

# **Agromorphological Characterization of Amaranth Accessions**

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# **Abstract**

A sound fundamental knowledge of the agro morphological characteristics of amaranth accessions is crucial for promoting their use in food. This review aims to assess the genetic diversity within the collection to understand the range of genetic variation. Thirty-two agromorphological parameters of Ten amaranth accessions were analyzed in Samanko conditions. Wide variability was present concerning leaf pigmentation (PigF), flower density index (IDF), seed color (CH), and Branching index (IR). Statistical analysis showed significant differences among accessions for morphological characters such as number of plants, leaf width, stem height, terminal inflorescence length of stem, axillary inflorescence length, 50% flowering and 1000 grain weight. It appears that the Axillary Inflorescence (LIA) and the Length of the Terminal Inflorescence are correlated  $(r = 0.82)$ . Length of the inflorescence terminal of the stem (LITT) and the Length of the Terminal Basal Branches (LITBT) are associated ( $r = 0.75$ ). Stem height presents a robust correlation ( $r = 0.447$ ) with 50% flowering date. The length of the inflorescence Axillary was also highly correlated with the length of the inflorescence terminal of the stem  $(r =$ 0.904\*). Ascending hierarchical classification revealed three distinct classes: C1: Madira 1, Madira 2, A2004, A2002; C2: TP5-sel, N'gourouma, Akeri, AC-NL; C3: AHTI, Akeri. "Akeri." "TP5-sel," and "N'gourouma" are associated with the weight of 1000 grains, the length of the terminal branches, and the shape of the terminal inflorescence. "Poly" and "AHTI" are quite similar and are associated with the parameters of the length of basal branches and length of a terminal inflorescence of the stem "Madira 1"; "Madira 2", "A2004", "A2002" on the correlation circle indicates that the height of the plants and the 50% flowering date. These results indicate a high possibility of genetic diversity among the amaranth accessions within the collection. The data can be exploited in future breeding programs to improve the species.

#### **Keywords**

Accession, Amaranth, Morphology, Characterization

# **1. Introduction**

African leafy vegetables contribute greatly to achieving food and nutrition security especially in times of famine and natural disasters [\[1\].](#page-16-0) According to the work of [\[2\],](#page-17-0) if people consumed leafy vegetables in their daily diet, it could improve their nutritional status, as these vegetables have high levels of micronutrients. Indigenous leafy vegetables are essential for food and nutrition security. Several studies have shown that African leafy vegetables offer many benefits, high micronutrient content, and beneficial medicinal propertie[s \[3\]](#page-17-1) [\[4\].](#page-17-2) In addition, these vegetables have interesting agronomic performances, such as a short growth period (some crops are ready to harvest within 3 to 4 weeks after planting) and low input requirements [\[5\].](#page-17-3) Amaranth is recognized as one of the most nutritious traditional leafy vegetables due to its high protein content and its richness in lysine, which is usually a limiting amino acid in cereals [\[6\].](#page-17-4) Amaranth has nutritional benefits, and it can be used in Nutritional Value Bakery Products. [\[7\].](#page-17-5)

Amaranthus (family *Amaranthaceae*) collectively known as amaranth is a cosmopolitan genus of annual or short-lived perennial plants consisting of approximately 60 species. These species can be divided into cereals and vegetable amaranths based on their use for human consumption [\[8\].](#page-17-6) Amaranthus sp. is one of the neglected leafy vegetables. Amaranth is rich in protein, fiber, and minerals, with higher contents than cereal[s \[9\].](#page-17-7) However, amaranth is affected by many factors that limit its production. Some of the major constraints on the production of leafy vegetables include poor quality seed, lack of production technologies, and poor marketing and processing strategie[s \[10\].](#page-17-8) Low research investment is due to the status of the indigenous plant. Pests and diseases of Amaranth also pose serious threats to its sustainable production. Leaf miners, leaf rollers, cutworms, aphids, and flea beetles [\[11\].](#page-17-9) Severe infestations can lead to complete yield loss as the leaves become unsuitable for marketing and consumption [\[12\].](#page-17-10) Several research studies have been published on various aspects of amaranth with the aim of valorizing it and showing its importance, including its transformation and use in Guatemala. Mexico, and Peru [\[13\],](#page-17-11) the development of genetic material and agronomic surveys in Mexico [\[14\],](#page-17-12) and the nutritional and anti-nutritional composition of amaranth leaf vegetables in India [\[15\].](#page-17-13) A comprehensive analysis of published results on Amaranthus spp., including aspects such as composition, and antioxidant properties. Applications and processing were conducted b[y \[16\].](#page-18-0) The work o[f \[17\]](#page-18-1) in Belgium provided insights into seed characteristics chemical composition, and technological properties of amaranth. Despite these studies, amaranth remains relatively unknown and neglected in the Sahelian region of Africa. This limited awareness likely contributes to the low collection and understanding of its genetic

variation in this area. African leafy vegetables like amaranth, can be grown under harsh climatic conditions with low external inputs. It is therefore important to explore the possibility of increasing production and marketing such vegetables in Mali. This will not only contribute to improved livelihoods of poor communities, but it will also indirectly contribute to improved nutrition and conservation of these valuable genetic resources. In efforts to be sustainable, it is important to understand the diversity of Amaranth in Mali in order to produce, market, and conserve it. For an indigenous crop with little information, a comparison of various accessions would be an important step in the identification of those morphological characteristics. According to various reports, the genus of amaranth exhibits a high degree of morphological diversit[y \[11\]](#page-17-9) [\[18\]](#page-18-2) [\[19\].](#page-18-3) The interplay between local environmental conditions and the agro-morphological characteristics of Amaranth in Mali underscores the importance of context-specific agricultural practices. Understanding these relationships can aid in selecting suitable accessions for cultivation that are resilient to local climatic challenges while maximizing nutritional benefits and food security. This knowledge is vital for promoting sustainable agricultural practices that leverage local biodiversity and traditional knowledge systems.

