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Full Length Article

Agro-Morphological Characterization of Cassava (*Manihot esculenta*) Landraces in Vietnam

Dam Thi Thu Ha[1], Le Thi Thu Trang[1], Hoang Thi Nga[1], La Hoang Nhat Minh[3], La Tuan Nghia[1], Hoang Thi Hue[1], Tran Dang Khanh[2] and Phuong Diep Vien Ta[1*]

[1]*Plant Resources Center, An Khanh, Hoai Duc, Hanoi, Vietnam*
[2]*Agricultural Genetics Institute, Hanoi, Vietnam*
[3]*University of Science and Technology of Hanoi, Hanoi, Vietnam*
[*]For correspondence: phuongvien28@gmail.com
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Abstract

Cassava (*Manihot esculenta* Crantz) is an important source of dietary fiber, vitamins and minerals. This study represents the first comprehensive evaluation of a large collection of local cassava accessions from all eight agro-ecological zones in Vietnam. The analysis revealed that local cassava accessions of Vietnam had morphological diversity and there was a significant difference among the studied accessions with genetic distances ranging from 1 to 15.52. A majority of local cassava accessions in Vietnam exhibited the following morphological characteristics: straight growing habit of young stems (83%), cylindrical plant shape (56%), silver-green stem exterior color (56.5%), no branching (55.5%), prominent foliar scars (83.5%), serrate stipule margins (57%), seven lobes leaves (85%), oblanceolate central leaflet (53%), light green young leaves (67%), light green leaf vein (60.5%), purple petioles (55%) and 75–90 degrees angle of petiole insertion (75.5%). This research will contribute to the development of a systematic database for local cassava germplasm in Vietnam, aiding in the preservation and conservation of this genetic resource for the future. Evaluating the agro-morphological characters and yield of local cassava accessions will provide breeders baseline data for selection, breeding, and evaluation, and facilitate the management of the valuable and diverse germplasm currently available in Vietnam.

Keywords: Agronomical; Cassava accessions; Morphological; Variability; Dendrogram

Introduction

Cassava (*Manihot esculenta* Crantz) is one of the most versatile crops, well-adapted to various terrains, particularly in arid regions, highlands, and sloping areas in Africa and Asia (Ceballos *et al*. 2004; Tumuhimbise *et al*. 2014; Daemo *et al.* 2023). Cassava is used as food, livestock feed, and increased income for farmers in challenging and border regions (Howeler 2014; Amelework and Bairu 2022). In Southeast Asian countries, especially Thailand, Cambodia, and Vietnam, cassava is primarily grown for its starch. Cassava is renowned for being cultivated with relative ease. Over the past 50 years, extensive research has focused on agronomic characteristics, cultivation practices, and the development of high-yielding, high-quality cassava varieties tailored to various ecological zones, with the aim of achieving optimal yields and promoting sustainable development (Malik *et al*. 2020; Peuo *et al*. 2021; Yuanjit *et al*. 2023).

Preliminary research suggested that the local cassava germplasm in Vietnam exhibited significant morphological diversity. However, local cassava varieties were progressively being supplanted by high-yielding improved varieties, a trend witnessed not only in Vietnam but also globally. In the absence of conservation efforts, there is a risk that the genetic diversity of cassava landraces could erode over time, driven by increasing population pressures and climate change (Lamprecht 2015).

Over the years (from 1992 to the present), the Plant Resources Center – the leading authority in plant genetic resource conservation – has collected cassava accessions from 33 provinces across the three regions including the North, Central, and South of Vietnam. However, these cassava germplasm have not yet been comprehensively and systematically evaluated (Ha *et al.* 2015; Ocampo *et al.* 2021).

Assessing the agronomic and yield characteristics of local cassava varieties is essential for providing breeders with a foundational basis for selection, hybridization, and varietal evaluation. This approach enables the effective

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management of valuable and diverse genetic resources available in Vietnam and informs the development of suitable cassava breeding methodologies. This is also the primary goal of the research team to assess and disseminate data on the morphological characteristics and yield of 200 local cassava varieties in Vietnam. It contributes to the breeding programs, exploitation, and sustainable development of local cassava genetic resources in the future.

