	Bungara	Bungara A	Mkuranga	Purple white flesh
4.	Bungara/Bungala/ Bokoboko	Bungara B	Mkuranga, Kibiti	Purple flesh tubers with rice aroma
5.	Mgenda	Mgenda	Kibiti, Mkuranga	Cream flesh
6.	Kinana	Kinana	Kibiti, Mkuranga	Small tuber with hairy skin, white flesh
7.	Vigenda	Vigenda	Kibiti	White flesh
8.	Cheupe/Mkandab we	Cheupe C	Kibiti, Mkuranga	White flesh
9.	Bungala/Mwekund uganda	Bungara C	Unguja	Red peels/purple flesh

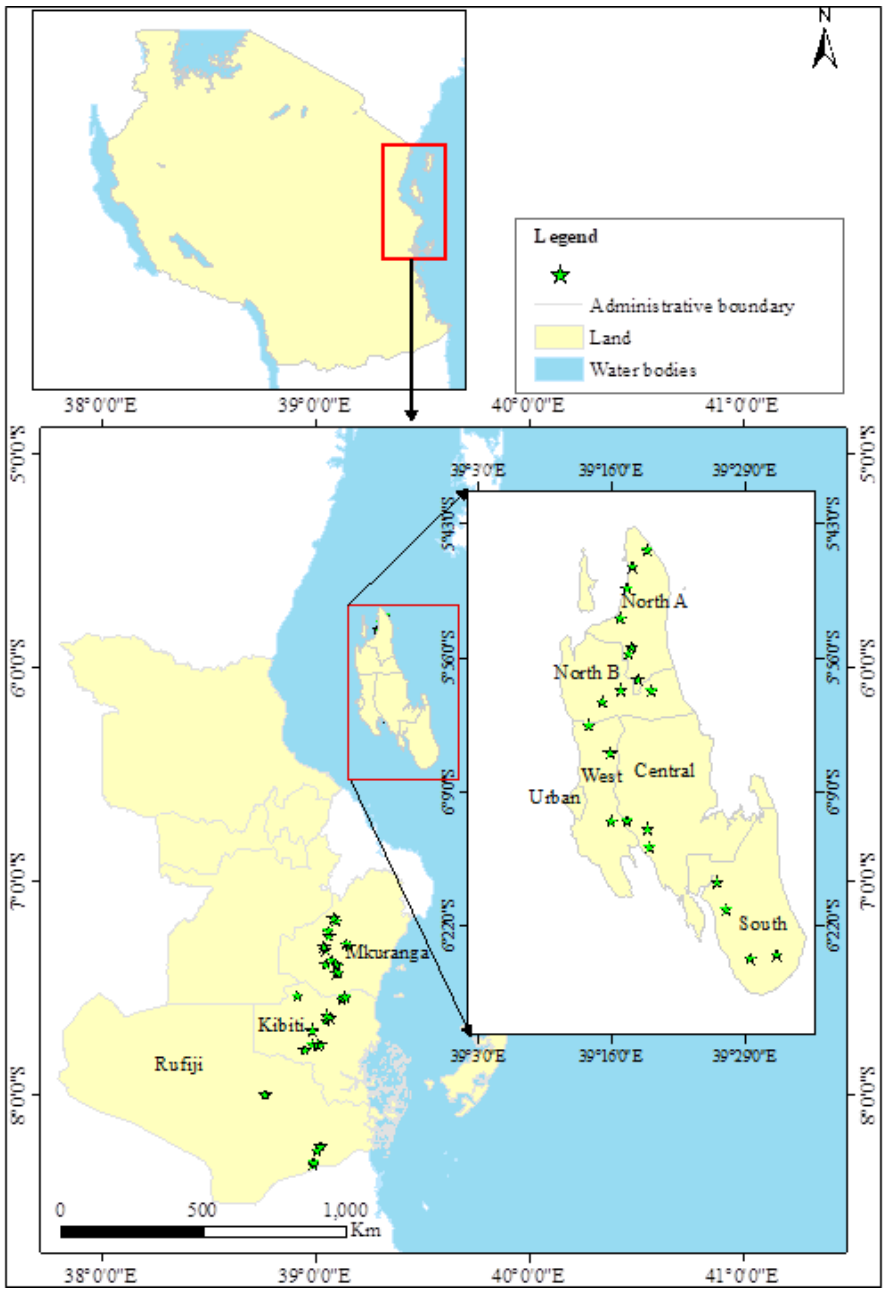

Figure 1: Map of Tanzania showing the study sites where water yam samples werecollected.