Research indicates that local climatic conditions, such as temperature and rainfall patterns, significantly affect the growth and yield of Amaranth, influencing traits like plant height and leaf size [\[20\].](#page-18-4) Furthermore, soil characteristics, including nutrient availability and pH levels, play a crucial role in determining the morphological traits of Amaranth plants, as fertile soils generally support better growth outcomes [\[21\].](#page-18-5) Additionally, water availability is critical; regions with consistent rainfall or effective irrigation systems tend to produce healthier plants with superior morphological characteristics [\[22\].](#page-18-6) The importance of local agricultural practices cannot be overstated, as traditional methods such as crop rotation can enhance soil health and biodiversity, positively impacting plant growth [\[23\].](#page-18-7) Lastly, the genetic diversity of Amaranth accessions allows for better adaptation to specific local conditions, contributing to resilience against environmental stresses [\[20\].](#page-18-4) Such information is important for selecting better accession for breeding programs. Breeders and farmers need agro-morphological characterization and a lot of information and amaranth accessions efficient for increasing productivity [\[18\].](#page-18-2) So far, no distinctive study has been undertaken in Mali to characterize amaranth accessions. The current study was undertaken to characterize amaranth accessions using agro-morphological traits under agro-climatic conditions of Samanko. The results are useful in future genetic studies to improve this climate-resilient and highly nutritive underutilized crop species.

#### **2. Materials and Methods**

## **2.1. Experimental Site**

The assessment was carried out during the 2023 rainy season in Mali, specifically at the World Vegetable Center experimental station in Samanko [\(Figure 1\)](#page-3-0). This

site receives an annual rainfall of 1047 mm and features nutrient-rich clay soil with high water retention capacity but it can be difficult to work with, especially when dry. The average temperature at the site is  $27.7^{\circ}$ C. [Figure 1](#page-3-0) illustrates the location of the experimental site and the general distribution of rainfall in this area

<span id="page-3-0"></span>

Figure 1. Map of the study area.

## **2.2. Amaranth Accessions Collections**

Ten amaranth accessions were collected from different zones in Mali.

## **2.3. Experimental Design**

A set of ten amaranth accessions were evaluated in a single environment using a randomized complete block design with three replications. The randomization of the experimental design was facilitated using the Breeding Management System (BMS) software. Each experimental plot consisted of lines measuring 5 meters in length. Planting densities were set at 50 cm  $\times$  50 cm between and within rows, respectively.

## **2.4. Data Collection**

Data on both quantitative and qualitative traits were collected. The quantitative traits included plant height (cm) (from ground level to the highest tip of the inflorescence) measured using a measuring tape, stem girth (cm) (between the 3<sup>rd</sup> and  $4<sup>th</sup>$  node) measured using a caliper, leaf length (cm), leaf width (cm), and inflorescence length (cm) measured using a measuring tape. The days to 50% flowering were determined by counting the days from planting to when 50% of the plants in the net plot flowered. Qualitative traits such as inflorescence color, plant growth habit, leaf pigmentation, petiole pigmentation, flower density index, stem color, inflorescence spininess, seed color, and the attitude of the terminal inflorescence were

visually observed on all plants within the gross plot.

#### **2.5. Data Analysis**

XLSTAT 2014.5.03 software was used for the analysis of variance. The Fisher test was used for the comparison of means when the analysis of variance revealed significant differences among the genotypes at a 5% probability threshold [\[24\]](#page-18-8) [\[25\].](#page-18-9)

Principal component analysis (PCA) was performed to reduce the dimension of the variables, to study the variability within the plant material, and to determine the different groups derived from the analysis. The multidimensional statistical method used to analyze the relationships between agromorphological parameters and yield components was principal component factor analysis (PCA), conducted using XLSTAT 2014.5.03 software on centered-reduced data. Thirty-two agromorphological parameters and yield components were analyzed, with these parameters serving as active or explanatory variables in the Principal Component Analysis (PCA). The variables included the number of plants raised, growth habit, main stem height, branching index, the average length of basal branches, average length of terminal branches, stem pigmentation, leaf length, basal branch length, length of the terminal branches; leaf length; leaf width; flowering date; length of the terminal inflorescence of the stem, length of the terminal inflorescence of the branch, weight of 1000 grains and length of axilary inflorescence, leaf pigmentation, prominence of the veins, attitude of the terminal inflorescence.

Additionally, ten varieties were used as supplementary variables: "Madira 1" "Madira 2." "A2004" "A2002", "N'gourouma", "Akeri", "AC-NL", "Poly" and "AHTI". An ascending hierarchical classification (CAH) was applied to construct groups of varieties according to agro-morphological characteristics and yield components using XLSTAT 2014.5.03 software.

# **3. Results**

#### **3.1. Agro-Morphological Characteristics of Amaranth Accessions**

#### **3.1.1. Qualitative Characteristics**

#### Variability for morphological traits and their frequency distribution

Variability for morphological characters and their frequency distribution is shown in [Table 1.](#page-5-0)

Growth habit: it was observed that 60% of the accessions studied produced an erect growth habit with 40% showing a semi-erect growth habit.