Materials and Methods

Materials

The study was conducted from February 2022 to December 2022 at the experimental field, under controlled irrigation, located at the Plant Resources Center, An Khanh, Hoai Duc, Hanoi, Vietnam (21°00' N; 105°43' E). 200 cassava accessions were collected from 39 out of 64 provinces across the three regions including Northern, Central, and Southern Vietnam, representing all eight agro-ecological zones in Vietnam. These accessions have been currently preserved in the Field Genebank at the Plant Resources Center, Hanoi, Vietnam (Table 1). This research included 46% cassava accessions from 15 provinces in Northern Vietnam, representing three ecological zones, *viz.* the Northwest, Northeast, and Red River Delta. The North Central, South Central, and Central Highlands regions accounted for 29% of the total accessions studied, while the Southeast and Southwest regions made up 25%.

Methods

The experiment was arranged in blocks of non-repeating sequence, each accession being one block. Plot size was 6 m x 5 m (10,000 plants ha^{-1}). Plant spacing was 1 m × 1 m, both within and between rows. Fertilization as organic manure or organic microbial fertilizer, and a total of 80 kg ha^{-1} N, 60 kg ha^{-1} P_2O_5, and 80 kg ha^{-1} K_2O were applied. Fertilization was split into three applications. Basal fertilization with all the organic manure or organic microbial fertilizer and one-third of N, one-third of P_2O_5, and all the K_2O were applied. The first top-dressing was conducted with one-third of N and one-third of P_2O_5 in tandem with weeding and hilling at 40–50 days after planting. The second top-dressing with the remaining of N and P_2O_5 were applied combined with weeding and hilling at 80–90 days after planting.

Accessions were characterized by recording morphological characters (descriptors) based on the Characterization and Preliminary Evaluation Form for Cassava of the Plant Resources Center, the guidelines of IPRGI and the list of selected morphological characters by Fukuda *et al*. (2010). The observed agro-morphological traits are detailed in Table 2.

The data, including means, maximums, minimums, Coefficient of Variation (CV %), and Standard Deviation (SD), were processed using Excel 2013 software. Morphological trait data were analyzed using NTSYSpc version 2.1 software (Rohlf 2000). The data were used to construct distance matrices from the Euclidean distances:

$$E_{i,j} = \sqrt{\sum_k (x_{ki} - x_{kj})^2}$$

Where E_{ij} is the Euclidean distance between two individuals i and j, X_{ki} is the value of the k^{th} trait for individual i, and where X_{kj} is the value of the k^{th} trait for individual j. The similarity matrix was used for cluster analysis based on the UPGMA (Unweighted Pair Group Method with Arithmetic Mean).

Table 1: List of origin of cassava landrace accessions used in this study

Agro-ecological zone	Origin	No. of accessions	Existence (%)
Northwest	Dien Bien	6	17.5
	Hoa Binh	9	
	Lai Chau	10	
	Son La	10	
Northeast	Bac Giang	5	24
	Cao Bang	8	
	Ha Giang	3	
	Lang Son	4	
	Lao Cai	3	
	Phu Tho	2	
	Quang Ninh	12	
	Thai Nguyen	3	
	Tuyen Quang	1	
	Yen Bai	7	
Red River Delta	Ha Noi	7	4.5
	Thai Binh	1	
	Ninh Binh	1	
North Central Coast	Ha Tinh	2	14
	Nghe An	7	
	Quang Binh	1	
	Quang Tri	8	
	Thanh Hoa	6	
	Thua Thien Hue	4	
South Central Coast	Binh Dinh	7	12
	Da Nang	1	
	Phu Yen	2	
	Quang Nam	6	
	Quang Ngai	8	
Central Highland	Dak Nong	1	3
	Gia Lai	4	
	Lam Dong	1	
Southeast	Binh Thuan	1	19.5
	Dong Nai	27	
	Binh Duong	3	
	Binh Phuoc	1	
	Tay Ninh	7	
Mekong River Delta	Ben Tre	2	5.5
	Ca Mau	1	
	Kien Giang	2	
	Long An	1	
	Tien Giang	5	
	Total	200	100