Morphological characterization

Morphological characterization of the nine water yam local cultivars was done by measuring 26 agro-morphological characters (Table 2) described in the International Plant Genetic Resources Institute (IPGRI/ IITA, 1997) manual. Data on leaf and stem characters were taken four months after planting (MAP) and tubers nine MAP. During characterization, the leaves and stems of each yam cultivar were taken and displayed on white paper. Then the measurement of quantitative characters such as leaf length, stem diameter and petiole

length was done by using a ruler. Qualitative characters included stem colour, presence or absence of wings and spines, twinning direction, stem cross-section shape at the base, the position of leaves, leaf colour, vein colour, margin colour, leaf shape and petiole colour and were visually scored and recorded according to the manual scale. Yam tubers were uprooted nine months after planting, cleaned with tape water to remove the adhering soil, and displayed on the clean white papers. Qualitative characters such as tuber shape, outer and inner skin colour, flesh colour, and tuber relationship were visually scored according to the same IPGR manual scale. Tuber lengths were measured using a ruler, while tuber weights were measured using a weighing balance (GOLDEN GLOBE®, UK). The average measurements of at least three tubers were taken for analysis. Two seasons of planting and morphological data collection were done in 2016/2017 and 2017/2018, respectively and the data were averaged.

Table 2: The twenty six yam morphological traits used for morphological characterization in this study (IPGRI/ IITA, 1997).

S/n	Trait	Classes (codes)
		Stem descriptors
1	Stem colour	1- Green; 2- Purplish green; 3- Brownish green; 4. Purple
2	Wings	1- Present; 2- Absent
3	Spines	1- Present; 2- Absent
4	Twining direction	1- Clockwise; 2- Anticlockwise
5	Stem diameter	1- < 0.4 cm; 2- 0.4-0.6 cm; 3- > 0.6 cm
6	shape at base	1- Octagonal; 2- Round
		Leaves descriptors
7	Position of leaves	1- Alternate; 2- Opposite
8	Leaf colour	1 – yellowish; 2 – pale green; 3 – dark green; 4 – purplish green; 5 – purple
9	Leaf vein colour (upper surface)	1 – yellowish; 2 – green; 3 – pale purple; 4 – purple
10	Leaf vein colour (lower surface)	1 – yellowish; 2 – green; 3 – pale purple; 4 – purple
11	Leaf margin colour	1 – green; 2 – purple
12	Leaf shape	1- Cordate; 2- Sagittate
13	Number of leaflets	1- One; 2- Three
14	Petiole length	1- Short (5 cm); 2- Medium (5-10 cm); 3- Long (> 10 cm)
15	Petiole colour	1- Green; 2- Brownish green; 3- Purple
		Tuber descriptors
16	Underground tubers	1- Present; 2- Absent
17	Tuber shape	1- Elongate; 2- Irregular (not uniform), 3- oval
18	Tuber outer skin colour	1- Brown; 2- Yellow, 3-purple
19	Flesh colour	1- White; 2- Yellow; 3- Purple;4- Purple with white; 5-

		White with purple
20	Inner skin colour	1- white; 2- cream; 3 Purple; 4-Deep purple; 4- White
21	Number of tuber	1-One; 2-2-5 few, 3- Several Above 5;
22	Tuber size	1- Small; 2-Intermediate; 3-Large
23	Relationship	1- Complete separate and distance; 2-Complete separate but close together; 3- Fuse at neck
24	Sprout at harvest	1- Yes; 2-No
25	Tuber weight	3 plants per assession (kg)
26	Tuber Length	1-Short (< 20cm); 2-Medium(< 40cm); 3 Long (>41cm)

Molecular characterization

DNA extraction of leaf samples of 128 *D. alata* genotypes was performed according to Sharma et al. (2008)with slight modifications. DNA extraction buffer was slightly changed by increasing the concentration of NaCl from 2.0 M to 2.8M while the concentration of other components was maintained, as described by Sharma et al. (2008). At least three fresh leaves (about 300 mg) per genotype were used for DNA extraction.The integrity of all DNA samples was analysed on 1% agarose gel stained with ethidium bromide. The concentration and purity of all DNA samples were determined using a Thermo scientific NANODROP 2000 Spectrophotometer (Cecil Instruments, Cambridge, UK) at A260 nm and A280 nm. DNA samples were normalised to 25 ng/µl working concentration.

Initially, 32 SSR markers were screened for polymorphism using at least 50 yam samples. Out of these, only 10 polymorphic SSR markers (Table 3) were used to assess the genetic diversity of 128 yam samples. PCR was carried out on Gene Amp PCR System 9700 (Applied Biosystems, USA). PCR reaction mix included 25 ng/µl of DNA, 12.5 µl of one taq 2X master mix with standard buffer (NEB inc®) and 0.5 µl of each forward and reverse primer. PCR conditions included an initial denaturation of 3 min at 95 °C, followed by 35 cycles of 30 seconds at 95 °C, 1 min at the annealing temperature defined for each primer pair used (Table 1) and 1 min at 72 °C. The final extension was 7 min at 72 °C and the final hold at 4 °C.

The PCR products were scored on 2% agarose gel electrophoresis stained with ethidium bromide in 1X TAE buffer run at 100 V for 1 hour and visualised on trans-UV light and photographed in UVPDigDoc-IT imaging system (Upland, CA, USA). Fragment size scoring was done manually by comparing the size with a reference standard of 100 bp DNA ladder (AXYGEN Biosciences), which ranges from 100 bp to 3000 bp. PCR fragments on the gel were scored for presence (1) or absence (0).