Stem and leaf pigmentation: Stem and leaf colour of the study accessions (60%) were mainly green.

Petiole pigmentation and Inflorescence Pigmentation: Accessions with white flowers had mostly green stems (60%) and low petiole pigmentation (53.3%).

Flower Density Index: half of the accessions had an average flower density index (50%).

Branching index: more than half of the accessions (60%) had erected ports. Seed color: the lovers presented three colors of seeds.

Stem pubescence: half of the accessions had pubescent stems.

Branching index (IR): Branching index score ranged from 1 to 3. The IR score ranged from 1 to 3. Sixty percent of the accessions had an IR score of 3.



<span id="page-5-0"></span>Table 1. Variability for morphological traits and their frequency distribution in amaranth in Samanko conditions.

#### Principal coordinate analysis

The four main axes (axis 1, 2, 3 and 4) explained 35.073; 20.94; 12.402 and 11.415% of the total variation respectively for the accessions giving a cumulative total variation of 79.831% [\(Table 1\)](#page-5-0). Axes 1, 2, 3 and 4 contributed 5.962, 3.660, 2.108 and 1.940 of the eingenvalues respectively, grown accessions at Samanko conditions [\(Table 2\)](#page-6-0).

Thorn in the leaf axil, Seed Shape, Seed Shell Type, Seed color, Pod dehiscence, and Flower Type were the main traits that contributed positively to PC1 for the accessions [\(Table 2\)](#page-6-0). It was also observed that Axis 1 is strongly associated with traits such as the attitude of the terminal inflorescence  $(r = 0.003)$ , the shape of the terminal inflorescence ( $r = 0.0001$ ), and the shape of the leaves ( $r = 0.005$ ).

Attitude of the terminal inflorescence; Presence of axillary inflorescence, Shape

of the edge of the limb, Growth habit, Flower two characters Presence of axillary inflorescence Shape of the edge of the limb were important for PC3. Petiole pigmentation, stem pigmentation, pubescence of the stem and branching index contributed positively to PC4. Density Index was the most important character that contributed to the second principal component. Nearly all the characters that made significant contributions to a particular principal component were important contributors to another principal component [\(Table 2\)](#page-6-0).



<span id="page-6-0"></span>Table 2. Eigenvalues<sup>a</sup>, eigenvectors<sup>b</sup> and percentage of variation explained by the first four principal components for 10 amaranth accessions.

<sup>a</sup>Eigenvalues indicate the amount of variance explained by each principal component; <sup>b</sup>Eigenvectors are the weights in a linear transformation when computing principal components; 'Values in bold correspond for each variable to the factor for which the squared cosine is the largest.

[Figure 2](#page-7-0) shows the genetic relationships based on the first (axis 1 and 2) and second (axis 1 and 3) planes of the PCoA. Generally, the separation between accessions was high, with the first and second planes clearly separating accessions. The accessions were also clearly separated by the second plane (axis 1 and 3) and the first plane (axis 1 and 2) [\(Figure 2\)](#page-7-0).

<span id="page-7-0"></span>

#### Red font point: accessions



#### **3.1.2. Quantitative Characteristics**

[Table 3\(A\)](#page-9-0) and [Table 3\(B\)](#page-9-0) reports agro-morphological parameters of Amaranth accessions such as: Number of plants (NP), stem height (HT), Basal branch length (LMBLB), length of the terminal branches (LMBLT), Leaf length (LoF), Leaf width (LaF), Fowering date (DF50%), Length of the terminal inflorescence of the stem (LITT), Length of the terminal inflorescence of the branch (LITB), Axillary inflorescence length (LIA) and Weight of 1000 grains (P1000).

Number of plants (NP) and stem height (HT): The statistical analysis showed a significant difference between accessions for the number of plants and stem height at the 5% level. The AHTI accession recorded the largest number of plants, followed by A2004. Madira 1. Madira 2, and N'gourouma. The accession Poly recorded the lowest number of plants. The average number of plants was  $60 \pm 5.8$ , ranging from 34 to 64. Stem height varied from 95.5 cm to 247.2 cm. with an average of 159.04 ± 33.45 cm. The Madira 2 and Madira 1 accessions exhibited significantly greater plant heights, with Akeri accessions following closely behind, showing no statistically significant difference from these two accessions.

Basal branch length (LMBLB) and length of the terminal branches (LMBLT): The mean basal branch length (LMBLB) ranged from 26.6 cm to 126.6 cm. with an average of  $59.43 \pm 22.69$  cm. while the average length of the terminal branches (LMBLT) varied between 8.7 cm and 41 cm. with an average of  $23.05 \pm 7.87$  cm. Statistical analysis showed a significant difference in both LMBLB and LMBLT among the accessions at the 5% level. However, no significant difference was observed between the Akeri, AHTI, Poly, and TP5-sel accessions, all of which recorded the highest values for LMBLB and LMBLT. On the other hand, Nadira 1 and Madira 2 accessions recorded the lowest LMBLB and LMBLT, with no significant difference compared to the A2004. A2002, and AC-NL accessions for these

variables.

Leaf length (LoF) and Leaf width (LaF): The statistical analysis showed significant differences between amaranth accessions for leaf length (LoF) at the 5% level. The LoF varied from 4.75 cm to 16.3 cm. with an average of  $12.3 \pm 2.76$  cm. The Madira 1 accession recorded the highest value. followed by Madira 2, N'gourouma, and A2004, which were statistically similar. Highly significant differences were observed among the accessions for leaf width (LaF) at the 5% level. The LaF ranged from 3.9 to 10.7 cm, with an average of  $6.9 \pm 1.84$  cm. The TP5sel and N'gourouma varieties recorded the highest LaF values, followed by the Poly variety. In contrast. Madira 1 and A2002 had the lowest LaF measurements, with no statistically significant difference between them.