Results

Stem morphological characteristics of cassava accessions

The statistical results on the morphological traits of the cassava stem varieties indicate that local cassava germplasm

Table 2: List of morphological descriptors for characterization of cassava used in this study

No.	Traits observed	Variants	Period
		Leaf morphology	
1	Color of unexpanded apical leaves	3.Light green 5. Dark green 7. Green purple 9. Purple	6–7 months after planting
2	Color of first fully expanded leaf	3. Light green 5. Dark green 7. Green purple 9. Purple	6–7 months after planting
3	Pubescence of young leaves	1. Absent 3. Little pubescence 5. Moderate pubescence 7. High pubescence	6–7 months after planting
4	Number of leaf lobes	3. Three lobes 5. Five lobes 7. Seven lobes 9. Nine lobes 11. Eleven lobes	6–7 months after planting
5	Shape of central leaflet	1. Oblanceolate 2. Linear 3. Elliptic 4. Pandurate 5. Lanceolate 6. Combination of above	6–7 months after planting
6	Color of leaf vein	3. Light green 5. Dark green 7. Green purple 9. Purple	6–7 months after planting
Petiole morphology			
7	Petiole length	1. Petiole absent 3. Short (5–10 cm) 5. Medium (15–20 cm) 7. Long (25–30 cm)	6–7 months after planting
8	Petiole color	3. Light green 5. Dark green 7. Green purple 9. Purple	6–7 months after planting
9	Distribution of anthocyanin	1. Absent 2. Top part 3. Central part 4. Totally pigmented	6–7 months after planting
10	Angle of petiole insertion	1. No petiole 3. 15–30º 5. 45–60º 7. 75–90º	6–7 months after planting
Stem morphology			
11	Growth habit of young stem	1. Straight 2. Zig-zag	6–7 months after planting
12	Shape of plant	1. Compact 2. Open 3. Umbrella 4. Cylindrical	6–7 months after planting
13	Color of stem exterior	1. Silver green 2. Light brown or orange 3. Dark brown	6–7 months after planting
14	Levels of branching	0. No branching	6–7 months after planting
15	Angle of branching	1. No branching 3. 15–30º 5. 45–60º 7. 75–90º	6–7 months after planting
16	Prominence of foliar scars	3. Little prominence 5. Moderately prominent 7. Prominent	6–7 months after planting
17	Stipule margin	1. Laciniate 2. Serrate 3. Entire 99. Other	6–7 months after planting
		Root morphology	
18	Position of root	1. Tending toward vertical 2. Tending toward horizontal 3. Irregular	Harvest
19	Extent of root peduncle	1. Absent 3. Short 5. Intermediately 7. Long	Harvest
20	Root shape	1. Conical 2. Conical-cylindrical 3. Cylindrical 4. Fusiform 5. Irregular 6. Combination of above	Harvest
21	Root constrictions	1. Absent 2. Present	Harvest
22	Texture of root epidermis	3. Smooth 5. Medium 7. Rough	Harvest
23	External color of storage root	1. White or cream 2. Light brown 3. Dark brown	Harvest
24	Color of storage root cortex	1. White or cream 2. Yellow 3. Pink 4. Purple	Harvest
25	Color of storage root pulp	1. White or cream 2. Yellow 3. Pink	Harvest