Table 3: Description of SSR markers used to assess the genetic diversity in this study.

Sn	Name	Forward	Reverse	Tm	Bp	Reference
1	Dab2D06	AACATATAAAGAGAGATCA	ATAACCCTTAACTCCA	51	165-176	Tostain et al. 2007
2	Da1F08	AATGCTTCGTAATCCAAC	CTATAAGGAATTGGTGCC	51	166-176	Tostain et al. 2007
3	Dpr3B12	CATCAATCTTTCTCTGCTT	CCATCACACAAATCCATC	56	127-170	Tostain et al. 2007

4	Dab2EO7	TTCCCTAATTGTTCCTCTTGTTG	GTCCTCGTTTTCCCTCTGTGT	51	104-183	Tostain et al. 2007
5	Dpr3D06	ATAGGAAGGCAATCAGG	ACCATCGTCTTACC	51	125-170	Tostain et al. 2007
6	Dpr3F04	CCCATGCTTGTAGTTGT	TGCTCACCTCTTTACTTG	51	81-131	Tostain et al. 2007
7	Dab2CO5	TCCCTCCCCATAGAAACAAAGT	TCAAGCAAGAGAAGGTG	56	178-193	Tostainet al. 2007
8	YM30	GGTCCTCTTCTATCCCAACAA	CACGTATTAACTCCATCTATCCAA	55	200-390	Otooet al. 2009
9	Da1A01	AACTATAATCGGCCAGAGG	TGTTGGAAGCATAGAGAATT	55	212-225	Otoo et al. 2009
10	Da1C12	GCCTTTGTGCGTATCTGA	AATCGGCTACACTCATCTC	55	140-160	Otoo et al. 2009

Collection of demographic and water yam production constraints data

A field survey was conducted between June 2017 and October 2018. The study population was 134 smallholder farmers growing yam in Pwani region (117) and Unguja Island (17). The study areas were chosen based on the informal records of yam production. The purposive sampling method was used to select villages and households in the study areas where the number of respondents depended on the number of available yam fields. Only one member of the household was interviewed. Primary information of respondents, including gender, age and education level, was recorded. A structured questionnaire was used to capture information on yam production status, disease and management, source of planting materials and major yam production constraints in the study areas. Data was collected using a semi-structured questionnaire comprising closed and open-ended questions.

Data Analysis

The unweighted pair-group method with arithmetic means (UPGMA) clusters based on morphological and molecular data were generated using Past 3.14 version software(Hammer et al. 2001) using classical algorithm dissimilarity matrix at Jaccard similarity index. Principal component analysis (PCA) was performed based on Past 3.14 software, and then the four first axes (with positive Eigenvalue) were recorded based on the dissimilarity index. A two-dimension scatter plot was plotted for the 128 collected samples using scores of the first two principal components. The polymorphism information content (PIC) for each marker was calculated according to Peakall and Smouse (2012). The Statistical Package for Social Scientists (SPSS, Version 24) was used to analyze the social-economic data.

Results

Morphological variations among water yams

The results showed that 73 % (19/26) of the evaluated characters were polymorphic, while 27 % (7/26) were monomorphic. Yam leaf colour varied from pale green to dark green. The leaf vein, petiole, and margin were green or purple. The nine water yam cultivars had the same cordate leaf shape, anticlockwise twinning direction, presence of underground tubers, octagonal stem and presence of wings. The tubers had various shapes. Of the 9 cultivars evaluated, 4 had

elongated, 3 had irregular, and 2 had oval shape (Figure 2). Tuber's exterior colour was brown (6) or purple (3). The tuber's inner skin was white (5) and purple (4). The flesh colour of a central cross-section of tubers was cream (1), purple white (2), white purple (2), white (3) and purple (1), as shown in Figure 2. Tubers of the evaluated yam varieties had different sizes and weights ranging from 0.3 kg to 4 kg.

Cluster analysis using morphological descriptors classified the nine water yam local cultivars into two main clusters (I and II) with three subclusters: A, B and C (Figure 3). Cluster I had seven cultivars, and Cluster II had two cultivars. Subcluster A had five cultivars which had either cream or white flesh: two cultivars, *Vigenda* and *Kinana*, from the Kibiti district and *Mgenda*, *Cheupe* A and *Cheupe* B, collected from the Mkuranga district. Subcluster B had one purple-white and one white cultivar: *Bungara* A and *Cheupe* C, respectively, both from the Mkuranga district. Subcluster C had purple flesh cultivars: *Bungara* B from Mkuranga and *Bungara* C from Kibiti.