Fowering date (DF50%): The 50% flowering date ranged from 40 to 60 days. with an average of  $50 \pm 6.26$  days. Akeri, Madira 2, A2004. Madira 1, AC-NL and A2002 all exhibited statistically similar results, recording the longest time to reach 50% flowering. These were followed by the N'gourouma variety. AHTI. Poly. and TP5-sel accessions recorded the shortest time to reach 50%flowering compared to the other accessions.

Length of the terminal inflorescence of the stem (LITT) and Length of the terminal inflorescence of the branch (LITB): The average length of the terminal inflorescence of the stem (LITT) was  $5.609 \pm 5.28$  cm. with a range of 1.3 to 21.3 cm. The length of the terminal inflorescence of the branch (LITB) varied from 3.2 to 22 cm with an average of  $8 \pm 4.849$  cm. Statistical analysis showed a highly significant difference between accessions for both LITT and LITB. AHTI and the accession Poly recorded the highest LITT, followed by Akeri and TP5-sel accessions, which were statistically similar. The lowest LITT values were recorded by Madira 1. Madira 2, N'gourouma. A2004, A2002 and AC-NL which were statistically equal. The Poly variety recorded the highest LITB, followed by AHTI, Akeri and TP5-sel accessions. No significant difference was noted between A2004, Madira 1, AC-NL, A2002, N'gourouma and Madira 2 accessions which recorded the lowest LITB values.

Axillary inflorescence length (LIA): Statistical analysis showed a highly significant difference in LIA among the accessions at the 5% significance level. Axillary inflorescence length varied from 2.35 cm to 17.8 cm. with an average of 7.23  $\pm$  4.65 cm. AHTI and Poly accessions recorded the highest LIA averages of 14.76 and 14.56 cm. respectively. They were followed by Akeri and TP5-sel accessions which were statistically similar. The lowest values were recorded by N'gourouma, Madira 1, Madira 2, A2002, A2004 and AC-NL which were statistically similar.

Weight of 1000 grains (P1000): The weight of 1.000 grains ranged from 0.83 to 1.57 g  $(\pm 0.213 \text{ g})$ , with an average weight of 1.11 g. Statistical analysis at the 5% significance level indicated a highly significant difference between the varieties. The AHTI and Poly varieties had the highest average 1.000-grain weights and were statistically similar. The Akeri variety followed them. In contrast, the N'gourouma. A2002, A2004, Madira 1 and AC-NL varieties recorded the lowest 1000grain weights.