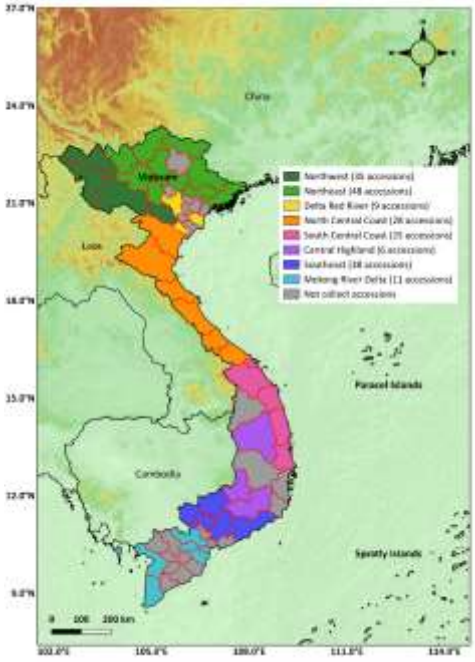

Fig. 1: Map of cassava accession collection locations in Vietnam
(Symbol colors specify the agro-ecological zones. The map was created using QGIS 3.22.9-Białowieża (QGIS Development Team 2022). Geospatial data used for the creation of this map include international (The Humanitarian Data Exchange 2022a) and subnational (The Humanitarian Data Exchange 2022b) administrative boundaries, distributed under ODC-ODbL and CC BY-IGO licenses respectively. The black lines are the national boundaries. The red lines represent the provincial boundary. The light green to reddish brown shaded background represents elevation, retrieved from (The World Bank 2020), distributed under a CC-BY 4.0 license)

in Vietnam exhibited several prominent characteristics including stem straight growing habit (83% of accessions), cylindrical shape (56% of accessions), silver-green stem (56.5% of accessions), no branching (55.5% of accessions), foliar scars prominent (83.5% of accessions), and margin of stipules serrate (57% of

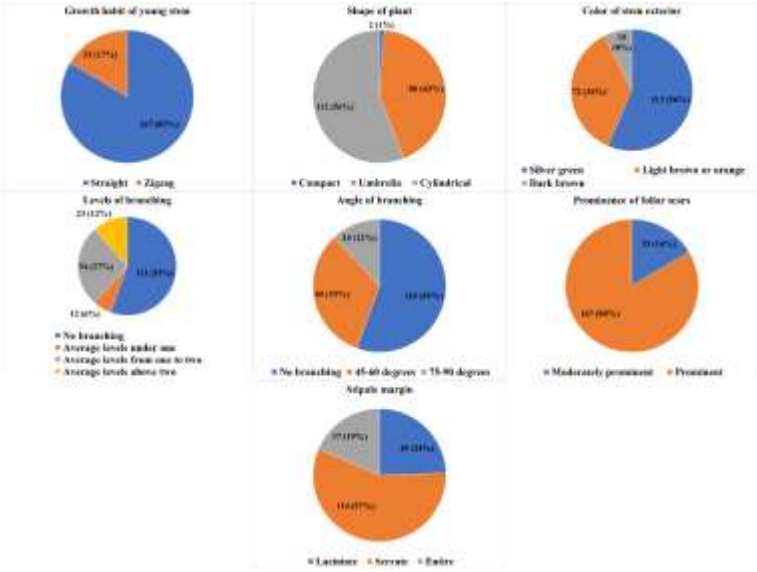

Fig. 2: The variability of stem morphological characteristics of 200 local cassava accessions of Vietnam

Fig. 3: Images of typical stem exterior color of some cassava accessions studied and their vernacular name with respect to A: silver green stem, B: light brown or orange stem, C: dark brown stem

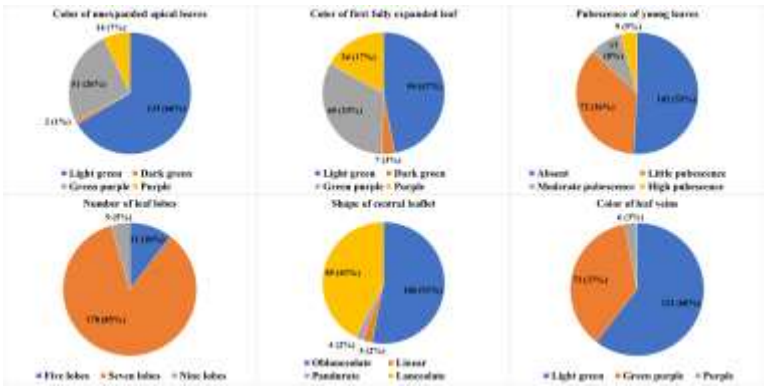

Fig. 4: The variability of leaf morphological characteristics of 200 local cassava accessions of Vietnam

accessions) (Fig. 1, 2). In addition, some morphological traits of the cassava stem were observed at a relatively low frequency (below 10%) among the 200 local cassava varieties studied. Particularly, the stem morphology with a compact shape was found in merely 1% of the accessions, including the S60 (Khoai mi Nhat) and S96 (trang Bac Thai) (Fig. 3). Similarly, only 7.5% of the accessions exhibited a dark brown stem exterior, *viz.* S61 (Khoai mi gon dia phuong), S116 (My gon), etc.