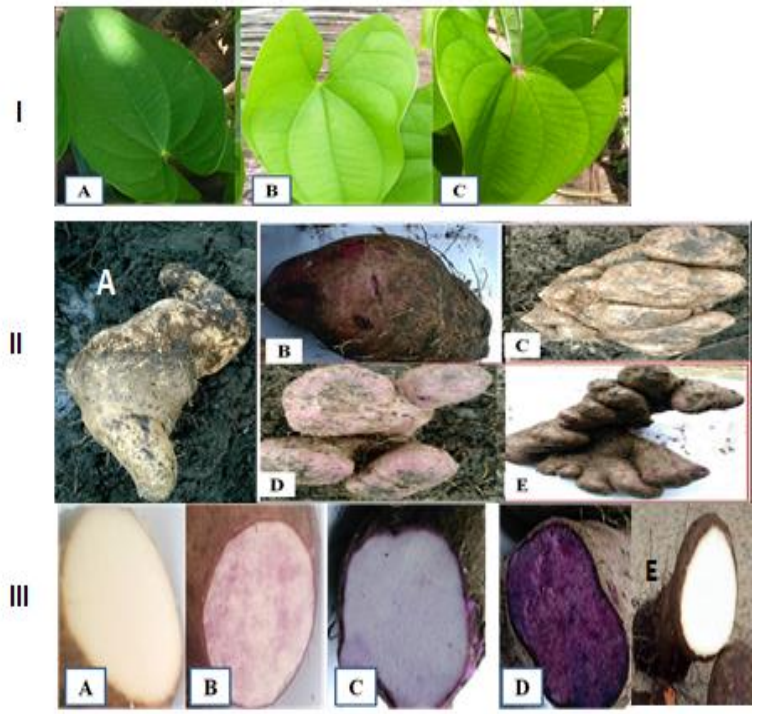

Figure 2: Variations of leaf and vein colour (I), tuber shapes (II) and flesh colour (III) amongthewater yam cultivars evaluated in this study. IA=dark green leaf; IB: Pale green without purple vein colour; IC: pale green with purple vein; IIA-IIE are various tuber shapes; IIIA:cream,IIIB: purple white, IIIC: white purple, IIID: purple and IIIE: white flesh colour.

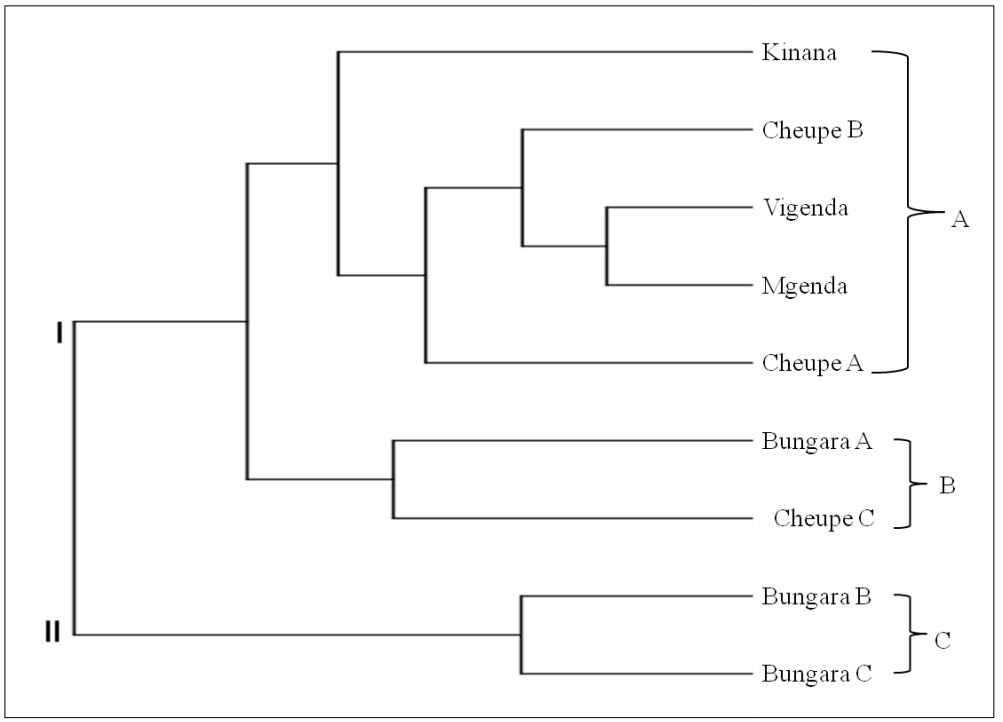

Figure 3: UPGMA cluster analysis of nine water yam local cultivars based on morphological characters. I and II are the major clusters, while A, B and C are subclusters.

Genetic diversity of the128 water yam genotypes

Ten SSR markers used in this study were highly informative, with a mean of 0.56 polymorphic information content (PIC) ranging from 0.3 to 0.9 (Table 3). YM30 was the most informative marker (PIC = 0.9), followed by Dab2C05 and Da1A01 (PIC =0.8), while Da1F08 and Dpr3D06 were the least informative markers (PIC =0.3). The SSR markers used in this study revealed a total of 40 alleles. YM30 recorded the highest number of alleles (7), while Dpr3D06 and Dpr3F04 recorded only two (Table 4 and Figure 4).

Table 4: Genetic diversity parameters of ten SSR markers used in this study.