(A)											
Amaranth accessions	<b>NPL</b> Htige		<b>LMBLB</b>	<b>LMBLT</b>	LoF	LaF					
Akeri	$56.66 \pm 4.04$ <sup>ab</sup>	$180.56 \pm 13.4$ <sup>ab</sup>	$57.767 \pm 22.31$ <sup>abc</sup>	$30.767 \pm 4.17$ <sup>a</sup>	$13.356 \pm 1.22$ <sup>abc</sup>	$7.47 \pm 0.87$ <sup>c</sup>					
<b>AHTI</b>	$64.00 \pm 0.01$ <sup>a</sup>	$136.53 \pm 3.11$ <sup>c</sup>	$62.70 \pm 6.39$ <sup>abc</sup>	$30.233 \pm 9.66^a$	$11.917 \pm 1.15$ <sup>bc</sup>	$6.95 \pm 1.1$ <sup>c</sup>					
Poly	$49.33 \pm 13.42^b$	$125.43 \pm 26.54^{\circ}$	$87.33 \pm 34.35^{\circ}$	$32 \pm 5.02^{\text{a}}$	$8.8 \pm 4.29$ <sup>d</sup>	$9.26 \pm 0.5^{ab}$					
N'gourouma	$62.67 \pm 0.57$ <sup>a</sup>	$179.05 \pm 10.88$ <sup>ab</sup>	$84.43 \pm 14.19$ <sup>ab</sup>	$24.83 \pm 1.24$ <sup>ab</sup>	$12.917 \pm 1.17$ <sup>bc</sup>	$9.53 \pm 1.12^a$					
Madira 2	$62 \pm 2.64^{\circ}$	$210.2 \pm 52.59^{\circ}$	53.167 ± 20.46 <sup>bc</sup>	$14.07 \pm 5.91$ <sup>c</sup>	$14.41 \pm 0.86$ <sup>ab</sup>	$6.55 \pm 0.17$ <sup>c</sup>					
TP5-sel	$59.33 \pm 4.04^a$	$143.13 \pm 9.82$ <sup>bc</sup>	$67.50 \pm 18.2$ <sup>abc</sup>	$28.47 \pm 4.52$ <sup>a</sup>	$10.28 \pm 4.84$ <sup>cd</sup>	$10 \pm 2.13^a$					
A2004	$63.33 \pm 1.15^a$	$147.43 \pm 24.04$ <sup>bc</sup>	$43.53 \pm 13.65$ <sup>cd</sup>	$18.48 \pm 7.19$ <sup>bc</sup>	$12.08 \pm 0.9$ abcd	$6.01 \pm 1.14$ <sup>cd</sup>					
Madira 1	$61 \pm 3^a$	$188.13 \pm 3.65^{\circ}$	$36.96 \pm 16.17$ <sup>c</sup>	$16.40 \pm 0.95$ <sup>bc</sup>	$15.97 \pm 0.29^{\text{a}}$	$4.28 \pm 0.33$ <sup>d</sup>					
<b>AC-NL</b>	$61 \pm 2.64^{\circ}$	$146.46 \pm 25.52$ <sup>bc</sup>	$40.4 \pm 9.70$ <sup>c</sup>	$18.03 \pm 3^{bc}$	$12.56 \pm \text{abcd}$	$6.56 \pm 0.85$ <sup>bc</sup>					
A2002	$58.67 \pm 0.57$ <sup>a</sup>	$133.46 \pm 19.07$ <sup>c</sup>	$60.56 \pm 17.09$ <sup>abc</sup>	$17.26 \pm 0.96$ <sup>bc</sup>	$10.58 \pm 1.94$ bcd	$4.76 \pm 0.69$ <sup>d</sup>					
Pr > F	0.055	0.003	0.041	0.001	0.043	< 0.0001					
Signification	$\mathcal{S}$	$\mathcal{S}_{\mathcal{S}}$	$\mathcal{S}_{\mathcal{S}}$	$\mathcal{S}_{\mathcal{S}}$	$\mathcal{S}_{\mathcal{S}}$	<b>THS</b>					
$CV(\%)$	7	18	29	29	24	17					
(B)											
Amaranth accessions	<b>DF50%</b>		<b>LITT</b>	<b>LITB</b>	<b>LIA</b>	P1000					
Akeri	$54 \pm 0.01$ <sup>a</sup>		$6.80 \pm 1.47^{\rm b}$	$11.40 \pm 0.52^b$	$7.917 \pm 1.45$ <sup>bc</sup>	$1.34 \pm 0.03^b$					
<b>AHTI</b>	$40 \pm 0.01$ <sup>d</sup>		$13.27 \pm 2.75^{\circ}$	$13.13 \pm 2.26^b$	$14.76 \pm 0.66^{\circ}$	$1.4 \pm 0.07$ <sup>ab</sup>					
Poly	$42.33 \pm 4.04$ <sup>cd</sup>		$15.96 \pm 5.2^{\text{a}}$	$17.03 \pm 4.3^a$	$14.56 \pm 3.59^{\circ}$	$1.47 \pm 0.11^a$					
N'gourouma	$49.33 \pm 4.04^b$		$2.967 \pm 0.47$ <sup>c</sup>	$4.98 \pm 0.75$ <sup>c</sup>	$5.81 \pm 2.24$ <sup>cd</sup>	$0.93 \pm 0.04^{\rm ef}$					
Madira 2	$57.33 \pm 2.3^a$		$2.643 \pm 1.45$ <sup>c</sup>	$4.25 \pm 0.54$ <sup>c</sup>	$3.46 \pm 0.99$ <sup>c</sup>	$0.97 \pm 0.04$ <sup>def</sup>					
TP5-sel	$45 \pm 1.15$ <sup>c</sup>		$6.78 \pm 2.51^{\rm b}$	$11.23 \pm 2.83^b$	$10.07 \pm 2.99^{\rm b}$	$1.67 \pm 0.04c$					
A2004	$54.66 \pm 1.15^a$		$1.85 \pm 0.2$ <sup>c</sup>	$4.717 \pm 0.95$ <sup>c</sup>	$4.45 \pm 1.62$ <sup>d</sup>	$0.947 \pm 0.12$ <sup>ef</sup>					
Madira 1	$55.33 \pm 1.15^a$		$1.967 \pm 0.47$ <sup>c</sup>	$4.80 \pm 1.05$ <sup>c</sup>	$5.23 \pm 2.08$ <sup>cd</sup>	$0.90 \pm 0.01$ <sup>e</sup>					
<b>AC-NL</b>	$55.33 \pm 1.15^a$		$1.78 \pm 0.56$ <sup>c</sup>	$3.62 \pm 0.36$ <sup>c</sup>	$2.65 \pm 0.36$ <sup>d</sup>	$1.07 \pm 0.04$ <sup>def</sup>					
A2002	$54 \pm 0.01^a$		$2.05 \pm 0.68$ <sup>c</sup>	$4.82 \pm 0.81$ <sup>c</sup>	$3.36 \pm 1.26$ <sup>d</sup>	$1.03 \pm 0.05$ <sup>d</sup>					
Pr > F	< 0.0001		< 0.0001	< 0.0001	< 0.0001	< 0.0001					
Significant		<b>THS</b>	<b>THS</b>	<b>THS</b>	<b>THS</b>	<b>THS</b>					
CV(%)	12		92	60	62	19					

<span id="page-9-0"></span>Table 3. (A). Agro-morphological parameters of Amaranth accessions. (B). Agro-morphological parameters of Amaranth accessions.

Means followed by the same letters in a column are not statistically different at  $P < 0.05$ . Abbreviation: LITT: Length of the inflorescence terminal of the stem (cm); LITB: Length of the terminal inflorescence of the branch (cm); DF 5%: Date 5% flowering; P1000: Weight of 1000 grains (g); LIA: Length of inflorescence Axillary (cm).