Leaf blade characteristics

Leaves of local cassava varieties in Vietnam had several typical morphological characteristics that accounted for a large

percentage, particularly 85% of local accessions had 7-lobed leaves (Fig. 4). Over 50% of the studied accessions had the light green color of young leaves (67%), young leaves without pubescence (51%), light green leaf veins (60.5%), and central leaflet oblanceolate (53%). Meanwhile, other morphological characteristics were less common. Merely 1% of accessions had dark green young leaves, 3.5 % had dark green first leaf, 4.5% had high pubescence on young leaves, 4.5% had 9-lobed leaves, 3% had purple leaf vein, 2% had central leaflet pandurate, and 2.5% had central leaflet linear. Certain morphological traits were not observed in the studied cassava collection, including central leaflet elliptic, dark green leaf veins, and leaves with three lobes.

Leaf petiole characteristics

The leaf petiole of studied accessions was observed based on 4 primary morphological descriptors, *viz.* the petiole length, petiole color, the distribution of anthocyanin pigmentation in the petiole, and the angle of petiole insertion to stem. Majority of accessions had petiole lengths ranging from 25 to 30 cm, accounting for 78.5%. A 21% accessions had medium lengths petioles ranging from 15 to 20 cm. Only the S195 (Man tun) accession had short petiole, i.e., from 5 to 10 cm (Fig. 5).

Local cassava accessions in Vietnam predominantly had petioles with a purple color (55%), followed by petioles with light green and greenish-purple colors, at 22 and 17%, respectively. Only 6% of accessions had dark green petioles, such as S8 (San nep), S19 (San Binh quy), S24 (Men co), S166 (San xanh Cao Bang), and S191 (Min co) (Fig. 6).

Additionally, accessions with 75–90° petiole insertion angles were predominant (151 accessions) were dominant, while 49 accessions were 45–60°. Out of all the accessions studied, 59.5% had anthocyanin distributed throughout the petiole, 28.5% had no pigment observed, 10.5% had pigment at the petiole tip, and 1.5% had anthocyanin pigment in the middle of the petiole.

Root morphological characteristics

Roots of most accessions studied were horizontal, accounting for 99.5%. Only accession S117 (My An Do) had a vertical root orientation. Additionally, 97.5% of accessions had white or cream pulp, while the remaining 2.5% had yellow pulp. Thus, accessions with yellow root pulp could be targeted for nutritional content analysis to develop high-quality products with enhanced nutritional value for domestic markets and exports. Additionally, over 50% of studied accessions had root constrictions absent (71.5%), root peduncle short (69.5%), the texture of root epidermis rough (57.5%), and storage root cortex pink (56.5%) (Fig. 7). Particularly, accession S17 (Trang Thai Lan) witnessed the diversity in the root shape morphology in both fusiform and cylindrical, accounting for 3.5 and 6% of the accessions, respectively (Fig. 8). Meanwhile, none of

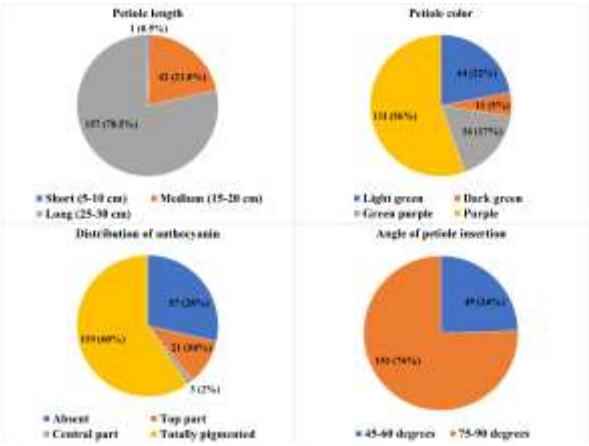

Fig. 5: The variability of leaf petiole morphological characteristics of 200 local cassava accessions of Vietnam

Fig. 6: Images of typical leaf shapes and petioles of some cassava accessions studied and their vernacular name

the 200 accessions evaluated had storage root cortex purple, root pulp pink, and position of root irregular.