No.	Primer name	Bp	No. of allele	Allele Frequency	PIC
1	Dab2D06	100-300	6	0.5	0.7
2	Da1F08	100-200	4	0.5	0.3
3	Dpr3B12	200-300	3	0.4	0.5
4	Dab2E07	100-300	3	0.5	0.5
5	Dpr3D06	100-200	2	0.7	0.3
6	Dpr3F04	100	2	0.7	0.5
7	Dab2C05	100-300	5	0.8	0.8
8	YM30	100-300	7	0.5	0.9
9	Da1A01	200-300	5	0.4	0.8
10	Da1C12	100-200	3	0.5	0.6
Mean			**4**	**0.55**	**0.56**

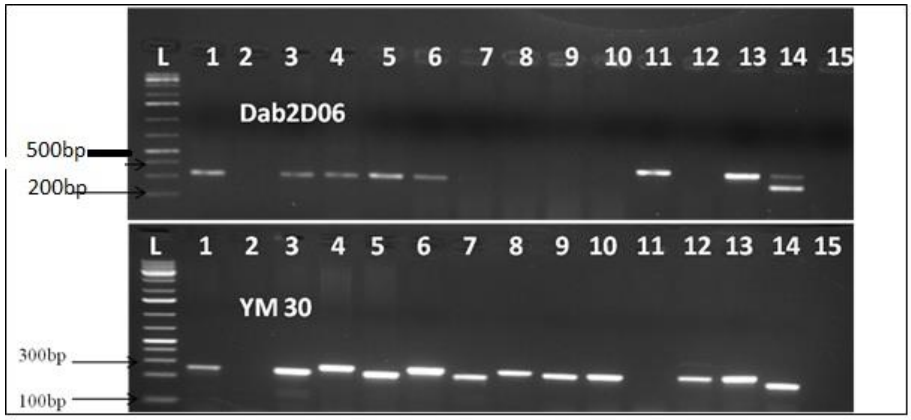

Figure 4: Representative picture of agarose gel electrophoresis used to score for thepresence/absence of fragments. Dab2D06 and YM30 are SSR markers, L is a 100bpDNA ladder, and the numbers represent yam samples.

Cluster analysis

Based on ten relatively high polymorphic SSR markers, the 128 water yam genotypes clustered into two major clusters (I and II) at 0.2 dissimilarity index. Five subclusters, A, B, C, D and E, were formed (Figure 5). Cluster I had two subclusters with 21 genotypes from Pwani and Unguja Island. Cluster II had three subclusters with 67 genotypes from Pwani and Unguja Island. Subcluster C had 7 genotypes, all from Unguja Island. Some genotypes collected in one district, e.g. MK4 and MK8 (from Mkuranga), KB4, KB5 and KB6 (from Kibiti) and UNG43 & UNG52 (from Unguja) clustered as duplicates. Other duplicates were from different study areas, e.g. RF8, KB40, UNG11 and UNG19 (from Rufiji, Kibiti and Unguja, respectively) and RF14 and UNG44 (from Rufiji and Unguja, respectively).

Principal component analysis (PCA) indicated that the first two principal components accounted for 27.59% of the total variation, with PCA1 and PCA2 contributing 13.42% and 12.07%, respectively. Clusters and duplicates observed in the dendrogram agreed with those observed in the PCA scatter plot,

especially for subclusters C, D and E (Figure 6).