# **3.1.3. Correlation Analysis between Variables Quantitative and Accessions**

The correlation between the different variables is presented in [Table 4.](#page-10-0) There were significant ( $P < 0.05$ ) and positive correlations between most of the quantitative variables [\(Table 4\)](#page-10-0). The correlation analysis reveals highly significant correlations. From the conformity test of the correlation coefficients. It appears that the Axillary Inflorescence (LIA) and the Length of the Terminal Inflorescence are correlated ( $r = 0.82$ ). Length of the inflorescence terminal of the stem (LITT) and the Length of the Terminal Basal Branches (LITBT) are correlated ( $r = 0.75^*$ ). Leaf length (LoF) is correlated with stem height ( $r = 0.36*$ ). The 50% flowering date presents a very strong correlation with stem height ( $r = 0.447$ <sup>\*</sup>). Length of the inflorescence terminal of the stem was highly correlated with Leaf length ( $r =$ 0.414\*). Length of inflorescence Axilary was also highly correlated with Length of the inflorescence terminal of the stem  $(r = 0.904<sup>*</sup>)$ . A negative and very highly significant correlation ( $r = -0.824$ ) emerges between the Length of the Axillary Inflorescence (LIA) and the 50% flowering date. There is also a negative correlation between the weight of 1000 grains and the 50% flowering date  $(r = -0.664^*)$ . Meanwhile in Length of the inflorescence terminal of the stem and stem height recorded a highly significant ( $p < 0.05$ ) negative correlation ( $r = -0.393$ <sup>\*</sup>) as well as between Leaf length and Date 50% flowering ( $r = -0.466*$ ) and between sheet width and Length of the terminal inflorescence of the branch ( $r = -0.459$ <sup>\*</sup>).

Variables	<b>NPL</b>	Htige	<b>LMBLB</b>	LMBLT	LoF	LaF	<b>DF50%</b>	<b>LITT</b>	<b>LITB</b>	LIA	P <sub>1000</sub>
<b>NPL</b>	1										
Htige	0.092	1									
<b>LMBLB</b>	$-0.181$	$-0.054$	1								
<b>LMBLT</b>	$-0.207$	$-0.234$	$0.493*$								
LoF	0.369	0.639	$-0.071$	$-0.336$							
LaF	$-0.269$	$-0.072$	$0.466*$	0.526	$-0.394$	1					
<b>DF50%</b>	0.229	0.447	$-0.424$	$-0.635$	0.470	$-0.466$	1				
<b>LITT</b>	$-0.223$	$-0.393$	0.415	0.751	$-0.420$	0.414	$-0.808$				
<b>LITB</b>	$-0.337$	$-0.400$	0.375	0.728	$-0.459$	0.443	$-0.790$	0.922	1		
<b>LIA</b>	$-0.138$	$-0.328$	0.517	0.737	$-0.222$	0.423	$-0.824$	0.904	0.849	1	
P <sub>1000</sub>	$-0.323$	$-0.396$	0.349	0.739	$-0.372$	0.438	$-0.664$	0.874	0.902	0.839	

<span id="page-10-0"></span>Table 4. Relationship among variables.

\*Correlation is significant at the P > 0.05 level. Abbreviation: NPL: Number of plants raised; LITT: Length of the inflorescence terminal of the stem; HT: stem height; LMBLB length of basal Branches; LMBLT: Length of terminal branches; LITB: Length of the terminal inflorescence of the branch; DF50%: Date 50% flowerin; LIA: Length of inflorescence Axilary; LaF: sheet width; LoF: Leaf length, P1000: Weight of 1000 grains.

#### **3.1.4. Principal Component Analysis**

The principal component analysis is presented i[n Table 5.](#page-11-0) To capture the majority of the information in [Table 5.](#page-11-0) It is necessary to consider the third and fourth axes, which together account for 80.07% of the total variability

- Axis 1, which explains 43.04% of the variability, is primarily influenced by the variable's presence of axillary inflorescence, length of the terminal inflorescence of the branch, length of the terminal inflorescence of the stem, shape of the leaf, length of the terminal branches and Weight of 1000 grains. This axis contrasts varieties with a prominent Length of inflorescence Axilary, Terminal inflorescence of the branch, Length of the inflorescence terminal of the stem, 50% flowering date, Leaf pubescence, length of terminal branches, Number of plants raised, Growth habit, Stem height, Weight of 1000 grains, Seed Shape Seed Shell Type, Seed color and significant pod dehiscence.
- Axis 2, which accounts for 15.33% of the variability, is primarily explained by the variable related to Terminal Inflorescence shap, Shape of the edge of the limb, Branching index, Pod dehiscence and Flower Density Index.
- Axis 3, which accounts for 11.804% of the variability, is characterized by the

variables Terminal Inflorescence shap, presence of axillary inflorescence and Leaf length.

• Axis 4, which accounts for 9.903% of the variability, is characterized by the variables petiole pigmentation, length of basal branches, sheet width, stem pigmentation and pubescence of the stem.

[Figure 2](#page-7-0) shows correlations between the variables and the factors. The first axis accounts for 43.04% of the total variation, while the second axis explains 15.33% of the total variability.

<span id="page-11-0"></span>Table 5. Correlations between variables and factors.





<sup>a</sup>Eigenvalues indicate the amount of variance explained by each principal component; <sup>b</sup>Eigenvectors are the weights in a linear transformation when computing principal components; 'Values in bold correspond for each variable to the factor for which the squared cosine is the largest.

# **3.2. Ascending Hierarchical Classification (AHC)**

[Figure 3](#page-13-0) illustrates the dendrogram showing the relationships among the ten amaranth accessions. The hierarchical clustering analysis (HCA) revealed three distinct clusters: C1, C2 and C3.