Morphological variability of local cassava accessions in vietnam

The clustering analysis results of cassava accessions was based on morphological traits using NTsyspc 2.0 software. The analysis revealed that 200 local cassava accessions were distributed into two main groups with a genetic distance of 10.3 (Fig. 9). A total of 43 accessions were in Group I, which was divided into two major subgroups: IA and IB.

In subgroup IA, only accession S192 had significant differences in leaf morphology traits compared to other accessions classified in the same subgroup, including nine lobes, green-purple young leaves, and purple leaf veins. Eleven other accessions grouped under subgroup IA were mainly identified by the light green color of unexpanded apical leaves and first fully expanded leaf, seven lobed leaves, light green leaf veins and petioles, the absence of distribution of anthocyanin, the angle of petiole insertion ranged from 75 to 90 degrees, umbrella shape of plant, stem exterior silver green, and foliar scars moderately prominent.

There were 31 accessions in subgroup IB. At a genetic distance of 7.41, subgroup IB was divided into two minor subgroups IB_1 and IB_2 with 22 and 9 accessions, respectively. Accessions under subgroup IB_2 had some

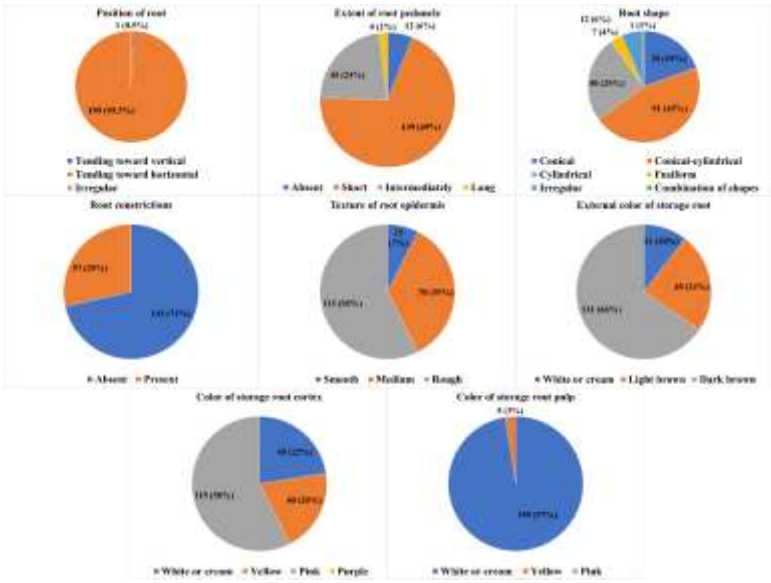

Fig. 7: The variability of root morphological characteristics of 200 local cassava accessions of Vietnam

Fig. 8: Images of root morphological characters of some cassava accessions studied and their vernacular name

common characteristics including, central leaflet oblanceolate, the unexpanded apical leaves light green, the first fully expanded leaf green purple, petioles light green, the distribution of anthocyanin absent, shape of plant cylindrical, no branching, and stipule margin serrate.

Meanwhile, accessions under subgroup IB_1 were characterized by light green color of unexpanded apical leaves, first fully expanded leaf and leaf veins, leaves with five to seven lobes, the light green petioles without pigmentation, and cylindrical non-branching stems. In this subgroup, at a genetic distance of 6.58, accessions S14, S57, S85, S143 and S172 had significant differences in the shapes of central leaflets, which were lanceolate compared to the oblanceolate shape of the remaining accessions.

With the accession number 2.6 times larger than Group I, Group II was divided into two major subgroups IIA and IIB with a genetic distance of 10.07. Subgroup IIA had the largest number of accessions i.e., 104. This subgroup was further divided into two minor subgroups, including IIA_1 and IIA_2 with 64 and 40 accessions, respectively. Analysis of morphological traits showed that subgroup IIA_1 was represented by branching stem phenotypes, while the accessions in subgroup IIA_2 had non-branching stems. Depending on their needs, farmers may choose accessions with either straight (non-branching) or branching stems. Accessions with straight stems are often preferred for intercropping and ease of harvesting, thus are more commonly selected for production.