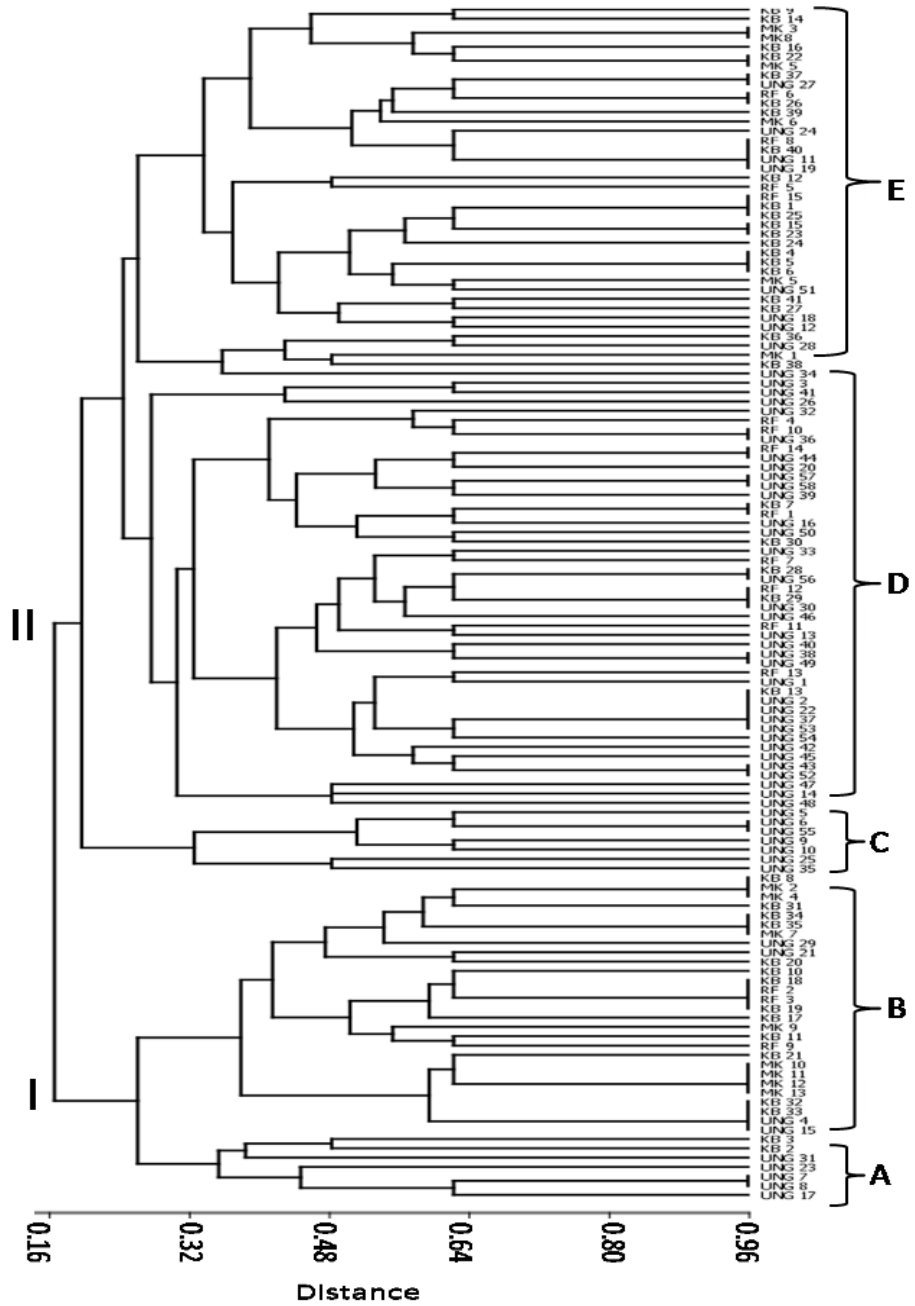

Figure 5: Cluster analysis of 128 water yam genotypes based on UPGMA method at Jaccard'ssimilarity index. I and II represent major clusters while A to E represent subclusters.

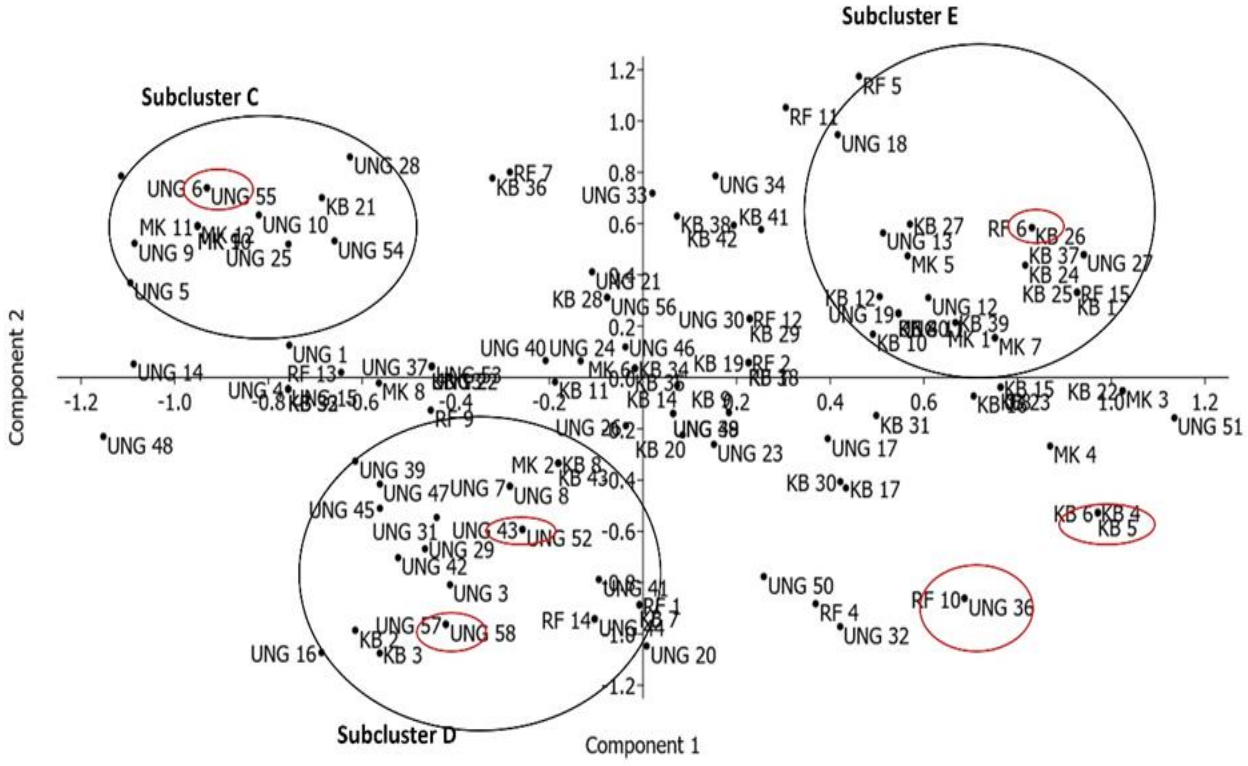

Figure 6: A two dimensional principal component analysis (PCA) analysis of 128 water yamgenotypes from Pwani and Unguja Island.Redcycles represent duplicates.