- Class 1 (C1) is located in the negative part of Axis 1 and is characterized by several key features. Varieties in this class have a long flowering period, with 50% of plants reaching flowering at an average of 55 days. The average stem height is 169.58 cm. and leaf length measures 13.33 cm. These varieties also exhibit a good attitude of terminal inflorescence. The accessions classified under C1 include "Madira 1". "Madira 2", "A2004" and "A2002". Additional Traits: These accessions have petioles with more or less pigmentation. Their leaves are more than 2 cm longer than those of the third class and they are 36.6 cm taller than Class 3 varieties.
- Class 2 (C2) is situated in the central part of the two axes and exhibits the following characteristics: The flowering date is relatively average with 50% of plants flowering in about 51 days. The average stem height is 162.304 cm. and leaf length measures 12.28 cm. Varieties in this class are noted for their prominent veins and strong stem pigmentation. Accessions in C2 include "N'gourouma", "Akeri", "TP5-sel" and "AC-NL". Additionally these accessions have average 1000 grains weights and basal branch lengths of 13 cm. Their leaf widths are 2 cm greater than those in Class 1 and 1 cm less than those in Class 3.
- Class 3 (C3) is located in the positive part of Axis 1 and is characterized by the following traits: The accessions in this class include "Poly" and "AHTI". Notable features are the lengths of inflorescences, such as axillary inflorescence and terminal inflorescence of the stem. The leaf shape is described as good and the growth habit is strong. Varieties in this class have a high 1000 grains weight and well-represented seed hulls. They exhibit average basal and terminal branch lengths of 31.11 cm and 75 cm. respectively. Additionally the flowering date is notably shorter at 41 days for 50% flowering and the average stem height is 130.98 cm. Additional Traits: These accessions have 1.5 times more weight of 1000 grains compared to Class 1 accessions.

<span id="page-13-0"></span>

Figure 3. Dendrogram showing the relationship among the ten accessions.

#### **3.3. Relationship between Variables and Accessions**

[Figure 4](#page-13-1) highlights the relationships between the different yield component variables and the accessions. The circle of correlations between variables and the genotypes in the main plan 1 - 2.

- Close Relationships:
- o Accessions "Akeri", "TP5-sel", "AC-NL" and "N'gourouma" are closely associated with several variables: the weight of 1000 grains (P1000), stem pigmentation (PigTi). petiole pigmentation, average length of the terminal branches (LITBT), and the and the shape of the leaf (Ffe).
- <span id="page-13-1"></span>o Accessions "Poly" and "AHTI" are also closely related. They are associated with the average length of basal branches (LMBLB), growth habit (Por ti), length of the terminal inflorescence of the stem (LITT), and length of the terminal inflorescence of the branch (LITB), presence of axillary inflorescence (LIA).



Red font: character; Green font: accessions. Abbreviation: NPL: Number of plants raised; LITT: Length of the inflorescence terminal of the stem; HT: stem height; LMBLB length of basal branches; LMBLT: Length of terminal branches; LITB: Length of the terminal inflorescence of the branch; DF50%: Date 50% flowering; LIA: Length of inflorescence Axilary; LaF: sheet width; LoF: Leaf length. Ti: Petiole pigmentation, AIT: Attitude of the terminal inflorescence, FIT: Terminal Inflorescence shap, PIA: Presence of axillary inflorescence, proemi ner: prominent ribs, FBLi: Shape of the edge of the limb, Ffe: Leaf shap, FBLi: Leaf pubescence, PigFe: Leaf pigmentation, EAF: Thorn in the leaf axil, PigTi: Stem pigmentation, FBLi: Pubescence of the stem, Port Ti :Growth habit, IR: Branching index, FG: Seed Shape, TypInf: Seed Shell Type, CG: Seed color, DehGou: Pod dehiscence, IDF: Flower Density Index, TypInf: Flower Type. P1000: Weight of 1000 grains.

Figure 4. Relationship between amaranth accessions and variables.

Furthermore, the position of the accessions "Madira 1"; "Madira 2", "A2004", "A2002" on the correlation circle indicates that the height of the plants, the number of plants, the length of the leaves, the 50% flowering date, the color of the pods, the pigmentation of the stems are among the quantitative and qualitative characters those which explain the variability of the accessions the most.

# **4. Discussion**

The major objective of this study was to assess the genetic diversity within the collection to understand the range of genetic variation [\[20\].](#page-18-4) The study of [\[5\]](#page-17-3) revealed a significant level of morphological diversity between and within amaranth accessions from Malawi. Significant variation was observed for all the agro-morphological parameters among the ten amaranth accessions from Samako in Mali. This wide range of variation for qualitative and quantitative traits is consistent with the findings o[f \[5\]](#page-17-3) and [\[26\].](#page-18-10) Overall, there was a clear separation of the field grown accessions from Samanko conditions based on the dissimilarity of the qualitative characters. This diversity is due to amaranths variability in mating behavior and genetic material, adaptability to different growing conditions, and a wide range of variability in sexual systems, from monoecy to dioecy. This could be due to character expression being more vivid in the Samanko field. [\[27\]](#page-18-11) and [\[26\]](#page-18-10) reported differences between accessions with respect to characters like stem colour, leaf colour and seed colour. [\[28\]](#page-18-12) reported wide genetic diversity for qualitative traits among 18 amaranth germplasm collections. [\[29\]](#page-18-13)[-\[31\]](#page-18-14) also reported polymorphism for these traits among different amaranthus accessions. In the present study, the two qualitative traits, namely, dense inflorescence index and Leaf pigmentation, showed high variability among the species. The diversity of Eighteen Amaranth germplasm collections in the Philippines and found a wide genetic variability among them for qualitative traits reported by [\[28\].](#page-18-12)