Subgroup IIB contained 53 accessions, further divided into IIB_1 and IIB_2 with a genetic distance of 8.9. In this subgroup, varieties in branch IIB_1 had either purple or green-purple leaf veins, while most accessions in branch IIB_2 had light green ones. Additionally, the results revealed that the pair of accessions S82 and S83, which had the lowest value of genetic distance under branch IIB_1, were both collected from Northeast (Cao Bang province), sharing several morphological similarities but distinguishing only in root shape (accession S83 with cylindrical and S82 with conical-cylindrical).

Discussion

Assessment of the agro-morphological traits and yield of cassava landraces will provide breeders with a basis for

selection, breeding, and evaluation work, as well as for the valuable, diverse, and effective management of cassava germplasm currently available in Vietnam. The study revealed several significant differences in the morphological characteristics of 200 accessions from 8 agro-ecological regions in Vietnam, with genetic distances ranging from 1 to 15.52. The accessions were grouped independently of the agro-ecological zones of collection. As a result, the differences in climatic and natural conditions across the agro-ecological zones of Vietnam did not affect the clustering of cassava accessions. Previous studies on cassava collections in Indonesia (Roslim *et al.* 2016) and Côte d'Ivoire (N'Zue *et al.* 2009; Kouakou *et al.* 2023) also showed that morphological variability is not according to their origin. This demonstrates cassava varieties' dynamic exchange and movement between ecological zones in Vietnam. The high-quality varieties suited to social cultivation practices are favored and continuously exchanged among regions (Missihoun *et al.* 2012; Ferguson *et al.* 2019).

Over many years of cultivation, farmers have used typical morphological characters such as the color of stem, leaf, and root to identify and name varieties (Agre *et al.* 2016). According to Karim *et al.* (2020), qualitative traits are usually controlled by few genes with major effects. These traits are easily observable, there by making differentiation and identification of genotype easier. This is also indicated in the study of Elias *et al.* (2001) that some morphological traits related to the color of stem, leaves, and pulp of the root show the least intra-varietal differences and can be easily used as markers for farmers to varietal classification. However, the overlap of varieties potentially appears when different cassava varieties possess the same name or, conversely, a single variety could have different names (Elias *et al.* 2001; Zue *et al.* 2014).

In this study, several accessions shared the same name, such as 12 accessions called "San do", 5 accessions named "San", and 5 accessions named "San nep", etc. These accessions showed similarities in the variability of several morphological characteristics, including the shape of central leaflet lanceolate, oblanceolate, or pandurate. The color of the first fully expanded leaf, leaf vein, and petiole were light green, green purple, or purple. The growing habit of the young stem was either straight or zigzag. The shape of the plant was umbrella, or cylindrical. The color of the stem exterior ranged from silver green, light brown, and orange, to dark brown. Some accessions had branching while others had no branching.

Morphological assessment data of 200 accessions originating from 8 agroecological zones of Vietnam revealed the morphological characteristics diversity of accessions across all zones. The majority of local cassava accessions in Vietnam exhibited the following morphological characteristics (in % of total accessions): straight growing habit of young stems (83%), cylindrical plant shape (56%), silver-green stem exterior color (56.5%), no branching