Demographic and production constraints

Female respondents were 58%, and males 42%. More than half of the respondents (55.2%) were aged between 46 and 61 years, with the majority (69.4%) having primary education. The major ranked constraints to yam production were drought (29.1%) and lack of planting materials (23.13%). Next to the two major constraints were insects, lack of planting materials and drought (9.7%), poor market and low productivity (5.22) and poor market and planting materials (5.22). Other constraints scored below 5% (Table 5). Most farmers (98%) were unaware of any disease or pest that attacks yam, while only 2% mentioned root rot and anthracnose diseases. Farmers also ignore the poor quality of tubers attacked by root rot disease and reuse the same materials due to scarcity of planting materials. Due to the lack of improved varieties and planting materials, 80.6% of the respondents use the same materials inherited from their great-grandparents, 15.6% obtain seed yam (planting materials) from their neighbours, while 3.7% source it from nearby villages (Table 6).

Table 5: Main constraints to yam production in Pwani and Unguja Island.

Constraint	Frequency	Percent
Land scarcity	2	1.49
Wild animals	3	2.23
Unavailability of planting materials	31	23.13
Poor Market	6	4.47
Insects	2	1.49
Drought	39	29.10
Weeds	4	2.98
Weeds and poor market	5	3.73
Insects, planting materials and drought	13	9.70
Land scarcity and poor market	6	4.47
Insect and unavailability of planting materials	1	0.74
Weeds and inadequate rainfall	4	2.98
Poor market and low productivity	7	5.22
Poor market and planting materials	7	5.22
None	4	2.98
Total	**134**	**99.93**

Table 6: Sources of seedyams (planting materials) in Pwani and Unguja Island.

Source	Frequency	Percentage
Inherit from parents	108	80.60
From neighbours	21	15.67
Nearby village	5	3.73
Total	**134**	**100**

Discussion

Yam being cultivated informally for decades in Tanzania, understanding its morphological and genetic diversity; and production constraints may enhance its promotion for better productivity. The analysis of morphological descriptors suggests that the tuber flesh and leaf colour were the main descriptors for clustering. Our results also indicated that the dominant yam flesh colours were cream, white and purple. Most cultivars with purple flesh and purple inner skin colour suggest they are closely related as they clustered together. From all morphological descriptors evaluated in this study, no descriptor characterized the local cultivars according to the district or location from which they were collected, a similar result to Massawe and Temu (2022). The most distinguishing morphological features observed in this study were the presence of wings, leaf shape and vein colour, tuber shape, presence of bulbils and inner flesh colour. Most of these distinguishing features were also reported (Jyothy et al. 2017, Massawe and Temu 2022), and they can be used to differentiate *D. alata* from other *Dioscorea* spp. where genetic tools are limited. The cultivars with white flesh were named interchangeably between the farmers within and across the study areas. For instance, some farmers call all white cultivars 'Bungara'/'Bungala' while others claim the name is for purple varieties only. However, morphological traits clustered most white-fleshed cultivars away from purple except for Bungara A, which clustered with Cheupe C.

Ten SSR markers used in this study and the detected 40 alleles are enough to estimate the genetic diversity within the population (Tostain et al. 2007, Otoo et al. 2015). YM30, Dab2D06, Dab2C05 and Da1A01 were the markers that gave a higher number of alleles in this study, similar to Muthamia et al. (2013) and Obidiegwu et al. (2009). Dpr3D06 and Dpr3F04 markers gave the lowest number of alleles (2), suggesting their monomorphism, a similar observation to Massawe and Temu (2023). Our results differ from Obidiegwu et al. (2009), who also used the same SSR markers but reported more alleles (9 for Dpr3D06 and 6 for Dpr3F04).The differences in allele number reported in our study might be due to variations within the yam genotypes used in our study or the SSR scoring method of presence/absence of a band.

The PIC values of 0.5 to 0.9 represent more diversity, while the PIC less than 0.5 represent narrow genetic diversity (Peakall and Smouse 2012). The mean PIC value of 0.56 recorded in this study revealed relatively high genetic diversity of yam in the study areas. High genetic diversity might be attributed to the nature of yam being vegetatively propagated, which usually maintains a high level of heterozygosity (Siqueira et al. 2012). It might also be due to spontaneous hybridization attributed to the common traditional practice of farmers selecting planting materials for plant improvement (Obidiegwu et al. 2009).Massawe and Temu (2023) reported almost a similar genetic diversity values (0.53) within water yam samples collected elsewhere in the country.