Traits such as the number of plants, leaf width, stem height, length of the terminal inflorescence of the stem, and axillary inflorescence length. 50% flowering and weight of 1000 grains exhibited significant variation. This variation is likely due to genetic differences among the accessions evaluated since they were grown in the same environment. Similar results were reported by [\[5\].](#page-17-3) This observation contrasts wit[h \[32\],](#page-19-0) who attributed vegetative trait diversity to environmental factors. Traditional African vegetables such as Amarant sp, and spider plant, are tolerant of abiotic stresses such as low soil moisture and heat conditions. In a study conducted in South Africa, amaranth was found to be more heat tolerant [\[33\].](#page-19-1) For [\[34\],](#page-19-2) temperature also affects the production of amaranth biomass, under high temperatures, amaranth produces more biomass than in cool times, which indicates that the local environment can influence amaranth production. Considering the various environmental conditions, explains the existence of a long list of diversity of amaranth in the area. Significant variations in days to 50% flowering also point to genetic differences among the accessions. This was reported by [\[35\],](#page-19-3) who demonstrated that early flowering in amaranth is governed by a single gene, with

the dominant allele determining earliness in flowering. [\[36\]](#page-19-4) suggested that late flowering could be advantageous for vegetable amaranths, providing farmers with a longer harvesting period for the leaves.

This section will help to understand the relationships between different agromorphological traits and how they influence each other which is crucial for breeding and improving amaranth species.

Understanding the associations between agro-morphological parameters is crucial for the selection process. The present study found narrow differences in correlation coefficients indicating a strong inherent association among traits. The 1000 Grain Weight (P1000) exhibited a highly significant and positive association with the length of the terminal inflorescence of the stem suggesting the potential for simultaneous improvement of these traits. The highly significant and positive correlation between the number of days to 50% flowering and the height of the plant indicates that plants that have a long cycle are those that have a larger size. Indeed these plants, given the length of their cycle have more time for vegetative development unlike those whose cycle is short. A negative and highly significant correlation ( $r = -0.82$ ) was observed between the length of the axillary inflorescence and the 50% flowering date. This indicates that plants that have a short cycle are those that have a greater Length of the Axillary inflorescence. Indeed, these plants given the length of their Axillary inflorescence have the advantage of having a large number of flowers and a good reproductive capacity. Additionally a negative correlation was found between the weight of 1000 grains and the 50% flowering date. This indicates that plants with a long cycle have light weights of 1000 grains. In a context of low rainfall, water requirements could not be met, which has consequences on grain filling. Hence light grain weights. Similar findings were reported by [\[5\]](#page-17-3) and [\[37\],](#page-19-5) who noted negative and significant associations between grain yield and traits such as leaf length, plant height, stem girth, days to 50% flowering, and leaf yield. This section will help to understand the relationships between different agro-morphological traits and how they influence each other which is crucial for breeding and improving amaranth species. Understanding the associations between agro-morphological parameters is crucial for the selection process.

The dendrogram indicated genetic diversity within and between cluster groups. It showed variations in quantitative characters such as stem height, leaf length, axillary inflorescence length, terminal inflorescence length, growth habit, high 1000 grain weights, and seed hulls, High variability in qualitative characters, such as recovery terminal inflorescence attitude, prominent veins, and strong stem pigmentation was also evident. The grouping of varieties into three clusters based on agro-morphological characters reflects the diversity of the sample studied. This study suggests that the genetic material has significant potential for improvement. Accessions with desirable traits can be crossed with other promising accessions to produce the desired hybrids. For [\[38\],](#page-19-6) this difference between group amaranth may be due to their collection from diverse geographical zones.

Plant height, days to flowering, leaf length, and individual leaf width are all key contributors to amaranth's leaf yield, as are a variety of other yield elements. [\[39\]](#page-19-7) supported similar findings. The observed high plant height in the 80% (C1 and C2) leaf yielder accession might be due to the inherent genetic variation, strong light competition, and partition of more assimilate for stem elongation. Similar findings were observed by [\[40\]](#page-19-8) in amaranths and taller plants outcompete weeds more successfully than shorter ones [\[41\].](#page-19-9) Similarly, the leaf area is crucial in determining the yiel[d \[42\].](#page-19-10) 

The sixty percent (60%) of accessions (C2 and C3) with high 1000 grain weight and very good inflorescence had the greatest leaf width and leaf shape. These accessions probably had the best leaf area. In addition, these accessions had very high petiole and stem pigmentation. When water availability increased, plants were able to perform photosynthesis more efficiently [\[43\].](#page-19-11) The leaf area intercepts sunlight, absorbs CO2 and inorganic nitrogen, and performs photosynthesis and biomass accumulation, among other factors, determining yield, as reported by [\[44\].](#page-19-12)

# **5. Conclusion**

This study highlighted the extent of quantitative and qualitative variations among the ten amaranth accessions. The observed differences in agro-morphological parameter correlation coefficients and the identification of three distinct classes (C1: Madira 1, Madira 2, A2004, A2002; C2: TP5-sel. N'gourouma. Akeri. AC-NL; C3: AHTI, Akeri) provide ample opportunities for plant breeders to improve amaranth production. The identified genetic diversity within these accessions offers valuable insights for future breeding programs aimed at enhancing the yield and quality of amaranth.

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# **Conflicts of Interest**

The authors declare no conflicts of interest regarding the publication of this paper.

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