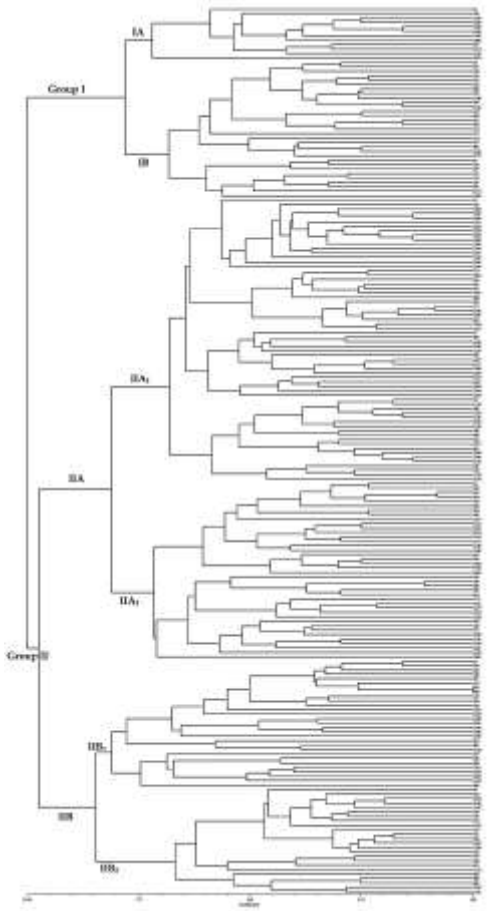

Fig. 9: Dendrogram (UPGMA) of 200 local cassava accessions based on morphological characters using NTSYSpc version 2.0 software.

(55.5%), prominent foliar scars (83.5%), and serrate stipule margins (57%). The leaves had seven lobes (85%) and the central leaflet was oblanceolate (53%). The young leaves and the leaf vein were light green accounting for 67 and 60.5%, respectively. The petioles were purple (55%) and the angle of petiole insertion was 75–90 degrees (75.5%). In addition, the characteristics of three-lobed leaves, dark green leaf veins, or elliptical-lobed leaf shapes were not observed in this report. This is in line with the previous studies on common cassava varieties in Vietnam, which also do not report the presence of the characteristics of 3-lobed leaves (Ha *et al.* 2015; Nguyen *et al.* 2021).

For root morphological characteristics, the low percentage of cassava yellow pulp in this report has also recorded by Mezette *et al.* (2013); Lebot *et al.* (2015); Karim *et al.* (2020); Doubi *et al.* (2021). Generally, varieties with yellow root pulp have higher carotenoid and amino acid content compared to those with white or cream pulp (Nagib Nassar *et al.* 2007). Thus, accessions with yellow root pulp could be targeted for nutritional content analysis to develop high-quality products with enhanced nutritional value for domestic markets and exports.

In contrast, other studies worldwide have reported different morphological characteristics compared to the cassava collections in Vietnam. For instance, Nadjiam *et al*. (2016) studied 59 cassava cultivars from 5 growing regions in Chad and reported that 74% of the cultivars had elliptic-lanceolate central lobes, 83% of the cultivars had greeny-yellowish stems, and no green petiole observed. Similarly, Doubi *et al*. (2021) did not record certain morphological diversities observed in the Vietnamese collections when studying 180 cassava cultivars from 13 regions of Côte d'Ivoire.

Conclusion

Local cassava accessions in Vietnam witnessed the diversity of the morphological characteristics observed. Their morphological variability was not according to their origin and clustered independently with the difference in natural conditions of agroecological zones. Furthermore, this study highlighted the vibrant exchange of cassava varieties in the local communities. The research will contribute to the establishment of a systematic database of local cassava germplasm in Vietnam, contributing to the preservation and conservation of this genetic resource for the future. Additionally, when combined with genetic data, the classification of cassava varieties will be described more clearly, providing breeders with a solid foundation for developing new varieties suited to the current climate change conditions.

Acknowledgements

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Author Contributions

Dam Thi Thu Ha, Hoang Thi Nga and La Tuan Nghia designed the experiment. La Hoang Nhat Minh and Phuong Diep Vien Ta collected the data. Le Thi Thu Trang and Hoang Thi Hue statistically analyzed and visualized the data. Dam Thi Thu Ha, Le Thi Thu Trang, Hoang Thi Hue and Phuong Diep Vien Ta interpreted the results. Dam Thi Thu Ha and Phuong Diep Vien Ta drafted the manuscript. Dam Thi Thu Ha, Phuong Diep Vien Ta, La Tuan Nghia and Tran Dang Khanh thoroughly revised it. All authors have contributed to the manuscript and approved this final version.

Conflicts of Interest

All authors declare no conflict of interest.

Data Availability

Data presented in this study will be available on a fair request to the corresponding author.

Ethics Approval

Not applicable to this paper

Funding Source

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