Cluster analysis based on SSR markers largely suggests that farmers from the study areas grow similar *D. Alata* genotypes. However, subcluster C had only genotypes from Unguja, indicating the limitations of sharing planting materials between the Tanzania mainland and Unguja Island. The cluster analysis also showed some duplicates regardless of different local names given and different sites of collection. For instance, the similarity between RF8, KB40, UNG11 and UNG19 in cluster II, subcluster E. Presence of duplicates might be attributed to the same yam genotype having different names in different communities due to the history of genotype or informal nature of exchange materials among farmers (Tostain et al. 2007, Otoo et al. 2009).

Generally, the PCA plot demonstrated trends similar to the cluster analysis revealed in the dendrogram. They both agreed with each other to a greater extent, especially for

subclusters C, D, and E and the observed duplicates. However, the PCA plot displayed a different scattering of subclusters A and B genotypes. This observation might be attributed to the total variation contributed by component 1 and component 2, or the genotypes are distantly related hence the possibility of more subgroups among the genotypes in subclusters A and B.

This study revealed that the main yam production constraints were drought and lack of planting materials (seed yam), followed by poor markets. Due to climate changes, rain patterns are irregular and sometimes not enough annually. The mean annual rainfall in Unguja is about 900 mm and about 1200 mm in Pwani (Mzava et al. 2020, Hamad et al. 2023). Genetic characterization of yam germplasm for drought-tolerant traits that can be included in the yam breeding program to release drought-tolerant varieties would reduce the severity of drought constraint. A lack of improved varieties accompanies the unavailability of planting materials since farmers have no choices; they plant what they have inherited from their great-grandparents. The scarcity of planting materials has also been reported (Idumah and Owombo 2019, Mignouna et al. 2023). The nature of the crop aggravates the problem of quality seed yam since the edible part is also the same source of planting material. Very little research has been done on yam in Tanzania; hence technologies like minisetts for seed yam production are unknown to most farmers. Other countries, such as Nigeria and Ghana, have overcome the problem of planting materials through advanced techniques like minisetts technology and plant tissue culture, which turns to rapid mass multiplication of planting materials (Aighewi et al. 2015).

Other constraints include poor market, weeds, insects and pests, wild animals and low productivity. Poor or unreliable yam market is due to informal production systems managed outside the country's formal market and economic channels. The Holy Month of Ramadhan is the only season of very high demand for yam when farmers are sure of selling their yam produce, especially in Unguja. Otherwise, farmers rely on local markets to sell fresh, boiled or roasted tubers beside the road and around primary school premises.

Cultural operations and weeding are very demanding and provoked by the old age of most yam farmers in the study area. Further, due to the nature of this crop, weeding becomes more difficult since most farmers do not practice stacking. The creeping nature of yam makes the task harder. Fasusi et al. (2022) and Mulualem et al. (2022) also reported high labour charges in yam production as among the major constraints to production in Nigeria and Ethiopia, respectively. Insects and pests constraints were ranked low, probably because farmers do not know if they affect yield. Some varieties, especially the aromatic ones, are highly favoured by wild animals such as warthogs and thus disliked by some farmers.

Farmers awareness on yam dieases in the study areas was shallow, and most farmers do not practice any disease management. Only 2% mentioned anthracnose and root rot diseases. Anthracnose disease of yam is a fungal disease caused by *Colletotrichum gloeosporioides* and considerably impacts yam production worldwide (Aduramigba-Modupe et al. 2012). Our finding is different from, for instance, Udemezue and ELC (2017), where farmers in Anambra, Nigeria, had good knowledge of pests and several diseases affecting yam.

Except for Unguja Island, where introduced varieties from Nigeria were encountered at Zanzibar Agricultural Research Institute (ZARI) Kizimbani, all yam cultivars grown by farmers were indigenous. Among the *D. alata* local cultivars, Bungara was richly available in many fields. This variety is favoured by its good agronomic traits disclosed by farmers as tolerance to drought and fewer management requirements. At the same time, this variety is widely available, hence relatively easy to obtain planting materials. During the field survey, most white yam varieties' local names were interchanged across different communities/households. Lack of research, promotion and release of new or improved varieties may have contributed to the low

distinction of the local varieties. The relatively high genetic diversity observed among the *D. alata* genotypes may be used to improve the yam germplasm in the country through breeding.

Conclusion

Relative high genetic diversity observed within the water yam genotypes used in this study may be used to select cultivars with novel or good agronomic traits and use them to improve the genetic pool of yam germplasm in the study areas and beyond. The main yam production challenges in the study areas and probably in the country are drought, unavailability of planting materials and poor market. It is high time now for root and tuber crop programs to include yam in the national breeding and conservation programs. Doing so will help address the challenges yam farmers face in the country by releasing improved varieties and promoting the crop for more production. The release of improved varieties tolerant to drought and the use of advanced technologies for seed yam will improve the current underpinning constraints.Therefore, we recommend establishing yam breeding programs coupled with extension officers and farmers training on modern yam's production and management systems. As global climate changes challenge and threaten various crop production, research on orphan crops such as yam must be strengthened.

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